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Ectomycorrhizal fungi of *Salix Rotundifolia* Trautv. II

Impact of Surface Applied Prudhoe Bay Crude Oil on Mycorrhizal Root Respiration and Cold Acclimation

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ABSTRACT. Ectomycorrhizal root tips of *Salix rotundifolia* Trautv. removed from Barrow. Alaska tundra treated with 5 or 12 l/m² Prudhoe Bay crude oil on 1 July 1975 showed decreased respiration rates within 48 hr after surface application of oil. Oil treated roots continued to have depressed respiration rates throughout the summer. The following summer, respiration rates of the 5 l/m² oil treated roots were higher than controls. With respiration of the 12 l/m² treated roots only 20% below controls. However, during the summer, respiration rates declined very rapidly, probably due to water stress caused by drought conditions. The third summer, respiration rates of all root samples were quite similar, with all rates low, probably due to continued water stress.

Viable root biomass declined from year to year in the oiled soils. Analysis of cold acclimation by Arrhenius plots of respiration rates shows losses in cold acclimation after oil treatment. Ectomycorrhizal roots of *S. rotundifolia* from the oil impregnated soils of a natural oil seep at Cape Simpson, Alaska showed a minimum loss in respiration rates and cold acclimation after exposure to fresh crude oil.

RÉSUMÉ. Des bouts de racines parasitées par les champignons de Salix Rotundifolia, provenant de la toundra Alaskienne de Barrow, ont été traitées sur la base de 5 ou 12 litres par mètre carré, de brut de Prudhoe Bay. On observait une décroissance dans le rendement respiratoire pendant 48 heures après application en surface du pétrole. Les racines traitées au pétrole continuaient d'avoir une respiration réduite pendant tout l'eété. L'été suivant, les rendements respiratoires des racines traitées au pétrole, à 5 litres par mètre carré, la respiration des racines n'était que 20% sous la normale. Cependant, au cours de l'été, les rendements respiratoires déclinaient très rapidement, probablement à cause d'un manque d'eau excessif, causé par des conditions de sécheresse. Le troisième été, les rendements repiratoires de tous les échantillons de racines étaient tout à fait semblables, à un niveau bas, dû au manque d'eau continuel.

Le biotope des racines, en bon état, se réduisait d'année en année dans les sols traités au pétrole. Après traitement au pétrole, les indices de résistance au froid, dans les rendements respiratoires, suivant la méthode des reports arrhenius, montrent des pertes. A Cap Simpson, des racines parasites de Salix Rotundifolia, venant des sols imprégnés de pétrole, à partier d'un suintement naturel, montraient une perte minimale dans le rendement respiratoire et dans la résistance au froid, après exposition dans due pétrole pur.

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INTRODUCTION

A common yet poorly understood plant adaptation to the cold, low energy, and nutrient conditions of the tundra is that of the mycorrhizal symbiotic complexes between plant root systems and soil fungi. Katenin (1964)

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investigated 188 tundra plant species and found 57% mycorrhizal. Bliss (1973) states that the majority of the plants he examined in the Canadian Arctic possess a mycorrhizal association. Miller *et al.*, (1974) examines 28 vascular plant species at Barrow, Alaska finding 18 to have a mycorrhizal association.

The adaptive significance of this root fungal symbiotic relationship under conditions of a short growing season has been discussed by Meyer (1973). The ectotrophic situation increases the functional surface area of the root system for mineral and nutrient uptake. This increased input enables the plant to complete its annual growth and reproduction cycle in a shorter period of time. Hattingh *et al.*, (1973) demonstrated that mycorrhizae act to increase the effective phosphate uptake by the plant under low soil phosphate conditions. These mycorrhizae act not only as primary agents for phosphate uptake, but also as major sinks of incorporated phosphate. Mikola (1969) has shown that, in cool climates where mineralization of organic nitrogen is hampered, mycorrhizal fungi are very important in mobilization and reduction of nitrogen at levels adequate to support a plant's growth.

In areas where climate limits or disrupts plant growth during a normal growing season, the mantle, or fungal sheath, plays a significant role in continued plant success or viability (Harley, 1975). The mantle acts as a specialized storage organ for minerals and nutrients such as nitrogen, phosphates, and glycogen which can be mobilized during times of plant need, such as the beginning of the growing season, when light might not be sufficient to support adequate carbon fixation for growth. The findings of Ling-Lee *et al.* (1975) showing accumulation and degradation of polyphosphate granules in the mantle support this role of the ectotrophic fungal mycorrhizal mantle. It is apparent, then, that the role of the mycorrhizae in Arctic tundra plants represents a significant component for plant growth and success.

With the advent of oil exploration and utilization in the Arctic, these mycorrhizal associations take on even more significance. Studies from temperate climates in industrial or coal mine bespoiled lands show that plants in such soils are invariably mycorrhizal. Schramm (1966) and Meyer (1968) showed that mycorrhizal plants growing on anthracite coal spoils exhibited normal growth and physiognomy, whereas those not mycorrhizally associated were stunted and showed signs of nitrogen deficiency. *Betula* and *Salix* utilized in this study which have representatives in the Barrow tundra, *Betula* and *Salix*, were shown to require an ectotrophic mycorrhizae for successful growth in the coal spoils. The Arctic representatives of these genera have ectotrophic mycorrhizae in tundra habitats (Katenin, 1964; Miller, *et al.* 1974).

Oil exploration has stimulated work on the effect of oil on Arctic tundra plants (Deneke *et al.*, 1975; Freedman and Hutchinson, 1976; McCowan *et al.*, 1973; McCowan, 1974). These studies have dealt almost exclusively with the above ground plant biomass, and have shown that direct contact with fresh crude oil kills leaves and causes premature senescence while exposure to oil in the soil adversely affects leaf enzyme and pigment systems, and root nutrient and water uptake.

The long term impact of oil on plant root respiration and biomass has only recently been examined (Linkins *et al.*, 1978b). This work has shown that root respiration rates are quickly depressed upon exposure to oil, and remain so for at least two years following exposure to oil. Respiratory quotients in these oil exposed roots are lowered from values indicative of carbohydrate based oxidative metabolism. Linkins *et al.*, (1978a) have also shown that at 1 °C respiration rates of oil exposed roots are significantly depressed below those noted in control roots. This finding is significant in light of the annual reduced root biomass for roots in oiled soils (Linkins *et al.*, 1978b) and the observation by Deneke *et al.*, (1975) and McCowan, *et al.*, (1973) that viable buds on rhizomes declined, as well as standing above ground biomass, for plants in oiled soils one winter after exposure to oil. These data suggest that roots in oiled soils for at least three years after an oil spill may be susceptible to increased winter kill.

Antibus and Linkins (1978) have shown that *Salix rotundifolia* Trautv. at Barrow, Alaska has an ectomycorrhizal association. The present paper examines the impact of surface applied oil on the ectomycorrhizal roots of *Salix rotundifolia*. The same parameters of general respiration, respiratory quotient, and viable root biomass were examined to determine if the ectomycorrhizal habit affords the roots any benefits over what has been determined in non-mycorrhizal roots (Linkins *et al.*, 1978a, b). Roots of *Salix rotundifolia* found in the weathered oil impregnated soils at the Cape Simpson oil seep were examined to see if they show an adaptation to fresh oil. Finally, the presence of cold acclimation in these mycorrhizal roots was examined in the presence of oil to determine if oil related losses in cold acclimation or chilling resistance could account for annual losses in viable root biomass (Antibus and Linkins, 1978; Linkins *et al.*, 1978a, b).

MATERIALS AND METHODS

Plot Description and Sampling: The Arctic willow, *Salix rotundifolia*, is a dominant vascular plant in areas of the U. S. I.B.P. Tundra Biome site #1 at Barrow, Alaska (Murray and Murray, 1973). The soils in this site are relatively dry polygonal tundra soils with a thin (2 cm) organic layer underlain by mineral soil (Everett 1974,1978). This soil is typical of the high center polygon center soils where similar oil studies were conducted (Everett, 1978; Linkins *et al.*, 1978a, b).

Plots approximately 1 × 2 m were delineated in areas where *S. rotundifolia* comprised greater than 90% of the vascular plant cover. Prudhoe crude oil at ambient temperature was applied to the surface of the *Salix* plots at a 5 or 12 l/m² level on 1 July 1975. Another plot within the I.B.P. Tundra Biome site #1 with *Salix rotundifolia* as the dominant vascular plant was delineated and treated with ambient Prudhoe crude oil at 12 l/m² on 9 July 1977. *Salix* plots were designated: control — no oil; 12 l/m², 1975; 5 l/m², 1975; and 12 l/m², 1977.

Salix rotundifolia is also the predominant vascular plant in the dry oil impregnated soils immediately surrounding the mid and upper areas of the natural oil seep /2 at Cape Simpson, Alaska. The oil in these soils is primarily highly weathered tars and asphalts (R. M. Atlas, pers. comm.).

Root samples from the control, 5 and 12 l/m² oiled plots at Barrow were taken at 5.0 × 4 cm soil blocks. In all cases, soil samples were wrapped in plastic and immediately transported to the Naval Arctic Research Laboratory at Barrow. Soil was then stored at 5-7 °C until used.

Roots were washed and picked from soil samples and placed in sterile soil water at 10 °C for use in respiration studies (Barnard and Jorgenson, 1977; Boyer *et al.*, 1971; Harley, 1969; Linkins *et al.*, 1978a, b; Mikola, 1967). Roots for respiration studies were chosen as described by Antibus and Linkins (1978) and were similar to the Class A or I described by Barnard and Jorgensen (1977).

Respiration studies were conducted in a refrigerated Gilson Differential Respirometer. Oxygen and carbon dioxide flux were determined using the direct KOH method (Umbreit *et al.*, 1964). Respiratory quotients were calculated from O_2 and CO_2 data (White *et al.*, 1973).

Weekly respiratory data for roots from the Barrow plots were done at 10 °C as previously described (Linkins *et al.*, 1978a, b). Evaluation of respiration at 26, 21, 16, 10, 5, 3 and 0.5 °C was done on 1 g fresh weight samples of roots. Runs were made either from 26 to 0.5 °C or from 10 to 26 and 10 to 0.5 °C. All runs were done in duplicate using triplicate samples. Samples were equilibrated at each temperature for at least 2.5 hrs after the new temperature was attained. The maximum time for any series of temperatures in a run was 30 hrs. After each series of a run, flask temperatures were equilibrated at 10 °C and respiration ratios compared to known 10 °C rates from fresh roots. A minimum of 5 respiration readings were taken at each temperature.

RESULTS AND DISCUSSION

Figures 1a, b, c show the effect of 5 and 12 l/m² oil on root respiration of *S. rotundifolia* for three seasons after surface application. Within 48 hrs after oil application, root respiration was reduced 22% and 56% for 5 and 12 l¢m² treated soils respectively. At the end of the season, depression was 80% below controls with no significant difference between oil treatments (Figure 1a). This general seasonal decline in control respiration was most likely due to decreasing soil temperatures (Everett, 1978) and the accompanying initiation of plant senescence (Everett, pers. comm.).

The second season oil treated roots had respiration rates higher than expected, with roots from the 5 l/m² soils having higher respiration rates than the controls (Fig. 1b). As the season progressed, respiration rates of the oiled roots declined far more rapidly than did the controls. At the end of the season, respiration rates of oiled roots again were an average of 80% lower than controls.

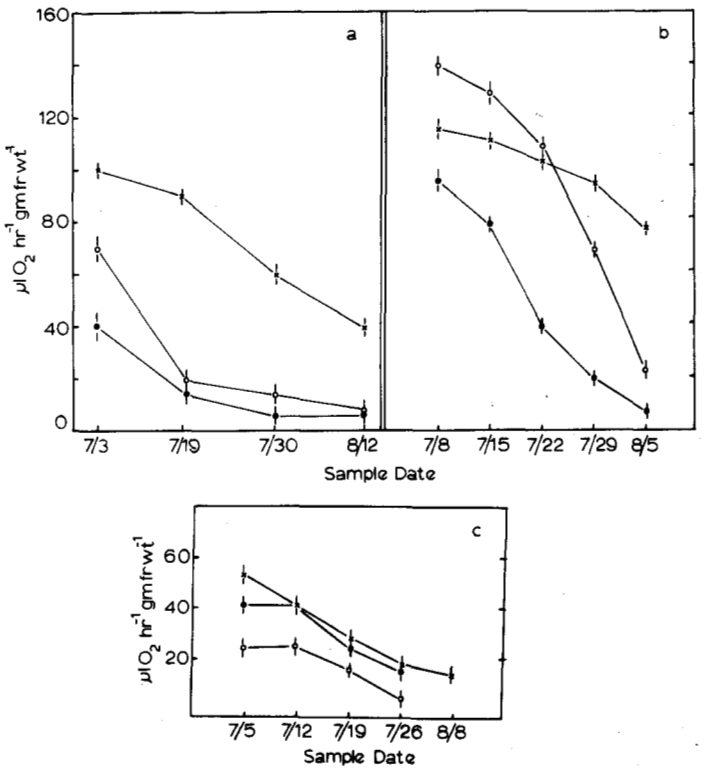

FIG. 1. a) Mycorrhizal root respiration during the 1975 field season. x - - - - x, control plot; o - - - - o, 5 l/m² plot; ● - - - - ●, 12 l/m² plot.
b) Mycorrhizal root respiration during 1976 field season. x - - - -x, control plot; o - - - - o, 5 l/m² plot; ● - - - - ●, 12 l/m² plot.
c) Mycorrhizal root respiration during 1977 field season. x - - - - x, control plot; o - - - - o, 5 l/m² plot; ● - - - - ●, 12 l/m² plot.

July and August declines in overall root respiration rates of control plants is attributed to the unusually low water potentials of these normally moist soils (Everett, 1978; Barnard and Jorgenson, 1977; Kramer and Kozlowski, 1960) and the normal seasonal declines in temperature (Everett, 1978). Whether the rapid decline in oiled root respiration rates is due solely to temperature is not known, since the drought conditions in 1976 had a significant impact on soil water potentials (Everett, 1978). The unusual decline in late season after early season high respiration rates is probably due to temperature (as is discussed later and supported Fig. 1a) and decreased soil water potential.

The effect of reduced soil water potential on root respiration is demonstrated in Figure 1c. Soil temperatures in 1977 were not significantly lower than in either 1975 or 1976 (Everett, 1978), yet control root respiration rates were much lower than in either previous year (Fig. 1c). The only major difference in the soil environment at Barrow in 1977 was that the drought condition which extended from 1976. In 1977, the *Salix* control plot had 45%

water by volume, the 5 l/m² had 15%, and the 12 l/m² had 4%. The overall seasonal decline in root respiration is probably due to continued drought stress, plus normally decreasing soil temperatures (Everett, 1978). In 1977, however, oiled roots never showed the large reductions in respiration rates relative to control roots. In fact, the 12 l/m² oiled roots were not significantly different in respiration rate from the control (Fig. 1c). Roots oiled at the 5 l/m² level paralleled the general trends in respiration rates, but at slightly lower rates.

Evaluation of the respiratory quotient (RQ) for roots from oiled soils revealed a significant decrease for all roots during all years. The RQ for control roots of *Salix* ectomycorrhizae was 0.85(0.10), while all roots from oiled soils had 0.65(0.15). Roots of *S. rotundifolia* from oil impregnated soils at Cape Simpson had RQ values of 0.68(0.09).

Respiratory quotients give a general evaluation of the substrate for oxidative metabolism, with values 0.9 - 1.0 indicative of carbohydrate catabolism while values around 0.66 are indicative of fatty acid catabolism via B oxidation (White *et al.*, 1973). The lower RQ values for the *Salix* roots from Cape Simpson or the non-mycorrhizal roots of *Carex aquatilis*, Wahl. or *Dupontia fischeri* R. Br. from Cape Simpson or the oil spill plots at Barrow (Linkins *et al.*, 1978a; Linkins, 1978) suggest that root metabolism may be permanently altered from the general 0.9-1.0 situation for tundra plants (Linkins *et al.*, 1978a, b) when oil of any state of weathering is present in the soil. The presence of the oxygen requiring enzyme aryl hydrocarbon hydroxylase in these roots (Linkins, 1978) could possibly account for lowered RQ values, but cannot preclude the possibility of aliphatic hydrocarbon utilization by the roots since they have the basic metabolic pathways which can contribute to straight chain oxidation (Hunt, 1972). However, specific enzymological work needs to be completed before aliphatic hydrocarbon utilization by plant roots can be considered as contributing to lowered RQ values for roots in oil contaminated soils.

If lowered RQ values are indicative of altered root metabolism and the occurrence of hydrocarbon modifying enzymes, both of which could be considered adaptations to the presence of oil in the soil, then plant adaptation to oil is incomplete. Roots of *Carex aquatilis* and *Dupontia fischeri* showed seasonal declines in viable root biomass for three seasons after exposure to oil, despite lowered RQ values (Linkins *et al.*, 1978a, b). Table 1 shows that the same trend in declining viable root biomass after exposure to oil is present for the ectotrophic mycorrhizal roots of *S. rotundifolia*.

The degree of impact of oil on viable root biomass is less severe in the *Salix* ectomycorrhizal roots (Table 1) than that noted for non-mycorrhizal roots in the similar soils of the high center polygons (Linkins *et al.*, 1978a, b). In the high center polygon habitats there were no observed viable roots after application of oil, and higher plant growth due to survival was only noted for *Petasites frigidus* (L.) Fries during the second year after oil application. The nature of the thick, protected root of *Petasites frigidus* and the alterations in the mantle structure in *Salix* ectomycorrhizal roots after exposure to oil (Antibus

and Linkins, 1978) suggests that the increased survival of *Salix* roots is due to a large degree to the physical protection afforded by the fungal mantle.

The trend in the annual decrease in viable root biomass for *Salix* mycorrhizal roots (Table 1) or non-mycorrhizal roots of *Carex aquatilis* and *Dupontia fischeri* (Linkins *et al.*, 1978a, b) suggests that the presence of crude oil may reduce the ability of the roots to survive in the tundra environment. Low temperature stress has been shown to cause a greater reduction in respiration rates of oil exposed roots of *D. fischeri* and *C. aquatilis* (Linkins, *et al.*, 1978a). Observations by Deneke *et al.*, (1975) and McCowan *et al.*, (1973) of reduced numbers of buds on rhizomes and decreased numbers of seemingly viable rhizomes and roots of plants in oiled soils after a winter season further suggest that these roots or rhizomes may be susceptible to cold and winter kill after exposure to oil.

The specific influence of oil on the mycorrhizal roots of *S. rotundifolia* can be evaluated by measuring respiration rates over a 26 to 0.5 °C range. Arrhenius linear transformations of these data are a convenience and useful means of evaluating the impact of oil on respiration rates at different temperatures. A linear constant slope of an Arrhenius plot indicates a proportional decrease in the activity of an unaltered system with temperature, while a change in the slope above or below a given temperature indicates a change in the efficiency of the system measured beyond the indicated temperature (Kumamoto *et al.*, 1971; Park, 1970). Consequently, an increased slope below a given temperature would indicate an increased energy of activation or Q_{10} for the system measured.

Figure 2a is an Arrhenius plot of respiration rates of *S. rotundifolia* ectomycorrhizal roots from Barrow 1975 control and 12 l/m² oil treated plots 2.5 weeks after exposure to ambient Prudhoe Bay crude oil. Control roots show no change in the slope of declining respiration rates with declining temperatures, supporting the opinion that these roots are cold acclimated or cold hardy. Roots from the 12 l/m² oil plots, on the other hand, not only show reduced respiration rates at all temperatures, but also an increased slope in declining respiration below 4-5 °C.

Arrhenius plots of root respiration rates were done in 1977 on roots from the 1975 control and 12 l/m² oiled plots and 12 l/m² oil treated plot treated 2.5

TABLE 1. Seasonal mean viable ectomycorrhizal roots of *Salix rotundifolia* from control and 5 and 12 l/m² oiled plots at Barrow, Alaska. Data presented as dry weight/2.5 x 4 cm soil core. Standard deviation in parentheses.

	Control	5 l/m²	12 l/m²
1975	0.20 (0.12)	0.56 (0.19)	0.54 (0.20)
1976	0.70 (0.12)	0.42 (0.08)	0.43 (0.10)
1977	0.62 (0.10)	0.37 (0.08)	0.27 (0.06)

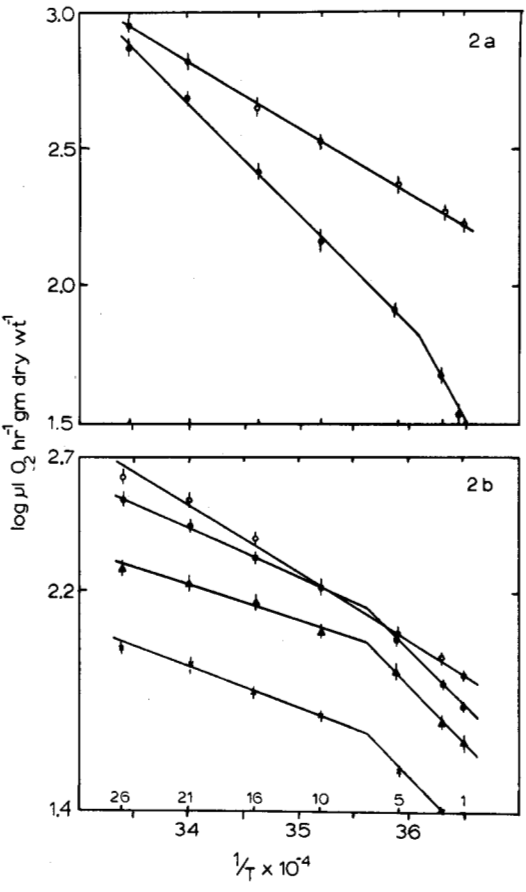

FIG. 2. Arrhenius plots of respiration rates of mycorrhizal roots of *Salix rotundifolia* from Barrow, Alaska, over a 26-0.5 °C temperature range. Plots represent linear regression analysis with coefficients of determinations of $r^2 = 0.89$ or better.

a) Respiration rates for 1975. o - - - - o, control roots; ● - - - - ●, roots from 12 l/m² oiled plots.

b) Respiration rates for 1977. o - - - - o, control roots; ● - - - - ● roots from the 1975 12 l/m² oiled plots; ▲ - - - - ▲ roots from a 1977 12 l/m² oiled plot; x - - - - x roots from a control plot treated with a 12 l/m² oil treatment in a respirometer flask.

weeks prior to the 1977 respiration rate evaluations (Fig. 2b). Control roots again show a constant slope equivalent to the slope from the 1975 control roots. Roots from the 1975 12 l/m² oil treatment plots still showed respiration rates lower than controls and an increased slope below 6 °C. However, the degree of depression and increase in slope were very much reduced from 1975 levels. Roots treated with oil in 1977, 2.5 weeks prior to respiration rate evaluation showed depression in respiration rates and an increase in slope below 4-7 °C. The impact of oil in 1977 seems initially to be less severe on root respiration rates at any temperature.

Ectomycorrhizal roots of *S. rotundifolia* from the Cape Simpson oil seep, which have a higher percentage of mantles formed by *Cenococcum graniforme*,

(Sow.) Ferd and Winge. were also evaluated with Arrhenius plots of respiration rates (Figs. 3a, b). Oil treated roots from Cape Simpson were treated *in vitro* in the respiration flask with a 12 l/m² equivatent of Prudhoe Bay crude oil.

Control roots from Cape Simpson in 1975 and 1977 gave a constant slope for the line describing declining respiration rates with declining temperature, as did control roots from Barrow (Figs. 3a, b). Comparison of respiration rates for the control roots from the two sites show that they are quite similar (e.g.: at 10 °C in 1975 they were 316 and 338 μl 0_2/hr/g dry wt for Barrow and Simpson roots respectively and in 1977 they were 156 and 170 μl 0_2/hr/g dry wt for Barrow and Simpson roots respectively). The 1975 values for *Salix* at 10 °C are slightly lower than those noted for temperate grown loblolly pine ectomycorrhizal roots of a similar type Class A type I, and considerably lower than the pine root respiration rates at 15 °C (Barnard and Jorgensen, 1977). However, if the loblolly pine root respiration rates are extrapolated down to 5

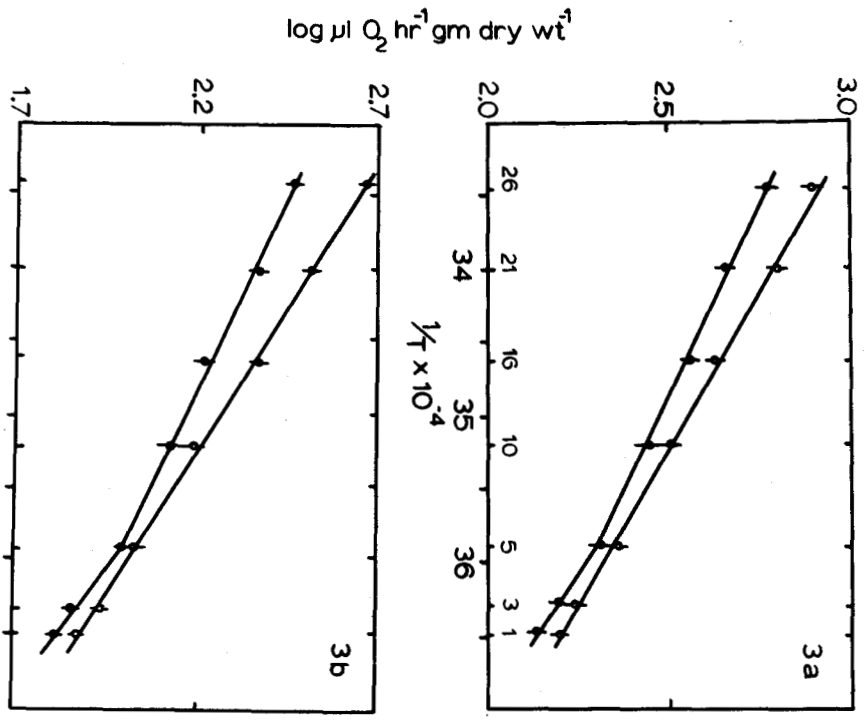

FIG. 3. Arrhenius plots of respiration rates of mycorrhizal roots of *Salix rotundifolia* from the Cape Simpson oil seep #2 at Cape Simpson, Alaska, over a 26-0.5 °C temperature range. Plots represent linear regression analysis with coefficients of determinations of $r^2 = 0.91$ or better.

a) Respiration rates for 1975. o - - - - o, control roots; • - - - - •, roots treated in the respirometer flask with 12 l/m² crude oil.

b) Respiration rates for 1977. o - - - -o, control roots; • - - - - •, roots treated in the respirometer flask with 12 l/m² crude oil.

°C (175 μl 0_2/hr/g dry wt) the 1975 respiration rate values for *Salix* (230 μl 0_2/hr/g dry wt) are considerably higher. The lowered respiration rates for 1977 roots are probably a reflection of decreased soil water potentials, as previously discussed. *In vitro* addition of oil to roots from Cape Simpson showed only a relatively moderate impact on respiration rates (Figs. 3a, b) when compared to the impact on roots from Barrow control plots (Fig. 2b). Even though there was a definite increase in the slope of respiration rates below about 5 °C, there was no significant difference in the slope of the control or *in vitro* treated roots in this 5-0.5 °C range.

Oil, then, significantly reduces root respiration rates in *S. rotundifolia* roots at all temperatures. Evaluation of the data with Arrhenius linear transformations reveals that there is an undermining of the cold acclimation or cold hardiness of the roots below 5-7 °C. This loss in cold acclimation or cold hardiness would affect root metabolism, energy generation for growth, mineral uptake, etc., throughout the growth season since soil temperatures below 5-7 °C are encountered daily during mid-summer July and August (Miller *et al.*, 1974).

Early agricultural work has shown that the lighter components of oil primarily disrupt the cell at the membrane by dissolving or disturbing the hydrophobic fatty acid area of the membrane (Baker, 1970). Cold acclimation or cold hardiness in plants has been correlated with changes in the sugar, protein, amino acid content of the cell or changes in the fatty acid composition of the membrane (Garber and Steponkus, 1976; Ilker *et al.*, 1976; Lyons, 1973; Steponkus, 1971; Steponkus *et al.*, 1977; St. John and Christiansen, 1976). All agree that the mechanisms of cold acclimation or cold hardiness are to varying degrees associated with the cell membrane, be it due to addition of specific proteins and cryoprotectants such as sucrose or alteration in the degree of unsaturation of membrane fatty acids and maintenance of membrane fluidity at lower temperatures.

At present, we cannot ascribe a specific site of mechanism for oil's undermining the cold hardiness of *S. rotundifolia* ectomycorhrizal roots. Oil undoubtedly affects the root cell membranes either dissolving or disturbing the unsaturated fatty acids involved in membrane fluidity and cold hardiness. However, the significant lowering of the root's RQ could be associated with loss in cryoprotectant accumulation at the root membrane. Oil could also directly lower enzyme substrate affinity at lower temperatures and thereby decrease cold hardiness.

Regardless of the specific mechanism involved, Arrhenius plots of respiration rates show that cold hardiness is decreased in these roots (Silvius, *et al.*, 1978). Yet, even though no specific mechanism for oil perturbation is apparent, the similarities of all slopes for oil treated roots in 1977 for *S. rotundifolia* (Fig. 2b) and *Carex aquatilis* (Linkins, 1978) suggest a common mechanism of oil perturbation on at least respiration. The duration of exposure only affects the magnitude of losses in respiration, not the slope of the losses with temperature.

CONCLUSION

The impact of crude oil on the respiration rate and survival of the ectomycorrhizal roots of *S. rotundifolia* in the dry tundra soils is less than that noted for the non-mycorrhizal roots of *C. aquatilis* or *Luzula confusa* Linde b. (Linkins *et al.*, 1978a, b) in similar dry soils. These ectomycorrhizal roots do not possess characteristics which make them immune to crude oil in the soil as their losses in respiration rates, cold and probably drought hardiness, and viable root biomass are similar to non-mycorrhizal roots in oil contaminated soils (Linkins *et al.*, 1978a, b) Even the acquisition of aryl hydrocarbon hydroxylase activity, reduced RQ values, biomass stabilization, and cold hardiness after prolonged existence in oiled soils is similar to non-mycorrhizal roots in similar habitats (Linkins, 1978; Linkins *et al.*, 1978a, b). The decreased impact of oil on ectomycorrhizal roots may be due mainly to their buffering effect in reducing the amount of oil reaching the living root, much as Wein and Bliss (1973) have suggested in how the bud scale may protect a bud from oil. The impact of oil on the mycorrhizal mantle, but not the *Salix* root, has been shown by Antibus and Linkins (1978). Whether the mycorrhizal fungus imparts any physiological advantage to the higher plant root under conditions of oil exposure is not known.

Despite the fact that the ectomycorrhizal habit does not seem to impart any unique oil tolerant or utilization characteristics, this habit may prove extremely useful in revegetation of drier soils contaminated with crude oil. If the ectotrophic fungus *Cenococcum graniforme* from the Cape Simpson oil seep *Salix*, which possess oil tolerance and utilization potential (Antibus and Linkins, 1978; Linkins, 1978) as well as maintenance of cold and drought hardiness, can effectively be associated with the wide range of known host plants for *C. graniforme* (Trappe, 1964), then the number of oil contaminated habitats which can be successfully revegetated will be greatly expanded.

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