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THE BIOLOGY AND CONTROL OF EHRHARTA VILLOSA, SOUTH AFRICAN PYPGRASS.

A thesis presented in partial fulfilment of the requirements for the degree of Master of Science in Plant Biology at Massey University.

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#### ABSTRACT

The biology of an adventive weed, Ehrharta villosa (F. Schult) was investigated at Turakina beach, Rangitikei, in order to understand its means of spread and to discover a way of controlling the plant. Two populations have been found at Turakina, the smaller being on the dune system and the other in a nearby pine plantation.

Studies of the seed biology showed that although viability was high (ca. 80%), the numbers of seed produced per  $m^2$  was low (8 to  $102 \text{ seeds/m}^2$ ). Dispersal of these seeds was limited to within 5m of the source population and no seeds were discovered in the soil seed bank. Circumstantial evidence suggests seed predation to be a contributing factor to this. Germination tests in sand showed seeds were capable of emerging from depths of up to 6cm.

Stages in seedling development were described. Pypgrass displays a tall habit with internodes which elongate soon after germination, but its seedlings are less vigorous than most other grasses and weeds. The morphology and growth pattern of pypgrass allows it to have a smothering effect on other vegetation.

The potential for vegetative spread by rhizome fragments was investigated by burial and reexcavation of fragments. Pypgrass is capable of reproduction from rhizome fragments of varying lengths. Mapping a 400m<sup>2</sup> area of the advancing front of one population showed that over one year pypgrass had spread between 4.1 and 9.0m. Species associated with pypgrass were recorded at

the beginning and the end of the study to give some indication of the effect of pypgrass density on those associated species. In quadrats where pypgrass was most dense, fewer species overall were found.

Dune species including pypgrass were tested for the presence of mycorrhizal fungi. Pypgrass proved to have the greatest percentage mycorrhizal infection (88.9%) with the other species having significantly lower percent infection. Mycorrhizal association may give an advantage to pypgrass by allowing greater uptake of water and minerals compared with other plants.

Leaf anatomical studies confirmed pypgrass is a C3 plant and other features such as sunken stomata and inrolled leaves may be of adaptive value in a coastal dune habitat.

Different methods for control of pypgrass were considered and it was decided that herbicide was the best option, because of the large area involved and the nature of the underground rhizomes. Field trials were used to evaluate haloxyfop for control of pypgrass. A single application did not completely control pypgrass, regardless of time of application. Two, split, applications also did not achieve complete control of leaf and rhizome, however split applications ensured tiller regeneration remained low throughout the trial.

Haloxyfop can generally be used selectively among dicotyledonous plants and monocotyledons that are not in the family Poaceae, but

it can harm some of these monocotyledons. Pot trials on dune monocotyledons associated with pypgrass demonstrated that marram was the only species significantly affected by haloxyfop, and even this plant was not completely killed. The trial established that haloxyfop would not adversely affect any native monocotyledonous plants growing in the area of pypgrass.

This study has gathered the necessary information to decide on a course of action. Pypgrass is at present confined to the Turakina area in two discrete populations. Use of herbicide (haloxyfop) in a number of split applications would prevent regrowth from rhizomes. Regeneration of pypgrass by seed after herbicidal control is not likely, allowing eradication to be an achievable aim.

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#### 1. INTRODUCTION

Ehrharta villosa (F.Schult) (pypgrass) was introduced to New Zealand in trial plots to stabilise the sandhills at Turakina Beach. It has spread from the site of its initial planting and is now well established in the Koitiata Domain reserve. A second population of pypgrass has been found in a nearby pine forest about 1km away.

Pypgrass in New Zealand grows as a dense sward which appears to choke out other dune plants to form a virtual monoculture (Plate 1.1). It appears that pypgrass grows more as an open herb in its native country, South Africa, allowing other dune plants to grow through it (Prof. A. MacLachlin, pers.comm).

In summer pypgrass has very little leaf tissue, whereas in winter it produces much more foliage (Plates 1.2 and 1.3). Pypgrass can climb up other plants to a height of at least two metres (Plate 1.4), and appears to climb by bending its stems at the nodes and loosely twisting itself around the support (pers.obs).

The Koitiata reserve has been recommended by the Department of Conservation (DOC) to be a protected natural area, due to the number and conservation value of the native dune plants contained within it. However, pypgrass has established as a dense sward and appears to be choking out most of these plants. Forest, river and urban boundaries protect the reserve from the devastating effects of introduced mammals, therefore, the reserve



Plate 1.1 Pypgrass creates a virtual monoculture on the dunes at Turakina Beach.



Plate 1.2 Pypgrass in summer. The leaves brown off leaving pypgrass with very little leaf tissue.



Plate 1.3 Pypgrass in winter. Compared to summer, pypgrass now has relatively lush foliage.

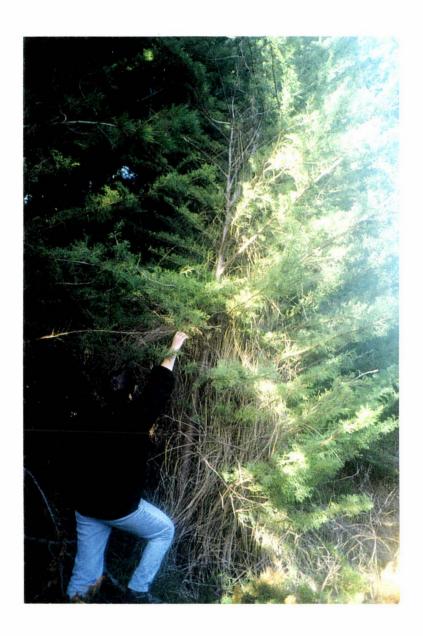


Plate 1.4 Pypgrass climbing in a macrocarpa tree. It is reaching a height of at least two metres here.

could only benefit from the removal of a weed with such an aggressive nature.

It may be possible to eradicate pypgrass because of its confinement to the Turakina area. Through the findings of this study, pypgrass spread may be halted before the Koitiata reserve is decimated further, and the area loses its many precious dune species.

The methods that this study employed to meet this objective were headed by two aims. The first and perhaps most important was to discover the best method of control of pypgrass at Turakina. If eradication is to be achieved, pypgrass must be killed and regeneration must not occur. The other important aim was to study the biology of pypgrass to discover what makes it so successful at Turakina and what is its means of spread.

A study of the seed biology was undertaken to determine its potential to spread by seeds. The study included counts of numbers of seeds produced, numbers dispersed away from source plants, numbers incorporated into the soil seed bank and the germination capabilities of the seed. Langer (1979), also working on grass species stated that a measure of seed production involves firstly, numbers of seeds produced per infloresence and secondly, the number of fertile tillers per unit area.

An examination of seed dispersal starts with an inspection of Pypgrass seed morphology to determine the dispersal mechanism for the seed. A seed trap is useful for determining the pattern of deposition over a set time and space (Werner, 1975). Johnson & West (1988) evaluated different seed traps, and it was from this work that a design was chosen to be used in the Turakina dune environment.

Seed bank studies have been carried out in lacustrine dune systems (Leck et al, 1989; Mack, 1976; Watkinson, 1978). However these workers have failed to find significant dune seed banks. The present study evaluated the seed bank of pypgrass.

The seeds of a sand dune plant may germinate under a great depth of sand. An investigation into the capabilities of pypgrass seed to germinate from different depths of burial will also allow an examination of the features of the seedling growth stages of pypgrass.

The potential of pypgrass to regenerate from rhizome fragments was also investigated. Rhizome fragments of many grass species can produce shoots and roots following detachment from the parent plant.

Morphological followed stages in seedling growth. Leaf anatomy of pypgrass growing at Turakina can be compared with a previously published description of pypgrass leaf anatomy (Ellis, 1987). Studies of leaf anatomy may reveal adaptations of the plant that allow it to succeed in its environment.

The rate of vegetative spread of pypgrass over a one year period was assessed by mapping an area at the advancing front of the population. Associated species in the area were also recorded.

Recent literature suggests that mycorrhiza have an important role to play in the success of sand dune plants, as mycorrhizal plants have been shown to be tolerant of drought and saline soils (Rozema et al, 1986; Jackson & Mason, 1984; Ernst et al, 1984). Dune species including pypgrass were tested for the presence of mycorrhizal fungi, since it could be an important factor in the vigorous growth of the grass compared to that of other dune plants.

Use of translocated herbicides is traditionally the best way of controlling perennial weeds. They are required for control of underground parts of a plant, such as the extensive rhizome system in pypgrass. Compared with physical and biological control techniques they offer the best chance of eradicating pypgrass from the area. Herbicide trials were undertaken on pypgrass in the field as part of the present study. Pot trials were used to evaluate the effect of herbicide on other dune monocotyledons in the area.

## 2. PYPGRASS BIOLOGY

#### 2.1 DESCRIPTION

Schult. f. (family Poaceae, subfamily Ehrharta villosa Bambusoideae, tribe Ehrharteae) is a perennial, rhizomatous grass native to Southern Africa. Τt can be differentiated morphologically from other Ehrharta species by very large, profusely hairy, bearded and aristate spikelets, and by a suffrutescent habit, with culms woody at the base and with reduced leaf blades (Gibbs Russell, 1987).

The leaf anatomy is distinguished from other *Ehrharta* species by the absence of a distinct midrib, adaxial semi-radiate mesophyll with the abaxial chlorenchyma palisade-like in arrangement, rectangular long cells and stomatal apertures which are overlapped by four cuticular flanges projecting from the two adjacent interstomatal cells (Ellis, 1987).

Ehrharta villosa is commonly known as "pypgrass" in New Zealand and Australia but is also known as "muggie grass" in South Africa.

## 2.2 DISTRIBUTION

# 2.2.1 South Africa.

Pypgrass is found on sea dunes from Lambert's Bay to Port Alfred in its native country (figure 2.1). The villosa group grows most commonly on seaside dunes in South Africa, but has been collected as far as one kilometre inland, although still in sandy soil.

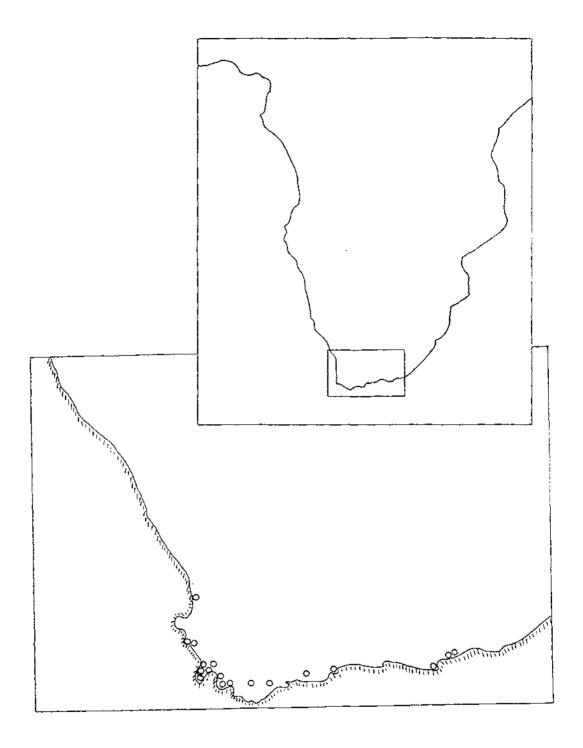


Figure 2.1 The distribution of pypgrass in South Africa.

Pygrass is found up to 1km inland from Lambert's Bay to Port

Alfred.

In South Africa, pypgrass flowers from October to December (Gibbs Russell, 1987).

There are two varieties of Ehrharta villosa - var. maxima and var. villosa, as well as Ehrharta thunbergii in the Villosa species group. All these are presumed to hybridise with each other as three intermediate forms have been reported in South Africa (Gibbs Russell, 1987).

#### 2.2.2. Australia.

Pypgrass has become fairly widespread in Australia. It is found in every state except Australian Capital Territory, Queensland and the Northern Territory (Scott and Delfosse, 1992). It has in the past been planted in certain areas of Australia as a sand binder and has now become naturalised in Victoria, South Australia and Western Australia (Vickery, 1975).

# 2.2.3. New Zealand.

Pypgrass was introduced to New Zealand 20-25 years ago by a scientist in the Plant Materials Section of the former Ministry of Works. The aim was to stabilise the sandhills at Turakina Beach, near Wanganui (South 40°4'30"; East 175°8') (Figure 2.2). A small number of tillers were originally planted in the dune area (the "Beach population"). The population increased in this area and by some means also spread to occupy a larger area (approximately 1km away) in the pine plantation, with very little pypgrass in between. It has not been recorded elsewhere in New Zealand.

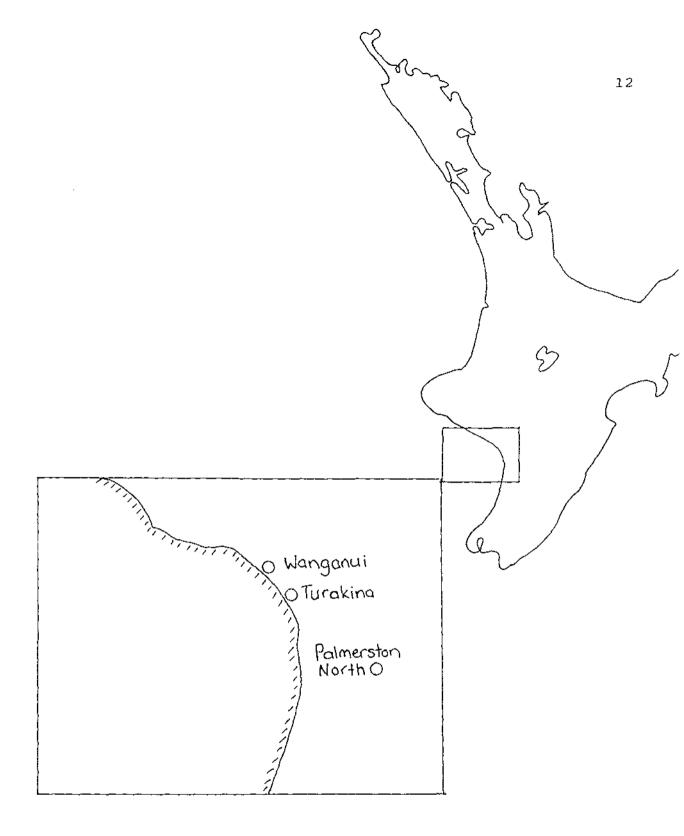


Figure 2.2 The distribution of pypgrass in New Zealand.

Pypgrass is found only in the North Island, at Turakina
Beach.

# 2.3 ECOLOGY OF THE AREA

Sand dunes are a difficult habitat for plant life being unstable, frequently dry near the surface, and with a high salt content from wind and spray. As well as this, sand dunes are prone to severe leaching making them very deficient in plant nutrients (Ranwell, 1972). The dryness of the dunes also make it extremely difficult for plants to establish. The primary source of water for dune plants comes from rainfall, since the water table in a dune just three or four metres high can make no contribution to the moisture requirements of plants rooting to depths of only about one metre (Ranwell, 1972).

Where wind has eroded sand down to the level of the water table, flat bottomed hollows (dunes slacks) are left. Dune slacks are much different to the dunes, mainly in water relations, and zones found in the vegetation which reflect may be fluctuations in the water table (Johnson, 1989). In a wet slack the water table never falls below one metre from the surface during all seasons and in fact may become flooded periodically due to rising ground water in the winter. Any plants can have their roots within reach of water at all times. In a dry slack habitat the water table lies between one and two metres below the soil surface, which means shallower rooted species are beyond the influence of the water table but deep rooted species will be in reach of water through all seasons (Ranwell, 1972).

At Turakina most of the sand dune area is cut off from the sea by the Turakina river (Figure 2.3), and a sand spit is extending

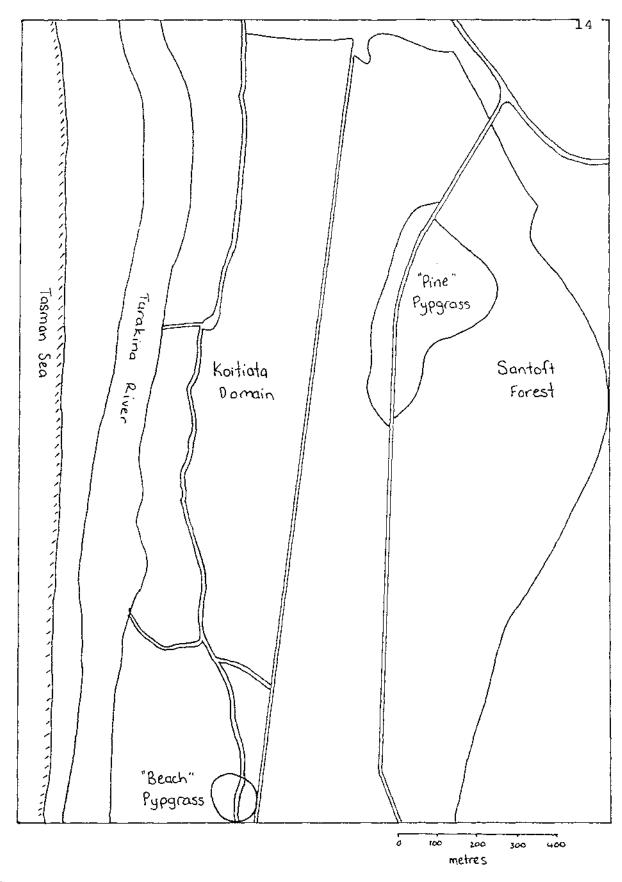


Figure 2.3 The Turakina beach area. Pypgrass is found in the Koitiata Domain and the Santoft Forest.

along the right bank of the river (Ravine, 1984). The foredunes are low (6m) and extend inland for 50 to 100m. A sandplain continues inland from this point to the artificial boundary of pine forestry (Santoft Forest). Tidal flats are also present at Turakina. These support two distinct vegetation communities: one of bachelors' button (Cotula coronopifolia) and some Limosella lineata which is regularly submerged, and another of predominantly glasswort (Sarcocornia quinqueflora), some shore primrose (Oenothera stricta) and halfstar (Selliera radicans). This area is separated from the first by very low dunes and would only be submerged by spring tides or floods.

The foredune is dominated by spinifex (Spinifex sericea) on the river side and by spinifex and marram (Ammophila arenaria) on the inland side. The inland side also has pingao (Desmoschoenus spiralis), club sedge (Isolepis nodosa) and tauhinu (Cassinia leptophylla). In the south of the dune area there is much flax (Phormium tenax) and Olearia solandri with a few cabbage trees (Cordyline australis), scattered manuka (Leptospermum scoparium) and club sedge. This community is not found elsewhere in the ecological district since further north there are fewer shrubs, more marram and much more tall fescue (Festuca arundinacea) (Ravine, 1984).

Behind the foredunes are the sandy flats on which is planted the Santoft forest. These flats or "plains" extend down the coast from the Whangaehu River to Waikanae (Ogden, 1974). Pypgrass

occurs mainly on the dune slack system and the plains behind the foredunes (Figure 2.3).

# 2.4 WEED POTENTIAL OF PYPGRASS

#### 2.4.1 Growth Habit

Scientists in Australia report that pypgrass grows there as an open "herb" and allows native plants to grow through. In South Africa, pypgrass is a primary successional species soon taken over by other species (Professor A. McGlauchlin, University of Port Elizabeth, South Africa (pers.comm)). Here the grass grows as scattered plants, with an open habit. In New Zealand however, pypgrass shows a tendency to grow in a solid mass.

The Department of Conservation Protected Natural Areas report on the Koitiata area, states that pypgrass is aggressive and has formed a dense sward over approximately one hectare of the dune. It is smothering indigenous dune plants as well as marram (Ammophila arenaria) and boxthorn (Lycium ferocissimum). The behaviour of pypgrass in New Zealand highlights the plant's weediness as it appears never to display a smothering growth habit in its native country, South Africa.

# 2.4.1.1 Personal Observations.

It has been observed at the site that stems which have fallen over and are lying along the ground are capable of growing roots at the nodes allowing the axillary bud to grow upwards contributing more tillers to the population.

.77

Pypgrass has been observed to climb upon any available vegetation and will grow up around and even between the branches of other plants as can be seen in the photos of pypgrass with tree lupin (Lupinus arboreus), boxthorn (Lycium ferocissimum) and macrocarpa (Cupressus macrocarpa) (Plates 2.1-2.3). Pypgrass appears to climb by twining its stems loosely around other vegetation by bending at the nodes. Plants have been observed to reach a height of over 2 metres, by climbing in this fashion.

Pypgrass creates a mass of interwoven rhizomes below the soil surface. Excavation of these rhizomes has revealed them to extend horizontally for many metres. At least half a metre of topsoil is occupied by the rhizome mass (Plate 2.4).

## 2.4.2 Affiliated Species.

At least two other species of *Ehrharta* have been recorded as weeds in New Zealand. *Ehrharta longiflora* (annual veld grass) is a weed of waste areas and orchards and nurseries (Vickery, 1975), while *Ehrharta erecta* (veld grass), which belongs to the same species group, is listed as common in the southern North Island and Marlborough on light sandy coastal soils (Upritchard, 1993).

Scott and Delfosse (1992) reported that Ehrharta species are important weeds of conservation areas in Australia, but have not been studied extensively. They are closely related to crop species and two native species in Australia, therefore were not recommended as good candidates for biological control.



Plate 2.1 Pypgrass climbing in a tree lupin plant.

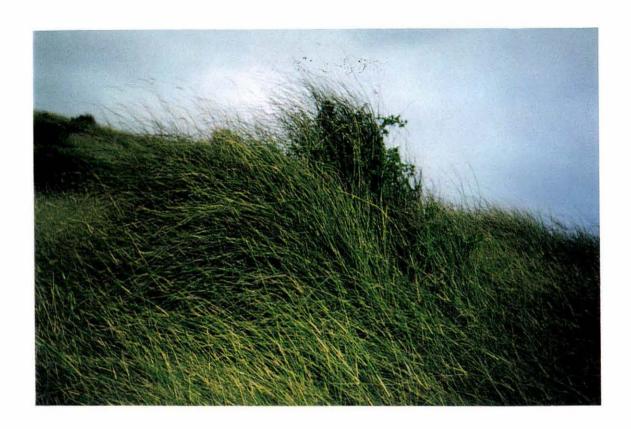


Plate 2.2 Pypgrass smothering boxthorn.

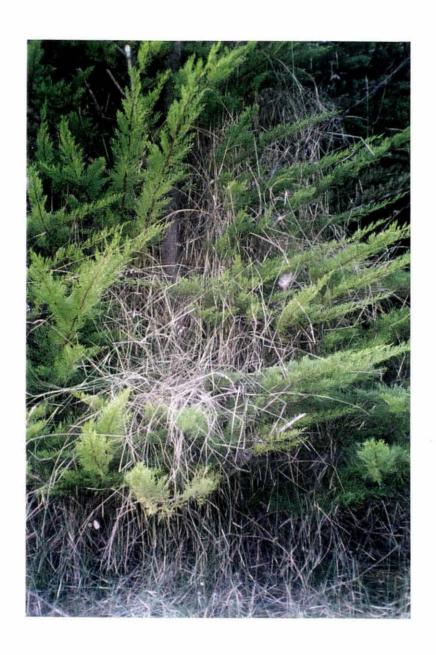


Plate 2.3 Pypgrass climbing a macrocarpa tree.



Plate 2.4 Rhizome mass of pypgrass. At least 50cm of topsoil is infested by the rhizomes.

need scale

In South Africa the weed potential of pypgrass has not gone unnoticed, being recognised as a general ruderal weed with the undesirable characteristics of being competitive for space, light, water and nutrients.

# 2.4.3 Polyploidy.

A unique feature of pypgrass is its apparently high ploidy level. Spies et al (1989) confirmed a basic chromosome number of 12 for the genus, and a chromosome number of 120 for pypgrass. Meiosis in pypgrass was normal, which is surprising in view of the high ploidy level. Normal meiosis indicates that each chromosome paired correctly, that is, the chromosomes had been replicated ten times to produce the decaploid. However only one specimen was successfully analysed and it constitutes the first ever reported chromosome number for the species (Spies et al, 1989).

Polyploidy can infer an ecological adaptability of the species to new environments and conditions (Chapman and Peat, 1992). A polyploid plant may even have enhanced adaptative abilities compared to other species in the genus due to the fact that because there are more loci there is an increased possiblity of mutation (Haas and Streibig, 1982). Furthermore, the alleles not involved in the mutation remain active, so any mutations need not lead to lethality as they do among diploids (Haas and Streibig, 1982). Polyploidy, especially to the level shown in pypgrass, can produce a plant with "luxuriant growth and strong competitive power" (Glauninger and Holzner, 1982). This occurs as polyploidy physically forces the plant to produce larger cells bearing all

the chromosomes, therefore the entire plant will be larger than its non-polyploid counterparts. Certainly, *Ehrharta villosa* is the largest of the *Ehrharta* genus (Gibbs Russell & Ellis, 1987).

### 3. SEED BIOLOGY

#### 3.1 SEED PRODUCTION

#### 3.1.1 Introduction

In general, a grass plant continues with vegetative growth until it is mature enough for the onset of reproductive development. In perennial grasses the flowering of an individual reproductive shoot is followed by the death of that shoot. Therefore the survival of the parent plant depends on the generation of further vegetative tillers, and so there is always going to be only a certain quota of fertile tillers at any one time (Chapman and Peat, 1992). Even in annual grasses only a proportion of tillers are likely to become fertile (Langer, 1980).

Seed' production starts with spikelet formation. The number of spikelets produced in the first place will dictate the final number of seeds produced. The primary factor involved in spikelet formation is the length of season and is controlled by photoperiod. Other factors involved are temperature, light, water and nutrient supply, especially nitrogen (Langer, 1979).

Similar diverse parameters control the ultimate number of fertile florets produced per spikelet. Examples include genetics - a preprogrammed 'disposition' of each spikelet to yield few or many seeds and environmental influences - reproductive structures are very sensitive to temperature injury (Langer 1979). Water stress

<sup>1:</sup> Seed is used here for convenience. In fact the "seed" is a grain (fruit containing a seed with the testa and fruit wall fused).

at a critical stage of development can cause floret sterility, but mineral supply has the most dramatic effect on, and is directly related to floret fertility (Langer, 1979). The final percentage of fertile florets formed in each inflorescence can be variable, ranging from as low as 25 per cent in some grasses to more than 90 per cent in field grown cocksfoot (Dactylis glomerata) and timothy (Phleum pratense) (Langer, 1979).

A calculation of seed production will allow an estimation of the number of potential seedlings that could be produced by the population. For a measure of seed production the number of seeds per seed head, and the number of seed heads per unit area are required (Langer, 1980).

### 3.1.2 Methods.

### 3.1.2.1 Seed Number per Seed Head.

In South Africa pypgrass flowers from October to December. At Turakina pypgrass has been observed flowering from early October to mid January, with the peak in late December to early January. It has also been observed that the beach population is more exposed to strong winds than the pine population. This would be expected to influence seed dispersal and floret fertility in a wind pollinated plant such as pypgrass. Also, an environment where there is strong wind almost constantly, is likely to become water stressed. The beach population is growing in nearly pure sand whereas the pine population is growing where there is a great deal of leaf litter predominantly from pine (Pinus radiata) and other plants such as boneseed (Chrysanthemoides monilifera)

and tree lupin (Lupinus arboreus). The soil in which there is a greater density of leaf litter would be expected to be more fertile than pure sand, possibly leading to greater plant fertility.

The number of seeds produced per seed head by pypgrass was investigated over three flowering seasons by collecting seedheads and counting the number of seeds forming in each of them.

In the summer of 1993 inflorescences were collected from both populations of pypgrass at two different times. In the first collection (late December 1993) 157 inflorescences were collected from the beach population but none collected from the population of pypgrass in the pine forest. In the second collection (January 1994) seed heads were taken from both the beach and pine populations.

In the summer of 1994 seed heads were once again collected at various times throughout the flowering period. On 27 December 1994 inflorescences were collected from both the beach and pine populations. On 9 January 1995 more seed heads were collected from both populations.

On 17 January 1996 seed heads were collected from both populations for the last time.

# 3.1.2.2 Density of Fertile Tillers.

The number of fertile tillers per square metre was estimated by placing six,  $lm^2$  quadrats at random within both populations and counting tillers producing seed heads. Quadrats were placed either in the open or over a lupin bush, to try to determine if the observation that pypgrass was flowering more prolifically around the lupin bushes was correct. This observation may have been simply due to the pypgrass climbing in the lupin, making it appear to be concentrated around it. Quadrats were placed over bone-seed plants in the pine population to clarify this. Quadrats were also randomly placed in the pypgrass under the pine trees, since it was observed that there appeared to be fewer numbers of fertile tillers in the shade.

# 3.1.2.3 Total Seed Production.

A simple calculation produces an estimate of total seed production by pypgrass. The average number of seeds produced per seed head, multiplied by the average number of seed heads per square metre, gives the average number of seeds produced per square metre. This calculation was performed for both populations.

# 3.1.3 Results

# 3.1.3.1 Seed Number per Seed Head.

11 December 1993: Of the 157 spiklets (3,140 florets) from the beach population 17 had one seed each. The rest of the florets were empty. The seeds collected were soft and bright green,

indicating that they were unripe and the empty florets had not yet produced seed, not that the other seed had already dispersed (Table 3.1). No seed heads were collected from the pine population of pypgrass.

18 January 1994: This time 30 flowering heads were examined from the beach population and only 8 florets out of approximately 600, had fat ripe seeds. Thirty five infloresences were looked at from the population of pypgrass in the pine forest. From approximately 700 florets only 86 seeds were found. However most of the flowering heads had produced more than one seed each. There was, in fact, an average of three seeds per flowering head. The pypgrass that appeared to be flowering the most prolifically were twining amongst the lupin plants.

- 27 December 1994: During the examination of 47 seed heads from the pine population, 118 seeds were found. These seeds were generally unripe, soft and green. The number of seeds per seed head found in this population was approximately 2.5. From the beach population 51 seed heads were inspected but only 9 seeds were found. Again these seeds were not ripe. The number of seeds per seed head produced in this population was only 0.2 (Table 3.1).
- 1 January 1995: Seed numbers remained fairly constant from previous years in both populations. Again it was noted that the pypgrass flowering the most prolifically in the pine population

Table 3.1. The number of seeds found to be forming in each seed head of both beach and pine populations of pypgrass, from December 1993 to January 1995.

# BEACH POPULATION:

	# seed heads	# seeds	seeds/seed head
Dec 93	157	17	0.1
Jan 94	30	8	0.2
Dec 94	51	9	0.2
Jan 95	50	24	0.5
Jan 96	107	74	0.7

Average number of seeds/seed head 0.3

# PINE POPULATION:

	# seed heads	# seeds	seeds/seed head
Dec 93 Jan 94 Dec 94 Jan 95 Jan 96	NO- 35 47 51	D A 86 118 115 1068	T A 2.5 2.5 2.5 2.3 7.6

Average number of seeds/seed head 3.7

were the plants growing close to and twining in the tree lupin plants.

17 January 1996: 107 inflorescences were examined from the beach population and 74 seeds were found from 2954 florets. This gave a similar number of seeds found per seed head (0.7) as in previous years. However the pine population showed a dramatic increase in seed numbers, where 1068 seeds were found out of 141 seed heads (2337 florets). Seeds produced per seed head for this season were then 7.6 (Table 3.1).

# 3.1.3.2 Density of Fertile Tillers.

The results show that the two populations produced similar numbers of fertile tillers in the open and near tree lupin plants, and there were consistently more fertile tillers associated with tree lupin plants than found in the open. The number of fertile tillers was lower in the shade than in the open, and there were also fewer fertile tillers around a bone-seed plant compared to in the open (Table 3.2).

# 3.1.3.3 Total Seed Production.

Table 3.3 shows an estimate of the seed production per m<sup>2</sup>, for the two populations. Seed production figures for the beach population of pypgrass were much lower than for the pine population in all positions tested.

Table 3.2. Density of fertile tillers found in both populations of pypgrass, in December 1995. 'In the open' means that quadrats were placed in almost pure stands of pypgrass.

Population	Position of quadrats	Mean no fertile tillers	Standard deviation
PINE	In the open	42.3	15.5
	By lupin	114.2	28.5
	Under trees	7.2	3.4
	By bone-seed	22.8	14.5
ВЕАСН	In the open	33.2	9.7
	By lupin	112.5	37.0

Table 3.3. The number of seeds produced by pypgrass per m<sup>2</sup> in both the pine and beach populations. Average numbers taken over three summers from 1993 to 1996.

		<del></del>
Population	Position of quadrats	Seeds per m²
PINE	In the open By lupin Under trees By bone- seed	102.4 276.3 17.3 55.1
ВЕАСН	In the open By lupin	8.0 27.0

### 3.1.4 Discussion.

In South Africa, pypgrass flowers from October to December (Gibbs Russell, 1987). Pypgrass flowers from early October to mid January in New Zealand and so appears to have a slightly longer flowering season here.

As there seems to be no previously published data on pypgrass seed production, it is impossible to determine whether pypgrass has a higher or lower seed production in New Zealand than in South Africa. The level of seed production in pypgrass is extremely low in relation to many temperate grasses. these species eg Cortaderia, produce tens of thousands of seeds per seed head. However it is common for tropical grasses to have a very low seed production (Hill & Loch, 1993). Pypgrass itself does not originate in the tropics (Gibbs Russell and Ellis, 1987), but it displays similar qualities to many tropical grasses which are generally not very efficient seed producers. reported to shed readily, and as development is poorly synchronised, a low proportion of seed matures at any one time (Hopkinson and English, 1982; Hill and Loch, 1993). Some, eg pangola grass (Digitaria decumbens) do not produce any viable seed, and only establish new populations through vegetative material (Skerman and Riveros, 1989). It may be that pypgrass does not rely totally on seed and is adapted to a vegetative means of spread, such as is found in pangola grass.

The seed production of the beach population was lower than that of the pine population of pypgrass. Low seed production can be

attributed to floret infertility since similar numbers of fertile tillers are produced by the two populations.

The beach population may have very low floret fertility due to water stress. It is probable that there is significantly less available water on the dunes and in the dune slack than in the pine forest. The pine population of pypgrass is situated where there is much more leaf litter trapping soil moisture. Langer (1979) explained that water stress at a critical stage of development would result in infertile florets. The beach population is more exposed to strong winds than the pine population, and the drying effect of the wind on the beach population could compound the moisture stress in the pypgrass plants. However, this does not explain the difference between the pine population data for 1995 and 1996.

The pine population may have shown an increase in seed production in 1996 compared to other years because of increased levels of nitrogen in the soil. All tree lupin plants were noticed dying throughout winter in 1995 at both the pine and beach sites. Tree lupins are normally perennial plants but at this time a disease caused the death of most of the plants (D. Ravine, pers. comm.). As they decomposed the nitrogen that had been fixed by the plants would have been released back into the surrounding soil. As Langer (1979) pointed out, mineral supply has the single biggest effect on floret fertility.

If this release of nitrogen had influenced the seed production of the pine population, then why has the beach population been seemingly unaffected? Firstly, there was a lower density of lupin plants at the beach site than in the pine forest, since tree lupins were deliberately planted in the forest. Fewer tree lupins would indicate that lower amounts of nitrogen would be released. Secondly, the beach population may be greatly affected by the lack of water and unable to take advantage of any nitrogen released by tree lupin.

In both populations of pypgrass, the average number of fertile tillers per metre was found to be greater around the tree lupin plants than in the rest of the population. This may be due to a mineral enrichment of the soil directly below a tree lupin plant. Tree lupins fix nitrogen and as leaves senesce the nitrogen contained in them is released, fertilizing the soil with nitrogen-rich litter (Maron and Connors, 1996). Mineral supply has a strong influence on tiller fertility and nitrogen stimulates tiller and inflorescence production (Langer, 1980).

These relationships between seed production, soil fertility, and water remain to be tested at the study site.

If pypgrass was flowering more prolifically around the tree lupins simply because it can use them for support and the greater concentration gives a perception of increased flowering, then more fertile tillers might be expected around bone-seed plants as well. There have been shown to be fewer fertile tillers

around bone-seed plants compared to around tree lupin plants, and so the increased numbers of fertile tillers around tree lupins cannot be solely due to the ability of pypgrass to climb up the plants.

The numbers of fertile tillers were lower in the shade than in the open. This is in agreement with Langer (1980), who stated that low light intensities will lower tiller fertility.

### 3.1.5. Conclusions.

Seed production is very low in pypgrass, and this may simply be because pypgrass may be adapted to survival without relying on seed production to any large extent. The differences seen in seed production between the two populations are suggested to be caused by floret infertility and not tiller infertility, under the influence of two strong factors, water stress and mineral supply. Tiller fertility in the pine population is indicated to be affected by light intensity and it is suggested that mineral (nitrogen) may also be a limiting factor in both populations.

### 3.2 SEED DISPERSAL

# 3.2.1. Introduction

The success of any plant is dependent on its dispersal in such a way that it is spread through space and time. Indeed, most seeds do show both horizontal and vertical dispersion in the soil, as well as persisting for a fluctuating length of time after dispersal (Leck et al, 1989). Annual weed species such as wild radish (Raphanus raphanistrum) rely upon the dormancy and

longevity of their seeds to allow them to persist as weeds (Cheam and Code, 1995) but perennial species may not depend so heavily on seeds, since they persist vegetatively. If seed dispersal is important for the survival of the plant, the morphology of the seed will play a major role in determining the extent of the plant's dispersal in space.

It is necessary to determine the pattern of seed deposition, when considering the dispersive abilities of a plant population. Such a determination is difficult by counting seeds already on the ground, because numbers can be distorted by seed deposited in previous years and by natural losses (eg predation) (Werner, 1975). Therefore seed traps are useful for determining the pattern of deposition over a set time and space.

Seed traps measure seed rain, therefore, seed bank studies will show the difference between the input to the seed bank each season by seed rain and the actual material available for replacement of individuals within the population (Leck et al, 1989). This will then allow a more accurate estimation of potential future seedlings.

All the above studies are important components of an investigation into the ability of pypgrass to be dispersed by seed.

# 3.2.2. Morphology.

Pypgrass is differentiated morphologically from other grasses by large spikelets 10 to 18 mm long that have sterile lemmas with conspicuously bearded bases, profusely hairy sides and mucronate to aristate tips. The first and second sterile lemmas are of similar size (Gibbs Russell, 1987). A spikelet consists of a pair of glumes, a pair of sterile lemma and at the tip of the rachis, a fertile floret composed of lemma, palea and bisexual flower (Gibbs Russell and Ellis, 1987), (Figure 3.1). Gibbs Russell and Ellis (1987) also state that it appears (for the genus) the spikelet is shed as soon as the fruit is mature, a dispersal unit being formed as the spikelet is shed above the glumes (Figure 3.2).

The species in New Zealand displays the same morphology, and dispersal units have been observed on the sand surface and being blown by the wind at Turakina. An inflorescence and spikelet collected from the "beach" population of pypgrass is shown in Plate 3.1.

### 3.2.3 Mechanism of dispersal.

It would appear from the morphology of the seed that the dispersal mechanism for pypgrass is wind, and dispersal by this method has been observed at the study site. It has also been noted that birds and mice eagerly feed upon the caryopsis. However the seed is probably not dispersed by these agents since it has no protective covering or other morphological feature that may allow it to pass through these animals, unaffected.

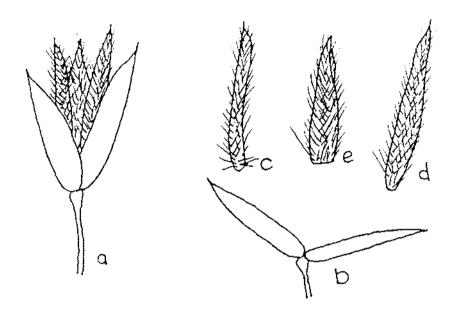


Figure 3.1 Pypgrass Floret. a: whole floret. b: glumes. c and d: sterile lemma. e: fertile lemma (seed inside).

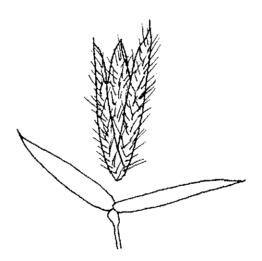


Figure 3.2 Dispersal unit of pypgrass. Lemma are shed above glumes.



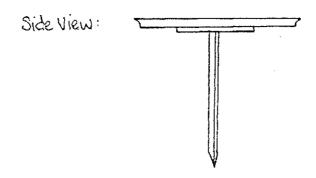
Plate 3.1 Pypgrass floret and spikelet collected from the beach population.

- 3.2.4. Seed Traps.
- 3.2.4.1 Introduction.

The primary objectives of the use of seed traps in this study were to attempt to determine the pattern of seed deposition of pypgrass, to make predictions as to where seedlings are likely to occur, and to determine how many seeds may be entering the seed bank each year.

Initially, a seed trap that was recommended for a dry seed such as is found with the dispersal unit of pypgrass, was used. This particular trap design made use of a sticky material spread over a plate or large petri dish to trap seeds that touched the surface. The plate was mounted on a rod that was pushed into the ground to the desired height (depending on surrounding vegetation) (Werner, 1975). A trap such as this (Figure 3.3) was used the first season of this study (summer of 1994-95) but it soon became apparent that birds were heavily predating the traps. It was observed that the dispersal units on the traps were only empty spikelets. Therefore the trap design was reconsidered.

Johnson & West (1988) also found Werner's trap to be unsuitable for the dry windy environments in which they were working, and presented five different trap designs which were compared for trapping ability, ease of seed resuspension by wind and ease of removal of seeds. From these results a decision was made on an appropriate trap for the environment and pypgrass seed. Johnson and West (1988) claimed that a funnel trap would be less likely to lose seeds as a result of flooding, resuspension by wind, and



Top View:
Grease applied
to inner area.

Figure 3.3 Diagram of Werner's plate seed trap.

seed predation by rodents, birds or insects, under field conditions. The funnel trap used was modified a little from Johnson and West's design so that resuspension by the wind and predation by birds was eliminated. The trap suggested makes use of a jar and a plastic funnel easily bought in many shops, but for financial reasons the design was altered and the trap constructed from a two litre plastic soft drink bottle as shown (Figure 3.4).

#### 3.2.4.2 Methods.

Forty funnel seed traps were placed around both populations of pypgrass in the following manner. Five traps were placed five metres apart in a straight line starting from each edge of the populations in each of the four main compass point directions. This allowed for detection of seed deposition up to 20 metres away from each population. The traps were checked every week for seeds, and any seeds found, were collected and tested for viability using the tetrazolium test described by Delouche et al (1962).

#### 3.2.4.3 Results.

Only two of the 31 dispersal units found in the traps contained a pypgrass seed. A positive tetrazolium test indicated that both these seeds were viable. Only two dispersal units were found five metres or more from either of the populations. These two were found in the same trap, five metres from the eastern edge of the beach population.

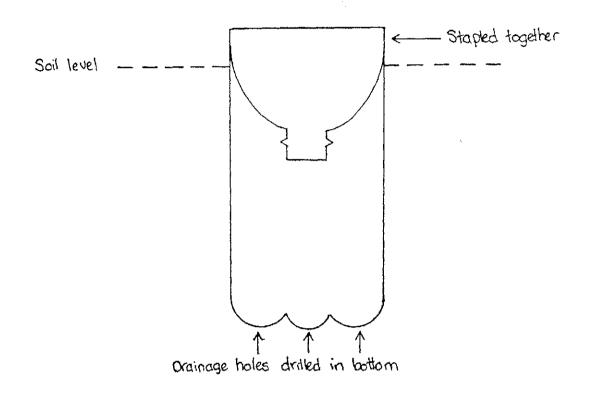


Figure 3.4 Diagram of funnel seed trap.

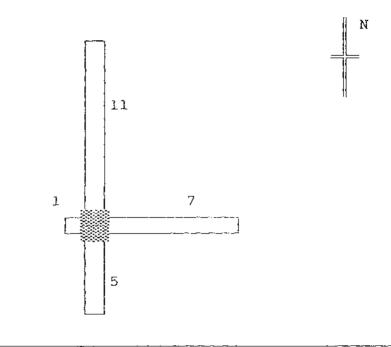
The pattern of deposition for dispersal units found is shown in Figure 3.5. This deposition pattern clearly shows the beach population to deposit most of its dispersal units to the north and east of the population, and very little to the west. The pine population deposits all of its dispersal units to either the north or south of the population.

From the dispersal units collected, the number of dispersal units that might be expected to fall per square metre is shown in Table 3.4. Since only two actual seeds were found in the seed traps it would be unwise to make calculations from this as to how many seeds would be likely to be found per square metre. The seed production data would prove to be a more reliable source of this information; however it is impossible to tell from seed production figures what proportion of the seed is predated prior to dispersal from the plant.

### 3.2.4.4 Discussion.

A positive test for seed viability using tetrazolium is for the embryo to show a pink colour (Delouche et al, 1962). Both of the seeds found in the seed traps showed this colouration after application of the chemical tetrazolium. No other seeds were found in the seed traps, suggesting that there were a higher percentage of empty dispersal units than full ones. This has in fact been shown to be true, since there has been found to be as little as one caryopsis formed for every ten seed heads (0.1 seed/seed head) (Section 3.1.3).

### BEACH POPULATION:



# PINE POPULATION:



Figure 3.5. Pattern of seed deposition for both populations of pypgrass. The shaded area represents the population of pypgrass and the bars from each side represent the number of seeds collected in seed traps placed 0m from each edge of the population.

Table 3.4. The total number of pypgrass dispersal units (with and without seed) found in the funnel seed traps, over the 1995-96 flowering season.

Population	Edge	# Dispersal units in traps	# Dispersal units/metre <sup>2</sup>
BEACH	North	11	1403
	South	5	638
	East	7	893
	West	1	128
PINE	North	4	510
	South	3	383
	East	0	-
	West	0	-

It would appear that pypgrass seed does not travel far in the wind, since only two dispersal units were found five metres away from the population, and no dispersal units were ever found in a trap any further away. However, newly dispersed seeds have been described as being widely and patchily dispersed in space (Alvarez-Buylla & Martinez-Ramos, 1990), therefore as distance from the population increases, chances of trapping seed decrease. As well as this other dune plants have been reported to disperse seeds only very small distances from the parent plant (Ehrenfeld, 1990). For example, the seeds of Lithospermum caroliniense, a perennial herb, are restricted to within 74cm of the parent plant (Westelaken & Maun, 1985). In any case the dispersal units of pypgrass can be said to be found up to five metres from the dense population.

The pattern of deposition of dispersal units at the beach site, where most dispersal units are deposited to the north and east of the population, was probably due to the prevailing winds being in those directions. The pattern in the pine population showed that no seeds were found in either of the westerly or easterly directions. This would be due to those edges of the pypgrass population being under the pine trees, where there was reduced seed production – as low as 7.2 fertile tillers per metre (Table 3.2).

### 3.2.4.5 Conclusions

Pypgrass seedlings are most likely to occur within five metres of either population, and most may be expected to be found to the north of the beach population. Dispersal and colonization of pypgrass to new areas by seed may play only a minor role in its spread.

### 3.2.5 Seed Bank.

#### 3.2.5.1 Introduction.

All viable seeds present on or in the soil constitute the soil seed bank. Seed banks consist of two components - a transient component is made up of those seeds which germinate within a year of initial dispersal and a persistent component made up of those seeds which remain in the soil for more than a year. The persistent component of the seed bank represents a reserve of genetic potential accumulated over time (Simpson et al, 1989).

Most previous studies have failed to find persistent seed banks in dune habitats (Mack, 1976; Watkinson, 1978; Boorman & Fuller, 1984), which Zhang & Maun (1994) suggested may be due to improper sampling techniques, inadequate sample size or improper depth of samples. They showed by artificially burying seeds, that for most dune species, a soil seed bank could potentially exist, and some species retained seeds on the plant creating an aboveground seed bank (Zhang & Maun, 1994). Their study did not take into account that seeds may not ever become buried naturally, even if they can survive it.

The importance of the seed bank directly relates to the likelihood of reestablishment of pypgrass by seeds already present in the soil, after removal of the adult plants.

#### 3.2.5.2 Methods.

The transient soil seed bank of pypgrass was sampled in February 1996 by taking sand from three places in both populations and sieving for seed. The amount of sand taken each time was 2500 square centimetres at the surface, taken to ten centimetres deep, as it had previously been established that no pypgrass seeds could germinate from this depth or greater (section 3.3). Seeds buried up to 10 cm would be the ones most likely to restore the pypgrass population within the first year of plant removal. Any pypgrass seeds found were to be checked for viability using the tetrazolium test described by Delouche et al (1962).

#### 3.2.5.3 Results.

No seeds whatsoever were found in any of the seed bank excavations. Some empty dispersal units were found in the soil of the pine population of pypgrass, but these are not included in the soil seed bank.

Additionally, no seedlings were at any time seen in either of the pypgrass populations during year round observations between October 1994 and February 1996.

### 3.2.5.4 Discussion.

Even though over one hundred seeds may be produced per square metre of the population, it appears that no seeds persist to form even a transient seed bank of pypgrass. Seed buried deeper than 10cm may survive under enforced dormancy and may germinate if dug up later. However at the time of testing the pypgrass popula-

tions had both just completed flowering and many seeds were expected to have been dispersed. No seeds were found suggesting heavy predation of the seed.

Sparrows remove seed from the inflorescence even before it has been released by the plant (pers. obs.). Initial seed traps, made with a greased plate to catch seeds, showed that birds, mice and rabbits living in the area had been feeding on the trapped seed as their footprints could be clearly seen in the grease.

Although pypgrass seed production is low, some seedlings were expected to be found at the site. This was not the case; no seedlings were ever sighted at Turakina. This observation appears to confirm that few or no seeds may survive to enter the seed bank, however it may be that conditions are simply not favourable for germination of pypgrass seed, or that condidions were not favourable over the period of study.

It may be possible to assess the potential seed bank of pypgrass by burying seeds to different depths artificially and retrieving them after various lengths of time. Zhang & Maun (1994) suggested that the fate of artificially buried seeds may reflect the behaviour of naturally dispersed seeds. Unfortunately this method cannot provide an accurate measure of the seeds natural distribution throughout different depths, nor can it account for the number of seeds which eventually form the seed bank (some may not survive to become buried). The failure to find pypgrass

seeds in the soil samples taken agrees with the results of others workering on sand dune plants (Mack, 1976; Watkinson, 1978).

#### 3.2.5.5 Conclusions.

It is suggested that heavy predation of the seed prevents the formation of a seed bank and so the occurrence of seedlings is extremely rare. It appears that seed dispersal in pypgrass has not been an important factor in the plant's survival and spread.

#### 3.3 EMERGENCE FROM BURIAL.

#### 3.3.1 Introduction

The seeds of a sand dune plant may become buried and later germinate from a depth. Studies of seedling emergence in sand dune species is considered important since it has been suggested that there is a relatively low chance of seeds remaining on the surface in a sand dune habitat (Maun & Lapierre, 1986). An investigation into the capabilities of pypgrass seed to germinate from different depths of burial was carried out and a comparison was made with other sand dune grasses.

To be capable of germinating, the seeds of pypgrass require a stratification period (Wells et al, 1986). Stratification consists of a seven day period at 5°C in moist conditions.

### 3.3.2 Methods

Seeds were stratified and buried in sand collected from Turakina at 0cm (surface), 1cm, 2cm, 4cm, 6cm, 8cm, and 10cm. Twenty

bag). The pots were initially placed on a spongy mat that provided water from below to the pots. However they were found to get too wet if left on the self watering mat in the glasshouse; the seeds rotted and the sand leaked from the pots. Therefore, the pots were kept in the glasshouse but watered manually every two days so that the sand in the pots remained just damp at the surface. Emergence dates and the number of seeds emerged were noted.

### 3.3.3 Results.

Table 3.5 shows the numbers of pypgrass seeds that survived to tillering after germinating from different depths of burial. All seeds left on the surface of the sand (no burial) germinated but twelve dried out almost immediately and died before any elongation of the primary leaf. Seven out of the eight remaining seedlings died before two leaves had formed. They appeared to dry out on the surface of the sand after the first leaf had unfolded.

Nine seeds buried at 1cm emerged over a range of three to seven days after burial and none of these seeds died after emergence. Out of the seeds buried at two centimetres, eleven emerged over six to nine days after burial. Eight seeds buried at four centimetres emerged between seven and sixteen days after burial, but only four of the twenty seeds buried at 6cm emerged in 16 to 21 days after burial.

Table 3.5. The numbers of pypgrass seeds that survived to tillering, after emerging from different depths of burial.

20 seeds were buried at each depth.

Depth of burial (cm)	Number of seeds emerged.	Number of seedlings survived to tillering.
0	20	1
1	15	9
2	19	11
4	8	8
6	4	4
8	0	0
10	O	0

Seedlings that were buried to a depth of either eight or ten centimetres did not emerge. When excavated at the end of the investigation (90 days), no seeds were found in the sand.

#### 3.3.4 Discussion.

All unburied seeds germinated but the poor survival of their seedlings could be due to damage incurred by the seed drying on the surface of the sand.

Two centimetres burial proved to be the depth from which most seeds emerged and survived to tillering. This could possibly be because the seeds were prevented from desiccating at shallow depths of burial, allowing more successful germination, such as has been suggested by Maun & Lapierre (1986). The emergence of a seedling from greater depths of burial can be affected by seed mass, soil type and compaction and growth form of the seedling (Maun & Lapierre, 1986). Seedling survival in sand dunes may be dependent on sand erosion or deposition, and desiccation (Maze and Whalley, 1992).

The seeds planted at eight and ten centimetres probably did not emerge from burial because they were not physically capable of extending the epicotyl to eight centimetres. That is, resources available in the seed were exhausted before the seedling could reach the surface of the sand. No seeds were found (even though they are quite large) when the pots were emptied at the end of the investigation. This probably indicates that the seed had first germinated, then died and rotted in the pot.

# 3.3.5 Conclusions

Pypgrass seeds develop poorly when unburied. Seeds can emerge from a depth of burial of up to six centimetres. Percent germination is highest when the seeds are left on the surface, however, the percentage of seedlings surviving to tillering is greatest from 2cm burial.

### 4. VEGETATIVE GROWTH

### 4.1 SEEDLING GROWTH

#### 4.1.1 Introduction.

A general pattern of grass growth for the majority of grasses has been described by Langer (1979). The first sign of growth is the emergence of the radicle which breaks through a protective sheath called the coleorhiza. Lateral rootlets then form off the growing primary root. At the same time, the first leaf elongates, eventually penetrating its own protective covering, the coleoptile.

Adventitious roots arise from the lowermost nodes of the first shoot and the tillers and contribute to water and mineral uptake. Thus, if the seed germinates below the surface of the soil the primary and adventitious root systems are generally separated through the elongation of one or more internodes which form a rhizome called the epicotyl (Figure 4.1).

During the vegetative stage of the grass life cycle, the meristematic zones are generally located near the soil surface. The stem apex sits just above the highest node, but since the stem is so highly contracted, its position is often described as at the base of the tiller (Langer 1979).

#### 4.1.2 Methods.

Seeds were stratified and planted at 1cm depth in sand collected from Turakina. Twenty seeds were planted, one seed to each pot

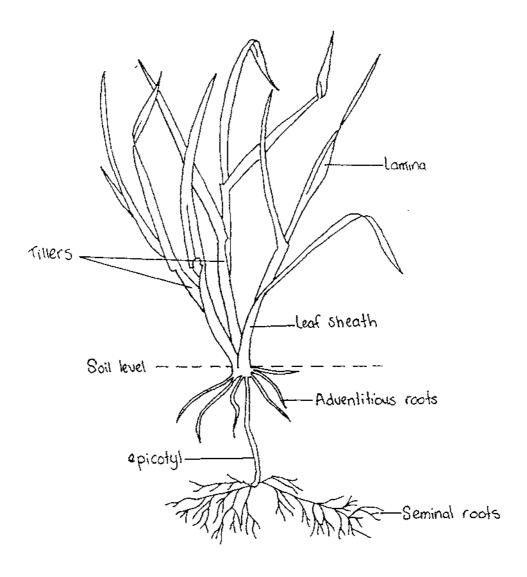


Figure 4.1. Diagram of a grass plant (from Langer, 1979).

(PB3 bag). The pots were found to get too wet if left on the self watering mat in the glasshouse, the seeds rotted and the sand leaked from the pots. Therefore, the pots were kept in the glasshouse but watered manually every two days so that the sand in the pots remained damp at the surface. Seedling height was measured daily after germination and the number of leaves counted.

## 4.1.3 Results.

Most of these seedlings had produced six leaves before tillering and had reached an average height of 410mm. When first germinated the seedlings grew quickly, and only slowed just before the second leaf emerged. At this point the seedlings went back into a stage of rapid growth, slowing once again before the third leaf emerged. This pattern continued as each leaf emerged, although the periods of rapid growth became less pronounced (Figure 4.2).

An interesting feature of the growth of pypgrass is that up until the third leaf emerges a pypgrass seedling has very short internodes (Figure 4.3) and looks like a typical bunch type grass. At the time of emergence of the fourth leaf the internodes elongate. As each subsequent leaf appears from inside the sheath of the previous leaf, the lamina extends, followed by the leaf sheath. The internode below then elongates, separating the two leaves (Figure 4.4). When tillers emerge, their internodes elongate immediately. Tillers do not go through the bunch stage (Figure 4.5).

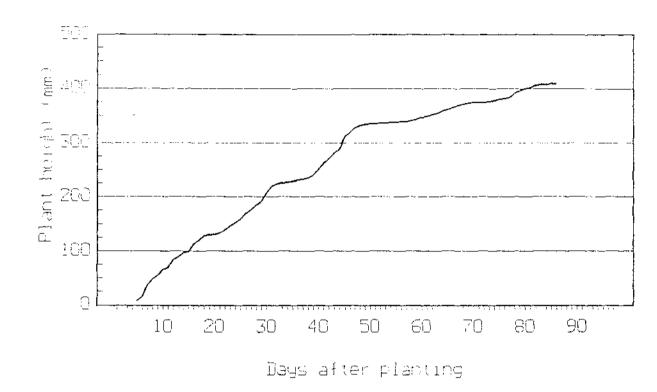


Figure 4.2. Change in pypgrass seedling height over time, when germinated 1cm below the surface and grown to tillering.

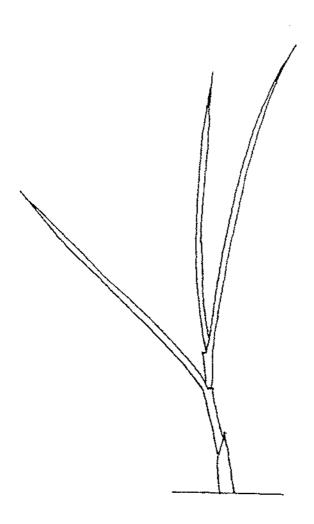


Figure 4.3 Pypgrass seedling at the three leaf stage.

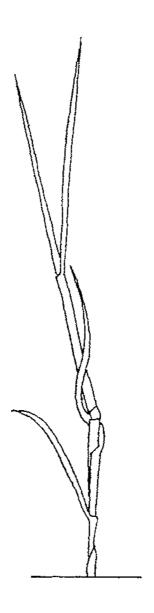


Figure 4.4 Pypgrass seedling after the three leaf stage but before a tiller has emerged.

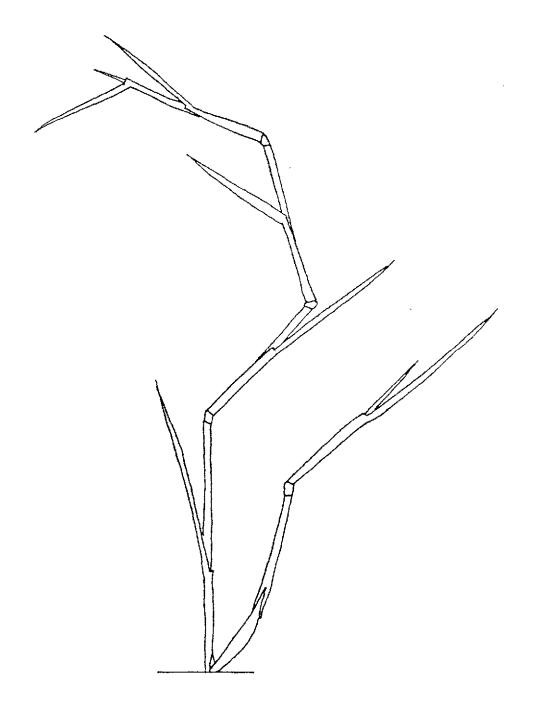


Figure 4.5 Pypgrass seedling with one tiller.

## 4.1.4 Discussion.

The actual growth form of pypgrass provides clues as to the affinities of the plant. Although its centre of geographic diversity is in a cool-temperate climate (section 2.1), it shows tropical affinities in its tall, elongated growth form. In temperate grasses such as oat (Avena sativa), elongation of the internodes between leaves does not occur except when the grass plant is preparing to flower (Langer, 1979). On the other hand internode elongation without floral initiation is common in most tropical grasses and bamboos (Chapman and Peat, 1992).

This characteristic tall growth form may suggest pypgrass seedlings would have an advantage of height over other seedlings. However, any advantage will not last long as the rate at which the plant develops is slow compared to other weeds. One example is wild radish (Raphanus raphanistrum), which can produce flowers only 48 days after germination (Cheam and Code, 1995). In the case of pypgrass, the two centimetre buried seedlings had produced only six leaves and one subsidiary tiller in 85 days. Most other grasses are also able to develop much faster than pypgrass. Perenial ryegrass (Lolium perene) will produce an average of 6.2 tillers by the time the plant is at the ten leaf stage (Langer, 1979). Pypgrass seedlings could not therefore be described as vigorous.

No seedlings have been found at Turakina, and it is unlikely that they were overlooked, since they are easily distinguished from rhizome-produced tillers. It appears, therefore, that pypgrass does not depend on seedlings for survival and spread.

#### 4.1.5 Conclusions.

The internodes of pypgrass elongate from the time of development of the fourth leaf. Although pypgrass is considered to be a cool-temperate grass, elongated internodes without floral initiation, are a common feature in tropical grasses.

Pypgrass will tiller when six leaves have been produced, but the seedlings are slow to develop, and lack the vigour of other grasses and weeds. Seedling development is not important in the success and spread of the pypgrass population.

# 4.2 REGENERATION FROM RHIZOME

# 4.2.1 Introduction.

Since seed production is very low in pypgrass (Section 3.1), regeneration from rhizome fragments may be an important factor in the survival and spread of the plant. Rhizome fragments of many species, couch (Agropyron repens) for example, can produce shoots and roots following detachment from the parent plant. According to Harris and Davy (1986) this can happen because fragmentation removes the rhizome buds from the direct influence of the parent plant. Most axillary buds on intact rhizomes remain dormant, but when the rhizome is cut they are released from apical dominance (Salisbury and Ross, 1985).

Firstly, a pot trial was set up, where three rhizome fragments containing either one two or three nodes were buried. These would be harvested each week for 20 weeks. The pots were placed outside and watered automatically by an overhead system. These fragments were found to rot as the sand in the pots became far too wet. Therefore, the trial was set up again in the pots. This time, they were placed in the glasshouse and were watered manually every two days so that the sand was damp, but not wet. These fragments also rotted within five weeks and not one of them grew shoots.

Owing to the failure of these attempts at assessing the regenerative potential of pypgrass rhizome fragments, a third (scaled down) trial was set up on 2 November 1995. This time of the year (end of spring) is probably not the optimum time for rhizome growth. For a perennial plant, the beginning of spring would be the time when most rhizome reserves would be mobilised to produce new tillers.

## 4.2.2 Methods.

Rhizomes that were excavated from the pine population of pypgrass at Turakina were cut into one to five node lengths. An area within the pine population was cleared and ten fragments of each size were left on the surface and another ten buried 5 cm below. After seven weeks, the fragments were recovered. The nodes growing were noted and measured. The temperature in the Turakina area over November and December 1995 averaged 13.8°C and 17.9°C respectively, with the maximum recorded temperature being 29.0°C

(21 Dec) and the minimum recorded temperature 3.0°C (3 Nov). Also over this period, 245mm rainfall was recorded (National Institute of Water and Atmospheric Research (NIWA)).

#### 4.2.3 Results

None of the rhizome fragments left on the surface grew. Seven out of the ten, one node fragments buried at 5cm produced a shoot. Three out of the ten, two node pieces produced one shoot, the same being produced from the four and five node pieces. Of the ten, three node pieces, two produced a shoot (Figure 4.6).

The lengths of the shoots produced by all fragments buried at 5cm ranged from 3mm to 162mm, but there was no correlation between rhizome fragment length and new shoot length.

By examining the leaves on the rhizome fragments it was possible to determine which end of the fragment had been closer to the rhizome apex. It was found that it was always either the first or second axillary bud closest to the "apex" end of the fragment that produced a new shoot. Seven out of the total of eleven shoots produced (from two to five node fragments), grew from the first axillary bud (see Table 4.1).

## 4.2.4 Discussion

The failure of all rhizome fragments left on the surface to sprout, was probably because they became too dry under the pine canopy.

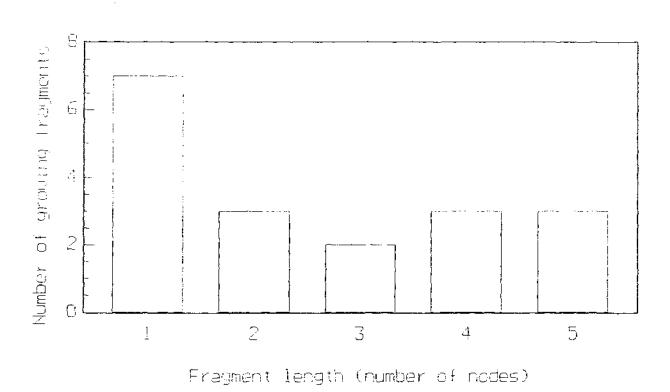


Figure 4.6 The number of fragments which produced a shoot. Ten fragments of each size (one to five nodes) were buried at 5cm, and each fragment produced only one shoot.

Table 4.1 The position of the axillary bud which produced a shoot, from the rhizome fragments with more than one node.

There were ten of each size of fragment (2 to 5 nodes) and each fragment produced no more than one shoot.

Position. (No. nodes from apical end)	Numbei rhizom 2				Total no. shoots
1	2	1	1	3	7
2	1	1	2	0	4
<b>∄</b> 3	-	0	0	0	0
4	-	~-	0	0	) 0
5	}		-	0	0

There does not appear to be any strong pattern emerging that would relate fragment length to either the number of shoots produced or the length of those shoots. Nyahoza et al (1974), working on Poa pratensis, state that rhizome buds have one of four fates. They can grow into secondary rhizomes, or into aerial shoots. However, they may also remain dormant or even abort. A rhizome bud that aborts may first grow a small distance, then die. Possibly, the very short shoots observed on the pypgrass rhizome fragments were in the process of aborting.

The one node pieces showed the strongest ability to regenerate pypgrass, however, they required burial to do so. From the observed drying out of the surface rhizomes, it appears that 5cm burial kept the fragments moist, and allowed them to produce shoots.

The two buds closest to the apex end of the fragment were always the ones to produce a shoot. A similar pattern is seen in *Poa pratensis* where, in the intact plant, buds furthest from the rhizome apex lost dormancy first, but the removal of the apex caused rhizome buds proximal to the apex end to produce shoots the most rapidly (Nyahoza et al, 1974). This pattern has also been seen in other rhizomatous grasses such as couch (McIntyre, 1970) and Johnson grass (*Sorghum halepense*) (Beasley, 1970).

This study does show that any piece of rhizome left in the soil may act as a source of colonization for pypgrass if it grows into a new plant. Langer (1979) states that rooted portions of

rhizomes and stolons are a means of vegetative propagation and are the basis of the success of couch as a persistent weed. Rhizomes with adventitious roots at their nodes are capable of dispersing over some distance as independent units (Langer, 1979). It is possible that regeneration by a rhizome fragment is a mode of dispersal for pypgrass, especially considering the poor regenerative ability of pypgrass from seed (Section 4.1).

#### 4.2.5 Conclusions

Rhizome fragments of pypgrass are capable of producing a new plant after being detached from the parent plant. They cannot do this if they dry out, but enough moisture can be retained to allow growth if the fragment is buried. The buds closest to the apical end most often produce shoots.

# 4.3 RATE OF VEGETATIVE SPREAD.

## 4.3.1 Introduction.

The rate of vegetative spread of pypgrass was estimated by mapping part of the boundary of the pypgrass population and repeating the exercise one year later.

# 4.3.2 Methods.

The area chosen for mapping contained a clear, sharp front of maximum density pypgrass, with scattered colonising plants up to the boundary of the population (Plates 4.1, 4.2). The mapped area was on the eastern edge of the 'beach' population, and covered 20m X 22m.

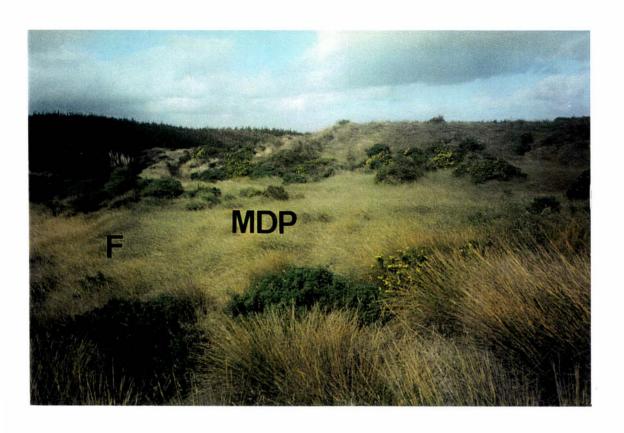


Plate 4.1 Study area of the beach population of pypgrass.

(September 1994), from the Northern edge of the study area.

The sharp front (F) of the maximum density pypgrass (MDP) can be seen to the left.



Plate 4.2 Study area of the beach population of pypgrass (October 1995), taken from the Northern edge of the study area. Note that scattered pypgrass can now be seen to the left (in front) of the maximum density pypgrass (MDP).

The area was marked out by driving a warratah standard into each of the four corners. Ninety degree angles for each corner were assured by using an optical square. Quadrats were laid down within the study area at positions determined as follows: In October 1994, the plant at the boundary of the population along the Om line (Figure 4.7) was found and its distance from the western edge recorded. A quadrat was placed within the pypgrass population, so that one edge lay on the Om line, and one edge touched the leading plant, just excluding it from the quadrat (Figure 4.7). Subsequent quadrats were placed at 3m intervals in the same line towards the maximum density pypgrass until a quadrat was laid within this area. Rows of quadrats were laid every four metres up to the 20m line. Within each quadrat the number of pypgrass tillers present was counted, and all other species present were also recorded.

In 1995 the quadrats were placed at the exact same positions as in 1994 to evaluate how those quadrats had changed. As well as this, new boundary plants were found and their positions noted in July 1995 (nine months after 1994 measurements) and October 1995 (one year after 1994 measurements). As in 1994, in each quadrat the number of pypgrass tillers, and all other species present, were recorded.

## 4.3.3 Results.

The measurements taken from the western edge to the last pypgrass plant found are shown in Table 4.2. The greatest distance that pypgrass appeared to move over the 12 month period was 9.0m and

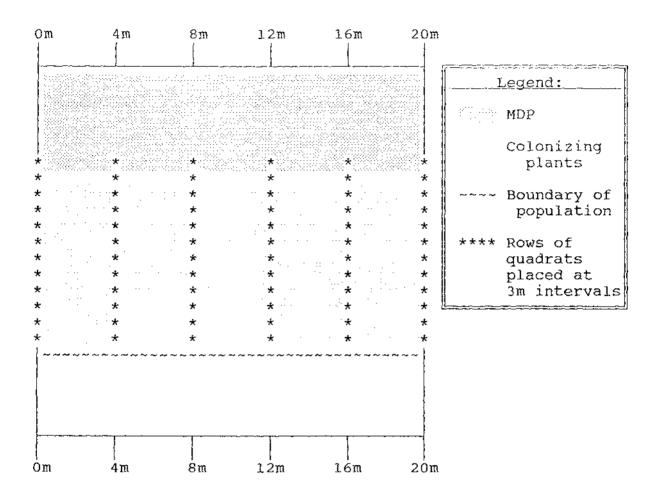


Figure 4.7 A diagram showing the marked out area of the boundary of the pypgrass population, in which quadrats were laid. Rows of quadrats started at the leading coloniser found in each line, and ended when back in the area of maximum density pypgrass (MDP).

Table 4.2. Distance from the western boundary of the study area to the outermost pypgrass plant for each of the six transects, measured in October 1994, July 1995 and October 1995.

	Distance along study area						
	Om	4 m	8m	12m	16m_	20m	
OCT 1994	16.6m	17.3m	18.lm	21.0m	22.Om	23.4m	
JUL 1995	20.1m	23.5m	24.2m	25.6m	26.1m	24.5m	
OCT 1995	21.4m	26.3m	26.4m	29.2m	29.4m	27.5m	
Total distance moved in 1 year.	4.8m	9.0m	8.3m	8.2m	7.4m	4.1m	
Average distance moved per month	O.4m	0.75m	0.69m	0.68m	0.62m	0.34m	

the shortest distance was 4.1m (Table 4.2). The rate of spread of pypgrass can be calculated from these figures. Given an average distance that pypgrass moved in one year of 7.0m then pypgrass rhizomes grew at a rate of at least 0.6m per month or 1.9cm per day.

The average number of pypgrass tillers/m2 is shown in Figure 4.8. The average maximum density of pypgrass (MDP) in 1994 was 108.6 tillers/m2 which fell to 0.5 tillers/m2 at the leading edge in 1994 (12m from MDP). In the same quadrats in 1995 however, the average maximum density of pypgrass had increased to 171.6 tillers/m² dropping to 13.2 tillers/m² 12m away (the leading edge now being close to 20m from the MDP). As the distance from the maximum density pypgrass increased, the number of tillers/m2 decreased in both years, but numbers of tillers/m² were consistently higher in 1995. Tiller numbers were assessed using square test which showed there to be significant a chi differences between the number of tillers counted in 1994 and in the same quadrats in 1995.

Differences in species composition between 1994 and 1995 can be seen in Figure 4.9 where species have been grouped together according to growth habit. In 1995 there were generally more species present than in 1994. The only significant differences were found in the numbers of different mosses and small dicotyledons. There were more mosses in 1995 than in the previous year. In 1994 no mosses were present for up to 6 metres from the MDP. There were also significantly more rosette plants

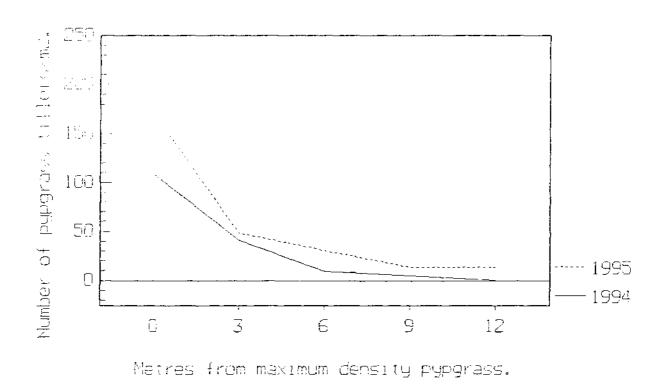
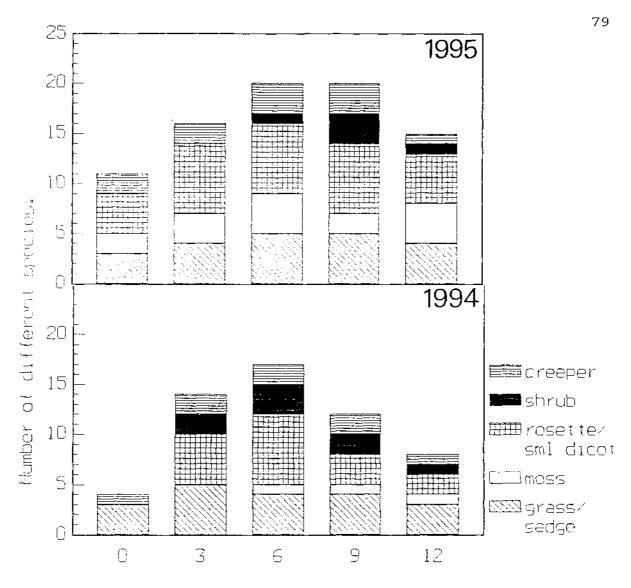


Figure 4.8 The average number of pypgrass tillers present per square metre in October 1994 and 1995. Measurements were taken at 3 metre intervals from the maximum density pypgrass.



Metres from maximum density pypgrass.

Figure 4.9 Changes in species composition in the study area quadrats from October 1994 to October 1995. Species have been grouped according to life form.

or small dicotyledonous plants in 1995 than 1994, however, there were fewer shrubs in the three metre and six metre quadrats in 1995 (Figure 4.9).

A relationship can be seen between the sharp drop in pypgrass tiller numbers and an increase in numbers of other species in both years (Figures 4.8 and 4.9).

The species composition also changed as the density of pypgrass increased over the study area. Eleven species were found growing at all densities of pypgrass. These included two mosses, the sedge Isolepis nodosa, two smaller grasses and the small dicotyledonous plants Galium aparine, Sonchus oleraceus, Meulenbeckia complexa, Hypochaeris radicata, Lactuca virosa and Sonchus asper.

Other species were found at medium to low densities and these were; the mosses, Bryum caespiticium and Campylopus introflexus; the sedge Isolepis cernua; and the dicotyledons, Parentucellia viscosa, Kanisa albida, Lupinus arboreus and Oenothera stricta. Some species were only ever found at low densities of pypgrass, including the grasses Festuca arundinacea and Ammophila arenaria, but most were larger dicotyledons such as Senecio elegans, Senecio glastifolius, Calustegia soldanella, Vicia hirsuta and Cerastium glomeratum.

In the dense pypgrass there was (by observation) considerably less weed growth. As density decreased the percentage of ground

cover by plants other than pypgrass increased dramatically. An estimation was made in 1994 to compare the quadrats in the maximum density pypgrass (MDP) with those at three metres. This estimation put pypgrass cover in the MDP at 85% and weed cover at 15%. Three metres from the MDP pypgrass cover was only 30% with weed cover being 60% and the remainder bare ground.

## 4.3.4 Discussion.

The distance that pypgrass had moved was recorded by noting the appearance of above ground tillers. However, the distance recorded by this method is not a true measure of total rhizome growth. To obtain a true measure rhizomes would have to be excavated, but any exposure of the rhizome apex to light will affect its natural growth (ie induce it to form a tiller). Therefore the rate of vegetative spread presented in Table 4.2 is an estimation of the actual rate of rhizomatous spread of pypgrass. Even so, pypgrass has been shown here to spread between 34 and 75cm a month by rhizome growth alone. As a comparison, couch rhizomes have been shown to be capable of growing at a rate of between 45cm and 120cm per month when grown from tillers in a pure stand (Tripathi and Harper, 1972).

Pypgrass tiller density in the maximum density pypgrass (MDP) was over twice that of tillers only 3 metres towards the advancing edge in 1994. In 1995 the MDP was more than three times as dense as tillers in the same position (3m away). It is between these

points that the sharp front of the pypgrass population is found, as shown in plates 4.1 and 4.2.

The number of different species was higher in 1995 than 1994 probably because of the dune slack flooding during the winter of 1995. This has clouded the results, as many species were present after the area had been flooded, and these may not survive when the dune area dries out. Species such as annual mouse-ear chickweed (Cerastium glomeratum) and the mosses Rhynchastegium tenuifolium, Bryum billardierei, and Campylopus introflexus were in this group of plants.

Grasses, sedges and creepers are the life forms that appear to be consistently unaffected. Pypgrass negatively affects the surrounding vegetation only when tiller numbers become very high (Figures 4.8 and 4.9). The thick rhizomes of pypgrass, with their many adventitious roots, may compete for space and water with the surrounding vegetation. This may allow only the most hardy of plants to survive with pypgrass. Competition may be the main method by which pypgrass establishes its near monoculture. Many of the other weeds present are dicotyledonous annuals, which rely on some amount of bare soil for the production of a new generation. If the soil has become covered by pypgrass, seeds of annuals may not germinate and if they do their seedlings are unlikely to establish, and they are subsequently excluded from the area.

It is possible that pypgrass achieves its near monoculture through an allelopathic effect. The rhizomes of pypgrass could produce and exude allelochemicals that suppress or inhibit the germination and growth of surrounding plants. Allelochemicals such as these have been reported in couch (Agropyron repens) rhizomes (Weston, 1986; Schulz et al, 1994). It has been found that low concentrations of these allelochemicals haven't any effect on other plants but high concentrations result in growth inhibition. Furthermore, dicotyledons are more susceptible than monocotyledons. The results gained from pypgrass would suggest this type of effect could be occurring at Turakina (since only the greatest density of pypgrass affects the surrounding vegetation and most plants able to survive with pypgrass are monocotyledons).

Changes in species composition may be related to phenomena other than the increased pypgrass tiller numbers. The drop in shrub numbers from 1994 to 1995 (Figure 4.9) can be attributed to the sudden death of lupin throughout the area. Moreover, the increased numbers of mosses in 1995 may have been heavily influenced by the flooding of the dune slack. However not all the described changes can be accounted for this way.

## 4.3.5 Conclusions.

A true measure of the rate of rhizome growth was not taken because of difficulties involved with excavating the deep rhizome system, without disrupting the natural growth of the rhizomes.

However, pypgrass has been shown to spread by rhizome growth by between 34 and 75cm per month.

Species growing on the dune could be affected by pypgrass. Competition for space and water by pypgrass could suppress the growth of other plants. Alternatively, an allelopathic interaction with pypgrass, may account for the changes seen in species composition. However, there is a clear possibility that unrelated (physical) factors have also influenced the species composition between 1994 and 1995.

# 5. OTHER ECOLOGICAL DATA

# 5.1 LEAF ANATOMY

## 5.1.1 Introduction.

The leaf anatomy of pypgrass has been described by Ellis (1987). He clearly illustrated the anatomy of all varieties in the villosa group. The villosa species group can be distinguished by the absence of a distinct midrib, adaxial semi-radiate mesophyll with the abaxial chlorenchyma palisade-like in arrangement. Stomatal apertures are overlapped by four cuticular flanges projecting from the two adjacent interstomatal cells (Ellis, 1987).

A comparison was made between this description of pypgrass anatomy and the anatomy of that growing at Turakina. Anatomical evidence of C3 or C4 photosynthesis, and of specialised adaptations, was sought.

# 5.1.2 Methods.

Pypgrass leaves were fixed in formalin-acetic-alcohol (FAA) on site immediately after collection. FAA was made according to Johansen (1940). The material was fixed for 48 hours before washing it in 70% ethanol, dehydrating it through a graded tertiary-butyl-alcohol (TBA) series (70% to 100%) and embedding it in paraffin wax. Transverse sections ( $10\mu m$ ) were cut on a microtome and these sections were stained in safranin and fast green, as described by Gibbs Russell and Ellis (1987)

#### 5.1.3 Results.

The leaf blade of pypgrass is inrolled loosely and has no distinct midrib, although in transverse section the median vascular bundle can be seen (Plate 5.1). Adaxial ribs are formed above each vascular bundle and small furrows exist between all ribs (Plate 5.2). No distinctive bundle sheath surrounds any vascular bundle (Plates 5.1, 5.2). The mesophyll of pypgrass has been described by Ellis (1987) as semi radiate in arrangement and this is particularly noticeable on the adaxial ribs (Plate 5.2). The palisade like arrangement of the abaxial chlorenchyma cells can also be seen in Plate 5.2.

Stomata are found only in the intercostal zones on the abaxial epidermis (the costal zones are equivalent to the ribs). Plate 5.3 shows a stoma with two of the four cuticular flanges overlapping it. Stomata are sunken below the surface of the epidermis and both guard cells and subsidiary cells are overlapped by cuticular flanges (Plate 5.3). Short spines are found on the ribs of the adaxial epidermis but are absent on the abaxial surface (Plate 5.2).

## 5.1.4 Discussion.

If pypgrass were a C4 species it may explain, in part, its success in the sand dune environment. In dry environments, C4 plants are more successful than C3 plants as their photosynthesis is twice as efficient at the conversion of the sun's energy into dry matter (Salisbury and Ross, 1985). The more rapid photosynthesis of a C4 plant also results in a lower water

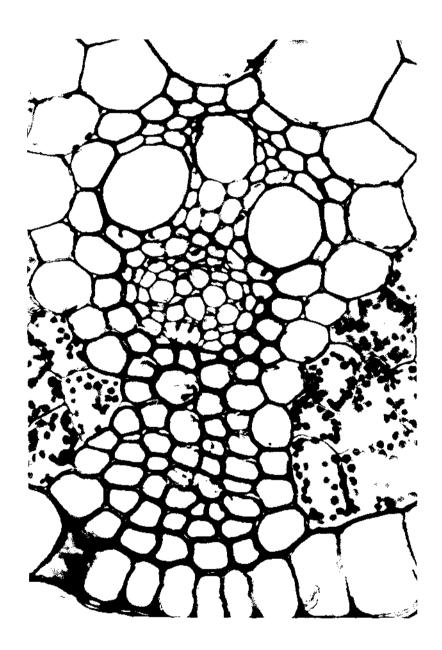


Plate 5.1 Leaf anatomy of pypgrass, with median vascular bundle in transverse section. Note that no bundle sheath surrounds any vascular bundle.

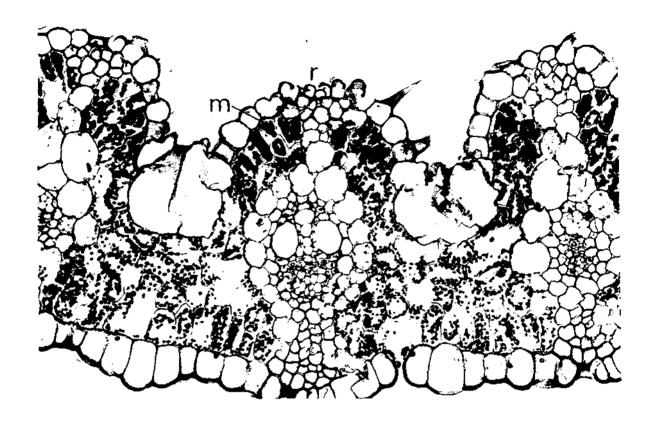


Plate 5.2 Transverse section of loosely inrolled pypgrass leaf.

Semi radiate mesophyll (m) is formed in each adaxial rib

(r). Note also the palisade-like arrangement of the abaxial chlorenchyma. Short spines are found only on the adaxial ribs.

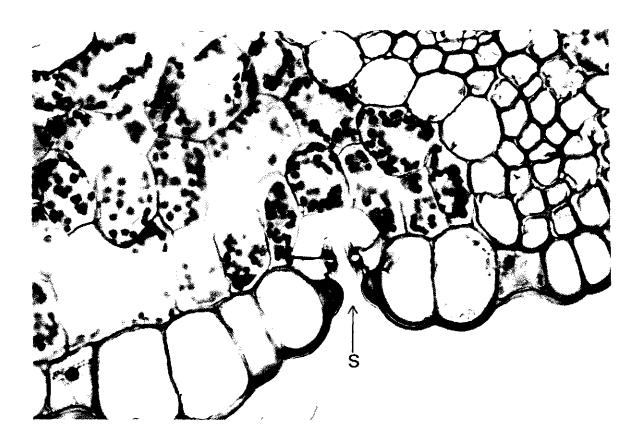


Plate 5.3 A stoma (s) of pypgrass in transverse section. Note the cuticular flanges covering both the stomatal guard cells and subsidiary cells.

requirement per gram of dry matter produced and photosynthesis occurs at a higher optimum temperature (30-47°C) than for a C3 plant (15-25°C) (Black, 1973).

The absence of a distinct bundle sheath containing chloroplasts confirms that pypgrass is a C3 plant (Salisbury and Ross, 1985). This is not unexpected in view of the taxonomic position of pypgrass, in the subfamily Bambusoideae (see Section 1.1) Langer (1979) states that rice and bamboo (also members of the Bambusoideae), have high photorespiration and lack the four carbon pathway, even though they are found in tropical environments. Therefore, on anatomical evidence, the C3 photosynthesis of pypgrass is not likely to provide an advantage over the other species in the dune environment at Turakina, which are also C3 plants.

One adaptive feature of the leaf anatomy of pypgrass is the distinctive stomata. These are sunken well into the abaxial epidermis of the leaf and partially covered by cuticular flanges (Ellis, 1987). This could reduce transpiration of pypgrass in a physiologically dry habitat.

## 5.1.5 Conclusions

This study of the leaf anatomy of pypgrass has confirmed that pypgrass growing at Turakina is homologous with that studied in South Africa. Pypgrass is a C3 plant with distinctive stomata, which are sunken and over-arched by four cuticular flanges. This arrangement of the stomata would be expected to reduce loss of

water from the leaf and be of selective advantage in a dry environment.

# 5.2 SOIL MYCORRHIZA

## 5.2.1 Introduction.

Sand dunes are a difficult habitat for plant life being unstable, frequently dry, leached of minerals and with a high salt content (see Chapter 2.3). Recent literature suggests that mycorrhiza have an important role to play in the success of sand dune plants, as mycorrhizal plants have been shown to be tolerant of drought and saline soils (Schenck, 1982.; Crawley, 1993).

A mycorrhiza is a highly specialized symbiotic association between a fungus and a plant root. Both partners in this relationship generally benefit from the association (Jackson and Mason, 1984). The occurrence of vesicular-arbuscular (VA) mycorrhiza is widespread among many plant species from many different habitats (Rozema et al, 1986). VA mycorrhizal fungi are so called as they form two distinct structures, vesicles and arbuscules. Vesicles are oval or sperical structures that can occur either intercellularly or intracellularly. Vesicles are assumed to be storage structures but may also act as survival units when the root dies (Jackson and Mason, 1984). Arbuscules, highly branched structures, are formed in cortical cells and look like little trees (arbuscule means 'dwarf tree'). An arbuscle creates an extremely large area of contact between root cell

cytoplasm and fungus, and the arbuscule branches are thought to be the main sites of nutrient exchange (Jackson and Mason, 1984).

If pypgrass makes an association with a mycorrhizal fungus, it could be an important factor in the vigorous growth of the grass compared to that of other dune plants. Mycorrhizal fungi with their extramatrical hyphae aid the plant in uptake of relatively immobile minerals, especially phosphorus, from the soil. The association works by the fungi effectively increasing the rhizosphere of the plant, extending the area of absorption beyond that of the root hairs (Schenck, 1982).

## 5.2.2 Methods.

Roots of pypgrass and other dune plants living in the same area as pypgrass were investigated for the presence of mycorrhizae. (Table 5.1). To test whether these plants were mycorrhizal, the roots of the plant were prepared and stained according to Phillips and Hayman (1970).

Unpigmented roots of a diameter less than 2mm were chosen. Roots were heated at 90°C for 1 hour in 10% KOH, then rinsed in water and acidified with dilute HCl. The KOH removes the host cytoplasm and most of the nuclei allowing the stain to penetrate readily so no stained host cytoplasm will obscure the fungal tissues. The roots were stained by simmering for five minutes in 0.05% trypan blue in lactophenol and the excess stain removed in clear lactophenol.

Table 5.1 Species Tested for Presence of Mycorrhiza. The species included in this investigation were from differing families, as can be seen in the table. All were collected from the Turakina dune area, where pypgrass is growing.

SPECIES TESTED	COMMON NAME	FAMILY
Ehrharta villosa	pypgrass	Poaceae
Phormium Tenax	flax	Agavaceae
Spinifex sericea	spinifex	Poaceae
Desmoschoenus spiralis	pingao	Cyperaceae
Isolepis nodosa	club rush	Cyperaceae
Lupinus arboreus	lupin	Fabaceae
Ammophila arenaria	marram	Poaceae

The roots were then cut into segments approximately one centimetre long and eight segments were mounted on a slide in water. Slight pressure on the coverslip flattened the roots ready for observation under a light microscope.

The best method for assessment of the levels of infection was formed by modifying the method of Giovannetti and Mosse (1980). An eyepiece micrometer grid of one hundred squares was used under the 10 X objective lens. This magnification was chosen as at this magnification the micrometer squares most closely approximated the size of a single cell in the root tips of all species tested. In an attempt at objectivity a mounted root segment was "lined up" with the objective lens while looking from the side of the microscope. Each square in the micrometer grid of one hundred, was scored "plus" or "minus" depending on whether hyphae, vesicles or arbuscles were sighted within that square. This was repeated six times for each species, averaged, and expressed as percentage infection.

## 5.2.3 Results

Hyphae, vesicles and arbuscles were observed in all species, but pypgrass and flax appeared to show the greatest percentage of mycorrhizal infection, 88.9% and 70.3% respectively. Spinifex, Isolepis and pingao all had a similar level of mycorrhizal infection (57% to 42%), whereas lupin and marram both had a much lower percentage infection than any other plant (9.8% and 7.8%).

The percentage infection is presented for all species in Figure 5.1. Species given the same letter are not significantly different. The margin of least significant difference is 22.2% as shown by the bar in the upper right corner of the graph. All species tested are in direct competition with pypgrass except pingao. Interestingly, the two plants which appear to some extent to be smothered or outcompeted by pypgrass are the two with the lowest percentage infection of mycorrhizal fungi (lupin and marram).

Each slide was scored blind; that is no identifying marks were on any slide except randomly assigned letters which were matched back to the correct species after assessment. This was to avoid any possible bias in objectivity of the assessor. It is possible there was a constant overestimate of the amount of infection, for two reasons. One, the darker colour of well stained root segments is apparent to the naked eye thus making an unbiased selection of root segments difficult; and two, more than one layer of cells is viewed under the grid so any infection sighted in whichever layer is counted. However, these two biases are consistent for all slides so although an overestimate may be produced the results are still able to be compared.

An example of the fungal material found in the cortex can be seen in the photographs of mycorrhizal tissue taken from slides made of pypgrass roots (Plates 5.4 - 5.6)

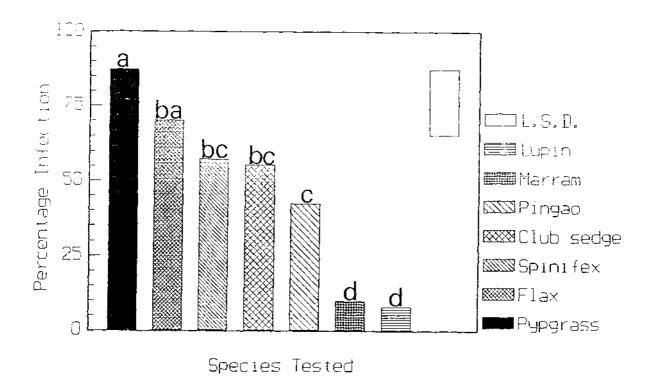


Figure 5.1 The percentage mycorrhizal infection found in the roots of some of the dune plants at Turakina Beach. The least significant difference is shown by the bar in the right top corner of the graph. Species sharing the same letter are not significantly different.

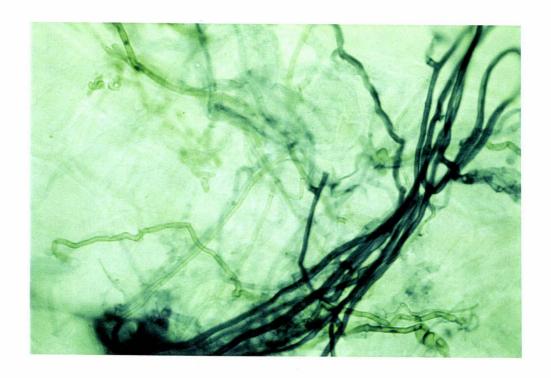


Plate 5.4 Extracelluar hyphae of the V A mycorrhizal fungus found in the roots of pypgrass. Magnification x10.

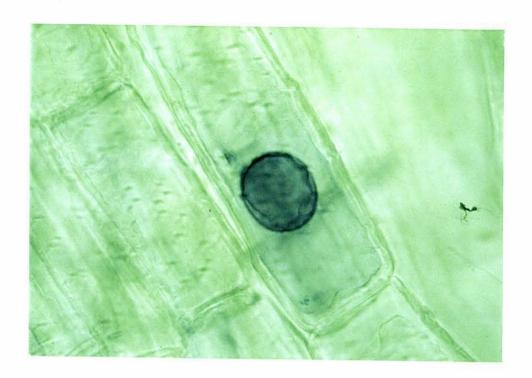


Plate 5.5 A vesicle of the V A mycorrhizal fungus found in the root cortex of pypgrass. Magnification x40.



Plate 5.6 Intracellular hyphae of the V A mycorrhizal fungus found in the roots of pypgrass. Magnification x40.

### 5.2.4 Discussion

If it is assumed that infection with mycorrhizal fungi does bestow on the plant an advantage in a water stressed and saline environment such as that of sand dunes, then the assumption can also be made that those plants with the greatest mycorrhizal infection can be expected to compete most successfully in that environment. Certainly the results gained are consistent with this statement. Those plants which are the most successful competitors in the dune environment at Turakina Beach are those which have a medium to large percentage infection of mycorrhizal fungi.

This could be purely due to improved water relations of the mycorrhizal plants. Some external fungal hyphae may increase the total absorptive surface of the root system, comparable to the effect of an increased number of root hairs (Rozema et al, 1986). This may facilitate increased uptake of water.

The success of mycorrhizal plants could also be attributed to increased uptake of micronutrients. Work done on another species of Ehrharta, E. calycina by Killham and Firestone (1983) with heavy metal uptake is consistent with other reports that V A mycorrhiza increase the uptake of micronutrients in plants growing under conditions of micronutrient (especially phosphorous) deficiency. Killham and Firestone (1983) also considered that mycorrhizal mediation of uptake may well have been associated with an increased effective surface area of the

infected root and the consequent increased volume of soil from which metals (or micronutrients) could be extracted.

Despite this the most obvious advantage pypgrass has over the other plants present on the dunes is its extensive rhizome system. This spreads for many metres horizontally and occupies the top layers of soil (at least 50cm). The rhizome itself will allow pypgrass to take advantage of any available water and nutrients that may be found quite a distance away. Having said that, a mycorrhizal infection can only enhance this ability.

There is evidence to suggest that levels of mycorrhizal infection fluctuate throughout the year, when conditions or temperatures become unfavourable for the fungi (Ernst et al, 1984). It could be that the levels of mycorrhizal infection detected in the dune plants were in a low phase of infection and that the plants with little fungal infection were tested at the wrong time of year. Testing the plants all through the year would be the next step to take into account any natural annual fluctuations.

The particular mycorrhiza that is associated with the dune plants tested is unknown, but work done on many different mycorrhiza show that their effect on their plant hosts is generally similar (Rozema et al, 1986; Jackson and Mason, 1984; Safir, 1987) so the identity of the mycorrhizal species is not essential.

Identification of a V A mycorrhiza is difficult to achieve. Vesicular arbuscular mycorriza cannot as yet be isolated and

grown on sterile media (Powell and Bagyaraj, 1984). Therefore fruiting bodies, that may allow the mycorrhiza to be identified, cannot be obtained this way; identification is based on morphology of spores isolated from soil. The fruiting bodies of mycorrhiza do however appear on or in the dunes but cannot be unequivocally traced back to a host. Therefore even if a fruiting body is found under a pypgrass plant it cannot be said that the pypgrass is the host.

Transplanting an already mycorrhizal plant into a pot and growing it until fruiting bodies form in the pot would be the one way that the fungus could be identified but it may take many months before a fruiting body is formed (Hall, 1984). At this stage all that can be said is that the fungal partner in each case was endomycorrhizal (Hall, 1984).

### 5.2.5 Conclusions

Overall it appears that pypgrass has a high level of mycorrhizal infection compared to other dune plants. This could give pypgrass a competitive advantage over the surrounding vegetation in two ways: firstly by improved water relations, and secondly by increased micronutrient uptake. Both of these may be achieved by the fungus effectively increasing the rhizosphere for the plant. Of course the extensive rhizome system of pypgrass will allow the exploitation of greater areas for water and micronutrient uptake than are available to most other plants.

#### CONTROL OF PYPGRASS

## 6.1 CONTROL OPTIONS

A number of possible methods could be assessed for control of pypgrass in the Turakina area. Three common ones are mechanical methods, biological control and use of herbicide. These options are outlined below.

### 6.1.1 Mechanical Methods.

These methods generally involve some kind of cultivation carried out at a particular stage of the plant's life cycle that will achieve the best result. The types of equipment that are used for weed control include ploughs (which bury plants and seeds), rotary cultivators (which break up root systems), harrows (which uproot plants ready for removal) and hoes (which sever leafy plant parts from their roots) (Gwynne and Murray, 1985).

Unfortunately, these methods have some obvious problems associated with them. It would be very difficult to use a plough or any large tillage equipment between the trees in the forest. On the dunes cultivation would be very damaging to the native plants and would disturb the soil to such an extent as to jeopardise the stability of the dunes. The pypgrass itself with its extensive rhizome system defies mechanical methods of removal, as any pieces of rhizome left in the soil will act as a source of further infection when each fragment grows a new plant (see section 3.2).

# 6.1.2 Biological Control.

Biological control involves the introduction of antagonists, for example insect pests or diseases, to weed populations. Biocontrol has several advantages over other types of weed control, including fewer harmful side effects, attack narrowed to one specific target weed, agents which are self perpetuating and costs non-recurrent (Wapshere et al, 1989). However, this method has its drawbacks. It will involve extensive research over a long period of time with no guarantee of success (Wapshere et al, 1989). Additionally, complete eradication is impossible by the biocontrol method. The most that can be achieved is an equilibrium where the weed is less of a pest, although still present (Crafts, 1975).

## 6.1.3 Use of Herbicide.

Use of translocated herbicides is traditionally the best way of controlling perennial weeds. They are able to control underground parts of a plant, this being the extensive rhizome system in pypgrass.

There are other specific problems associated with the use of herbicides in this situation. The dunes cannot be left in an unstable condition, that is, without some vegetative cover. It should be possible with the correct herbicide to kill the target plant and leave most of the desirable vegetation. This requires the use of a selective herbicide.

Problems occur with the use of any herbicide where a small number of desirable species may be affected, however careful selection of the herbicide will minimise this risk.

The dunes and forest are open to the public, therefore any herbicide regiems will have to be carefully managed to ensure public safety.

So although herbicides have some drawbacks, compared with physical and biological control techniques they offer the best chance of eradicating pypgrass from the area.

# 6.2 HERBICIDE SELECTION.

Haloxyfop was the herbicide evaluated for controlling pypgrass. This is a selective, translocated herbicide of relatively low toxicity, making it ideal for the current situation. It was sold by Dow Elanco as Gallant but is now marketed as Gallant NF and is an aryloxyphenoxypropionate herbicide, a class of herbicides known to control rhizomatous grass species (Gronwald, 1994)

# 6.2.1. Activity in the Plant.

Gallant contains the ethoxyethyl ester of haloxyfop. Although the haloxyfop molecule is biologically active by itself it is commercially available in the ester form to facilitate application and uptake (Hendley et al, 1985). According to Harris (1987) haloxyfop effectively controls both annual and perennial grass species. Haloxyfop behaves like many of the aryloxyphenoxypropionates and penetrates the foliage rapidly

where it is hydrolysed to the acid form of the molecule. This now active molecule is able to be transported by the plant (McCall, 1988). Haloxyfop is readily absorbed by the roots although most of the herbicide will be taken up by the leaves in mature plants (Harris, 1987).

Plant growth is halted after application due to necrosis of meristematic regions, but because the herbicide is slow acting, no symptoms are visible for several days (Harris, 1987). Subsequently mottled chlorosis appears and spreads in developing leaves, with anthocyanin production becoming apparent in mature leaves (Gronwald, 1994). Plants later wilt as their tissues desiccate. The actual time taken for complete kill depends on the maturity of the plants at time of application and on environmental factors.

# 6.2.2 Activity in the Soil.

Rahman et al (1983) showed that haloxyfop can remain active in the soil for 6 to 14 weeks, but this constitutes a relatively short residual life. The soil mobility of the herbicide was tested in a sandy loam soil by James and Rahman (1990). They concluded that haloxyfop is only slightly to moderately mobile in the soil. The average distance leached by haloxyfop was shown to be 6.4 cm with the greatest distance leached by haloxyfop in any of the experiments only 7.5cm. They concluded that haloxyfop is unlikely to move horizontally off site or vertically any deeper than the root zone. Haloxyfop may be more mobile in pure

sand, but the leachate is unlikely to seriously affect the surrounding susceptible plants.

### 6.2.3 Mode of Action.

It has been shown that haloxyfop inhibits fatty acid biosynthesis (Cho et al, 1986) at relatively low (10<sup>-7</sup> to 10<sup>-6</sup>M) concentrations (Secor and Cseke, 1988). In investigations using maize suspension cells, it was shown that the herbicide target is an acetyl-coenzyme A carboxylase (ACCase) isoform, that is encoded by a nuclear gene (Gronwald, 1994). It appears that the haloxyfop molecule competes for the binding site on ACC that would normally be occupied by carboxyl transferase but only does this in monocotyledonous plants (Taylor et al, 1995).

The ACCase enzyme catalyses the carboxylation step in fatty acid biosynthesis (Gronwald, 1994). Fatty acids are needed for the formation of lipids (phospholipids, glycolipids and sterols) by the endoplasmic reticulum (ER). Lipids are major components of, and required for the growth of, cellular membranes (Salisbury and Ross, 1985). Therefore, if fatty acids are not synthesised, no lipids can be formed and the cell and its components cannot continue to renew their membranes.

### 6.2.4 Selectivity.

Grasses are susceptible to haloxyfop but most rushes and sedges and all dicotyledons are known to be resistant (Harris, 1987). Secor and Cseke (1988) initially reported that selectivity was probably due to variations in ACCase sensitivity to the

herbicide. However, it was later shown that selectivity depends on structural differences between dicotyledon and grass ACCase (Gronwald, 1994).

Haloxyfop was presumed to be effective on *Ehrharta* species since it shows very good control of other grasses with strong rhizome systems such as couch (*Elytrigia repens*), Indian doab (*Cynodon dactylon*) (Hartley, 1984) and Mercer grass (*Paspalum distichum*) (Rahman and Sanders, 1990). However it will not harm *Pinus radiata*, tree lupin (*Lupinus arboreus*) or any (native) dicotyledonous sand dune plants.

### 6.2.5 Translocation.

Hendley et al (1985) showed that the amount of radio-labelled haloxyfop translocated from primary shoots of couch to tillers and rhizomes was low (about 1% of applied chemical was recovered from the rhizome). However this small amount was still injurious to tillers and effective in controlling regrowth from the rhizome. As a comparison Tardif and Leroux (1991) found that around 2.7 to 5.3% of applied glyphosate was translocated into the rhizome of couch with variability resulting from differences in the plant growth stage. Glyphosate gives good control of couch rhizomes, so these results show that the proportion of chemical translocated does not have to be high to be effective.

Haloxyfop appears to be translocated in the phloem of couch and continues to be imported into the target area for up to 24 days.

All haloxyfop translocated into the target area contributes to the herbicidal effect (Wilhm et al 1986).

Gaskin and Woon (1994) also tested the ability of haloxyfop to be taken up and translocated within oat (Avena sativa). Close to 62% of applied herbicide was taken up and 7.1% of absorbed herbicide was translocated out of the leaf. The low translocation (as with other herbicides) may be explained by the very nature of haloxyfop inhibiting its own translocation by causing a disruption of plant transport systems (Gaskin and Woon, 1994). In spite of this, enough herbicide reaches the assimilate sinks to be phytotoxic (Hendley et al, 1985).

# 6.3 FIELD TRIALS

#### 6.3.1 Introduction.

Field trials were carried out with haloxyfop to confirm its effectiveness against pypgrass and to determine which month would be the best for spraying pypgrass. Trials also sought to determine if two (split) applications would provide better control than one single application.

### 6.3.2 Methods.

The trial site was situated in Compartment 130 of the Santoft Forest (South 40° 4" 30'; East 175° 8") (Fig 6.1) located 1km from the Turakina Beach village of Koitiata. Trials were not carried out on the population of pypgrass within the dune area, as vegetation in this area is recommended for protection by the

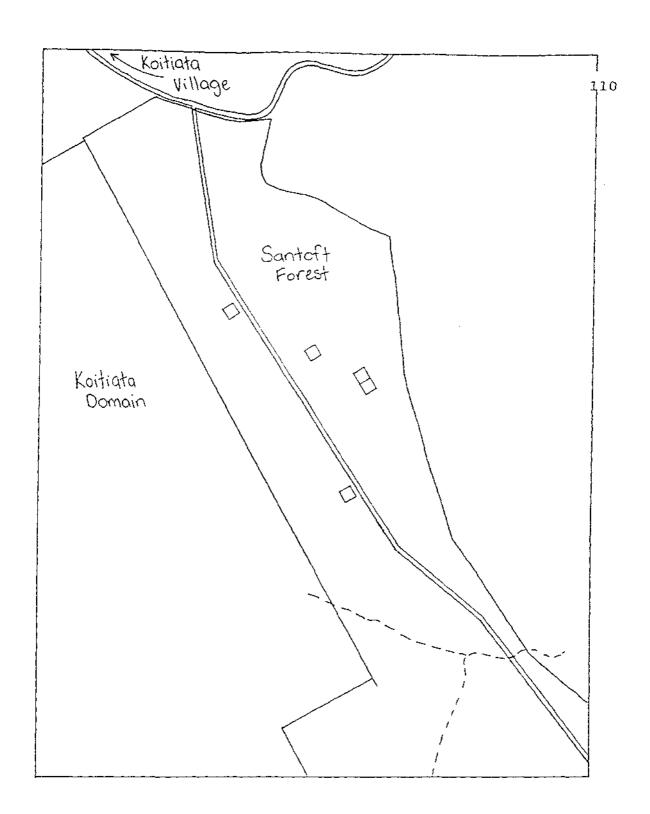


Figure 6.1. Compartment 130 of the Santoft Forest. The position of each trial block within the population of pypgrass is denoted by [].

Department of Conservation. The forest area also had more flat sites for locating plots, as well as reduced public access.

Five blocks, each containing eight trial plots (each 4.3m X 4m), were marked out in the area. This allowed for five replicates of each treatment, using a randomised block design. The herbicide that was used for the trial was Gallant (ethoxy ethyl ester of haloxyfop) which is no longer commercially available. Gallant NF (methyl ester of haloxyfop) was introduced in 1996, and has replaced the old formulation. Herbicide was applied at 1.0 kg.a.i./ha (except in Treatment 7 where it was applied at 0.5 kg.a.i./ha) with a propane powered precision sprayer set at 200kPa. The spraying oil Uptake was added at a rate of 1ml per 100mls spray mix (1%).

Treatments were applied between the 10th and 20th of every second month, as weather permitted. Temperature and relative humidity readings were recorded directly after application of haloxyfop as an assessment of conditions at the time of spraying (Table 6.1). No rain fell within 2 hours of application of haloxyfop on any day it was applied. There was a frost in the morning just before the August application (Treatment 4).

Assessments were made every two months for the duration of the trial. A visual assessment of live pypgrass cover was taken by scoring each plot from 0 to 10. "0" signified that no live pypgrass was visible in the plot and "10" represented dense coverage of pypgrass. The data collected from the vegetation

Table 6.1. Haloxyfop treatment of pypgrass in the Santoft
Forest made at various times throughout 1995.
Temperature and relative humidity readings were taken
directly after application of haloxyfop.

Treat	Date(s) Sprayed	Kg a.i. per ha	Day temp °C	Relative Humidity
1	10 Peb	1.0	25	64%
2	20 Apr, 14 Oct	1.0	24, 19	59%, 51%
3	20 Apr, 14 Oct	0.5	24, 19	59%, 51%
4	10 Jun	1.0	17	68%
5	19 Aug	1.0	15	66%
6	14 Oct	1.0	19	51%
7	19 Dec	1.0	23	48%
8	untreated control	_	_	

scores was analysed with an analysis of variance using the Student-Newman-Keuls multiple range test.

To help quantify the visual assessment, plots which had just received scores of 2, 5 and 8 each had four quadrats placed in them. These quadrats were then cut at ground level and the vegetation collected and sorted to exclude any material that was dead or not pypgrass. After drying at 85°C until the weight was constant, the final weight was taken and averaged. Plots scoring 2 had an average pypgrass dry weight cover of 783 kg/ha, those with 5 had 1706 kg/ha, and plots scoring 8 had an average cover of 2839 kg/ha. Photographs were also taken of each of the scores and are shown in Plates 6.1, 6.2, and 6.3.

Regrowth was assessed by inspecting the number of tillers emerging after control of the pypgrass in each plot. Plots were assessed for the first time when they had been treated two months previous, and were assessed every two months after that for the duration of the trial. They were assessed by randomly placing six quadrats of 1000 cm² (50cm X 20cm) in them and counting the number of regenerated tillers in the quadrat. This number was then averaged and expressed as the number of tillers found per square metre. This data was also analysed with an analysis of variance, using the Student-Newman-Keuls test.



Plate 6.1 Pypgrass in the pine population that had just received a score of 2 (0 = all dead, 10 = plants healthy).



Plate 6.2 Pypgrass in the pine population which had just received a score of 5 (0 = all dead, 10 = plants healthy).



Plate 6.3 Pypgrass in the pine population which had just received a score of 8 (0 = all dead, 10 = plants healthy).

#### 6.3.3 Results

# 6.3.3.1 Vegetation Scores

Table 6.2 shows the effect of haloxyfop on pypgrass as assessed by scoring for each treatment in February 1996. Figure 6.2 shows the effect haloxyfop had on pypgrass with each When sprayed in February or October, haloxyfop application. proves to have had maximum effect on pypgrass in two months. Every other treatment has had maximum effect on pypgrass by four months after spraying. A single application of haloxyfop did not achieve complete control of leaf and rhizome on any occasion, regardless of time of application.

Split applications also did not achieve complete control, however none of the plots began to recover from the spray during the trial period (Figures 6.2, 6.3). Considering the split applications, Figure 6.3 shows the effect the different rates had on pypgrass. There is a significant difference in effect between the two rates in the first two months after treatment, but after this time both rates control pypgrass similarly.

# 6.3.3.2 Regrowth Assessment

Irrespective of when haloxyfop was applied, there were very few new tillers present 2 months after a single application (Table 6.3). Four months after treatment however, tiller numbers had increased significantly, except where haloxyfop was applied in February or April. A split application ensured tiller regeneration remained low 4 months after the final application. The lower rate of haloxyfop (0.5kg a.i./ha) used in the split

Table 6.2. The effect of haloxyfop on pypgrass following application at various times throughout 1995, as assessed by scoring (0 = all dead, 10 = plants healthy) in February 1996. Means sharing the same letter are not significantly different at the 5% level.

Treatment	Average Score	Months Since Last Sprayed
1.February	0.7 b	12
2.Apr + Oct (1.0kg)	0.4 b	4
3.Apr + Oct (0.5kg)	0.5 b	4
4.June	1.3 b	8
5.August	1.3 b	6
6.October	1.1 b	4
7.December	2.6 b	2
8.Control	6.2 a	

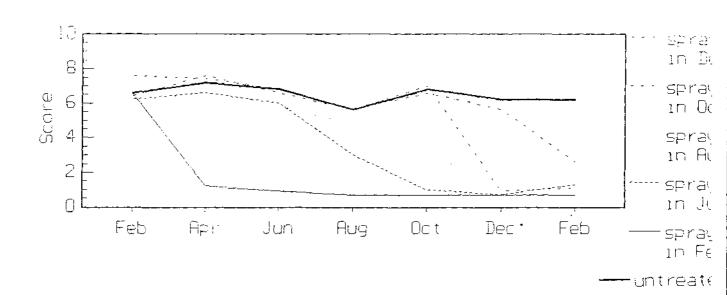


Figure 6.2. The effect of a single application of haloxyfop on pypgrass cover throughout 1995, as assessed by scoring (0 = all dead; 10 = plants healthy). No treatment was significantly different from any other (except the control) at the end of the trial (Feb 1996).

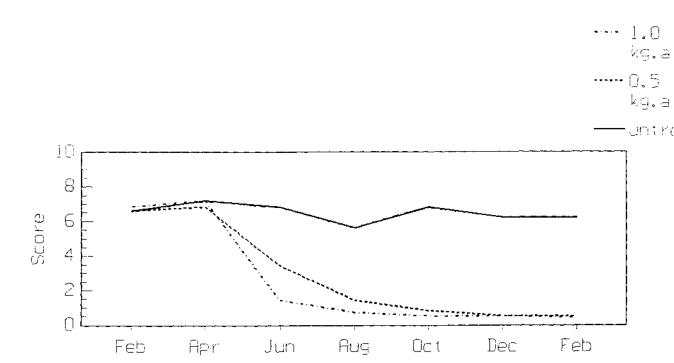


Figure 6.3. The effect of a split application (April and October) on pypgrass growth throughout 1995. Pypgrass growth was assessed by scoring where 0 = all dead and 10 = plants healthy. Both treatments were significantly different from the control but, neither treatment showed significantly greater overall effect from the other.

Table 6.3. Mean number of tillers regrowing per m² in treated plots, 2 months and 4 months after treatment. For treatments 2 and 3, the month underlined is the part of the treatment referred to in each case. Tiller numbers were counted using a randomly placed quadrat (6 replicates). No treatment was significantly different to any other 2 months after treatment, and means sharing the same letter 4 months after treatment, are not significantly different (p = 0.05).

	Number of new tillers/m <sup>2</sup>		
Treatment	2 months after treatment	4 months after treatment	
1.February	0.5	1.7c	
2. <u>Apr</u> +Oct (1.0kg)	0.6	0.7c	
3. <u>Apr</u> +Oct (0.5kg)	0.8	1.2c	
4.June	0.4	2.5bc	
5.August	3.2	5.0ab	
6.October	0.4	7.0a	
7.December	0.5	-	
2.Apr+ <u>Oct</u> (1.0kg)	0.5	0.5c	
3.Apr+ <u>Oct</u> (0.5kg)	3.0	2.5bc	

application did not allow significantly more regrowth than the higher rate (1.0kg a.i./ha) as can be seen in Table 6.3.

### 6.3.4 Discussion.

It appears that one strong dose of haloxyfop is not enough for complete control of pypgrass regardless of application time. Split applications also did not achieve complete control, although none of the plots fully recovered from the treatments. A single application of haloxyfop in February or April minimised tiller regeneration, as did split applications at both a high (1.0kg a.i./ha) and a lower rate (0.5kg a.i./ha).

Tiller regeneration was counted as though all tillers had regenerated from within the plot itself, but of course during this time rhizomes from the edge of the plots were growing inwards and would have eventually started to produce tillers also. As previously reported (see section 4.3), rhizomes can grow laterally 34 to 75cm per month, meaning they can easily reach the middle of a 4.3 X 4m plot in 3 to 6 months. To allow for this, results for up to four months after treatment were the only ones considered, and in this way I attempted to only count tillers emerging from rhizome fragments left after spraying.

June and August appear to be unsuitable months to apply haloxyfop. It was noted that in August there was a frost in the morning just before application which may have affected the uptake of the herbicide by pypgrass. Caseley (1987) reported that severe frost, which caused foliage necrosis, generally

resulted in poor control by glyphosate irrespective of the plant species. Furthermore, low temperature decreases the rate of chemical and enzyme reactions, thus decreasing the rate of haloxyfop uptake by pypgrass. However, the day temperature had reached 15°C after herbicide application, so temperature should not have affected uptake from this point.

It is more likely that pypgrass is in a period of slow growth during the winter. If plant metabolism is low then the rate at which herbicide has an effect on the plant will be low also (Caseley, 1987).

Observations suggest October to December is when pypgrass is growing most rapidly and so would be transporting a lot of assimilates to the rhizome and fertile tillers. Late February through to April is when pypgrass has the most leaf available on the plant (pers.obs). It is during these two stages of the plant's life cycle that pypgrass appears to be more vulnerable to haloxyfop.

Results suggest there is little difference between 0.5kg/ha and 1.0kg/ha haloxyfop used in a split application for control of pypgrass. Therefore, if a complete kill of pypgrass was required, I would recommend using 0.5kg haloxyfop/ha to decrease costs and to use it sequentially at least three times (eg: April, October, April).

With haloxyfop being no longer available in the ethoxyethyl ester formulation that was used in this trial, the new formulation (methyl ester) may not require a series of applications. Data published by Hendly et al (1985), showed that the methyl ester of haloxyfop eliminated shoot regrowth from rhizomes of couch where the ethoxyethyl ester of haloxyfop controlled only 50% of rhizome regrowth. Thus the new formulation of Gallant, currently marketed, may be more effective against pypgrass rhizome regrowth than the formulation used in this study.

# 6.4 POT TRIALS.

#### 6.4.1 Introduction.

Pot trials were carried out on some of the plants that grow on the dunes with pypgrass. These trials were to determine if haloxyfop would affect the other dune monocotyledons sufficiently that using this herbicide would be detrimental to the dune community.

### 6.4.2 Methods.

The species tested are listed in Table 6.4. Flax was bought from a nursery as established plants at a height of about 70cm. The pingao plants were obtained from the Kaitoki Prison Native Tree Nurseries. Marram and club rush were collected from the site and transplanted into pots where they were monitored for 2 months to ensure they were growing vigorously at the time of spraying. Spinifex was unable to be tested because transplanted plants

Table 6.4. Pot trials with haloxyfop and some dune species found at Turakina. An untreated control group of each species was compared to the plants treated as shown.

Species Tested	Common Name	Family	Rates Applied (kg a.i./ha)
Ammophila arenaria	Marram	Poaceae	1.0, 0.5
Phormium tenax	Flax	Agavaceae	1.0, 0.5
Desmochoenus spiralis	Pingao	Cyperaceae	1.0, 0.5
Isolepis nodosa	Club Rush	Cyperaceae	1.0

would not grow satisfactorily in pots and could not be obtained otherwise.

Haloxyfop was applied to each species using a laboratory pendulum sprayer similar to that described by Weise (1977). The sprayer used compressed air to propel the spray and the plants were positioned at the bottom of the pendulum arc, ensuring constant speed of the sprayer and even coverage of herbicide. The herbicide was applied at rates equivalent to 0.5 and 1.0 kg a.i./ha, at an application pressure of 200kPa, and there was also an untreated control. The spraying oil Uptake was added at a rate of 1ml per 100mls spray mix (1%).

Five plants each of flax, pingao and marram and four plants of club rush were used in each treatment. They were kept outside in sheltered conditions both before and after spraying on the 19th of February 1996. The plants were watered twice daily by an overhead sprinkler system and temperatures for the four months around the time of spraying fluctuated between 28.7°C and 3.4°C. All plants were potted in polythene bags (PB 6), flax and pingao were grown in commercial potting mix, with marram and club rush in beach sand fertilized by Osmocote.

Plants were scored in much the same way as for the pypgrass trial, and given a score from 0 to 10 where 10 meant the plant was completely healthy and 0 meant the plant was dead. A score of 5 would be given to a plant that had about half of its leaves dead or showing signs of irreversible damage. The data collected

was analysed with an analysis of variance, and means that were significantly different were separated using the Student-Newman-Keuls multiple range test.

#### 6.4.3 Results.

Marram, the only member of the Poaceae family tested, was the only species which was significantly affected by the haloxyfop two months after spraying (Table 6.5, Plate 6.7). The 1.0 kg a.i./ha application rate had no more effect on the species than 0.5 kg a.i./ha. However, even marram was not killed by the haloxyfop, though this was consistent with observations made of marram in the field plots (Section 6.3) where recovery from haloxyfop application occurred frequently. Scores for flax, pingao and club rush were unaffected by the haloxyfop applications (Table 6.5, Plates 6.4 - 6.6).

## 6.4.4 Discussion

As mentioned earlier haloxyfop controls many grass species but will not affect broadleaf plants and most rushes and sedges (Harris, 1987; O'Conner, 1993). The trial confirmed this by showing flax, pingao and club rush to be unaffected by haloxyfop. Marram was the only species affected by haloxyfop, which was expected in view of its taxonomic position. In spite of this, marram was not completely killed by either rate of haloxyfop, and will completely recover from the effects of the herbicide.

Spinifex is also in the Poaceae family so is expected to be affected by haloxyfop. However since it was not tested in this

Table 6.5. The effect of haloxyfop on some dune monocotyledons found at Turakina. Effect was assessed by scoring where 0 means plant completely dead; and 10 means plant healthy. Means in the same row sharing the same letter are not significantly different at the 5% level.

Species	Time of assessment	Untreated	0.5 kg/ha	1.0 kg/ha
Marram	At treatment	7.6a	7.6a	8.0a
	After 2 months	7.6a	5.6b	6.0b
Flax	At treatment	8.0a	8.0a	8.0a
	After 2 months	8.2a	7.8a	7.8a
Pingao	At treatment	7.8a	8.0a	8.0a
	After 2 months	8.6a	9.0a	9.0a
Club rush	At treatment After 2 months	8.3a 8.3a	-	9.0a 9.0a



Plate 6.4 Flax plants after the final assessment of the pot trial. The plant on the left (a) is from the control group. the other two plants pictured were given either 0.5 (b), or 1.0 (c) kg.ai/ha haloxyfop as part of the pot trial. It can be seen that neither of the treated plants has been damaged.



Plate 6.5 Pingao plants after the final assessment of the pot trial. The plant on the left (a) is from the control group. the other two plants pictured were given either 0.5 (b), or 1.0 (c) kg.ai/ha haloxyfop as part of the pot trial. It can be seen that neither of the treated plants has been damaged.



Plate 6.6 Isolepis plants after the final assessment of the pot trial. The plant on the left (a) is from the control group. the other plant was given 1.0 (b) kg.ai/ha haloxyfop as part of the pot trial. The treated plant has not been damaged.



Plate 6.7 Marram plants after the final assessment of the pot trial. The plant on the left (a) is from the control group. the other two plants were given either 0.5 (b), or 1.0 (c) kg.ai/ha haloxyfop as part of the pot trial. Both of the treated plants have been damaged, though not severely.

trial and no published data on its susceptibility could be found, it is unknown as to what extent it is likely to be affected.

Overall, if the population of pypgrass at Turakina Beach is to be controlled using haloxyfop then the dune area would not be left completely devoid of plants capable of keeping the sand dunes stable. The only plants that would appear to be affected by haloxyfop would be any introduced grass on the dune, including marram (which are not species recommended for protection) and the native sand grass, spinifex. Despite this, spinifex would be little affected since it only grows on one edge of the pypgrass population. Herbicide drift would be the only concern, but this could be avoided by using the herbicide only on days which are not windy.

A further aspect to consider is the use of a well understood translocated herbicide, glyphosate. Glyphosate is known to control a wide range of monocotyledonous and dicotyledonous plants as it is non-selective. Glyphosate will almost certainly control pypgrass, however it will also kill any other plant that the spray touches.

Glyphosate would be cheaper to use than haloxyfop (Many perennial grasses are controlled at 6L/ha Roundup (active ingredient, glyphosate) eg couch, which would cost \$89.00/ha. Use of Gallant (active ingredient, haloxyfop) at 5L/ha would cost \$211.00/ha) and it may be useful for clearing the tracks in the pine plantation however care would be needed to ensure none of the

spray or spray drift reaches the trees. Although glyphosate could be used cautiously in the pine plantation it would not be suitable for a broad spraying regime on the dunes, as all desirable plants would be destroyed along with pypgrass leaving the dunes with no stabilising vegetation. So although glyphosate would be more economical to use than haloxyfop, its usefulness would be severely limited by its selectivity.

## 6.5 CONCLUSIONS

It appears that haloxyfop is capable of controlling pypgrass but it would require a number of sequential applications. Furthermore, the native vegetation in the Koitiata domain would be safe from the effects of haloxyfop. Haloxyfop therefore shows good potential to be a useful herbicide in this situation.

## 7. CONCLUSIONS

Ehrharta villosa (pypgrass) belongs in the family Poaceae, subfamily Bambusoideae, and the tribe Ehrharteae. It is a perennial, rhizomatous grass native to Southern Africa. Pypgrass is widespread in South Africa and Australia but is found only at Turakina in New Zealand. Two populations are present, one in the Koitiata Domain, and the other in the Santoft Forest.

The plant community in the Koitiata domain at Turakina beach is important as it is not found elsewhere in the ecological district.

The growth form of pypgrass in New Zealand is that of a smothering sward whereas in South Africa, pypgrass grows as an open herb. It is this growth habit that appears to be affecting the plants on the dunes at Koitiata.

At least two other species of *Ehrharta* have been recorded as weeds in New Zealand and one particularly weedy characteristic of pypgrass is its apparently high ploidy level.

The seed production of the New Zealand populations of pypgrass was investigated and was found to be very low. This means pypgrass may not rely on seed for reproduction. The differences seen in seed production between the two populations can be attributed to floret infertility, under the influence of two strong factors, water stress and mineral supply.

Tiller fertility is affected in both populations by mineral supply, and light intensity. Both populations appear to be similarly affected by mineral supply.

Pypgrass seedlings are most likely to occur within five metres of either population, and most will be found to the north of the beach population. However, heavy predation of the seed seems to prevent a seed bank forming, so no seedlings may be found at all. It appears that seed dispersal in pypgrass is not an important factor in the plants survival and spread.

A germination trial showed pypgrass seeds develop best when planted at only one to two centimetres below the surface of the soil. However, seeds can emerge from burial at six centimetres. The internodes of pypgrass elongate, from the time of development of the fourth leaf. Although pypgrass is considered to be a cool-temperate grass, elongated internodes without floral initiation, are a common feature in tropical grasses.

Pypgrass will tiller when six leaves have been produced, but the seedlings are slow to develop, and lack the vigour of other grasses and weeds. Seedling development was not found to be important in the success and spread of the pypgrass population.

Studies of the leaf anatomy of pypgrass have confirmed that pypgrass is a C3 plant with distinctive stomata, which are sunken and over-arched by four cuticular flanges. This arrangement of

the stomata would be expected to reduce loss of water from the leaf and be of selective advantage in a dry environment.

The ability of pypgrass to regenerate from rhizome was investigated. Rhizome fragments of pypgrass are capable of producing a new plant after being detached from the parent plant. They cannot do this if they dry out, but enough moisture can be retained to allow growth if the fragment is buried. The buds closest to the apical end most often produce shoots.

A study of the edge of one population was undertaken to determine the rate of vegetative spread of pypgrass. A true measure of the rate of rhizome growth was not taken because of difficulties involved with excavating the deep rhizome system, without disrupting the natural growth of the rhizomes. However, it was found that pypgrass can spread by rhizome growth by between 34 and 75cm per month.

Species growing on the dune could be affected by the spread of pypgrass. Competition for space and water by pypgrass rhizomes is the most likely factor in suppressing the growth of other plants. Alternatively, an allelopathic interaction with pypgrass, may account for the changes seen in species composition. However, there is a clear possibility that unrelated (physical) factors have also influenced the species composition between 1994 and 1995.

Mycorrhiza may have an important role to play in the success of sand dune plants, as mycorrhizal plants have been shown to be tolerant of drought and saline soils (Schenck, 1982., Crawley, An experiment to determine the level of mycorrhizal infection of pypgrass and dune plants associated with it showed that pypgrass has a high level of mycorrhiza associated with it, compared to other dune plants. Mycorrhiza could give pypgrass an advantage over the surrounding vegetation in two ways: firstly improved water relations, and secondly by increased micronutrient uptake. Both of these may be achieved by the mycorrhiza effectively increasing the rhizosphere for the plant.

Finally, options were explored for the control of pypgrass on the dune. Use of herbicide was discovered to be the best choice of control methods.

The herbicide chosen for trials with pypgrass was Gallant, known to control rhizomatous grass species.

Gallant contained the ethoxyethyl ester of haloxyfop. According to Harris (1987) haloxyfop effectively controls both annual and perennial grass species. Haloxyfop penetrates the foliage rapidly where it is hydrolysed to the acid form of the molecule. This now active molecule is able to be transported by the plant.

It has been shown that haloxyfop works by inhibiting fatty acid biosynthesis. The herbicide target is an acetyl-coenzyme A carboxylase (ACCase) isoform. The haloxyfop molecule competes for the binding site on ACC that would normally be occupied by

carboxyl transferase but only does this in monocotyledonous plants (Taylor et al, 1995). This selectivity of haloxyfop depends on structural differences between dicotyledon and monocotyledon ACCase (Gronwald, 1994).

Haloxyfop appears to be translocated in the phloem and the proportion of chemical translocated does not have to be high to be effective.

Field trials on pypgrass in the Santoft forest showed that haloxyfop is capable of controlling pypgrass but it would require a number of sequential applications.

Pot trials were carried out on some of the monocotyledonous plants living in contact with pypgrass. This trial established that native vegetation in the Koitiata domain would be safe from the effects of haloxyfop. In this situation, haloxyfop has the capacity to be a useful herbicide against pypgrass.

The Department of Conservation (DOC) is about to start a herbicide regime based on these findings in an attempt to control the beach population of pypgrass.

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