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Boojum RESEARCH LIMITED

DEVELOPING A TECHNIQUE FOR RECLAMATION OF HIGH IRON SULPHUR TAILINGS BY ESTABLISHING A MARSHLAND

PHASE 1

SELECTION OF SUITABLE PLANT MATERIAL AND METHODS OF ESTABLISHMENT

BY

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FOR

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EXECUTIVE SUMMARY

Boojum Research Ltd. has conducted laboratory and field investigations related to the establishment of semi-aquatic vascular plants on perched pyrrhotite tailings (Fault Lake) at Falconbridge. Seeds from a variety of semi-aquatic plants were collected from the Falconbridge Conservation Area and elsewhere. The seeds were subjected to different pre-germination treatment procedures prior to germination trials. Air-dried seeds (a grass, a sedge and cattails from acidic, alkaline and control habitats) held at room temperature, showed best germination in tap water; seeds of 2 other sedges and 1 rush failed to germinate following any of the pre-germination treatments.

Seeds showing positive results in tap water were further assayed for germination on pyrrhotite from Fault Lake. The pyrrhotite was either amended with dewatered sewage sludge, with $Ca(OH)_2$ (pH 3.0, 7.0 or 10.0) or left unamended (pH 2.3 - 2.5). No seeds germinated on sewage-amended, pH 3.0-amended or unamended pyrrhotite. Excellent germination (50 to <100%) occurred with the grass and a sedge on pH 7.0 - 10.0-amended pyrrhotite. In contrast, cattail seeds collected from three different habitats (acid, alkaline and control) showed considerably lower germination rates (ca. 10%).

The germination results for the semi-aquatic grass and a sedge suggest that direct germination of at least some semi-aquatic vascular plants on moist, pH 7.0 - 10.0, amended pyrrhotite may be a feasible method for introducing certain species of wetlands plants into the Fault Lake experimental cells.

Field and laboratory experiments with cattail seedlings and mature plants were carried out under contract within Inco/Rats program and are relevant in the context of the overall objective. These results suggest that mature root crowns of cattails, those which bear one or more rhizome primordia, stand a better chance of surviving transplanting onto either acidic or alkaline tailings than do seedlings and younger plants.

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1.0 INTRODUCTION

According to Nawrot (1985), interest in converting abandoned mine drainage areas into wetlands dates back about 30 years. In coal mining regions of the United States, the reclamation of mine sites into areas of natural wetlands has been successful. Symposia focusing on the various technologies required to create wetlands are being held with increasing frequency (Brooks et al., 1985).

It has been demonstrated that with careful planning reclamation scenarios for surface mined lands and waste areas are less costly to implement and maintain as wetlands, than if the areas are reclaimed conventionally. Environmental problems related to acid generation may be reduced once a wetland has become established. Accumulation of organic detritus on the bottom of the bog reduces O_2 penetration into underlying acid-generating waste and hence, reduces acid generation. Furthermore the aquatic ecosystem has the capacity to remove many toxic elements from the water and thereby improve the effluent water quality. In spite of the great and continuing interest in employing wetlands for the close-out of coal mine areas, the application of the same concept has rarely been extended to the chemically somewhat harsher conditions of pyritic and pyrrhotite tailings areas of the base metal industry in more northern climates of Canada.

It should be noted that the Falconbridge Conservation Area serves as a stellar Canadian example of the use of wetlands in a effluent treatment system. The settling area for the neutralized waters leaving the polishing ponds was previously a dead bog >160 hectares in area. The neutralized water was discharged over the acidic bog and within a short period of time (4-6 years) a variety of semi-aquatic plants invaded the area via natural plant dispersal agents. The Conservation Area now stands to the credit of Falconbridge Ltd. who maintain the wetlands and encourage its spreading through management of water levels during the year. The Conservation Area also provides a valuable source of semi-aquatic plant seed and root stock for use in establishing wetlands on other mine sites.

Although wetland management is well established, there are no methods for the successful establishment of wetland ecosystem for the extreme conditions of pyritic tailings and pyrrhotite. The aim of the CANMET/Boojum/Falconbridge joint research program was to develop methods leading to the establishment of a wetlands on an abandoned, impounded pyrrhotitic tailings pond (Fault Lake) at Falconbridge, Ontario.

The project goals for 1985, as outlined in the original proposal, underwent some modification due to unanticipated engineering problems encountered in constructing retaining ponds on the Fault Lake tailings. Therefore the emphasis had to be shifted from experiments designed to assess the survival and overwintering of semi-aquatic plants in experimental holes dug in the pyrrhotite, to laboratory experiments relevant to wetlands establishments, namely: semi-aquatic plant seed germination and establishment on amended and unamended pyrrhotite. These laboratory experiments provide information for use in subsequent field work on Fault Lake in 1986.

This report describes the results of experiments on semi-aquatic plant seed germination and establishment. The work carried out to obtain chemical data on leaching of the mine slimes and pyrrhotite from Fault Lake, as well as results on water percolation conducted on site at Fault Lake in July, 1985 have already been submitted as one of the interim reports 1 to 5.

2.0 MATERIALS AND METHODS

2.1 Seed collections from semi-aquatic plants

Seeds of semi-aquatic plants were collected from a number of sites. Three types of sedge seeds (Scirpus spp.) and one type of rush seed (Juncus sp.) were collected from the Falconbridge Conservation Area on 14 August 1985. A second collection of these seeds was attempted in early September but by then the seeds had been shed. The grass seeds (Bromus sp.) and acid and alkaline cattail seeds were collected on the Inco Levack property. Other cattail seeds were collected from a "control" site located near side road 65 toward Rudder, Ontario.

2.2 Pre-germination seed treatment

Seeds were maintained at room temperature (stored inside and outside a dessicator) or were frozen (2 months). Cattail seeds collected from plants growing in alkaline, acid and control sites were held at room temperature. Bonnewell et al (1983) reported no special temperature or drying pre-treatment for cattail seeds.

2.3 Methods for seed germination

2.3.1 Wick method

In most of the studies, a filter paper "wick" method was used. A small triangular piece of (Whatman #1) filter paper, about 6 cm long, was moistened with the test liquid (or the supernatant portion of a pyrrhotite slurry). Seeds were sprinkled on one side of the filter paper; this side was then pressed against the bottom of a 12.5 cm dia. petri plate. The petri plate was elevated about 1.0 cm on one side and sufficient liquid was added to immerse the lower tip of the filter paper triangle. The petri plate was covered.

Germination was examined after 1-3 weeks. In all the germination experiments, the physical conditions of light (60-75 $uE.m^{-2}.s^{-1}$), photo-period (12:12-L:D) and temperature (23^{o}C) were identical. The number of each type of seeds used in each experiment varied from 8-200, depending on supply.

2.3.2 Filter paper sandwich method

In one set of experiments with cattail seeds, an alternative method was tested. A strip of filter paper, the width of conventional microscope slide (2.5 cm) and twice the length (7.5 cm x 2 = 15) was moistened. Seeds were sprinkled on one half of the filter paper; the other half was folded over to cover the seeds and the paper was placed between two glass slides, much like a sandwich. The assembly was secured with a paperclip. The sandwich was placed in a petri plate. One end of the slide sandwich rested on small glass beads while the other end was immersed in the test liquid.

2.3.3 Direct germination on amended and unamended pyrrhotite

Pyrrhotite, having the consistency of sand, was treated in one of the following ways. A slurry was made by adding distilled water (1:3; v/w) and the pH was adjusted to 3.0, 7.0 or 10.0 by addition of dry $Ca(OH)_2$. The additions of $Ca(OH)_2$ were made over 2-3 days, in order to ensure stabilization of the pH. The initial pH of unamended pyrrhotite was 2.3-2.5.

In some experiments, 6% dewatered sewage sludge was obtained from the Humber Waste Treatment Plant (City of Toronto). The sludge was autoclaved (20 min; 120°C) prior to use. A ratio of 10 ml sludge to 6 gm (DW) pyrrhotite was employed.

The pyrrhotite was kept moist (but not flooded) in the germination experiments, seeds were directly sprinkled onto the pyrrhotite surface.

2.4 <u>Seedling establishment</u>

2.4.1 Early growth of seedlings - filter paper germinated stock

Seedlings of <u>Bromus</u> sp. were robust and pushed the filter paper triangle up toward the lid of the petri plate. A dilute (1 $g.L^{-1}$) commercial household nutrient mixture ("RX-15") was added and the seedling roots were kept moist (in a quasi hydroponic milieu). Two week old seedlings (ea. 2-5 cm tall) were transplanted into moist, unamended pyrrhotite or pyrrhotite amended with sewage sludge.

Seedlings of cattail were transferred to petri plates filled with moist sand. After 2 weeks, when the cattail leaves (1-2 cm long) were in an erect position, clump of seedlings (with sand immersed roots) were transplanted onto pyrrhotite as above.

2.4.2 Early growth of seedlings - direct germination on pyrrhotite

Transplanting of any plant necessitates disturbing the roots, temporarily upsetting the physiology of growth and development of the plant. In the case of transplanting from non-stressed seed germination conditions to the stressed milieu of moist pyrrhotite (amended or unamended), we decided to eliminate the transplant "trauma" and germinate directly onto pyrrhotite those seeds which showed the best results in the germination experiments.

2.5 <u>Cattail and Phragmites planting on mine slimes at Fault Lake</u>

Seeds from cattails growing along the "acid road" shoreline of the Conservation Area, were sown (July 24, 1985) around the perimeter of the "transient" pond at the mine slime end of Fault Lake.

A dozen young cattail plants and one Phragmites (Reed Grass) were transplanted from the same location in the Conservation Area to the transient pond on Fault Lake.

3.0 RESULTS

3.1 Seed collections

Seeds were collected twice towards the end of the summer. The results suggest that viable seeds may be collected from semi -aquatic plants (grass #1 and sedge #2) - exclusive of cattails - in August. By September, these seeds have been shed.

3.2 Pre-germination seed treatment

The embryo within the seed is dormant and in order to "activate" it, that is, to trigger it to germinate, a sequence of physical-chemical, external and internal factors must be brought into play. In the most simple case, seed germination may merely require that dry seeds be exposed to water. Often, however, the sequence of factors is much more complicated (cf. Bewley & Black, 1978 and 1982).

If semi-aquatic seeds are harvested in late summer for sewing on potential reclamation sites, one must know whether the seeds have a special requirement for germination. It was beyond the scope of the present study to explore any but the most commonly effective pre-germination treatments which promote seed germination: moisture, freezing and scarification, i.e., cutting through the seed coat in order to facilitate water penetration into the embryo.

The most straightforward germination test is to place seeds on a piece of filter paper moistened with tap water, in a covered petri plate. Primary roots and leaves will usually appear within one or two weeks. In the present experiments we decided to test several germination procedures because we were employing petri plates filled with pyrrhotite (amended and unamended) and the conventional method might not have been the most useful one.

3.2.1 Pre-germination seed treatment

Table 1 gives the data for the first experiment - exclusive of cattail seeds. A very small but persistent germination occurred in sedge species #1 in all treatments. A sizeable number of seeds from sedge species #2 (dessicator stored) germinated after 3 weeks. Grass species #1 also showed good germination in both room temperature groups.

A repeat of the germination experimenmt was carried out and the results in Table 2 confirm those in Table 1. Rush seeds, with a thick seed coat, showed no germination after three weeks. The seeds were then "scarified", a technique whereby the seed coat is carefully sliced along one edge in order to permit water entry into the seed. After 3 weeks, the scarified seeds had still not germinated.

TABLE 1

Effect of Pre-germination Protocols on Seed Germination

Experiment #1

	Number and % germination					
			In dessicator			
	Room Temp.		Room Temp.		Frozen	
Seed type	2 wks	3 wks	2 wks	3 wks	2 wks	3 wks
<u>Scirpus</u> spp.						
Sedge #1	1/104 (0.9%)	1/104 (0.9%)	2/78 (2.6%)	2/78 (2.6%)	1/27 (3.7%)	1/27 (3.7%)
Sedge #2	0/15	0/15	0/23	5/23	0/17 (21.7%)	0/17
Sedge #3	0/12	1/12 (0.8%)	0/12	0/12	0/10	0/10
<u>Juncus</u> sp.						
Rush #1	0/16	0/16	0/14	0/14	0/12	0/12
<u>Bromus</u> sp.						
Grass #1	6/24 (25%)	6/24 (25%)	3/36 (8%)	3/36 (8%)	0/24	0/24

All tests performed in petri dishes with triangular filter paper method and tap water.

TABLE 2

Effect of pre-germination protocols on seed germination

Experiment #2

Seed type	Room Temp. 2 wks	Number and % germination In Dessicator Room Temp. 2 wks	Frozen 2 wks
Scirpus spp.			
Sedge #1	33/183 (18%)	38/200 (19%)	1/150 (0.6%)
Sedge #2	31/ 56 (55%)	6/ 69 (9%)	0/ 56
Sedge #3	0/ 28	0/ 19	0/ 17
Juncus sp	0/ 40	0/ 30	0/ 36
Bromus sp	>50%	20/140 (14%)	11/100 (11%)

Germination of the cattail seeds is somewhat surprising in that the seeds from "environmentally stressed" populations growing in alkaline mine slimes and in an acid creek (pH 2.3) showed a higher percentage of germination than did seeds from the control population.

Table 3 gives the results for cattail seed germination experiments. It can be readily seen the seeds obtained from cattails growing in the pre-bog acid creek at Levack showed the best germination. In addition, the young seedlings were sturdy with thick green leaves. The next best performance was by the seeds collected from cattails growing on alkaline mine slimes at Levack. The young seedlings were less robust than those derived from the "acid seeds". Finally, the poor performance of the control seeds was evidenced by their wispy character: thin, limp leaves.

When compared with seeds from the control sites, the astonishing germination performance of the acid creek cattail seeds and, to a lesser extent, those seeds from alkaline cattails does make ecological sense in terms of species survival. That is, the two populations of cattails growing under physiological stress of either acid or alkaline conditions produce seeds which contain more stored nutrient materials (carbohydrates and protein) than do seeds from plants growing in less stressed ("normal"?) habitats. Under stressed conditions at Levack, the acid creek cattails were ca. 1.5 m tall whereas plants in an unstressed population (also at Levack) were in excess of 2 m tall. In other words, more C and N goes into seed production in stressed plants than into leafy, dry matter (Kalin, 1986 1986, RATS, Inco). The stored reserve materials in the seeds from the

TABLE 3

Cattail seed germination experiments

% Germination[2]

Cattail[1] Seed Source	Experiment (and date) #1 18-XI-85	#2 2-XII-85	#3 30-XII-85
Acid Creek	38	90	100
Alkaline Mine Slimes	5	54	80
Control Site	13	33	26

1 - T. latifolia from different sites
2 - 50-100 seeds per treatment

stressed population gives cattail plants a better chance of survival in new sites (removed from the parent) because a greater number of the seeds are capable of germination and the seedlings themselves are robust and sturdy individuals.

It should be noted, however, that seeds from the three cattail seed sources germinated with about an equal success rate on pH 7.0 amended pyrrhotite, with no demonstrable superiority of any seed type in winter (see section 3.3.2).

3.2.2 Comparisons of seed germination methods

The pre-germination treatments were assayed by germination in tap water using the simple wick method. It was felt that a modification of the wick method would be needed for seed germination tests where amended and unamended pyrrhotite was being used. The so-called sandwich method was designed to prevent direct contact of the seeds with the pyrrhotite surface. The filter paper only absorbed the supernatant of the pyrrhotite slurry. In other experiments (section 3.3.2) seeds were directly germinated on the pyrrhotite.

3.2.2.1 Wick vs. sandwich methods

As the results in Table 4 indicate, the "wick" method was far superior to the "sandwich method".

TABLE 4

Comparison of cattail seed germination techniques: "wick" vs. "sandwich" methods (7-XII-85)

Seed Source and pH of Medium	Number and % germination "Wick" Method	"Sandwich" Method
Acid seeds		
pH 3.0*	32/40 (80%)	4/79 (5%)
pH 7.0*	30/53 (57%)	4/59 (7%)
pH 10.0*	10/37 (29%)	1/52 (2%)
Control seeds**		
Distilled water	57/275 (27%)	11/174 (6%)

* pH of supernatant of pH-amended pyrrhotite.
** as indicated in Table 3, acid cattail seeds showed 90-100% germination.

In the sandwich method, only 5-8 seeds (out of 100-120) germinated after 1 week. It was interesting that most of the seedlings were located at the margins of filter paper with few in the water-logged, submerged portion of the filter paper slide sandwich. The distilled water germination data in Table 4 seem to eliminate the possibility that pH-amended pyrrhotite had a significant effect in depressing germination of seeds employed in the sandwich method. The other possibility is that the water trapped between the slides became partially anaerobic and seeds will usually not germinate under such conditions. The presence of the germinated seeds at the margin of the filter paper supports the O_2 deficit hypothesis.

In the filter paper wick method, where the seeds lay between the bottom of a petri plate and a piece of moistened filter paper, there was a lesser chance for anaerobic conditions to build up. We entertain this last hypothesis of germination inhibition caused by an anaerobic environment, inspite of the fact that the published method of germinating cattail seeds calls for seeds to be placed under a layer of moistened filter paper and then flooded with 1 cm of water (Bonnewell et al., 1983).

3.3.2.2 Germination on pyrrhotite

a) Cattails

One essential difference may be noted between the wick method and direct pyrrhotite germination protocols: the wick method adsorbed water overlying pH amended pyrrhotite (Table 4) and the seeds were not germinated in direct contact with the pyrrhotite. Seeds were germinated in direct contact with pH amended pyrrhotite in the experiment discussed below.

Tables 5 and 6 show the results of cattail seeds germinated directly on pyrrhotite. In Table 5, seeds of acid-creek cattails were used because of the superior germination ability. Their germination percentage was rather poor, however, in comparison with the results shown in Tables 4 (wick method) and 3.

TABLE 5

Direct germination (1 wk) of acid cattail seeds, sedge #2 and grass #1 seeds on pH-amended pyrrhotite

pH of $Ca(OH)_2$* amended pyrrhotite slurry	% Germination		
	Acid Cattails	Sedge #2	Grass #1
pH 3.0**	0/>50	0/50***	0/>100
pH 7.0	2/>50 (<4%)	0/50***	90-100%
pH 10.0	6/>50 (<11%)	0/50***	90-100%

* sufficient $Ca(OH)_2$ was added to change pH; amounts not recorded over 2-3 day period
** cattail and grass (but not sedge) seeds became covered with a layer of grey fungus
*** at the end of 2 weeks, >50% of these sedge seeds had germinated

The data in Tables 4 and 5 suggest that inspite of creating a favourable pH in the pyrrhotite slurry at the outset of the experiment, the chemical "climate" on the pyrrhotite surface - after the supernatant has been discarded - may continue to change back toward its initial acid generating character, once the neutralizing supernatant has been removed.

b) Grass #1 and sedge #2

In comparison with the cattail results from the direct pyrrhotite germination experiment (Table 5), excellent (90-100%) germination occurred at pH 7 and 10 for grass #1. Sedge #2 showed no germination after one week - normally these seeds take 2-3 weeks - and after 2 weeks >50% had germinated. It may be noted that these two species substantially outperformed cattails in the direct germination of seeds in pH-amended pyrrhotite.

TABLE 6

Germination of acid, alkaline and control cattail seeds on amended pyrrhotite

Cattail Seed Source	% Germination pyrrhotite (pH 7.0)	pyrrhotite + sewage sludge (pH 2.5)
Acid	10-12/ 100 (11%)	0*
Alkaline	10-12/ 100 (11%)	0*
Control	10-12/ 100 (11%)	0*

* none of the seeds germinated; the surface of the pyrrhotite became infested with a grey fungus (mold)

3.3 Seedling establishment

3.3.1 Experiment on pyrrhotite

Neither the young "acid" cattail seedlings, raised for 2-3 weeks in sand culture, the grass #1 seedling, nor sedge #2 seedlings were successfully transplanted onto moistened pyrrhotite (unamended or amended with sewage sludge). Within 3-5 days of being transplanted, the leaves became bleached (loss of chlorophyll), an indication of death. Although transplanting is a stressful physiological event for plants, it is likely that the acidity of the pyrrhotite killed the seedlings.

However, as noted above (section 3.3.2) seeds from these three plants will germinate on pH amended pyrrhotite (7 and 10) but not at pH 3. So it is not too surprising that seedlings could not be transplanted onto unamended pyrrhotite.

3.3.2 A perspective on cattail seedling survival and plant establishment - laboratory results and field observations

While the environmental (physical and chemical) factors which control cattail seedling growth and developing on tailings - and in the laboratory - still remain somewhat of an enigma, we are gradually becoming familiar with these plants through our observations of their behaviour, both in the field and laboratory. Some relevant observations are listed below which have been derived from the work supported by the Inco/Rats program:

1) Young cattail seedlings growing in alkaline mine slimes (Levack) were transplanted to the laboratory in a manner which did not disturb the seedlings (1 cm tall) or the surface of the mine slimes. They were placed

in aquaria and covered with 0, 1 and 5 cm of water. Within 2 weeks all of the seedlings were dead. Similarly, seedlings maintained in the Inco greenhouse also perished.

2) Early in the summer, the alkaline mine slimes in the vicinity of some cattail stands at Levack were carpeted with young cattail seedlings. By the end of the summer the majority of these were dead - with 3 exceptions:

a) cattails seedlings which received fertilizer (2 kg of Scotts' Grow Vegetables or Scotts' Grow Flowers) treatment persisted well into the Fall when the area became inundated with water and froze over.

b) cattail seedlings growing on a tongue of mine slimes extending out into the Levack mine water retention pond formed by the effluent from the mine. The pond water had a NO_3 concentration of 41 ppm which was probably derived from blasting powder.

c) small, scattered 100 cm^2 clusters of plants growing in depressions in mine slimes on Levack.

Observations a) and b) suggests nutrients are essential for seedling establishment and c) indicates that the moisture balance and exposure may be other important factors.

3) Six percent dewatered and sterilized sewage sludge was added to sand cultures of the "acid" cattail seedlings (section 2.4.1) and no increase in growth was noted. When a dilute solution of the household plant nutrient was added (3 ml "RX-15" (1 $g.L^{-1}$)), the seedlings died within a week. These results suggest, that although nutrients appear important, the time of application during cattail seedling establishment is critical.

4) When cattails (>1 m tall) are transplanted, regardless of the presence or absence of visible rhizome primordia, the leafy portion may die within 1-2 months. This does not mean that the entire plant has died. Juvenile plants nearly always emerge in close proxiomity to the parent plant. Only 2 out of 60 plants which were transplanted at Levack failed to produce juveniles whereas nearly all of the aerial foliage of the transplants (parent) died back, long before the normal fall senscence of the leaves occurred. All juveniles appeared to be produced from rhizomes as they emerged at some distance from the parent plants.

5) As discussed below (section 3.5), young cattails which probably contained few rhizome primordia in the the root crown, did not survive the transplanting from the Conservation Area to the transient pond on the acid mine slimes at Fault Lake. In only one case, did a new shoot emerge beside the dead parent.

6) In retrospect, our cattail transplanting success at Levack was considerably better than at Fault Lake - with one minor point of interest: at Levack, an initial group of 10 plants were all young ones and they took much longer to become established than did more mature transplants which possessed developing rhizomes. It should also be noted that the Levack transplanting did not involve a change in the sediment type, as did the

transplantings at Fault Lake.

7) Mature cattails with one or more rhizomes developing at the root crown were dug from the alkaline mine slimes (pH 8-9) and grown hydroponicallly in the pre-bog acid creek (pH 2.3) at Levack. The survival rate was greater than 50%. It is obvious that mature cattails can successfully tolerate a wide range of pH's at the root-rhizome region.

8) Mature cattails - those with one or more rhizome primordia - stand a considerably better chance of surviving transplanting onto alkaline or acidic tailings than do seedlings and younger plants.

3.5 Transplanting semi-aquatic plants onto Fault Lake

Only one of the 12 young cattails transplanted into the shallow (~10 cm) transient pond at Fault Lake produced a juvenile plant.

The single *Phragmites* plant produced several short juveniles and it will be interesting to see whether these juveniles, as well as the cattail, have overwintered and are present in the spring, 1986.

The cattail seeds which were liberally sown on the shore-side of the transient pond did not appear to germinate. No young plants were evident on that side of the pond. It might be of interest to check the shore in spring 1986 for germinating cattail seeds.

4.0 DISCUSSION

Establishment of a wetlands is predicated on either successfully seeding the area of intended colonization, or transplanting propagules, i.e., root stocks, rhizomes, cuttings, etc., of the desired plants. The decision of which method to employ is necessarily based on the peculiarities of the plants of interest.

In particular, the availability of seeds and ease of germination, *in situ*, on the wetland site determines whether this most cost-effective method can be implemented. Some wetland plants produce few seeds and often these seeds are difficult to germinate, e.g., *Phragmites*. Some seeds may germinate under mild conditions but not in an environment of low acidity and/or high solute and metal loadings. Ironically, however, in such cases (e.g. cattails) the mature plant may be able to successfully cope with such an environment and reproduce within such a milieu by other means, i.e., rhizomes (underground stems).

The alternative to planting seeds involves labour intensive transplanting procedures, as noted above for *Phragmites*. Warburton et al. (1985) would include cattails as well, since there are no other satisfactory options. Once the latter plants have become established, they may spread rapidly via rhizomes which produce juvenile plants at their termini.

Two collections of semi-aquatic plants were attempted: August and September. The August collection was used in the present experiments as most of the plants from which seeds had been collected in August were devoid of seeds in September.

It is well known that seeds frequently require subjection to special sequences of physical (environmental) factors before they will germinate. Such factors included low (or freezing) temperatures, special photoperiod requirements, digestion of the outer seed coat, etc. The list of possible factors is a long one and we were only able to try three of the most common requirements: drying, freezing, and scarification, i.e., slicing open the seed coat. Our results showed that the best germination was obtained when the seeds were merely dried at room temperature.

The single rush from which seeds were collected had a very thick seed coat. Frequently seed germination can be stimulated in thick coated seeds if the coat is sliced open and water is allowed to penetrate into the seed embryo. Unfortunately, this technique was not effective in the case of the rush seeds. Unravelling the particular (unique) requirements of seed germination for a given species of plants can frequently involve prolonged experimentation (Bewely & Black, 1972 and 1978). Such a commitment was quite beyond the limited scope of the present investigation.

In the present study with semi-aquatic seeds we found two plants: Bromus sp. and Scirpus sp., whose seeds can be successfully germinated directly on pH 7.0 and 10.0-amended pyrrhotite. We were surprised to find that, while cattail seeds collected from plants growing in an acid creek germinated with great efficiency (~100%) in tap water. These acid seeds germinated with only about a 10% efficiency on pH-amended pyrrhotite.

No seeds germinated on unamended pyrrhotite (pH 2.3 - 2.5) or on pyrrhotite amended with sewage sludge. Thus, the key to germination of the semi-aquatic plants with which we experimented lies in pH control. Apart from an elevated H^+ concentration, it is possible that inhibition of germination at low pH may also result from an uptake of heavy metals, i.e., Cu, Ni, Zn. At elevated pH's, these metals are mostly insoluble and therefore are not present in the water which the seeds imbibe from the moist pyrrhotite.

The results of these laboratory investigations are directly applicable to the proposed installation of a wetlands on the pyrrhotite tailings at Fault Lake. If the tailings can be amended with sufficient lime to bring the pH to near neutrality, seeds of Bromus sp. and Scirpus sp. (both available from the Conservation Area) can be sewn on moist pyrrhotite. Cattails and Phragmites, on the other hand, will probably have to be planted by hand. It is known from our other work with cattails growing under acid conditions, that pH amendment of the pyrrhotite may not be necessary for their propagation.

5.0 RECOMMENDATIONS

Based on results of the RATS/Falconbridge/Boojum Research in the field and laboratory, together with information gained from the RATS/Inco/Boojum Research at Levack, the following recommendations are suggested for wetland development on the Fault Lake pyrrhotite tailings. The recommendations assume that the chief requirement for a wetlands, i.e., continuous supply and retention of water, can be met at the Fault Lake site.

(1) Seeds of *Bromus* sp. (a grass) and *Scirpus* sp. (a sedge) can be sown on pyrrhotite, amended to pH 7.0 with lime, and can be expected to grow. While cattail seeds also germinated on pH 7.0 - amended pyrrhotite, we were unable to grow these plants beyond the seedling stage. The following procedure (2, below) is recommended for cattail propagation on pyrrhotite, on the basis of our field experience at Levack and recommendations by Warburton et al. (1985).

(2) Mature cattail root stocks can be dug from the Conservation Area and planted directly into pyrrhotite which has not had any pH adjustment. It should be noted, however, that best growth of cattails will not occur under these acidic conditions. The plants will be short in stature (<1 m tall). Pyrrhotite amended to pH 7 or above will enable plants to achieve a more robust character (>2 m tall).

6.0 REFERENCES

Bewley, J.D. and M. Black, 1978. Physiology and biochemistry of seeds. (1) Development germination and growth. 306 pp. (2) Springer-Verlag; Berlin-Heidelberg-New York.

Bewley, J.D. and M. Black, 1982. Physiology and biochemistry of seeds. Viability, dormancy and environmental control. 375 pp. Springer-Verlag; Berlin-Heidelberg-New York.

Bonnewell, V., W.L. Koukkari and D.C. Pratt. 1983. Light, oxygen and temperature requirements for *Typha latifolia* seed germination. Can. J. Bot. 61: 1330-1336.

Brooks, R.P., D.E. Samuel and J.B. Hill, eds. Wetland development on coal mine slurry impoundments: principles, planning and practices. Proceed. Conf. October 1985. Penn State Univ., State College, Pa. VII + 393 pp.

Nawrot, J.R. 1985. Wetland development on coal mine slurry impoundments: principles, planning and practices. In (Brookes, R.P., D.E. Samuel and J.B. Hill, eds) Wetlands and water management on mined lands. Proceedings, Conf. October 1985. Penn State Univ., State College, Pa. p. 161-172.

Warburton, D.B., Klimstra, W.B. and Nawrot, J.R. 1985. Aquatic Macropyte Propagation and Planting Practices for Wetland Establishment. In (Brookes, R.P., D.E. Samuel and J.B. Hill, eds) Wetlands and water management on mined lands. Proceedings, Conf. October 1985. Penn State Univ., State College, Pa. p. 161-172.

APPENDIX

PROGRESS REPORTS

Boojum RESEARCH LIMITED

SUMMARY OF ECOLOGICAL WORK

The objectives of the ecological work were:

1. Identification of suitable seed-root material for transplanting into the experimental cells.

2. Development of a methodoloy for the transfer of plant material from the source site to the tailings environment.

3. Test the effectiveness of ameliorating materials for experimental cells.

Five progress reports have been submitted to Falconbridge reporting on the details of the work carried out and the results achieved.

1st Progress Report, July 1985: Several versions of constructing experimental test cells were tried. It was concluded that it would be difficult to keep water on the pyrrhotite without amendments which are expected to seal the surface.

2nd Progress Report, September 1985: Exploratory transplanting experiments were implemented on site in a section of Fault Lake where water was retained over slimes. Potential results might only be expected in 1986. The construction of the experimental test cells was delayed and planned transplanting experiments could not be implemented.

Therefore emphasis was placed on applicable laboratory work. Seeds for germination experiments were collected and static leaching experiments in the laboratory were carried out to determine metal concentrations in stagnant water over pyrrhotite.

3rd, 4th and 5th Progress Reports October, November 1985 and January 1986: We addressed germination and seedling experiments with different experimental methods and have repeated selected experiments. The results are very encouraging since a relationship between the seed source (acid or alkaline) and the ability of the seeds to germinate has been confirmed.

We have identified seed sources with a high germination rate which furthermore produced robust seedlings. The ongoing work on establishment of seedlings on amended pyrrhotite should lead to results which could be tested in the experimental cells in 1986.

Transplanting methods have been developed for alkaline areas and acid areas during the Levack study. Although field experiments were not possible on the Fault Lake site, experience gained from the Inco site will benefit the introduction of wetland vegetation to the experimental cells in the next season.

Germination, establishment and transplanting experiments of wetland vegetation will be the main thrust of the 1986 work.

130 AMELIA STREET TORONTO ONTARIO M4X-1E6 1-416-963-0490

AN EXPERIMENTAL APPROACH TO BUILDING A BOG OR A WETLAND

START UP REPORT , JULY 1985

BY M.KALIN

FOR B.MICHELUTTI

FALCONBRIDGE LTD.

1.0 Introduction

Boojum Research Limited, within the framework of the RATS program, has initiated some preliminary field experiments pursuant to the objectives of establishing a wetland on the Fault Lake tailings. The following brief report describes the work carried out on 24 July 1985 by M. Kalin and R. Buggeln of Boojum Research Ltd., with assistance of Ted from Falconbridge Ltd.

2.0 Assessment of containers for growing aquatic plants on site

Falconbridge provided 5, 18′ (dia) x approx. 2-1/2" (long) thick-walled ABS pipe for use as containers for growing aquatic plants on the Fault Lake tailings. We envisioned sinking these pipes into the pyrrhotite and/or mine slime tailings and then filling the pipes to the desired level with tailings and water for experiments with aquatic plants.

2.1 Water percolation tests through mine slimes and pyrotite

2.1.1 Mine slimes

Three holes, 16-18" deep, were dug in the mine slime covered (East?) end of the Fault Lake tailings area. Two holes were close to the road, the third hole was close to the "transient" pond, formed from run-off water. Stakes with 1" graduations marked on them were driven into the holes. The holes nearest the road were filled with about 30 gallons of water; the pond-side hole with about 15 gallons. Loss of water from the first two holes began immediately. By the end of the day all holes were dry (period of 4-5 hrs). In comparison, the hole by the pond lost the water slower.

2.1.2 Pyrrhotite

One end of ABS pipe was coated with a thick layer of black mastic (roofing tar) and then placed tar end down on a flat area of solid (1/2 - 3/4", flat, shale-like bits) pyrrhotite. The tar was expected to seal the innterface between the pyrotite and the pipe. The pipe was carefully filled half way with broken pyrrhotite and then water was added to a level of about 6" above the pyrrhotite surface. Almost immediately, a rim of moist pyrrhotite appeared on the tailings as a perimeter around the pipe. As the tar seal was reasonably tight between the tar and pyrrhotite, saturated pyrrhotite remained in the ABS pipe. In 3 hours all of the water in the pipe had percolated into the tailings.

In a second experiment, we attempted to make a water tight "flower pot" by coating one inch of ABS pipe with tar, as above, and setting it tar side down on a piece of black polyethylene. An additional ring of tar was placed on the inside of the pipe to improve the seal. The pipe was then filled with pyrrhotite and water, as above. Unfortunately, the tar seal did not hold and numerous leaks soon appeared. Other flat materials, e.g. heavy gauge roofing felt, plywood, etc., were not tried out since all these would require additional labour.

2.1.3 Conclusions

1) The broken (1/2 - 3/4", flat, shale-like bits) pyrrhotite on the tailings is readily perimeable to water. Considerable thought must be given to the engineering techniques which will render the pyrrhotite area on the Fault Lake tailings less water permeable.

2) The mine slime region of the tailings appears to have greater water retention characteristics than the pyrrhotite. The transient pond may be expected to vary in size with rainfall, spring thaw, etc. There was some water retention in the hole dug near the edge of the pond - the latter now measures approx. 30′ x 80′ and with about 4-6" of standing water in it, indicating a subsurface "water table" in this areas (at least around the pond perimeter). The lateral extent, i.e. distance from the pond, of this water table should be determined.

3) The pH of the water in the "transient" was 3.5. Laboratory experiments with slurries (1:4 w:v) of slimes (or pyrrhotite) water showed a pH range of 7.1 - 7.6 for the slimes and 2.7 - 2.9 for the pyrrhotite. The acidity in the transient pond is therefore derived from rainwater run-off and/or through the pyrrhotite.

4) Alternative containers for the growth experiments with aquatic plants must be sought.

RATS/FALCONBRIDGE/BOOJUM

SECOND PROGRESS REPORT

ON

THE FAULT LAKE STUDY

PREPARED FOR

BOB MICHELUTTI

FALCONBRIDGE

BY

M. KALIN

SEPTEMBER 1985

OVERVIEW

1.0 INTRODUCTION

The Fault Lake study site at Falconb ridge is the central focus of the RATS work conducted by Boojum Ltd., Toronto, Ontario. The following report briefly outlines the progress of work initiated earlier: results of some laboratory experiments and field observations.

2.0 FAULT LAKE TRANSPLANTED BIOTA

On September 11, 1985, a visit was made to Fault Lake to check on the survival of the transplanted cattails and Phragmites. Two of the cattail plants (out of the initial dozen) are producing juvenile shoots but all of the parent plants are now dead. The Phragmites plant, although dead, also produced a juvenile shoot from its base.

The transient pond on the mine slimes has virtually dried up, although the mine slimes themselves appear to be water saturated.

The moss covering on the mine slimes (south side of pond) is very healthy with some evidence of growth into the so-called "gametophyte" form.

3.0 METAL LEACHING FROM MINE SLIMES AND PYRRHOTITE

The following experiment was conducted to determine the rate of dissolution of selected metals, present in Fault Lake pyrrhotite and mine slimes, into the water. 100 g of the solids were covered with 400 ml of tap water and either replaced with fresh tap water 3 times during the 38 day experiment (conducted at room temperature), or not replaced. There were three replicates for each treatment. The first treatment is referred to as "sequential leaching" and the second treatment, "continuous, long-term" leaching. Any evaporated water, however, was replaced under both conditions. pH and conductivity were measured periodically in all containers. Water samples from the three replicate containers were pooled for metal analyses. The pooled sample was split in half with one portion immediately acidified and the second portion vacuum-filtered through a membrane filter (0.45 um pore size) before acidification.

The results are shown in Figs. 1-4 and Tables 1-2.

3.1 Conductivity and pH

3.1.1 Sequential and continuous, long-term leaching

Figures 1-2, and 3-4 show the conductivity and pH, respectively, of the water overlying the mine slimes and pyrrhotite. It is readily seen that there is little change in either parameter after the water has been in place over the materials for a few days. Conductivities are expectedly higher in the pyrrhotite water than in the mine slime water.

Figure 1 gives the electrical conductivities of leach water in the sequential leaching experiment while Figure 2 shows the conductivity profile in the continuous, long-term leaching experiment. In general, the short-term data for the mine slimes match the continuous leaching data while those sequential leaching data for the pyrrhotite do not match, i.e., cannot be superimposed on the continuous leaching experimental data. It would appear that renewal of the leaching water removes fewer el ectrolytes from the pyrrhotite over time than are lost from the mine slimes over a similar time course. This result is possibly due to the coarseness of the shale-like pyrrhotite, and a small surface to volume ratio, as compared with the flour-like consistency of the mine slimes, i.e. with a large surface to volume ratio.

Figure 3 compares the changes in pH in leach water in the sequential leaching experiment for pyrrhotite and mine slimes. In the former water, the pH decreases from about 5-6 to about 3 within 48 hours. For mine slimes, there is an increase in pH from about 6.5 to 7.5 - again within 48 hours. The results in the long term, continuous leaching experiments (Figure 4) do not substantially differ from those shown in Figure 3.

3.1.2 Metal content of the leachates

Table 1 shows the metal content (total Fe, Ni and Cu) leached into the water from the pyrrhotite and mine slimes in the two treatments: (1) where the water was replaced 3 times, and (2) where the materials were initially covered with water and additions only made to replace that lost by evaporation.

Considering the pyrrhotite samples first, there was no difference between filtered and unfiltered water. There was more Cu and Ni leached from the material in the sequential leaching series, i.e., the sum of the metal concentrations for the 3 separate leach waters than in the containers with the long-term non-renewed water. There was about 25% more Ni and 100% more Cu in the sequential leachate than in the single, long term containers. The total iron content showed the opposite result for as yet unexplained reasons.

The mine slimes showed order of magnitude differences between unfiltered and filtered samples, with the former samples exhibiting the greatest differences. The greatest disparities appeared in the sequential leaching series for Fe and Cu and the long-term series for Fe. Ten to 100 times more metal was leached from the mine slimes in the sequential leachings than in the long-term ones.

3.1.3 Metal content of pyrrhotite and mine slimes

Table 2 gives the metal content in the pyrrhotite and mine slime solids as percent of the dry solid matter. Regardless of the treatment, sequential or long-term leaching, there were essentially no differences in the residual metal concentrations of the solids following the 38 day leaching experiments with the pyrrhotite and mine slimes. The former contained about 3 times the amount of Fe as the latter while the mine slimes contained about twice the Ni content of the pyrrhotite; Cu was essentially identical in each.

The amount of Fe dissolved from the pyrrhotite during the experiment was on the order of 0.05%; the dissolution of Ni from the mine slimes was on the order of 0.3% of that present in the sediments.

4.0 AQUATIC AND SEMI-AQUATIC SEED SSOURCES

Seeds from cattails, sedges and rushes were collected at the Conservation Area. They were air-dried and stored in paper bags for subsequent germination experiments this winter in the laboratory.

TABLE 1

Metal content in water covering pyrrhotite and mine slimes (ppm) in sequential and long-term leaching experiments

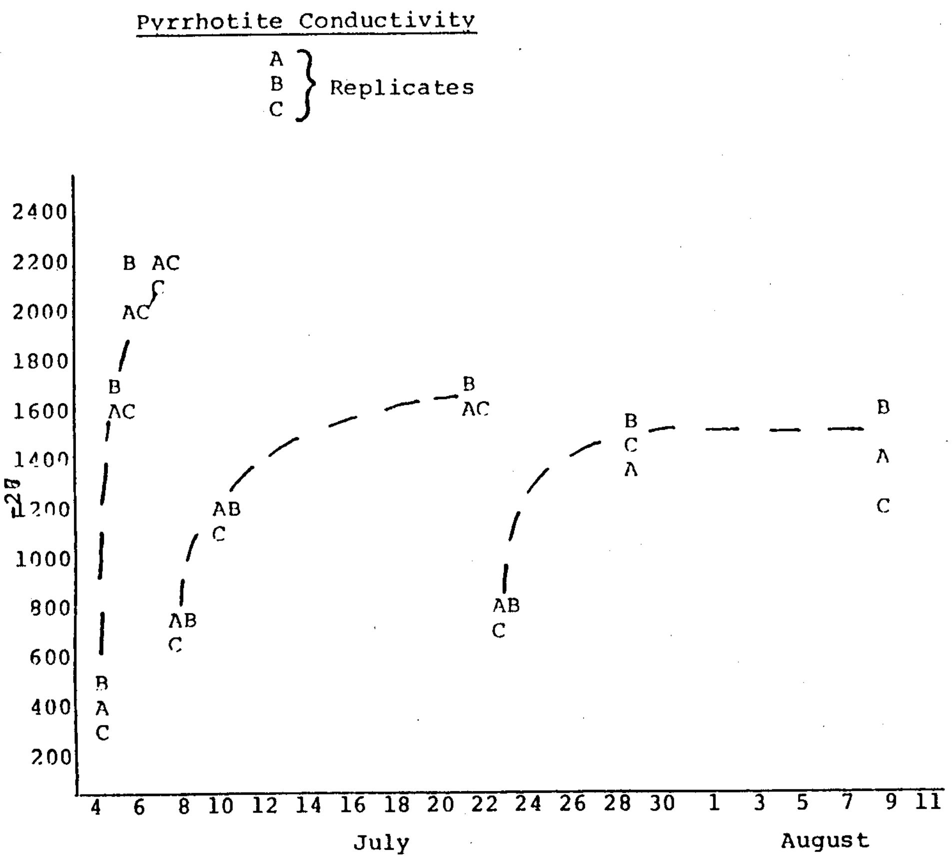
Metal		08 July* Filt. ppm	08 July* Unfilt. ppm	23 July Filt. ppm	23 July Unfilt. ppm	09 August Filt. ppm	09 August Unfilt. ppm	Total Metal conc. ppm	
pyrrhotite									
Fe	sequential	42.2	42.0	115	115	85	87.5	242	244.5
	long-term					344	375	344	375
Ni	sequential	12.1	12.1	2.9	2.93	1.49	1.49	16.5	16.5
	long-term							12.0	12.0
Cu	sequential	1.62	1.55	0.29	0.29	0.08	0.08	2.0	1.9
	long-term							0.93	0.84
mine slimes									
Fe	sequential	0.06	318	0.93	1.92	0.20	23.1	1.2	343
	long-term							0.31	0.42
Ni	sequential	2.11	12.5	0.29	0.33	0.18	2.05	2.0	14.9
	long-term							0.37	0.42
Cu	sequential	0.03	1.92	0.29	0.03	0.02	0.31	0.34	2.26
	long-term							0.02	0.03

* Experiment began on 3 July 1985

TABLE 2

Metal content in pyrrhotite and mine slime solids (% dry wt)

Metal		07 July	09 August
Pyrrhotite			
Fe	sequential	37.0	37.0
	long-term		37.2
Ni	sequential	0.19	0.17
	long-term		0.19
Cu	sequential	0.07	0.07
	long-term		0.07
Mine slimes			
Fe	sequential	13.8	14.0
	long-term		14.2
Ni	sequential	0.36	0.37
	long-term		0.30
Cu	sequential	0.09	0.08
	long-term		0.08



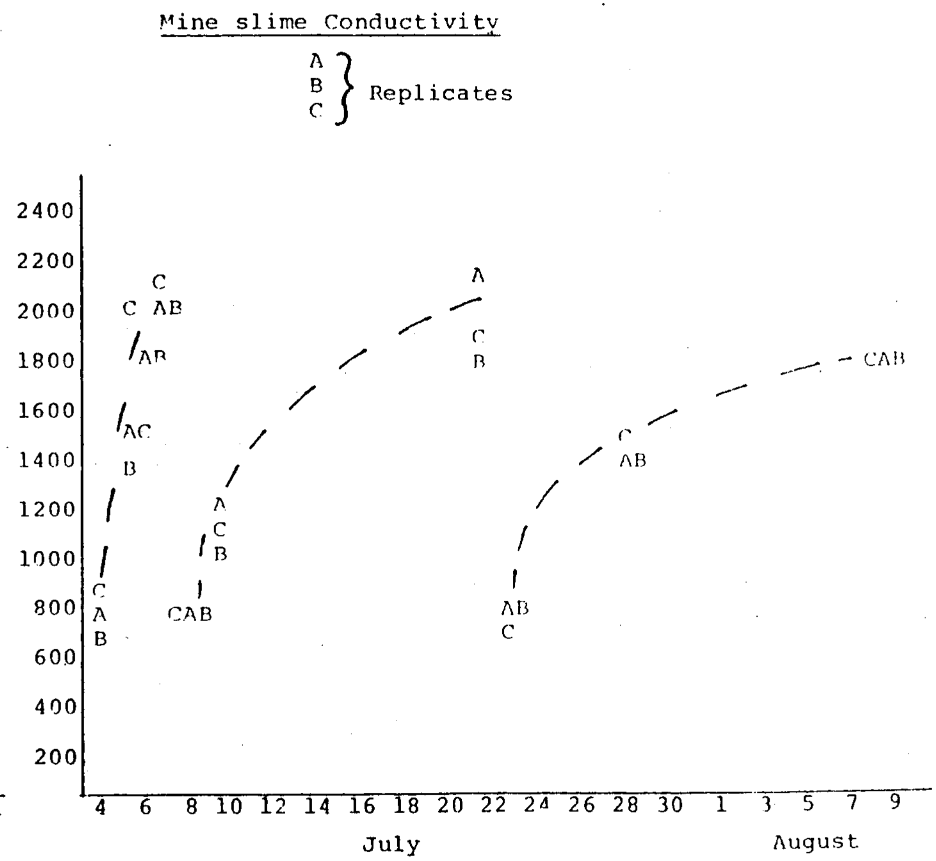


Figure 1 - Sequential leaching from pyrrhotite and mine slimes - conductivity uohm . cm^{-1}

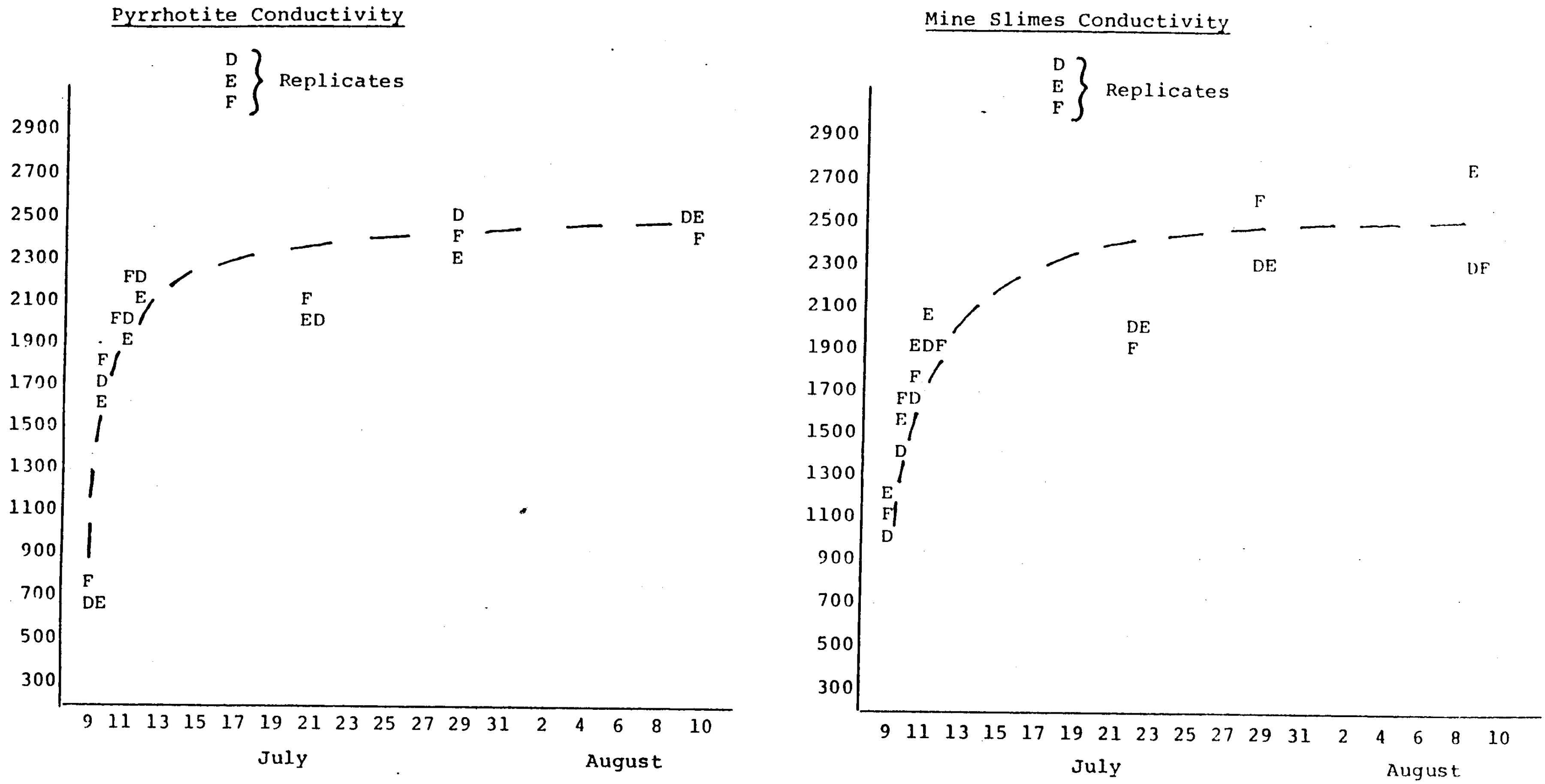


Figure 2 - Long term continuous leaching from pyrrhotite and mine slimes - conductivity uohms.cm^{-1}

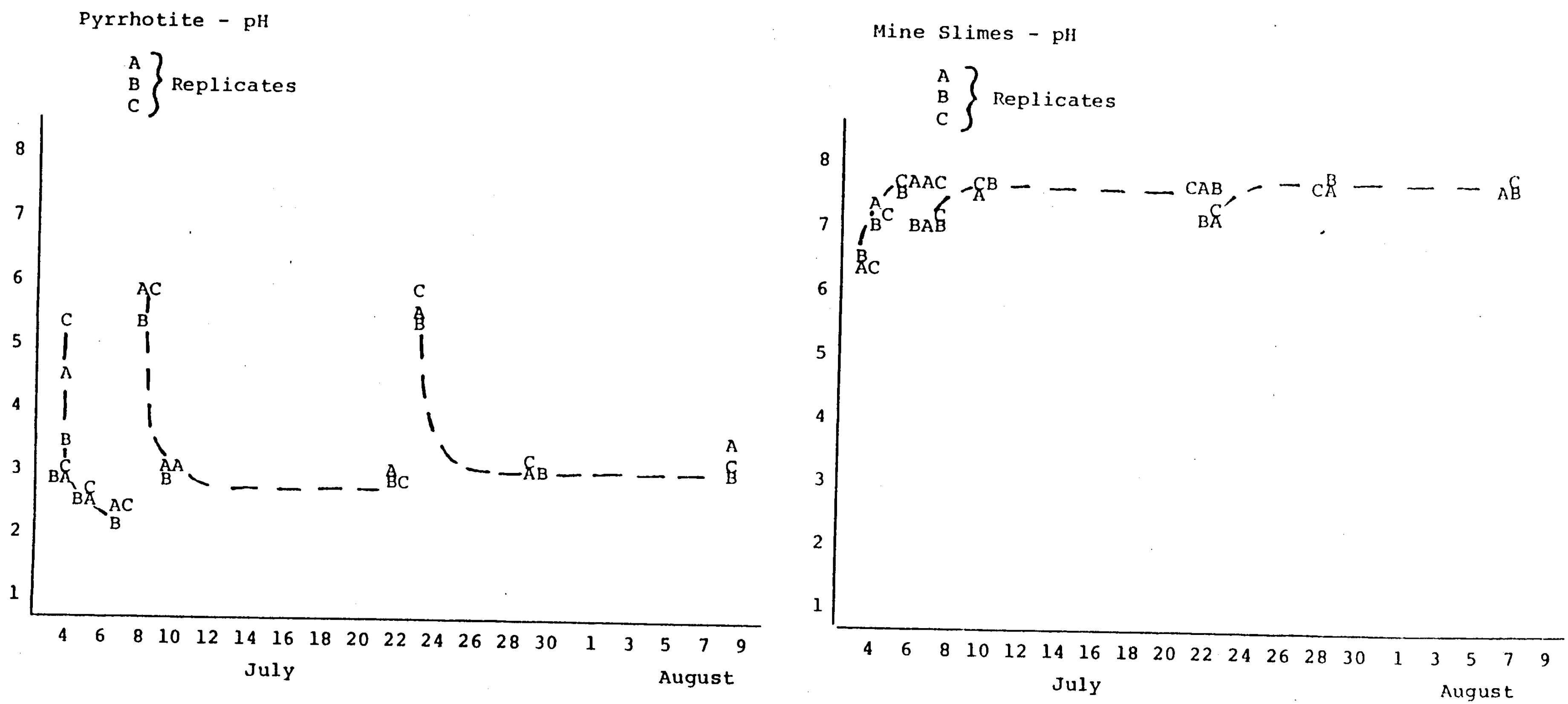


Figure 3 - Sequential leaching from pyrrhotite and mine slimes - pH

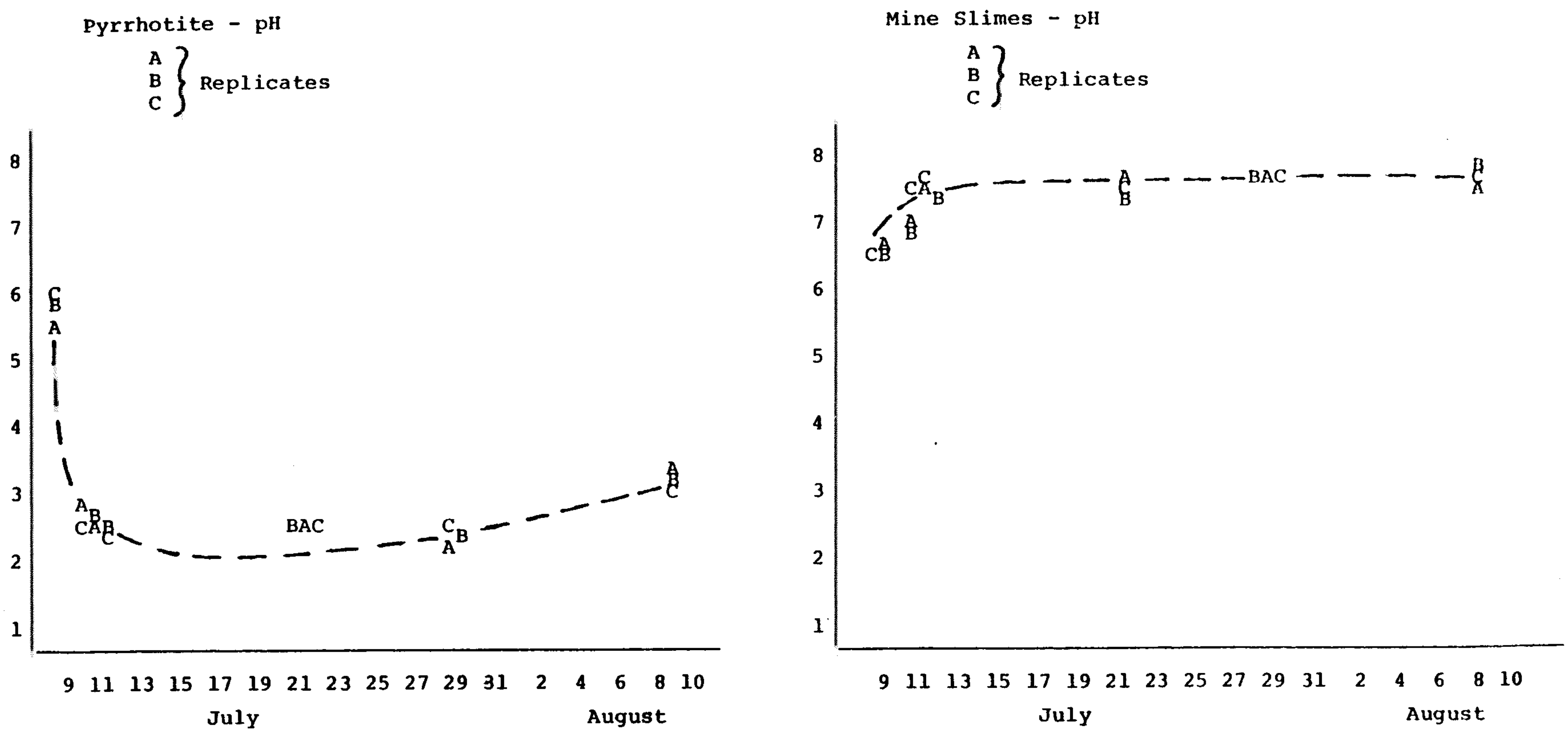


Figure 4 - Long term continuous leaching from mine slimes and pyrrhotite - pH

Boojum
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RATS/FALCONBRIDGE/BOOJUM

THIRD PROGRESS REPORT

ON

THE FAULT LAKE STUDY

PREPARED FOR

BOB MICHELUTTI

FALCONBRIDGE

BY

M. KALIN

NOVEMBER 1985

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Third Progress Report
Falconbridge/RATS/Boojum

1.0 INTRODUCTION

The major goal of ecological engineering, as promulgated by Boojum Research Ltd. with respect to the Fault Lake site, is the establishment of a wetland of semi-aquatic plant species on that site. Part of this task requires that we develop an understanding of the factors effecting both seed germination, and seedling establishment, of potentially suitable semi-aquatic plants. Experiments designed to obtain this information are being conducted at Boojum Research and will be ongoing for the next few months.

Emphasis has been placed on this aspect of Ecological Engineering because difficulties have been encountered by Falconbridge in preparing the field sites, i.e., study cells in Fault Lake. These cells were to have been used in the in situ testing of the overwintering ability of selected semi-aquatic plants. Our present focus on seed germination and seedling growth is directly relevant to the perpetuation of a wetland through the recruitment, establishment and persistence of siblings of semi-aquatic plants.

2.0 SEED COLLECTION

We have collected cattail seeds (both Typha latifolia and T. angustifolia) from a number of locations. A single collection of the latter species is from East Mine at Falconbridge. There are 4 collections of the former species from acidic, alkaline, high Ni (Strathcona area, Falconbridge, Onaping) and control locations. The question to be answered here is whether seeds from all of these sources are equally viable. In addition, we have collected seeds of a sedge, a rush (Juncus sp.) and reed grass (Phragmites communis); all of these are from the Falconbridge Conservation Area.

Inasmuch as the seasonal seed ripening patterns of these plants are unknown, several collections (late August & mid September) of sedge, rush and Phragmites seeds were made in an attempt to insure that we have viable seeds in our possession.

3.0 EXPERIMENTAL PLAN

The following outline, and accompanying Table 1, explains the experimental approaches which will be implemented by Boojum Research Ltd.

TABLE 1

EXPERIMENTAL PLAN FOR EACH SEED TYPE

EXPERIMENTAL PHASE	pH EFFECTS	PYRRHOTITE (+/- AMENDMENTS)
I Pre-germination seed treatment	-	-
II Seed germination		
Phase 1	x	-
Phase 2	-	x
III Seedling establishment	-	x

3.1 Pre-Germination Treatments

The seeds are being subjected to 3 pre-germination procedures. One group of seeds will continue to be stored in a dry condition at room temperature. Another group have been stored in a dessicator, again at room temperature, while a third group have been frozen.

3.2 Seed Germination Study

3.2.1 General Methodology

15-20 pre-treated seeds will be germinated at room temperature by pressing them into a moistened piece of filter paper, placing the filter paper seed-side down in a Petri plate and adding water to a depth of 5 mm. Emergence of the root from the seed is traditionally scored as seed germination and petri plates will be observed every few days until a standardized procedure can be developed for each plant species. We already know for instance, that 7 days is an acceptable incubation time period for our control cattail seeds. All treatments will be run in duplicate.

3.2.2 Effects of pH on Seed Germination

In these experiments, the seeds will be germinated under conditions in which the pH of the moistened filter paper is either 2.5 (3.0), 7.0 or 10.0.

3.2.3 Germination On Pyrrhotite Leachate (Plus Amendments)

In these experiments, a layer of crushed pyrrhotite (or pyrrhotite plus amendments such as lime or sewage sludge) will be placed in the bottom of Petri plates before adding the moistened filter paper "sandwich" containing the seeds and a small volume of water, sufficient to keep the filter paper covered with water.

3.3 Seedling Establishment and Growth

In this phase of the work, germinated seedlings from section 3.2.3 above, will continue to be observed after they are transferred from the filter paper directly to the underlying surface of the tailings surface (unamended or amended pyrrhotite). The water level will be maintained such that the seedlings are covered by no more than 5 mm of water so that gas exchange for photosynthesis and respiration will not be reduced by diffusion of gases through water.

3.4 Adjustments to Experimental Procedures

Depending on the various successes of the above experimental protocols, certain aspects may be changed, augmented, etc., as time progresses.

Boojum Research Limited

RATS/FALCONBRIDGE/BOOJUM

FOURTH PROGRESS REPORT

ON

THE FAULT LAKE STUDY

Prepared for

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By

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November 1985

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PROGRESS REPORT # 4

1.0 INTRODUCTION

Experiments, as outlined in Progress Report November 1985 are being conducted in the laboratory on semi-aquatic plant sees germination and seedling growth on amended and unamended pyrrhotite. The experimantal plan as laid out in Table 1 of the third progress report has been partially completed. Results of Phase I and part of Phase II are presented below, along with a description of on-going experiments.

2.0 RESULTS

2.1 Experimental Phase I - Pre-germination Seed Treatment

Seeds were maintained at room temperature (stored inside or outside a dessicator) or were frozen (2 Months). The germination test was set up following the procedure outlined in Section 3.2.1 of the third progress report. The results of the pre-germination treatments on seed germination are presented in Table 1. Cattail seeds collected from plants growing in alkaline, acid and control sites were held at room temperature. Bonnewell et al (1983) reported no special temperature or drying pre-treatment for cattail seeds.

TABLE I

Effect of Pre-germination Protocols on Seed Germination

	Room Temp		Room Temp In Dessicator		Frozen	
	2 Wks	3 Wks	2 Wks	3 Wks	2 Wks	3 Wks
SEED TYPE						
Sedge Species #1	1/104 (0.9%)	1/104 (0.9%)	2/78 (2.6%)	2/78 (2.6%)	1/27 (3.7%)	1/27 (3.7%)
Sedge Species #2	0/15	0/15	0/23	5/23 (21.7%)	0/17	0/17
Sedge Species #3	0/12	1/12 (0.8%)	0/12	0/12	0/10	0/10
Rush Species #1	0/16	0/16	0/14	0/14	0/12	0/12
Grass #1	6/24 (25%)	6/24 (25%)	3/36 (8%)	3/26 (8%)	0/24	0/24
CATTAILS						
Acid	11/29 (38%)	-	-	-	-	-
Alkaline	4/86 (4.7%)	-	-	-	-	-
Control	9/72 (12.5%)	-	-	-	-	-

A very small but persistent germination % occurred in sedge species #1 in all treatments. A sizeable number of seeds from sedge species #2 (dessicator stored) germinated after 3 weeks. Grass species #1 also showed good germination % in both room temperature groups. Germination of the cattail seeds is somewhat surprising in that the seeds from "environmentally stressed" populations growing in alkaline mine slimes and in an acid creek (pH 2.3) showed a higher percentage of germination than did seeds from the control population.

Rush seeds, with a thick seed coat, showed no germination after three weeks. The seeds were then 'scarified', a technique whereby the seed coat is carefully sliced along one edge in order to permit water entry into the seed. After one week, the scarified seeds had still not germinated.

2.2 Experimental Phase II - Seed Germination: pH and Pyrrhotite Amendment Effects

As noted in Section 3.2 of the third progress report, the effect of pH and both amended and unamended pyrrhotite on seed germination was going to be studied.

2.2.1 Unamended Pyrrhotite (pH 2.2)

Cattail seeds from plants growing in alkaline, acid and control sites were set up in germination experiments on pyrrhotite. No No seeds had germinated after two weeks.

The results are interesting as it is known from field studies that cattails growing on alkaline mine slimes (pH 9.5 - 10) can be grown hydroponically in acidic water (pH 2.5). The seeds used in the experiments originated from a population in a creek running over pyrrhotite. Wether the cattails had germinated in the creek is not known.

2.2.2 Amended Pyrrhotite

Experiments are currently underway to investigate the germination of control cattail seeds in pyrrhotite amended with sufficient $Ca(OH)_2$ to bring the pH of the overlying aqueous phase to 2.9, 6.9 and 10.2.

Six percent dewatered sewage sludge was obtained from the Humber Waste Treatment Plant (City of Toronto). The sludge was autoclaved (20 minutes, 120° C) prior to use. A ratio of 10 ml of sludge to 6 grams (DW) pyrrhotite (pH 2.5) was used for other germination experiments.

2.3 Experimental Phase III - Seedling Establishment

As noted in Section 3.3 of the third progress report, seedling establishment in pyrrhotite (amended or unamended) will be studied as an follow-up to the seed germination experiments.

2.3.1 Unamended Pyrrhotite (pH 2.2)

Week-old cattail seedlings (about 1 cm tall from Phase I above) were planted (submerged) in a small dish containing pyrrhotite and water 5 cm deep. Within two days, the seedlings were bleached white.

The water was decanted leaving a residual depth of about 2 mm. (grass # 1 seedlings from Phase 1 above) were planted in the moistened pyrrhotite. The leaves of those seedlings touching the side of the dish (for support) turned black within two days. Seedlings which remained erect without touching the acidic water were brown after one week.

These experiments will be repeated with cattails and other seedlings, ensuring that each seedling is self-supported in the pyrrhotite sediments.

3.0 Comments

As noted above in Section 2.2.1, we have found some interesting information on cattails - their undeniable presence in an acid creek yet the inability of their seeds to germinate under acidic conditions.

While we know that we can successfully transplant mature cattails into the acid creek, it will be interesting to see whether we can successfully transplant cattail seedlings into moistened pyrrhotite.

RATS/FALCONBRIDGE/BOOJUM

FIFTH PROGRESS REPORT

ON

THE FAULT LAKE STUDY

PREPARED FOR

BOB MICHELUTTI

FALCONBRIDGE

BY

M. KALIN

JANUARY 1986

1.0 INTRODUCTION

Experiments are continuing on semi-aquatic plant seed germination and seedling growth on amended and unamended pyrrhotite.

The experimental plan, as laid out in Table 1 of the Third Progress Report, has been partially completed and experiments in Phase I (Pre-germination seed treatment) and Phase II (Seed germination: pH and pyrrhotite amendment effects) are currently being repeated.

Experiments, as outlined in Phase III (Seedling establishment), have been partially completed and are being repeated.

Some interesting results have come to light from our recent investigations and these will be discussed in detail below.

2.0 RESULTS

2.1 Experiment Phase I - Pre-germination seed treatment

The experiment reported in Progress Report #4 (Table 1) was repeated and substantially similar results were achieved (after one week).

Again we found that Grass species #1 showed good germination regardless of the pre-germination conditions: (a) room temperature; (b) room temperature in dessication; or (c) frozen. As before, the next best performance was by sedge species #1; seeds of the two remaining sedge species and one rush species did not germinate.

Of particular interest were the repeats of the acid, alkaline and control cattail seed germination experiments (see Table 1). A superior germination performance occurred with seeds obtained from plants growing in an acid creek (pH 2.8). The seeds from plants growing on alkaline mine slimes (pH 8-10) showed the next best performance, while the control cattail seeds, collected from plants growing near a small infrequently travelled gravel road showed the poor germination.

Coincident with the germination results was the comparative robustness of the seedlings, i.e., length and thickness of the leaves: acid > alkaline >> control. The stature of the seedlings was later reflected in their ability to adapt to sand culturing (see Section 2.3 below).