EXPERIMENTAL OIL SPILLS ON MACKENZIE DELTA LAKES. II. EFFECT OF TWO TYPES OF CRUDE OIL ON LAKES 4C AND 8

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### EXPERIMENTAL OIL SPILLS ON MACKENZIE

DELTA LAKES. II. EFFECT OF TWO TYPES OF CRUDE OIL ON LAKES 4C AND 8

by

## N. B. SNOW

and

#### D. M. ROSENBERG

This is the **slxty-**sixth

Technical Report from the

Research and Development Directorate

Freshwater Institute

Winnipeg, Manitoba

Ceci est le soixante-sixième Rapport Technique de la Direction de la Recherche et Développement Institut des eaux douces Winnipeg, Manitoba The data for this report were obtained as a result of investigations carried out under the Government of Canada Environmental-Social Program, Northern Pipelines, Task Force on Northern Oil Development.

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#### ABSTRACT

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Snow, N. B., and D. M. Rosenberg. 1975. Experimental Oil Spills on Mackenzie Delta Lakes. II. Effect of Two Types of Crude Oil on Lakes 4C and 8. Fish. Mar. Serv. Res. Dev. Tech. Rep. 549: 19 pp.

The initial effects of two types of crude oil on selected physical, chemical and biotic parameters in two Mackenzie Delta lakes were studied together with changes in the composition of each oil.

Two adjacent lakes (4C and 8) in the central region of the Delta were partitioned by polyethylene sheeting to form impoundments of 0.1 and 0.3 ha respectively. 60 liters of Pembina crude oil were pumped onto the surface of the smaller impoundment (Lake 4C) on 6 August 1973 and 180 liters of Norman Wells crude oil were pumped onto the surface of the larger impoundment on the following day, 7 August 1973. Selected physical and chemical water parameters, periphyton, littoral zoobenthos and invertebrates contained in the surface film were sampled in the partitioned (oil-spill) and control areas of each lake, 2 months before and after each experimental spill. Rate of evaporation and changes in composition of each oil were followed subsequent to each spill by gas chromatographic analysis.

The lighter fractions of both oils, which are the most toxic components to aquatic invertebrates, evaporated within two hours following each experimental spill. Of the two oils used, Pembina crude appeared to be more acutely toxic to invertebrates than did Norman Wells crude.

An initial period of high mortality to zoobenthic, pleustonic and aerial invertebrates was replaced after two days by a period of decreased mortality lasting for two and three weeks in Lakes 4C and 8 respectively.

Massive growths of filamentous blue-green algae occurred on samplers placed in the oil-spill sections. Seston weight, particulate carbon and particulate nitrogen increased significantly in the oil spill area of each lake above control concentrations. Total dissolved nitrogen (TDN) also increased significantly (4 mMoles/m<sup>3</sup>) above control concentrations in response to each oil. These increases were not accounted for by a theoretical consideration of solution of the nitrogen contained in each oil either as a result of chemical breakdown or biodegradation. It would seem that oil stimulated nitrogen-fixing organisms were implicated in producing these changes in water chemistry, and this possibility is discussed.

It is hypothesized that the effects of crude oil on such lakes occur in three phases. The first is a period of acute toxicity which is of short duration (~ 2 days) and overlaps with a physically deleterious phase extending to several weeks. These effects may then be replaced by a chronic eutrophication, i.e. acceleration in rate of nutrient supply. The relative lengths and severity of each phase will be dependent upon such factors as volume of oil, type of oil and climate.

## RÉSUMÉ

Snow, N. B., and D. M. Rosenberg. 1975. Experimental Oil Spills on Mackenzie Delta Lakes. II. Effect of Two Types of Crude Oil on Lakes 4C and 8. Fish. Mar. Serv. Res. Dev. Tech. Rep. 549: 19 pp.

On a étudié les premiers effets de deux types de pétrole brut sur des paramètres physiques, chimiques et biologiques qui l'on a choisis dans deux lacs du delta du Mackenzie, de même que les changements survenus dans la composition de chaque genre de pétrole.

Par un barrage de polyéthylène, on a séparé deux lacs adjacents (4C et 8) dans la région centrale du delta afin de former des bassins de 0.1 et 0.3 ha, respectivement. Le 6 aout 1973, on a déversé 60 litres de pétrole brut Pembina à la surface du plus petit bassin et le 7 août 1973, on a déversé 180 litres de pétrole brut Norman Wells à la surface du bassin le plus grand. Deux mois avant et après chaque déversement expérimental, on a prélevé, dans les zones divisées (déversement de pétrole) et contrôlées de chaque lac, des échantillons de périphyton, de zoobenthos du littoral et d'invertébrés se trouvant sur la couche de surface et ce, dans les paramètres aquatiques d'un milieu physique et chimique choisi. Après chaque déversement, on a effectué une analyse chromatographique au gaz pour déterminer le taux d'évaporation et les changements survenus dans la composition de chaque genre de pétrole.

Les particules les plus légères des deux sortes de pétrole qui constituent les élèments les plus toxiques pour les invertébrés aquatiques, se sont évaporées dans les deux heures qui ont suivi chaque déversement expérimental. Des deux types de pétrole utilisés, le Pembina brut semble être beaucoup plus toxique pour les invertébrés que le Norman Wells brut.

A une période initiale de deux jours pendant laquelle le taux de mortalité des invertébrés zoobenthiques, pleustoniques et aériens s'est avéré très élevé, a succédé une période de deux à trois semaines pendant laquelle on a noté un taux de mortalité décroissant dans les lacs 4C et 8 respectivement.

Parmi les échantillons disposés dans les sections atteintes par le déversement de pétrole, on a remarqué la croissance massive d'algues filamenteuses bleu-vert. Le poids du seston, le carbone particulaire ainsi que l'azote particulaire ont augmenté de façon significative dans les zones de déversement de pétrole de chaque lac et ce, dans des concentrations telles qu'il était impossible de les contrôler. En réaction à chaque type de pétrole, l'azote dissous totalement (ADT) a également augmenté de façon significative (4 m Moles/m<sup>3</sup>) et ce, dans des concentrations qui ont rendu le contrôle impossible. À cause de la décomposition chimique ou de la biodégradation, il a été impossible d'enregistrer ces accroissements en faisant une étude théorique de la solution d'azote contenue dans chaque sorte de pétrole. Il semblerait que les organismes qui retiennent normalement l'azote et qui ont été stimulés par le pétrole sont en partie responsables des changements qui se sont produits dans la chimie de l'eau et l'on étude cette possibilité.

On émet l'hypothèse que les effets du pétrole brut sur ces lacs se sont produits en trois phases. La première est marquée par un haut degré de toxicité d'une courte durée (~2 jours) et chevauche une phase nuisible

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physiquement qui s'étend sur plusieurs semaines. Ces effets peuvent ensuite être remplacés par une eutrophication chronique, c.-à.-d. par l'augmentation rapide du taux d'approvisionnement nutritif. La durée et l'intensité relatives de chaque phase dépendront de facteurs tels que le volume de pétrole, le genre de pétrole et le climat.

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INTRODUCTION \*

The problem of oil pollution has been the subject of several reviews (e.g. Atlas and Bartha, 1973). Most of the current research effort is directed towards this problem in the marine environment.

In a previous study (Snow and Rosenberg, 1975), we investigated the effects of a northern crude oil on a small, whole lake ecosystem. A major problem encountered in this and any other whole ecosystem manipulation study was a suitable control. Even in the Mackenzie Delta, which has approximately half a million lakes, it was not possible to find two lakes at the same latitude which were sufficiently similar in terms of fauna and flora so that one could be a control system for an experimental study of the other. This necessitated relying upon one year's pre-spill data for the Lake 4 experiment, with which to compare post-spill effects. This was unsatisfactory as year-to-year variations in biomass and species life-histories are not known.

Drawing upon the experience of this first experiment, it was decided to carry out two more spills on adjacent lakes which would be partitioned to obtain experimental and control areas within the same lake. It was also decided to study the effects of two types of crude oil and concentrate upon the littoral benthos as we found that this biotic component was immediately affected by oil. We also expanded some chemical measurements and included changes in the composition of the spilled oil with time.

Both of the lakes chosen for the experimental spills differ from Lake 4 with respect to their flora and fauna (and also from each other) but in terms of physical and chemical regimes they are all similar (see Brunskill et al., 1973 and Campbell et al., 1975). They have similar maximum depths of 2-3 m, and small temporary inflows. Lake 8 has been connected to a larger adjacent lake since 1970.

### METHODS AND MATERIALS

The two lakes chosen for the 1973 oil spill experiments (Lake 4C and Lake 8) are adjacent to Lake 4 (see Map, Fig. 1). Both the former lakes had areas partitioned off by 6 mil. clear polyethylene sheeting. To ensure as complete a seal as possible, the sheeting was weighted along its bottom edge and buried in the sediment to a depth of approximately half a meter. The partition was supported by wooden posts and extended half a meter above the lake surface. Sampling stations were set up to the littoral zone on either side of this structure.

At 14:48 MST on August 6, 1973, 60 liters of Pembina crude oil was poured onto the experimental area of Lake 4C at its center. The partitioned area of this lake is approximately 0.1 ha.

<sup>\*</sup> The fifth in a series of 13 technical reports on ecological studies of aquatic systems in the Mackenzie and Porcupine drainages in relation to proposed pipeline and highway developments.

The Lake 8 spill was carried out at 15:00 MST on the following day. At this time,  $180 \ \ell$  of Norman Wells crude oil were pumped onto the surface of the experimental area of this lake, at its centre. The partitioned area of this lake is approximately 0.3 ha. (i.e. three times that of the Lake 40 experimental area).

Zoobenthos and bottom sediment samples from these experimental lakes were obtained using the modified Ekman-type grab and procedures described in Brunskill <u>et al.</u> (1973). These samples were taken prior to and immediately after each spill, then routinely at monthly intervals until freeze up. Single sampling stations were located either side of the polyethylene partition in the littoral zone of each lake.

Organisms at or near the water surface before the spills and in the oil layers following the spills, were sampled using a handskimmer of our own design. This device is a scoop which takes a 0.2 m wide sample and is fitted with a 400 µm mesh screen at its posterior end.

The two types of oil used were Norman Wells crude ( a northern crude oil) and Pembina erude (a blend of Albertan crude oils provided by Imperial Oil Ltd. as the closest approximation to Prudhoe Bay crude oil available at that time in the absence of the latter oil itself).

Both types of crude oil were sampled for chromatographic analysis prior to the spills, at two hourly intervals immediately following the spills and less frequently thereafter for a period of three weeks.

The oil samples were taken in 125 ml pyrex glass bottles with ground glass stoppers. These bottles were purged with nitrogen on site, prior to the samples being collected by hand, then stored in darkness at low temperature  $(-1^{\circ}C)$  before their shipment to Ottawa for analysis.

Oil samples were analyzed using the gas chromatographic procedure described by Scott (1974). This method employs SCOT columns and F.T. detectors. Oil losses by evaporation subsequent to the spills were determined by analysis of the fractions of samples collected from the lakes using a modified Hempel distillation (Scott and Chatterjee, 1974).

Water samples were taken using a Van Dorn sampler and all samples were preliminarily processed as soon as possible after their collection (usually within 24 hrs.). Dissolved oxygen samples were titrated by the Winkler method within an hour of collection. Most other samples were treated to allow their storage, and subsequent shipment for final processing in Yellowknife or Winnipeg. The physical and chemical parameters monitored included temperature, conductivity, pH, dissolved oxygen, major ions, seston and nutrients. For a detailed consideration of the sampling and analytical procedures see Brunskill et al. (1973).

Periphyton samplers were suspended one meter below the water surface in the experimental and control side of each lake. For a detailed description of these samplers and their operation, see Roeder, et al. (1975).

### RESULTS

The Pembina crude oil on the experimental section of Lake 4C was observed from the air at the time of the spill and was seen to spread evenly and slowly with an irregular leading edge and no evidence of a slick of faster moving components preceding the bulk oil as was apparent in the Lake 8 spill. At ground level, the separation of slick and bulk components did become apparent as a surface reticulation.

Two-thirds of the shoreline of the experimental area had been blanketed by the oil within the space of one hour and the whole margin of this area was similarly affected within two hours of the spill.

Within twenty-four hours, the thin, even oil film had piled up to form a thick layer amongst the emergent macrophytic vegetation (predominately <u>Menyanthes</u> sp.). Six weeks after the spill, the only evidence of the oil was a 'tide-line' along the polyethylene sheeting, and some pockets amongst decomposing marginal vegetation at water level.

The Norman Wells crude oil spilt onto Lake 8 spread downwind from the prevailing NW wind very rapidly and had reached the shore of the experimental area seven minutes later. Within thirty minutes, half of the shoreline of this area was affected. The shoreline of Lake 8 is quite different from that of either Lake 4 or Lake 4C. It has no emergent macrophytic vegetation. This is probably because of the greater magnitude of fluctuating water levels resulting from the connection of this lake with a much larger one (see Fig. 1). The spreading of this oil was similar to that observed on Lake 4 in the preceding year, but quite different from that of the Lake 4C Pembina crude. From the moment of contact with the water, a rapidly moving slick preceded the brown sludge of heavier components in their advancement towards the shore. A distinct sheen, distinguishable from the bulk oil was still discernible days later. Also, by this time, the bulk oil had affected the whole experimental area margin and was evident against the polyethylenc partition. Within the next two months, following a one meter drop in lake level, the bulk oil was largely stranded along the mud and moss of the shoreline.

Chromatograms of original Pembina crude oil and the oil two hours after the spill on Lake 4C are shown in Fig. 2 (A & B respectively). It is readily apparent from these two chromatograms that the lighter fractions of the oil (including aromatics and n-alkanes less than  $C_0$ ) have virtually disappeared in the two hours following the spill. The higher n-alkanes are identifiable in both chromatograms and are present in approximately the same quantities in each sample. The difference in peak heights between A and B is the result of different sample size. Chromatograms of original Norman Wells crude oil and that two hours after the spill on Lake 8 have a similar appearance to those of Pembina crude oil counterparts. Two hours following the experimental spill on Lake 4C, 21% by volume of the Pembina oil had evaporated and this increased to 26% after 4 hours. Evaporative losses after 16 hours were 30% and by 36 hours 39% of the oil had evaporated. Within 48 hours of the spill 45% of the oil had been lost. Very little evaporation appeared to have occurred after this time. The initial evaporation of the Norman Wells crude oil was greater than that of the Pembina crude used 24 hours earlier. Thirty minutes after the spill 27% by volume of the bulk

oil had evaporated. After 4 hours 32% of the oil had evaporated and this increased to 37% within 24 hours. Samples collected 48 hours after the spill showed evaporative losses of approximately 49% by volume and after 62 hours 52% of the oil was calculated to have been lost in this way. The oil continued to evaporate at a slower rate and there was no evidence for dodecane in the oil samples after 120 hours. All samples collected after this time contained some tridecane. Not all losses are evaporative as a finite amount of oil components will go into solution, but this is considered to be a minuscule fraction compared to the total volume of oil spilt.

The range of physical and chemical parameters measured during the open water season 19 3 for Lakes 4C and 8 are shown in Table I. The parameters presented in this table are those which showed no significant difference between the experimental (= oil spill area) stations, (L4C-1 & L8-1) and the control stations (L4C-2 & L8-2) following the spills on each lake.

These data indicate that the physical and chemical characteristics of each lake on both sides of the partitions underwent normal open water cycles.

Four of the parameters measured did show significant differences between the oil spill area of each lake and their controls. These parameters were: total dissolved nitrogen, particulate nitrogen, particulate carbon and seston weight. The results are presented graphically for the experimental and control stations of both lakes in Fig. 3 (Lake 4C) and Fig. 4 (Lake 8).

Seston values for the oil spill area of both lakes were higher than those of the control areas (Figs. 3 & 4). These differences are also seen in the particulate carbon and nitrogen values but particulate phosphorus levels at control and experimental stations in both lakes were not significantly different at any time following the spills.

Seston and particulate carbon and nitrogen reached their peaks in the oil spill area of Lake 4C three weeks after the spill. Secondary peaks occurred between five and six weeks after the spill. There was only a single corresponding peak in the oil spill area of Lake 8 and this also occurred three weeks after the experimental spill on that lake.

The variations between total dissolved nitrogen levels of control and experimental areas of Lake 4C were not as consistent as for Lake 8 values but at several sampling times showed a significant increase in the oil-affected area (Figs. 3 & 4).

Immediately following the experimental spills, and to date, there appears to have been no significant differences between littoral zoobenthos standing crops in the oil spill and control areas of either Lake 4C or 8. These data are currently being analysed and will be included in a subsequent report.

The zoobenthic fauna of each lake differed with respect to numbers of taxa represented but zoobenthos standing crops were similar at each sampling. These standing crops are in the range 1000-13,000 organisms/m<sup>2</sup> in both lakes. Chironomid larvae dominated the zoobenthos of Lake

4C whereas oligochaetes, gastropods and pelecypods were the most numerous zoobenthic organisms in Lake 8.

Data for organisms collected by the surface-skimmer in the oil slick of each lake are presented in Table II (Lake 4C) and Table III (Lake 8). Organisms collected by this device represent components of the littoral zoobenthos as well as aerial insects of terrestrial origin. Dipteran adults and larvae predominated in most of these samples, particularly those from Lake 4C. Surface skim samples taken from the control areas at the same sampling intervals are not included in Tables II and III because they consisted of single larval or adult insects at most, or more usually, no organisms at all.

The differences in composition between surface skim samples from the oil slick of both experimental spills appear to be largely attributable to the different zoobenthic fauna of each lake. It can be seen (Tables II & III) that corixids (<u>Callicorixa</u> sp.) were as numerous as dipteran adults and larvae (primarily Chironomidae) in Lake 8. Adult beetles also comprised a larger proportion of the samples from this lake than those from Lake 4C.

Whilst the oil was spreading on Lake 4C, and before it had affected the whole lake margin, large numbers of <u>Gerris</u> sp. were driven into embayments amongst emergent vegetation by the slowly advancing oil. These animals, adult beetles and a number of damselfly adults were observed to expire very quickly (within 1 - 2 hours) upon contact with Pembina crude oil. These organisms survived total immersion in Norman Wells crude oil for several hours during the Lake 4 experimental spill of 1972.

The entrapment of organisms by the oil slicks had a longer duration for the Lake 8 experimental spill (3 weeks) than for the Lake 4C spill (3 days). After these times, the skim samples contained no organisms at all or single sporadic emerging insects.

The periphyton samplers suspended in the oil spill areas of both lakes were covered by massive growths of algae, which were not observed on the samplers from the control areas. This growth was primarily filamentous <u>Oscillatoria</u> sp., other blue-green algae, and an admixture of diatom and green algal species.

### DISCUSSION

The direct harmful effects of crude oil to aquatic organisms can be manifested in two ways. Crude oil can poison such organisms (chemical toxicity) or may present a mechanical impediment to them by clogging vital processes such as respiration, feeding, etc. (physical effects). There may be indirect effects which will be discussed later.

In both of the experimental oil spills carried out on sections of Lakes 4C and 8, using two types of oil, the lighter fractions of each oil were

found to have virtually disappeared within two hours following the spills. It is in these fractions that most of the toxicity resides (Nelson-Smith, 1968; Ottway, 1971). The toxicity of each oil is therefore, likely to be of an acute nature. The toxic effects of oils on aquatic animals have been reviewed by Nelson-Smith (1970) and on plants by Baker (1971a). It is generally concluded that the majority of chemical toxicity resides in the lighter, low-boiling point fractions, i.e. the aromatic hydrocarbons (benzene, toluene, xylene, etc. and their derivatives) and phenolic compounds (naphthenic acids etc.). Mackay and Wolkoff (1973) have shown that low-solubility hydrocarbons and low vapour pressure compounds have "half lives" in solution of minutes or hours under laboratory or environmental conditions.

Considerable numbers of organisms were collected from the spreading oil slick during both experiments (see Tables II and III). It was noted that those taken from the Pembina crude oil slick in the collections up to 24 hrs following the spill were usually dead upon collection. Organisms taken from the Norman Wells crude oil slick over the same time span were usually moribund but alive. It seems certain however, that these animals would not survive their coating of oil. Pembina crude oil appeared to be more toxic to invertebrates than Norman Wells crude oil.

Under the experimental conditions, both types of crude oil used, exhibited an initial rapid evaporation which thereafter decreased steadily. This was the result of the fact that a large proportion of any crude oil is composed of components having low vapour pressures.

The high numbers of organisms taken in oil slick samples from both lakes during the first two days (Tables II and III) appear to coincide with the chemically toxic stage of oil-pollution. The lower numbers of organisms in oil slick samples from both lakes subsequent to the period of 2 days following the spill, and after the evaporation of the lighter oil fractions indicates a physically detrimental effect of the residual oil. Only the lighter fractions of crude oil have been found to be toxic to intertidal animals (Crapp, 1971).

There is almost certainly an overlap of each of these phases (chemical toxicity and physical effects) the duration of which probably depends upon factors such as initial composition of the oil and climatic conditions.

The fact that the duration of the "physical" phase varied for each oil used in our experiment (Tables II and III), being considerably longer for the Lake 8 experiment than for that on Lake 4C, probably results from the larger amount of oil spilt in the former experiment coupled with the lack of marginal vegetation which appeared to retain the oil spilt on Lakes 4 and 4C.

The different composition of aquatic organisms collected from the oil slick on each lake (Tables II and III) resulted from differences in each lake's fauna and not from selective toxicity of the different oils used in each experiment. The higher proportion of aerial skimming insects (adult odonates and flies) present in the Lake 4C oil slick samples result from the protected aspect of that lake coupled with the higher

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viscosity of Pembina crude oil which trapped such insects readily and firmly. The large amounts of marginal vegetation around Lake 4C harbours large numbers of water striders (gerrids) which occurred in significant numbers in the oil slick from that lake. These animals, and the emergent vegetation are absent from Lake 8 and hence from its oil slick samples. Lake 8 does, however, support a large population of another hemipteran, (Callicorixa sp.) and these organisms comprised significant numbers of the oil slick samples from that lake.

Aquatic organisms which make use of the water's surface film were the groups which appeared to be the most adversely affected by the presence of oil in each 'ake. These organisms were the gerrids, the corixids (both Hemiptera) and adult beetles of the genus <u>Haliplus</u> (Coleoptera). A mineral oil ("Malarial B") has been shown to kill other Hemiptera (<u>Anisops</u> sp., a notonectid) in concentrations which did not adversely affect any other insects (Desai and Rao, 1971).

The degree of toxicity and duration of the period of physical effects will depend upon the nature and composition of the crude oil. Within a given time, the rate of evaporation of Norman Wells crude oil on bake 8 was more rapid than that of the Pembina crude oil on Lake 4C. There was also a greater proportion of residue following evaporative losses from the Pembina crude than from the Norman Wells oil. When considering the effects of oilspills and estimating possible environmental repercussions, considerations must be given to the oil properties themselves.

Concomitant with the toxic and entrapment effect of the oils on each lake, changes were also occurring in the water chemistry, phytoplankton and periphyton as the result of the presence of crude oil. The increases in seston, particulate nitrogen (PN), and particulate carbon (PC) evident in the oil spill section of each lake during the third week following the spills appeared to be the result of microbial and phytoplankton blooms. At this time, no such bloom was apparent in the control section of each lake. An increase in seston would produce an increase in particulate elements which accounts for the observed increases in PN and PC but why there was no coincident increase in particulate phosphorus is not clear.

The bloom itself was relatively short-lived (2 weeks) and was not visually obvious at this time. The massive amounts of algae (primarily filamentous blue-greens) present on artificial substrates suspended in the oil spill section of each lake two months after the spills (see Roeder <u>et al.</u>, 1975) were not quantified but enormous. No such growth was evident on control substrates. It is important to note that these samplers were at no time actually in contact with the oil in either lake, therefore whatever the nature of the growth stimulant, it was present in the water itself. These data indicate that the presence of crude oil in a lake has a potentially eutrophying effect.

The only dissolved nutrient measured which showed any significant change in the water of the oil spill section of each lake when compared to levels in the control areas following the spills was total dissolved nitrogen (TDN). The fact that total dissolved phosphorus (TDP) showed no such increase at the same time does not necessarily rule out the possibility that more of this nutrient was also present in the oil spill areas as such an active element has a very high rate of recycling. Schindler <u>et al.</u> (1971) pointed out that the absolute amount of dissolved phosphorus measured at any instant in time in lake water is not a meaningful parameter in terms of nutrient dynamics.

An increase in TDN in the oil spill sections of the lakes could occur through two main processes. Firstly, by the dissolving of nitrogen contained in the oil itself, either by physical solution or via the intermediary of micro-organisms, phytoplankton or fungi. Secondly it may have resulted from increased nitrogen fixation by indigenous oil stimulated lake biota.

Assuming a specific gravity of 0.9 g/cc for the Norman Wells crude oil and an average nitrogen concentration of 0.35% by weight (data provided by Imperial Oil Ltd.), it was calculated that if all of the nitrogen in 180  $\ell$  of oil went into solution in the oil spill section of Lake 8 (volume  $\approx$  3750 m<sup>3</sup>) it would result in a TDN level of 150 µg/ $\ell$ . The observed mean increase in TDN in the oil spill section of Lake 8 following the spill, was 64 µg/ $\ell$  (4 mMoles/m<sup>3</sup>).

A similar calculation for the 60  $\ell$  of Pembina crude oil (nitrogen concentration 0.1% by weight) spilt onto the oil-spill section of Lake 4C (volume 1350 m<sup>3</sup>) gives a value of 40 µgN/ $\ell$  compared to the same observed mean increase of 64 µgN/ $\ell$ .

In the latter case, solution of all the oil-nitrogen does not account for the observed increase in TDN. In the case of the Lake 8 experiment, more than twice the observed increase in TDN could be accounted for by oil-nitrogen if all of this went into solution. If, however, the increase in PN following the spills, is added to the observed TDN increase, the gap diminishes. Furthermore, the assumption that all oil-nitrogen could or would go into solution under spill conditions is not a realistic one as nearly all oil-nitrogen is bound up in the heavier oil fractions (Ball, 1962). These fractions constitute the bulk of the residuum following evaporative losses and it is difficult to conceive of nitrogen from complex organic molecules such as pyrrholes, pyrimidoles, etc. being released without a complete breakdown on the residuum. Considerable amounts of residual oil remained around the margin of each oil spill section two months after the spills.

A laboratory experiment was carried out in which a known amount of Norman Wells crude oil was added to a known volume of 1) autoclaved, filtered lake water, 2) raw lake water and 3) distilled water in separate vessels. TDN levels in the water of each vessel did not increase during a sixweek period.

From a consideration of all these data, it would seem that solution of oil nitrogen alone would not account for the observed increase in TDN.

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It is well known that many heterocystous blue-green algae can fix nitrogen. A massive increase of blue-green algae was noticed on the samplets suspended in the oil spill section of each lake. It would be tempting to assume a causal relationship between this growth and the increase in TDN. The blue-green algae on our samplers, however, consisted primarily of <u>Oscillatoria</u> sp., a non-heterocystous alga. Some nonheterocystous blue-green algae are known to fix nitrogen, this is usually an anaerobic or microaerobic process. <u>Gloeocapsa</u>, a non-heterocystous unicellular blue-green alga, has been shown to fix nitrogen aerobically (Wyatt and Silvey, 1969) and the non-heterocystous filamentous bluegreen alga <u>Plectonema boryanum</u> Strain 594 also exhibits nitrogenase activity although this is microaerobic (Stewart and Lex, 1970). <u>Oscillatoria</u> itself is known to fix nitrogen, but only microaerobically (Stewart, 1973).

Levels of dissolved oxygen following the experimental spills in the oil spill section of each lake were never particularly low (10-13 mg/ $\ell$ ) and therefore, it seems unlikely that the increase in blue-green algae could produce the observed increase in TDN.

Therefore, it would seem that nitrogen fixing microbial organisms may have been responsible for the observed increase in TDN following the oil spills. Baker (1971b) reported growth stimulation of salt marsh grasses following oil pollution and suggested that nitrogen fixing methaneoxidising bacteria may be involved in this process.

Nitrogen fixation (as measured by acetylene reduction) by the sediment microflora of Lakes 4C, 8, two other Mackenzie Delta lakes and Shell Lake has been demonstrated by Knowles (unpublished data). Ethylene production was 64-371 n moles/g wet sediment/day and there was virtually no production in the absence of a carbon source (glucose).

It is hypothesized that crude oil spilt onto a Mackenzie Delta lake stimulated the growth of indigenous microorganisms capable of nitrogen fixation. The increase in this activity produced increases in TDN which accumulated in the water column, but the reason why no significant increases in either TDP or PP were not detected remains a problem for future study.

#### CONCLUSIONS

The lighter fractions of both Pembina and Norman Wells crude oil evaporated in the period of two hours following each experimental oil spill on Lakes 4C and 8 respectively. These fractions are the most toxic to invertebrates. Of the two oils used, Pembina crude appeared to be more acutely toxic to invertebrates than did Norman Wells crude.

The initial period of high mortality for zoobenthic, pleustonic and aerial invertebrates caused by each type of crude oil lasted for approximately two days. A decreased mortality occurred after this time which lasted for two weeks and three weeks in the case of the spills of Pembina crude and Norman Wells crude respectively. It is hypothesized that the initial two day mortality period corresponds to an acute chemical toxicity phase overlapping with a physically deleterious phase. This latter phase then accounted for the subsequent mortality. The differences in duration of the second phase were probably the result of the differences in the shoreline development of each lake.

Periphytic blue-green algae covered samplers suspended under the oilslick in each experiment during the two months following each spill. Such growth did not occur on samplers suspended from the lake surface in the control section of each lake. Significant increases in TDN occurred in the experimental section of each lake following the experimental oil spills. These increases could not be accounted for by a theoretical consideration of solution of the nitrogen contained in each oil either as a result of chemical breakdown or biodegradation. It was therefore, assumed that nitrogen-fixing microorganisms had been stimulated by the addition of oil. Such microorganisms are known to exist in Lakes 4C and 8. The periphytic blue-green algae which increased in abundance in the experimental section of each lake were non-heterocystous and were not considered to be nitrogen fixing types.

It is hypothesized that the addition of crude oil to sections of these lakes stimulated the growth of indigenous nitrogen-fixing microorganisms. The increase in this activity produced increases in TDN and TDP. Phosphorus was rapidly taken up by, amongst other members of lake biota, non-heterocystous blue-green algae, causing them to bloom periphytically, whilst TDN accumulated in the water column.

It may therefore, be possible to identify a third phase in the effect of crude oil on a lacustrine ecosystem: a chronic eutrophying effect.

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PARAMETER		LAKES			
		4C	8		
l'emperature	(°C)	1.0 - 22.0	1.0 - 11.3		
Conductivity	(µmho/cm at 25 <sup>0</sup> C)	151 - 240	150 - 305		
рH		7.2 - 8.0	7.25 - 8.75		
Dissolved $0_2$	(mg/1)	6.75 - 12.56	5.6 - 13.6		
Alkalinity	(Moles $HCO_3/m^3$ )	1.15 - 2.58	1.02 - 2.88		
TDF	$(mMoles/m^3)$	<0.05 - 0.83	<0.05 - 0.80		
Si	(mMoles/m <sup>3</sup> )	7.6 - 36.7	13.6 - 66.0		
so <sub>4</sub>	(mMoles/m <sup>3</sup> )	106 - 224	212 - 362		
C1	$(mMoles/m^3)$	68 - 359	130 - 418		
РР	$(mMoles/m^3)$	0.12 - 0.77	0.19 - 0.73		

Table 1. \* Range of physical and chemical parameters for Lakes 4C and 8 (May 27, 1973 - October 2, 1973).

\* For more complete data, see Brunskill <u>et al</u>. (1973, Vol. II Appendix IX).

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Taxon	Time After Spill (days					
	1/2	1	2	4	15	
Zygoptera adults ( <u>Enallagma boreale</u> )	15	_	_	-	-	
Hemiptera ( <u>Gerri</u> s <u>comatus</u> )	30	. —	-	_	-	
Hemiptera ( <u>Sigara</u> <u>fallenoidea</u> )	-	5	-	-	~	
Coleoptera (Adults)	40	20	-	-	-	
Trichoptera (Larvac)	-	5	-	5	_	
Tip <b>u</b> lidae (Adults)	-	5	-	-	-	
Chironomidae (Adults)	-	20	5	-	_	
Chironomidae (Larvae)	10	29	25	-	5	
Misc. Diptera (Adults)	40	50	10	-	-	
Culicidae adults ( <u>Aedes</u> sp.)	-	40	-	5	5	
Oligochaeta	-	-	5	-	-	
Gastropoda		_	20	-	~	
Pelecypoda	_ •	10	10	-	-	

Table II. Numbers of organisms per square metre trapped in the surface film following the oil spill on Lake 40 on 6 August, 1973. (- indicates no organisms in sample).

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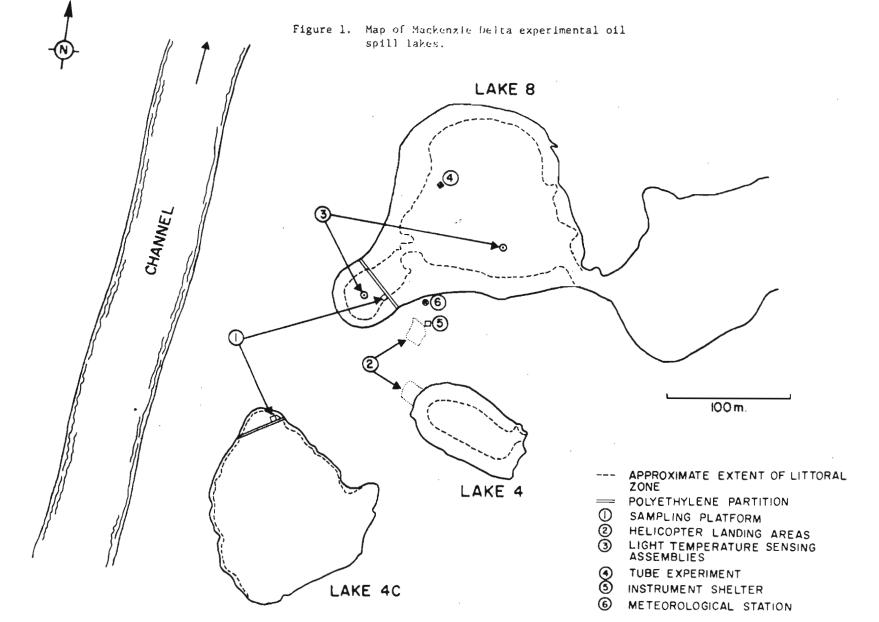
Taxon	Time After Spill								
	1/2 day	l day	2 days	4 d <b>a</b> ys	10 days	2 weks	3 wks	6 wks	7 wks
Zygoptera (nymphs)	5	-	-	-	-	-	-	-	-
Corixidae	40	155	25	20	-	-	5	-	-
Ephemeroptera (nymphs)	—	-	-		-	-	-	-	-
Coleoptera (adults)	-	75	5	5	-	-	15	-	5
Trichoptera (larvae)	-	_	10	10	5	-	-	-	-
Ch <b>iro</b> nomidae (adults)	5	25	-	-	-	-	-	-	-
Chironomidae (larvae)	-		10	10	75	15	5	-	-
fisc. Diptera (adults)	-	-	-	-	-	-	20	-	-
Amphipoda ( <u>Gammarus</u> <u>lacustris</u> )	-	5	-	-	<u> </u>	-	-	_	
Gast <b>ro</b> poda	5	45	-	-	-	-	-	5	-
Pelecypoda	-	5	_	-	-	-	-	-	-

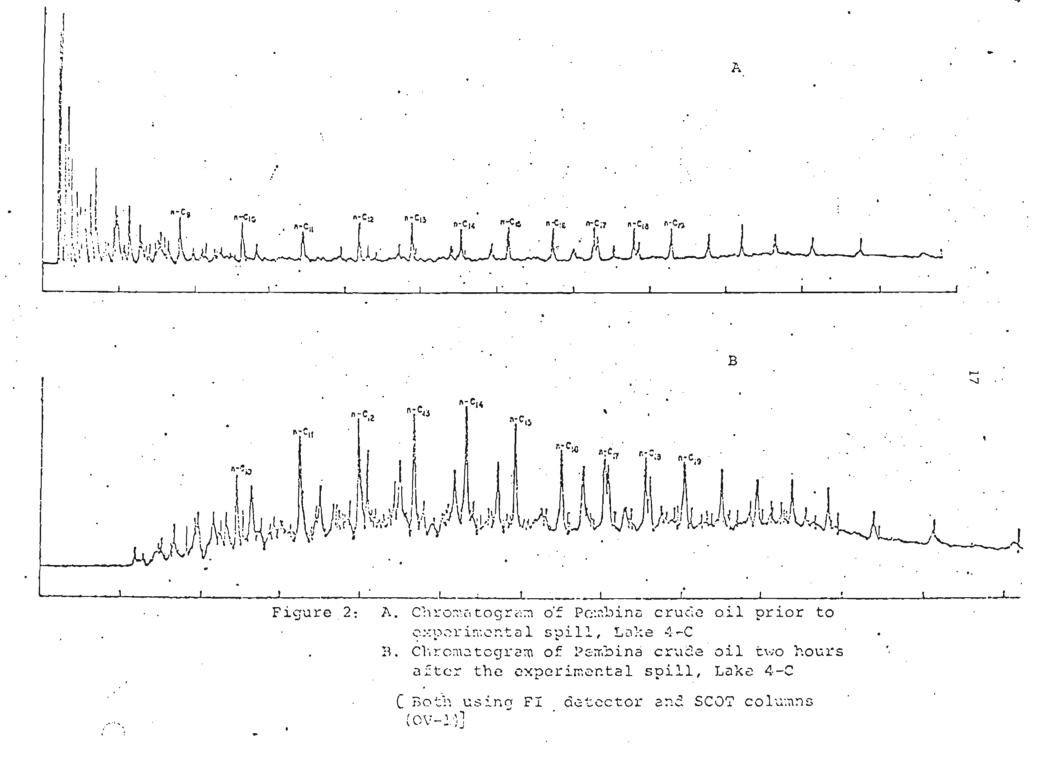
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Table III. Numbers of organisms per square metre trapped in the surface film following the oil spill on Lake 8 on 7 August, 1973. (- indicates no organisms in sample).

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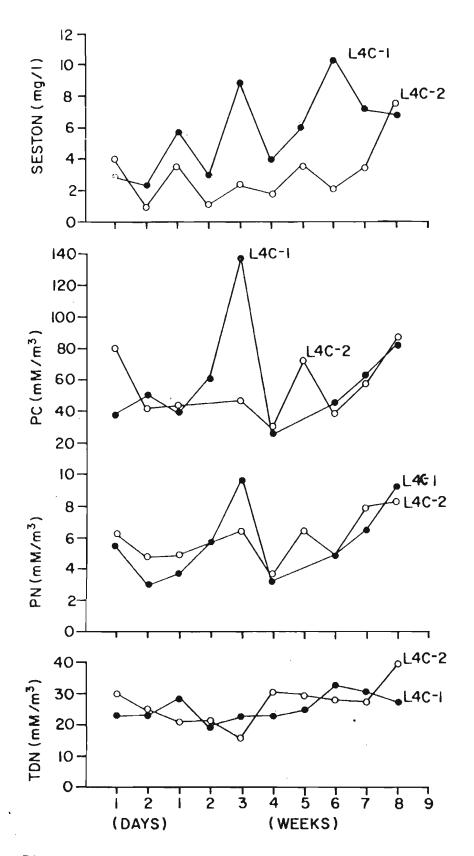


Figure 3: Total dissolved nitrogen (TDN), particulate nitrogen (PN), particulate carbon (PC), and seston weight values for Lake 4C oil-spill station (L4C-1) and control station (L4C-2) following the experimental oil spill on 6 August, 1973.

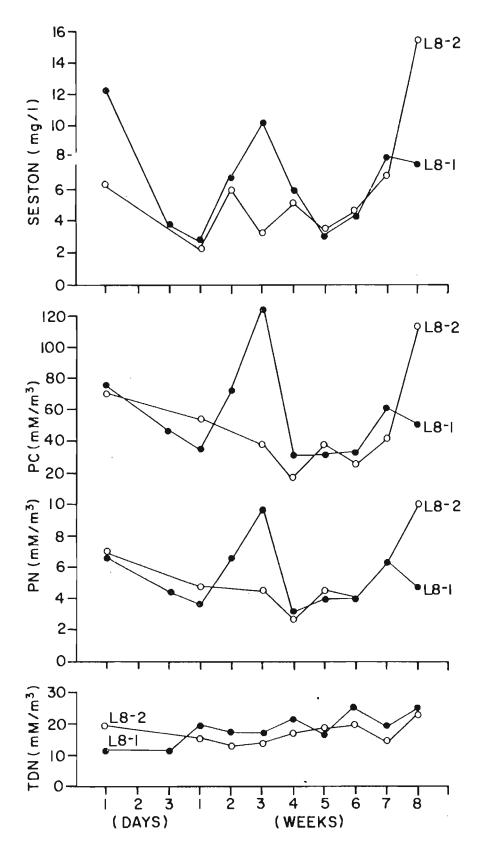


Figure 4: Total dissolved nitrogen (TDN), particulate nitrogren (PN), particulate carbon (PC), and seston weight values for Lake 8 oil-spill station (L8-1) and control station (L8-2) following the experimental oil spill on 7 August, 1973.