A stylized green illustration of a plant with three upright seed heads and several long, pointed leaves. The plant is positioned on the left side of the cover, set against a background of horizontal yellow and white stripes.

Reclamation Studies On Oil Shale Lands In Northwestern Colorado

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Reclamation Studies On Oil Shale Lands In Northwestern Colorado

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FOREWORD

This is a fourth-year progress report for the period June 1, 1979, to May 31, 1980. The results that follow represent an integrated approach to answering mined land rehabilitation questions associated with oil shale development in northwestern Colorado. Researchers from the disciplines of range science, ecology, soil science, agronomy, botany, mycology, and microbiology have been brought together to address some of the more pertinent problems associated with disturbed land reclamation. This report is intended to provide information to government agencies and private industry to assist in preparing (1) long-term development plans, (2) rehabilitation guidelines, and (3) approaches to reclaiming oil shale lands. Included herein are results and tentative conclusions from research in progress.

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ABSTRACT

The overall objective of this project is to study the effects of various reclamation practices on above- and belowground ecosystem development associated with disturbed oil shale lands in northwestern Colorado. Plant growth media that are being used in field test plots include retorted shale, soil over retorted shale, subsoil materials, and surface disturbed topsoils. The project was initiated in June 1976 and is presently reporting on fourth-year data from both field and laboratory studies.

The majority of research reported on within this report is being conducted on a 20-ha Intensive Study Site located near the focal points of oil shale activity in the Piceance Basin. The site is at an elevation of 2,042 m, receives approximately 30 to 35 cm of annual precipitation, and encompasses the plant communities most typical of the Piceance Basin.

Some of the more significant results that are reported herein are (1) a soil cover of at least 61 cm in conjunction with a capillary barrier provided the best combination of treatments for the establishment of vegetation and a functional microbial community, (2) aboveground production values for native and introduced species mixtures are

comparable after three growing seasons, (3) cover values for native species mixtures are generally greater than for introduced species mixtures, (4) native seed mixtures, in general, allow greater invasion to occur than introduced mixtures, (5) sewage sludge at relatively low rates appears to provide the most beneficial overall effect on plant growth of any fertilizer treatment tested, (6) cultural practices, such as irrigation, mulching, and fertilizing, have significant effects on both above- and belowground ecosystem development, (7) topsoil storage after 1.5 years does not appear to significantly affect general microbial activities but does reduce the mycorrhizal infection potential of the soil at shallow depths, (8) populations of mycorrhizal fungi are decreased on severely disturbed soils if a cover of vegetation is not established, (9) significant biological differences among ecotypes of important shrub species have been identified and will assist in selecting appropriate plant materials for reclamation, (10) a vegetation model is outlined which upon completion will enable the reclamation specialist to predict the plant species combinations best adapted to specific reclamation sites, and (11) synthetic strains of two important grass species are close to development which will provide superior plant materials for reclamation in the West.

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EFFECT OF PLANT SPECIES, SOIL MATERIAL, AND CULTURAL PRACTICES UPON PLANT ESTABLISHMENT AND SUCCESSION

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OBJECTIVES

This subproject has several related objectives all directed toward the establishment of a diverse, functional, and self-sustaining ecosystem in as short a time as possible. The inputs of this subproject will eventually serve to provide procedures for establishing an effective plant community on lands disturbed by mining activity. Various plant species mixtures, planting techniques, fertilization treatments, irrigation, and mulches are being tested on disturbed soils, retorted oil shale, and soil over retorted shale material. The specific objectives of the research include: (1) determining proper seeding practices of potentially usable mixtures of plant species and their relationship to cultural practices such as fertilizing, mulching, and irrigation, (2) determining the rate and direction of plant succession on disturbed topsoil, subsoil, overburden, retorted shale, and retorted shale overlain by soil as influenced by natural invasion, seeding mixtures, and cultural practices, and (3) monitoring vegetation reference areas to assist in determining revegetation success.

PROGRESS TO DATE

Retorted Shale Succession Study

The Retorted Shale Succession Study was initiated in 1977 to evaluate the effect of proposed surface disposal plans for retorted shale on plant growth and succession. To evaluate these plans, large-scale artificial profiles were constructed to simulate field conditions that would result from different disposal methods. The profile configurations are as follows:

1. Process shale to a depth of 61 cm without any surface covering
2. 30.5 cm soil over retorted shale
3. 61 cm soil over retorted shale
4. 91.5 cm soil over retorted shale

5. 61 cm soil over 30.5 cm rock capillary barrier over retorted shale
6. Soil check with no retorted shale in the profile

All shale treatments in the profiles received 61 cm of Paraho (direct mode process) retorted shale from the Anvil Points Facility near Rifle, Colorado. In addition, three seeding mixtures were studied consisting of diverse combinations of grass, forb, and shrub species in either an all native mixture, all introduced mixture, or a combination of the native and introduced mixtures. Fertilizer treatments consisted of two levels of nitrogen and phosphorus and a control. N and P were applied in combination at the following rates: Treatment 1--112 kg N/ha and 56 kg P/ha, Treatment 2--56 kg N/ha and 28 kg P/ha, and Treatment 3--0 kg N/ha and 0 kg P/ha.

Results and Discussion

The results presented herein are from data taken during the 1979 growing season. It should be noted that the plant community is two years old and still in the early stages of succession. Despite this fact, significant responses have been observed that may dramatically affect reclamation practices associated with the disposal of retorted shale.

Effect of Paraho Retorted Shale

The most significant result from this study is the complete failure of vegetation to establish on the shale-to-surface treatment. This treatment was drill seeded and fertilized but received no supplemental water for leaching or irrigation. This approach is consistent with our goal of minimizing the inputs of scarce resources.

The vegetation failure on the shale-to-surface treatment can be explained by the many chemical and physical problems associated with Paraho retorted shale as a plant growth medium. The retorted shale and soil used in this study were analyzed for those chemical properties which might have the greatest effect on plant growth (Table 1).

Results obtained from the soil analysis procedures on retorted shale are limited in their

Table 1. Chemical characteristics of Paraho retorted shale and soil used in the Retorted Shale Succession Study.¹

Chemical Characteristics	Retorted Shale	Soil Cover
<u>Measurement</u>		
pH	9.6	8.0
EC, mmhos/cm @ 25°C ²	7.0	1.6
SAR	14.0	6.8
<u>Cations meq/l²</u>		
Ca	21.0	3.3
Mg	3.9	2.1
Na	49.2	11.0
K	4.7	0.1
P, ppm	2.1	1.4
K, ppm	387.0	77.4
NO ₃ -N, ppm	4.0	11.4
Texture	Gravelly Silt Loam	Silt Loam

¹Values given are the means of 21 replications for retorted shale and 12 replications for the soil.

²Retorted shale analyses were on a 1:1 extract, and soil analyses were on a saturation extract.

applicability. These soil testing procedures are designed specifically for use on natural soil material. During the retorting process the raw shale is heated to extremely high temperatures (650-800°C) and considerable chemical and physical alteration of the shale takes place. This heating creates extreme conditions that cause many complex chemical reactions that cause unusual chemistry in the retorted media. Because of the problems associated with the analysis of retorted shale research work needs to be implemented to determine the usefulness of soil tests in evaluating retorted shale as a plant growth medium.

The pH of Paraho retorted shale is high and has a detrimental effect on plant growth. The California Fertilizer Association (1975) states that the pH affects nutrient availability and that at high pH values nutritional imbalances occur. Black (1968b) reported that deficiencies of phosphorus, iron, and zinc are fairly common at high pH, and this may result in reduced growth. In a review of the literature, Justice and Reece (1954) found that high pH values may inhibit seed germination.

The soluble salt content of the retorted shale, as expressed by electrical conductivity (EC), is considered high enough to present plant growth problems. A soil is considered saline when the EC of a saturation extract is greater than 4 mmhos/cm. Soluble salts increase the osmotic

potential in the soil solution, and this in turn increases the moisture stress on the plant. McGinnies (1960) found in his work with six range grasses (all of which were used in this study) that as moisture stress increased, total germination was reduced. Richards (1954) reported on several studies which found that increases in the osmotic potential resulted in decreased water availability to the plants and thus a decrease in plant growth.

The Sodium-Adsorption Ratio (SAR) indicates that the processed shale has a medium sodium hazard. Large amounts of sodium in a soil are known to have a deleterious effect on plant growth. Bernstein (1962) reviewed the literature and reported that, in general, as the sodium content in soils increased, plant yields decreased. Bower and Wadleigh (1949) found that high levels of sodium decreased the uptake of calcium, magnesium, and potassium by the plants and depressed yields. Soil dispersion, normally associated with sodic soils, should not be a problem with retorted shale because of its coarse texture.

The fertility status of the retorted shale is not favorable for plant establishment or growth. The material contains adequate amounts of potassium but is extremely low in nitrogen and phosphorus. A recent greenhouse study at Colorado State University indicates that high levels of nitrogen and phosphorus (112 kg/ha and 448 kg/ha, respectively) may be necessary for initial plant establishment.

The black color of the shale material resulted in surface temperatures in excess of 65°C during the first growing season. McDonough (1977) reported that the optimum temperature range for the germination of most desirable range species is between 10° and 30°C.

From the previous discussion, it can be seen that vegetation failed to establish on the retorted shale for a combination of reasons. This past fall the Shale-to-Surface Plot was reworked in hopes of making it more amenable to plant growth. Fertilizer applications at the rate of 112 kg N/ha and 448 kg P/ha were applied in combination to overcome nutrient deficiencies. The entire plot was reseeded with three mixtures, one of which was composed entirely of salt-tolerant species. A seed-free straw mulch was spread over the shale following seeding to reduce surface temperatures during the growing season. Results from this work will be monitored in 1980.

Effect of Soil Placed Over Retorted Shale.

The amount of topsoil placed over retorted shale had a pronounced effect on vegetation growth, and this is expected to become more significant as the communities develop. There was a substantial increase in production and cover of seeded vegetation from the 30.5 cm to the 61 cm of soil placed over retorted shale, but no general trend at the deeper depths after two years of growth. However, most reseeded areas take at least three years to become fully established. Thus, it is expected that soil depth may play a much greater role next year as the plant roots begin to fill the soil profile.

Across all soil-shale treatments only the native seed mixture is exhibiting decreasing

production and cover with decreasing soil depth. This response may in part be caused by the different root growth patterns of native and introduced species. While introduced species generally exhibit earlier tiller production and thus a branching growth pattern that concentrates initial root growth in the upper reaches of the profile, native species tend to put more growth into the primary root system which results in deeper root penetration of seedlings. Thus, native species may initially be more sensitive to soil depth over retorted shale than introduced species.

Other factors besides vegetation growth need to be considered in evaluating the soil depth treatments. Lindsay (1979) found that plants growing on the retorted shale study area are accumulating molybdenum at levels that are toxic to livestock. Molybdenum concentrations are highest in vegetation on the 30.5 cm soil cover over retorted shale and decrease with increasing amounts of soil over cover (Table 2). Hersman and Klein (1979) investigated in a laboratory study the effects of retorted oil shale additions on the microbiological characteristics of surface soils. With retorted shale present up to 10 percent by weight, significant reductions in nitrogen fixation rates, dehydrogenase activity, fungal populations, glucose mineralization, and ATP concentrations occurred. They concluded that soil depth over retorted shale must be sufficient to allow plants to establish a functional decomposer community in the plant root zone without being influenced by the underlying shale material.

When root samples were taken on the soil-shale treatments, it was observed that roots from all plants growing in soil cover displayed normal morphology while those growing into the shale were dramatically altered (Figure 1). Root penetration from the soil to the shale was limited to the upper 8 cm of the retorted shale, and root growth became extremely diffuse and less vigorous in the shale material. This same type of root growth is observed with plants growing in highly saline media. This root growth phenomenon will be monitored closely in succeeding growing seasons.

Effect of Seed Mixture

The continuing controversy over the use of native versus introduced species has increased the importance of seeding trials in reclamation studies. In this study the native seed mixture exhibited

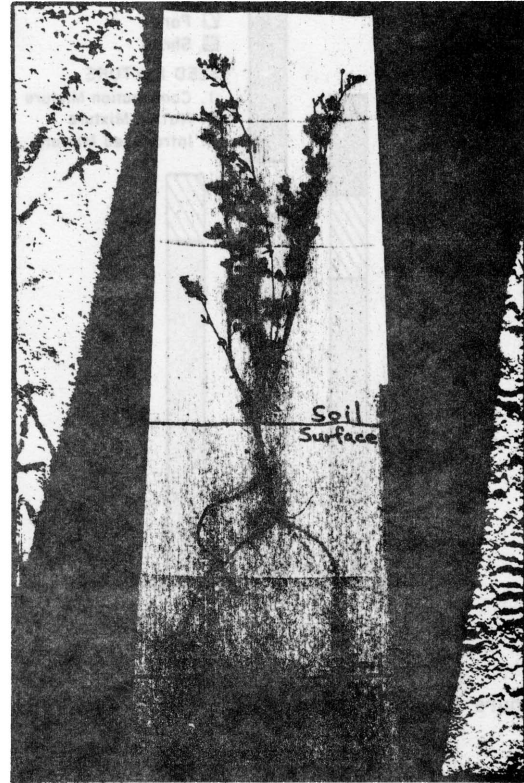


Figure 1. Root morphology of Ladak alfalfa following excavation from a soil-shale profile with 30 cm (1 ft) of soil over 60 cm (2 ft) of retorted shale.

significantly higher ($p < .0001$) percent cover values than either the introduced or the combination native and introduced seed mixtures (Figure 2). Breaking the total down by life form, it is observed that the native seed mixture produced a higher seeded grass cover than the combination mixture or the introduced mixture. Grass species which were primarily responsible for the high cover values in the native mixture were Rosana western wheatgrass (*Agropyron smithii*), Sodar streambank wheatgrass (*Agropyron riparium*), and beardless bluebunch wheatgrass (*Agropyron inerme*). Introduced grasses which showed good cover values included Nordan crested wheatgrass (*Agropyron cristatum*) and Oahe intermediate wheatgrass (*Agropyron intermedium*).

Table 2. Elemental analysis of vegetation growing on the Retorted Shale Succession Study.

Soil Cover Over Shale	Molybdenum (ppm)		
	Western wheatgrass (<i>Agropyron smithii</i>)	Utah Sweetvetch (<i>Hedysarum boreale</i>)	Fourwing Saltbush (<i>Atriplex canescens</i>)
30.5 cm	2.1	26.0	8.7
61.0 cm	0.7	9.7	9.0
91.5 cm	0.7	6.4	4.2
61.0 cm over 30.5 cm gravel	0.6	3.1	2.4
Soil check	1.2	2.9	1.1

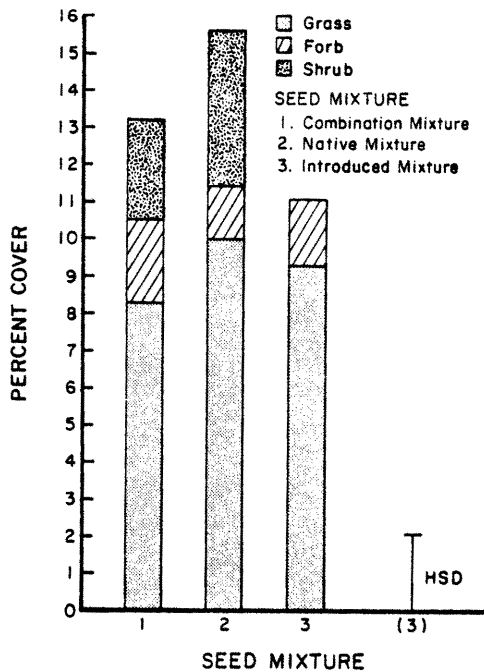


Figure 2. Mean cover of seeded species by life form for the three seeding mixtures across all fertilizer and soil-shale treatments.

The seeded forb cover demonstrated a different trend from the grasses in that the combination seed mixture produced significantly higher ($p < .005$) values than the native seed mixture. This was a result of the poor performance of the native forbs and the tremendous growth of Madrid yellow sweet-clover (*Melilotus officinalis*) in the combination seed mixture. This plant is an aggressive introduced species which establishes well on disturbed sites; but, since it is a biennial, it often drops out of the plant community after a few years.

The native seed mixture had a significantly greater ($p < .05$) seeded shrub cover than any of the three seed mixtures. The high shrub cover values in the native mixture can be attributed to the excellent growth of fourwing saltbush (*Atriplex canescens*) and winterfat (*Ceratoides lanata*). Although both these species were also present in the combination seed mixture, their growth appeared reduced by competition from Madrid yellow sweet-clover. Figure 2 also points out the complete failure of the introduced shrub species, Siberian peashrub (*Caragana arborescens*) and Russian olive (*Elaeagnus angustifolia*), to establish from seed. Research work needs to be implemented to improve the selection and establishment of both native and introduced shrub species.

Total cover of invading species (those not seeded) was significantly higher ($p < .01$) on subplots seeded with the native mixture than on subplots seeded with either the introduced or combination mixtures (Figure 3). Robertson and Pearse (1945) state that the amount of invasion in a seeded stand is an indication of how fully the established vegetation is utilizing the resources present. After two years the native seed mixture is still relatively open to invasion whereas the introduced seed mixture is more fully occupying all of the space

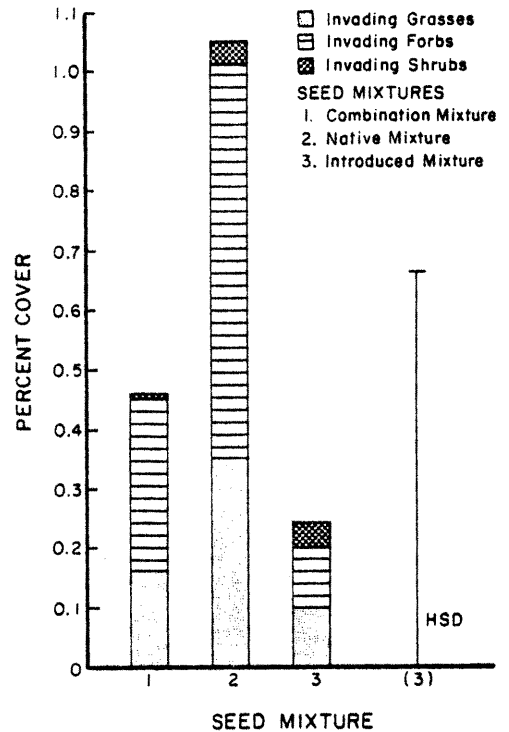


Figure 3. Cover of invading species by life form for the three seed mixtures across all fertilizer and soil-shale treatments.

and utilizing soil and moisture resources. This difference may be partially attributed to the concentration of roots near the surface by introduced species which leaves a smaller amount of area open to invasion. The primary invading species on the Retorted Shale Study are Russian thistle (*Salsola iberica*) and cheatgrass (*Bromus tectorum*).

Effect of Fertilizer

The addition of nitrogen and phosphorus fertilizer increased the biomass of seeded species across all three seeding mixtures (Figure 4). This increase in biomass with greater fertilizer additions was almost totally the result of increased grass production. Black (1968a) found that increased grass yield resulting from fertilization could be closely correlated with an increase in depth and surface area of root growth. This resulted in the grasses being able to extract more moisture from the subsoil.

Fertilization treatments resulted in a decrease in the biomass of seeded forbs. The control treatment (no fertilizer) had significantly greater ($p < .0004$) biomass than either of the nitrogen and phosphorus fertilizer treatments. This increase in forb biomass with no fertilization is presumably caused by a reduction in grass competition and the presence of nitrogen-fixing legumes. Hodder (1978), working on surface-mined lands in Montana, found that nitrogen-fixing legumes exhibited the greatest development under low or no fertilization treatments. It appears that under fertilized conditions nitrogen-fixing legumes lose a great deal of their competitive advantage.

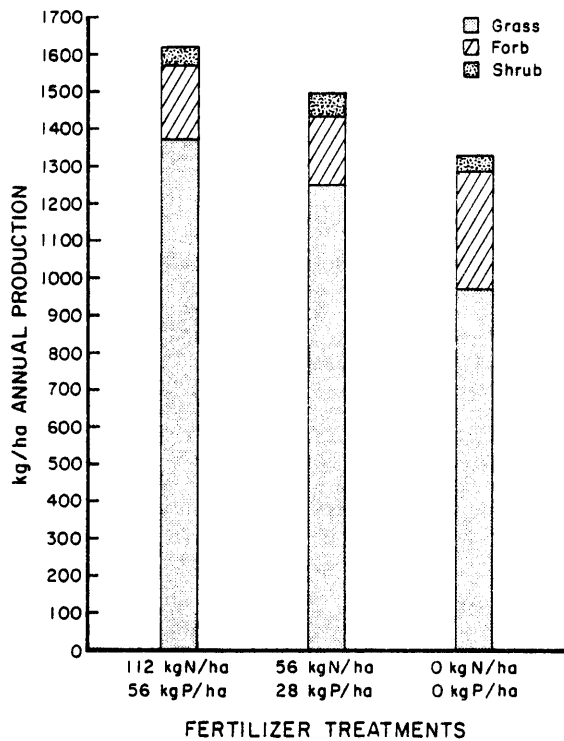


Figure 4. Current annual production of seeded species by life form for the three fertilizer treatments across all seed mixtures and soil-shale treatments.

Conclusions

The following conclusions are based on data taken during the second growing season. As the vegetation on the study develops successfully and roots fill the suitable plant growth medium, changes in these results may occur.

1. Paraho retorted shale cannot be directly revegetated within an environmentally acceptable time without large amounts of resources and management.
2. The most feasible approach to the surface stabilization of Paraho retorted shale is to cover the material with an adequate amount of suitable soil or geologic material.
3. Deeper soil coverings of 61 and 91.5 cm over retorted oil shale may be necessary to establish diverse and productive above- and belowground ecosystems and to minimize the harmful effects of toxic elements.
4. The native seed mixture is exhibiting the highest seeded species cover as a result of the excellent growth of native grass and shrub species.
5. Cover of invading species is highest on the native seed mixture and lowest on the introduced seed mixture perhaps as a result of less competition for moisture near the soil surface.

6. Fertilization with nitrogen and phosphorus increased grass production but resulted in a significant decline in forb production and had no effect on shrub growth.

Revegetation Techniques on Intensively Disturbed Soils

During the fall of 1976 a study was initiated to evaluate various revegetation techniques on a series of intensively disturbed plots. The plots were disturbed by thoroughly mixing the topsoil and subsoil material to a depth of 1 m. This level of disturbance was designed to simulate a plant growth medium that would result when topsoil was not conserved or simply not present in sufficient quantities during oil shale development.

The establishment of a diverse and functional ecosystem on this material was believed to initially require a certain level of cultural input. Revegetation of this heterogeneous topsoil-subsoil mixture is difficult as it presents a coarse, stony, and nutrient deficient seedbed and root zone profile. The objective of the study is to evaluate various cultural practices for their ability to aid in the reclamation of an intensively disturbed area. Seeding techniques being studied are as follows:

- Technique 1: Drill mixtures of grasses, forbs, and shrubs at a rate of 17 kg/ha.
- Technique 2: Drill mixtures of grasses, forbs, and shrubs at a rate of 19 kg/ha with an increased amount of shrub seed.
- Technique 3: Drill grasses and forbs at a rate of 17 kg/ha.
- Technique 4: Broadcast grasses and forbs at a rate of 29 kg/ha and lightly cover with soil.

Three seeding mixtures all containing a diverse combination of grasses, forbs, and shrubs were evaluated in the study. They are: (1) a combination mixture of native and introduced species, (2) a native species mixture, and (3) an introduced species mixture. Each of the three mixtures was seeded with the four seeding techniques above. Fertilizer treatments consisted of one level of nitrogen and one level of phosphorus in combination compared with a control. Wood fiber mulch was applied over the entire study at a rate of 2.2 MT/ha. There was no control for mulch comparisons.

Irrigation was implemented on one-half of the experimental plots during the first two growing seasons following seeding (1977 and 1978). The amount of water applied approximated the highest amount of precipitation that would be expected to occur at the site based on 20 years of previous data. In 1979 the irrigation treatment was discontinued as planned. Comparison of third-year data from the study should be used to demonstrate the effects of applying supplemental water to aid in vegetation establishment.

Results and Discussion

It should be emphasized that the results presented in the following section are after the third growing season. Thus, the plant communities established under the various cultural inputs are still in the early stages of succession and may be subject to change in subsequent years. Since it is generally accepted that it takes three years for seeded vegetation to become established, the results have considerable practical significance in the area of vegetation establishment.

Effect of Seeding Technique

The development of oil shale in the Piceance Basin will result in the disturbance of many shrub dominated communities. These communities provide essential forage and cover for mule deer during the winter and, in addition, are important habitat for many birds and small mammals throughout the year. Thus, it is important that shrubs be successfully reestablished on reclaimed lands.

Seeding Techniques 1 and 2 were utilized in this study to evaluate the effect of a higher shrub seeding rate on shrub establishment. It was expected that more shrubs would be established with Technique 2; however, Table 3 shows that only under non-irrigated conditions did Technique 2 produce a significantly greater ($p < .05$) number of shrubs with the combination mixture. Shrubs that performed well were fourwing saltbush and winterfat with lesser success being achieved with Stansbury cliffrose (*Cowania mexicana stansburiana*) and green ephedra (*Ephedra viridis*). The mean density of shrubs for all three seeding mixtures was higher under non-irrigated conditions as compared to irrigated conditions. The reason for this response is not clear at this time, but it may be related to the increased competition attributed to the grass component.

Table 3. Mean density of seeded shrubs (plants/m²) after three years in response to seeding technique, species mixture, and irrigation on intensively disturbed soil.

Seed Mixture	Seeding Technique			
	Non-Irrigated		Irrigated	
	1 ¹	2 ²	1 ¹	2 ²
Combination	4.56	6.88	2.80	3.00
Native	3.68	2.36	1.80	2.68
Introduced	0.24	0.00	0.00	0.12

¹Grasses and forbs drilled at a higher rate than shrubs.

²Shrubs drilled at a higher rate than in Seeding Technique 1.

Table 3 also points out the total failure of the introduced shrubs, Siberian peashrub and Russian olive, to become established regardless of seeding

technique or irrigation treatment. These findings suggest that if introduced shrub species are desired on reclaimed areas in the Piceance Basin, then (1) their seeding rates need to be increased or (2) they need to be established from transplants or (3) different shrub species with a greater chance of success need to be utilized.

With Technique 1 shrub cover was significantly lower ($p < .02$) on irrigated than on non-irrigated plots (Figure 5). Shrub density values were similar under both treatments and thus fail to explain the observed difference. However, the difference may be explained by an increase in seeded grass and forb density, biomass, and cover under irrigated conditions. This resulted in an increase in competition for the shrubs by the grass and forb component. Humphrey (1959) and McGinnies and Arnold (1939) found that grasses and forbs utilize water more efficiently than shrubs and that this may result in greater grass and forb production. Despite the increase in grass and forb density, cover,

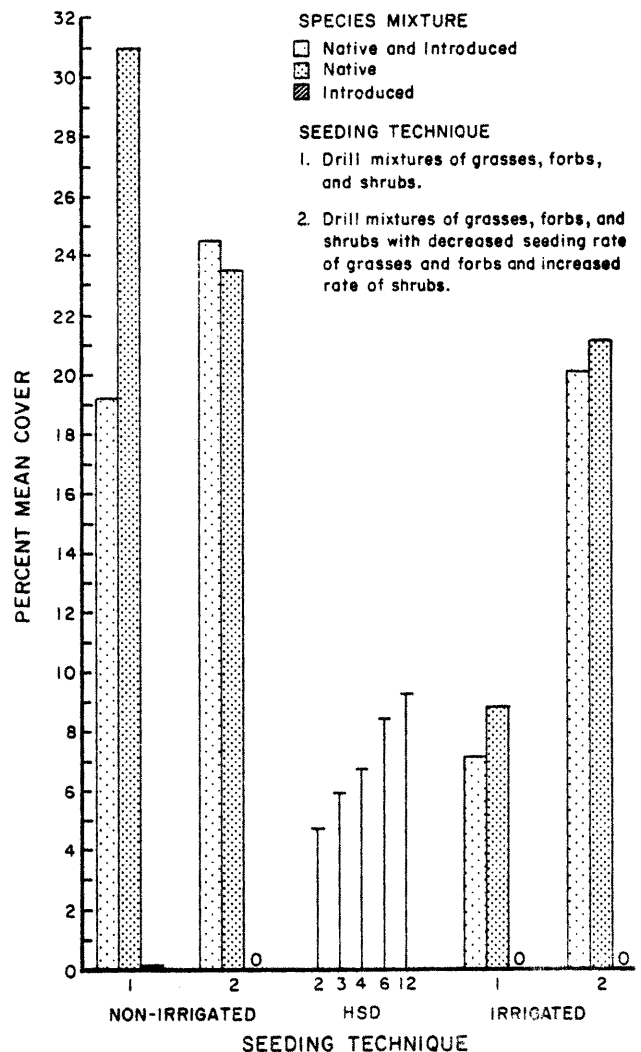


Figure 5. Mean cover of seeded shrubs in response to seeding technique, species mixture, and irrigation on Intensively Disturbed Soils.

and biomass a similar decline in shrub cover from non-irrigated to irrigated treatments was not observed on subplots seeded by Technique 2. This response with Technique 2 cannot be explained at this time.

Native shrubs, when seeded, provided at least 50 percent of the total vegetation cover after three years across irrigation treatments. Shrub cover was significantly higher ($p < .02$) on non-irrigated subplots seeded with the native mixture using Technique 1 (Figure 5) than under any other combination of treatments. The establishment of shrub cover proved most effective under this combination of cultural practices.

Seeding Techniques 3 and 4 were utilized in this study to evaluate the methods of drilling versus broadcasting. Drilling is acknowledged as the superior method for establishing vegetation because seed is planted and covered at a uniform depth. Broadcasting is considered to be less favorable because seed left uncovered at the soil surface is subject to environmental hazards such as desiccation and wind and water erosion. The drawbacks inherent with broadcasting can be partially overcome by doubling the seeding rate and using a supplemental treatment to cover seed (e.g., raking or harrowing).

Grasses and forbs seeded in Technique 3 were drilled. Grasses and forbs seeded in Technique 4 were broadcasted at approximately twice the seeding rate used in Technique 3. Following broadcasting, the seedbed was lightly raked to increase the probability of the seed being covered with soil. After comparing these two techniques, it was found that vegetation cover was not significantly different regardless of seeding mixture or irrigation treatment utilized. The density of total seeded species was significantly higher ($p < .0001$) on subplots which were broadcast seeded as compared with those subplots which were drill seeded (Table 4). These results imply that broadcasting may prove equally effective as drilling provided twice the seeding rate is used and supplemental treatments are applied.

Table 4. Density of total seeded species in response to seeding technique on intensively disturbed soils.

Seeding Technique ¹	Mean Density (plants/m ²)
1	49.68
2	41.84
3	65.60
4	75.24

¹1 = grasses and forbs drilled at a higher rate than shrubs; 2 = shrubs drilled at a higher rate than in Technique 1; 3 = grasses and forbs drilled; 4 = grasses and forbs broadcasted.

The success of broadcasting may be attributed to the introduction of the supplemental treatment with the existing rough microtopography. This

resulted in a large number of seeds being covered with an adequate amount of soil to insure emergence. Similar studies done by Hodder (1976) and Harper et al. (1965) indicated that a roughened seedbed provides a favorable microhabitat for seedling establishment.

Effect of Fertilizer

Fertilizer addition significantly increased ($p < .0001$) biomass of seeded species on both irrigated and non-irrigated plots. The increased biomass was a reflection of the inherent value of the added nutrients to plants. Also, Wight and Black (1978) and Viets (1962) found that supplemental nitrogen increased biomass by increasing the amount of water that became available for plant use.

The addition of N and P increased plant cover on both irrigated and non-irrigated plots (Table 5). However, plant cover on irrigated plots was not sufficiently greater on fertilized plots to be statistically significant.

Table 5. Mean percent cover of total seeded species in response to irrigation and fertilization on intensively disturbed soils.

Non-Irrigated Plots		Irrigated Plots	
NF	F	NF	F
12.44	19.26	17.13	18.17

NF = no fertilization

F = 112 kg N/ha, 90 kg P/ha

Across all three species mixtures, seeded grasses exhibited similar aboveground production with the addition of N and P (Table 6). Subplots seeded with the introduced seed mixture produced the highest biomass values followed by the combination seed mixture. The lowest biomass production occurred in subplots seeded with the native seed mixture.

Table 6. Mean biomass of seeded grasses in response to fertilizer on intensively disturbed soils.

	Combination	Native	Introduced
Non-fertilized	868.40	718.00	1060.20
Fertilized	1249.60	1050.00	1507.20

While the biomass of seeded grasses increased with fertilizer treatments, the density, cover, and biomass of seeded forbs decreased significantly with the addition of nitrogen and phosphorus fertilizer (Table 7). The majority of seeded forbs were leguminous species capable of nitrogen fixation. Cooke et al. (1968) and Cosper et al. (1967) found that in grass-legume pastures the legume component

decreased and the grass component increased with nitrogen and phosphorus additions. Thus, it would seem that nitrogen-fixing legumes lose much of their competitive advantage under fertilized conditions.

Table 7. Mean density, cover, and biomass of seeded forbs in response to fertilizer on intensively disturbed soils.

	Density (plants/m ²)	Cover (%)	Biomass (kg/ha)
Non-fertilized	3.52 ^a	1.99 ^a	72.40 ^a
Fertilized	1.68 ^b	0.93 ^b	48.49 ^b
	(p<.0001)	(p<.0008)	(p<.0894)

^{a,b} Means in the same row bearing different superscript letters are significantly different (p<0.1).

Effect of Irrigation

The density and biomass of seeded species on the irrigated plots were greater than density and biomass on the non-irrigated plots (Table 8). This increase in density and biomass can be attributed largely to an increase in the number and yield of seeded grasses with the addition of supplemental water. However, it should be noted that cover after three years of growth (of which the first two were irrigated) showed no significant difference in favor of the additional water.

Table 8. Mean density, cover, and biomass of total seeded species in response to irrigation on intensively disturbed soils.

	Density (plants/m ²)	Cover (%)	Biomass (kg/ha)
Non-irrigated	41	16	1,052
Irrigated	73	18	1,570
Native (undisturbed) vegetation	86	18	350

A comparison of the non-irrigated and irrigated plots with the native (undisturbed) vegetation showed the non-irrigated plots and irrigated plots to be higher in current annual production, similar in percent cover values, but lower in density than the native vegetation (Table 8). Current annual production in the non-irrigated plots shows a four-fold increase above production in the native vegetation. This can be explained by the dominance of grasses in the reclaimed plots which exhibit more annual production as opposed to the woody shrubs which predominated in the native vegetation community. After three growing seasons the reclaimed plots exhibited greater annual production than the native, undisturbed vegetation. However, additional time may be required for the plots to develop successional and to determine the effects of time on the reseeded vegetation.

Invasion of Plants

In comparing invasion on the irrigated and non-irrigated plots, the non-irrigated plots showed higher invader density. However, most invaders on the non-irrigated plots were small seedlings of Russian thistle which never attained maturity. Although density values differed greatly between the irrigated and non-irrigated plots, the cover values of invading species were similar for the two treatments.

Conclusions

These conclusions represent the results obtained from the 1979 growing season on the Revegetation Technique Study which was established in 1976.

1. The seeding technique with a higher shrub seeding rate produced greater shrub density on non-irrigated plots seeded with a combination species mixture.
2. Under irrigated conditions shrub cover increased only when shrub seeding rates were increased within the combination of native and introduced species mixtures.
3. The establishment of shrub cover proved most effective on non-irrigated subplots seeded with a native seed mixture using seeding Technique 1.
4. The density of total seeded species was higher on subplots which were broadcast seeded and lightly covered with soil as compared with those subplots which were drill seeded with one-half the rate of seed.
5. Fertilizer addition increased biomass of seeded species on both irrigated and non-irrigated plots.
6. The density and biomass of seeded species on the irrigated plots were greater than density and biomass on the non-irrigated plots, but percent cover was unaffected.
7. A comparison of the non-irrigated and irrigated plots with the native, undisturbed vegetation showed the non-irrigated and irrigated plots to be higher in current annual production, similar in percent cover, and lower in density than the native vegetation.
8. Density of invading species was higher on the non-irrigated than the irrigated plots, but cover values were similar between the two treatments.

Successional Study on Surface Disturbed Soil

The Successional Study on Surface Disturbed Soil was designed to determine the effects and interactions of seed mixture, fertilizer, and mulch as they relate to seeded plant community succession

on shallowly disturbed soil. This disturbance consisted of all existing vegetation being scraped off and the soil being ripped to a depth of 30 cm. The study was implemented to simulate minor disturbances that may result from activities occurring with oil shale development (e.g., road construction and staging areas).

The six seeding mixtures of native and introduced species used in this study ranged from simple grass combinations to complex grass-forb-shrub mixtures. The three fertilizer treatments used consisted of two levels of nitrogen and phosphorus and a control treatment. Wood fiber hydromulch was applied at the rate of 2.2 MT/ha following seeding in the fall of 1976.

Results and Discussion

These results are based upon 1979 data which were obtained near the end of the third growing season. Although three years is a relatively short period of time in terms of a successional study, the seeded vegetation is well established and preliminary results can be drawn. The dynamic nature of these plant communities, however, may alter these results in subsequent years.

Effect of Seed Mixtures

The native grass-forb-shrub mixture showed significantly higher ($p < .05$) total cover than the combination native and introduced grass-forb-shrub mixture. The native grass-forb-shrub mixture also exhibited the highest total cover across all seed mixtures (Figure 6). The higher total cover from this mixture can be attributed largely to the cover provided by the shrub component. This dramatic increase in shrub cover resulted from the growth of fourwing saltbush which was present only in the native grass-forb-shrub mixture.

The greatest biomass was produced by species seeded in the combination native and introduced grass-forb-shrub mixture (Table 9). The grass

Table 9. Total biomass of seeded and invading species.

Seed Mixture	Biomass of Seeded Species (kg/ha)	Biomass of Invading Species (kg/ha)
Native grass	1912	204
Introduced grass	1484	48
Native grass-forb	1407	142
Introduced grass-forb	1170	50
Native grass-forb-shrub	1286	171
Native and Introduced grass-forb-shrub	1685	88

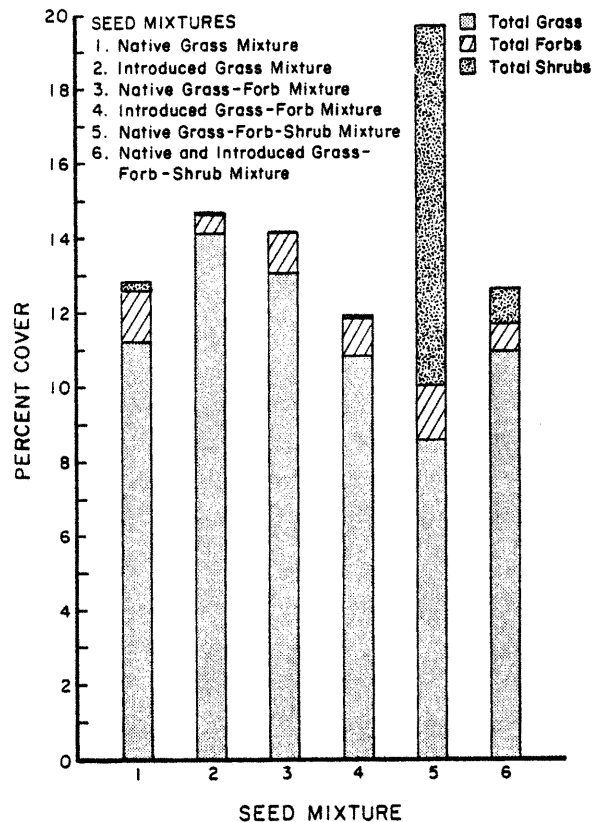


Figure 6. Total cover for seeded and invading species on the Shallowly Disturbed Successional Plot.

component of this mixture was significantly higher ($p < .0025$) than the grass component in the native grass-forb-shrub mixture (Figure 7). This result can be attributed to the presence of introduced, high yielding grass species in the combination native and introduced mixture which were not present in the native mixture.

The density of seeded grasses was higher in the native grass mixture than the introduced grass mixture. Cover, however, was greater in the introduced grass mixture than the native grass mixture even though the number of plants per plot was less (Table 10). This result can be partially explained by the fact that introduced grass species develop into plants which are generally more robust than the native grass species seeded (Hitchcock 1971).

Table 10. Density and cover of the native grass mixture as compared with the introduced grass mixture on the Shallowly Disturbed Successional Plot.

	Native Grass Mixture	Introduced Grass Mixture
Density (plants/m ²)	83.40 ¹	48.92 ¹
Cover (%)	10.56	13.56

¹HSD = 18.94

The limited nutrients and space in the subplot can accommodate fewer plants of the introduced species with greater efficiency as compared with the native species.

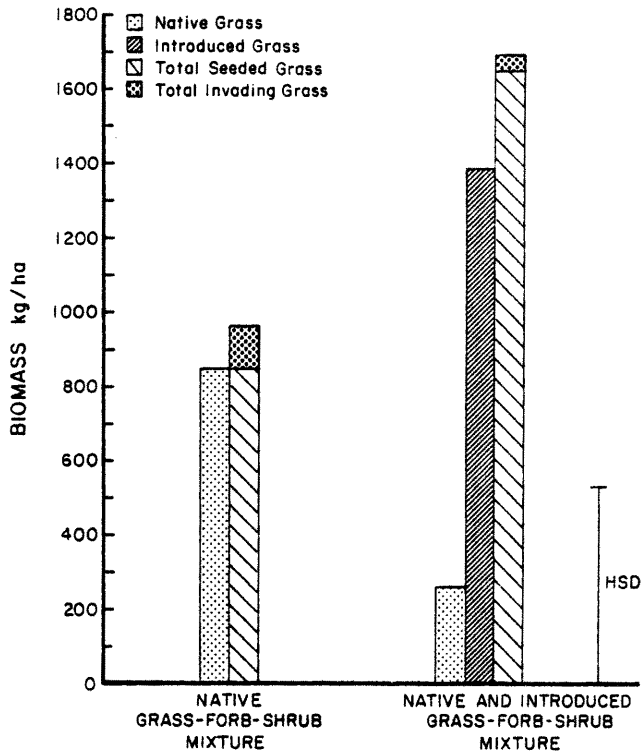


Figure 7. Total grass biomass of seeded and invading species on the Shallowly Disturbed Successional Plot.

It is generally accepted that introduced species will initially demonstrate higher production and cover over native species during early establishment. However, in subsequent years as the plant community closes and competition becomes more intense, native species may exhibit a greater competitive advantage. This advantage is a result of the inherent adaptability of native species to their environment. After three years of study, it appears that native species are approaching production and cover levels similar to those of introduced species. Terwilliger et al. (1974) discussed a native/introduced competitive response where native species gradually exerted a competitive advantage over introduced species in the same seed mixture. After a period of 15 years, Terwilliger reported crested wheatgrass as being only an occasional plant, whereas Indian ricegrass (*Oryzopsis hymenoides*) and Douglas rabbitbrush (*Chrysothamnus douglasii*) had become prominent species.

Effect of Fertilizer Treatments

Percent cover and biomass of seeded and invading species proved significantly greater under high fertilization when compared with the control (Figure 8). This response can be attributed primarily to an increase to the grass component in

each mixture (Table 11). Grasses appear to possess an inherent ability to respond to additions of N fertilizer as reported in similar studies by Lutwick and Smith (1979) and Black (1968).

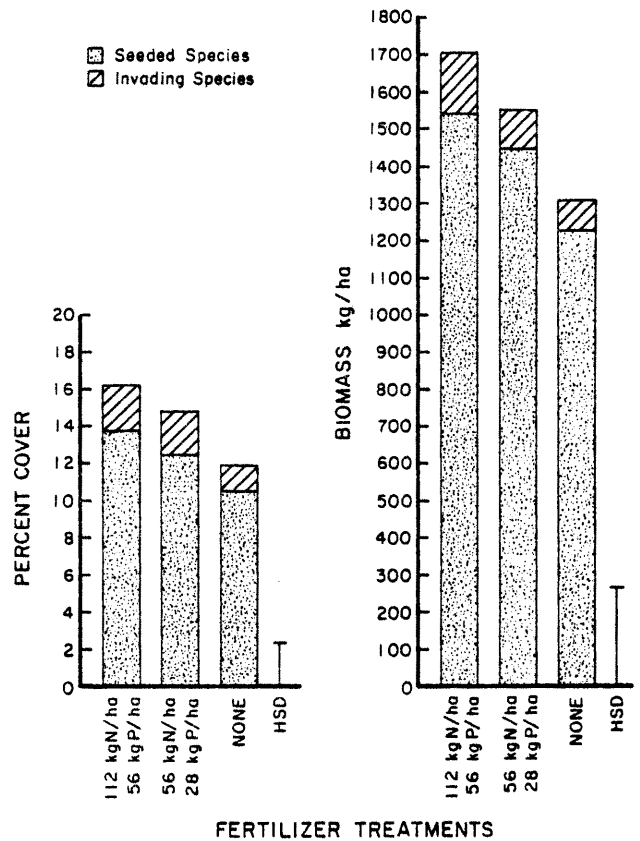


Figure 8. Fertilizer response of total seeded and invading species for the Shallowly Disturbed Successional Plot.

Table 11. Effect of fertilizer on biomass and percent cover of seeded and invading species by life form on the Shallowly Disturbed Successional Plot.

Life Form	Fertilizer Treatment (kg/ha)	Cover (%)	Biomass (kg/ha)
Grass	112 N, 56 P	11.96	1505
	56 N, 28 P	11.51	1396
	None	9.93	1230
Forb	112 N, 56 P	1.22	102
	56 N, 28 P	1.14	56
	None	0.68	31
Shrub	112 N, 56 P	2.02	100
	56 N, 28 P	2.17	108
	None	1.35	46

Seeded and invading forbs showed no significant cover or biomass response to fertilizer

treatments (Table 11). Both Hedrick (1964) and Cosper et al. (1967) have shown that forbs are generally depressed by N fertilization and increase production as N fertilization is reduced. This general lack of response by forbs to the addition of N may be the result of nitrogen not being a limiting factor for growth of forbs on this site. During plot construction the standing native vegetation was removed. However, most of the root material was left in place providing a source of organic nitrogen across all subplots as the organic material decomposed. In addition, the soil ripping done prior to seeding improved aeration and may have increased nitrification in the soil. This process of promoting nitrification through physical manipulations of the seedbed (such as, plowing) has also been reported by Buchman and Brady (1974). In succeeding years nitrogen may become a limiting factor; and, as a result a significant forb response may become apparent.

Effect of Mulch

The only significant effect attributable to mulch occurred with grass density. The mulched plots produced 52 plants/m² as compared with the unmulched plots which produced 64 plants/m². The mulching treatment had no significant effect on biomass or cover of seeded species or the amount of invasion that occurred. There was some speculation that the mulch may have absorbed and evaporated light showers which did not enhance the soil moisture relations.

Invasion of Plants

The total cover of invading species was significantly higher ($p=.05$) in the native grass-forb-shrub mixture as compared with the introduced grass mixture, the introduced grass-forb mixture, and the native and introduced grass-forb-shrub mixture (Figure 9). The general trend shows lower invasion cover in introduced and the combination (native and introduced) mixtures as compared with the native seed mixtures. Invading grasses and forbs were responsible for most of the invasion that occurred in each of the seeded mixtures. Cheatgrass was the most aggressive grass invader and Russian thistle and scarlet globemallow (*Sphaeralcea coccinea*) were the most prolific invading forbs.

As would be expected the greatest biomass for total invading species occurred in the high fertilization treatment (Figure 10), and the lowest biomass response of invading species occurred when no fertilizer was added. Forbs exhibited the greatest difference between fertilizer treatments, producing 92 kg/ha under high fertilization as compared with 24 kg/ha under no fertilization. Although most of the invading forbs were non-leguminous species, the fertilizer response was similar to the response exhibited by the seeded forbs (i.e., increased biomass production under high fertilization as compared with no fertilization).

Density and Cover Trends for 1978-1979

Total percent cover for seeded and invading species has remained relatively stable between the

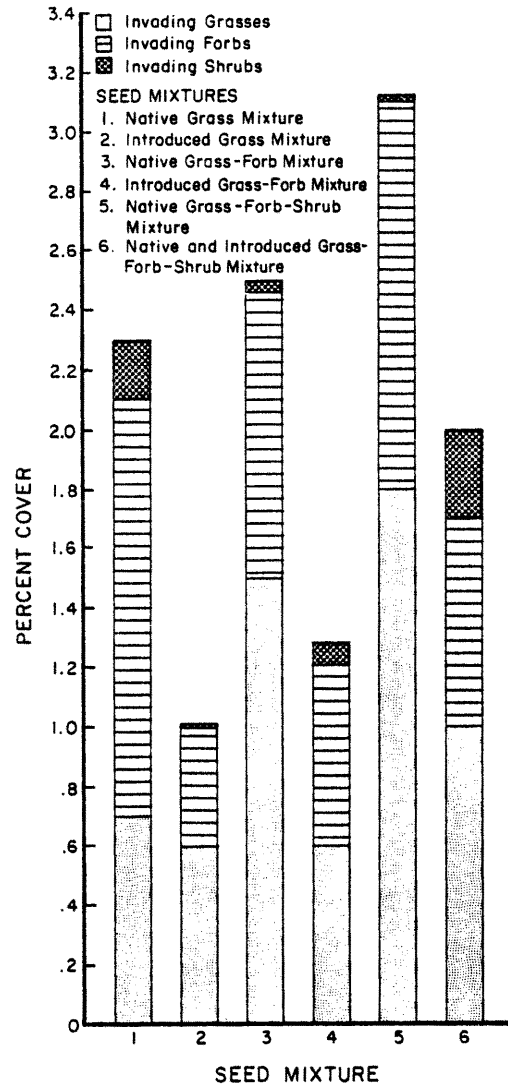


Figure 9. Total cover of invading species plotted against seed mixture on the Shallowly Disturbed Successional Plot.

two years of 1978 and 1979. Density, however, has shown a great increase in both seeded and invading species over all plots. The increase in density can be explained by the presence of many new seedlings in 1979 which indicates a favorable reproductive environment. The greatest density increase between the two years occurred in those seed mixtures containing only native species. This may suggest that native species are beginning to express a competitive advantage over introduced species (Figure 11).

The present trend, therefore, shows an increase in total density without a measurable change in cover. This result indicates that the seeded grasses and established invading species are beginning to regenerate, causing an increase in plant numbers composed of first-year seedlings.

Conclusions

The conclusions drawn from this study are based on results obtained during the third growing

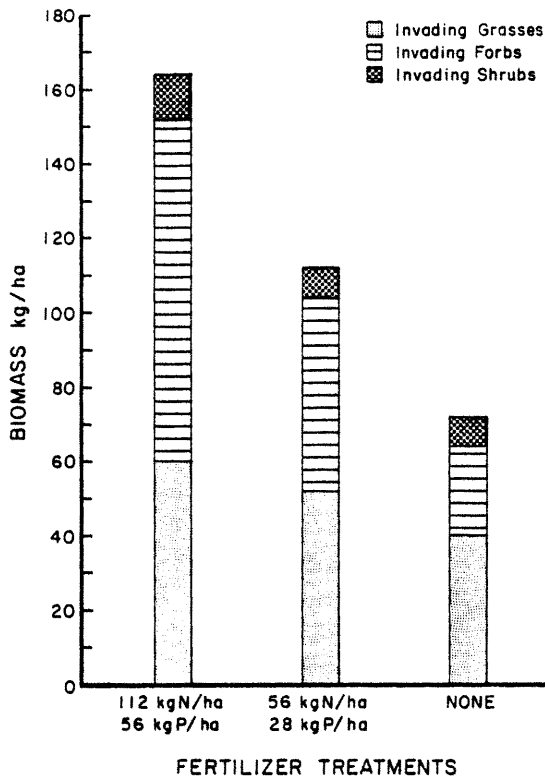


Figure 10. Total biomass of invading species under different fertilizer treatments on the Shallowly Disturbed Successional Plot.

season. In subsequent years the dynamic nature of these plant communities may offer data which result in findings that differ materially from those at this time.

1. The native grass-forb-shrub mixture exhibited the highest total cover across seed mixtures primarily because of the presence of fourwing saltbush.
2. The greatest aboveground production occurred with the native and introduced grass-forb-shrub mixture.
3. After three years it appears that native species are approaching production and cover values similar to those of introduced species.
4. Percent cover of seeded and invading species was significantly greater under high fertilization as compared with no fertilization.
5. Seeded and invading forbs exhibited no significant cover or biomass response to fertilizer treatments.
6. The mulching treatment generally had no significant effect on the seeded species or the amount of invasion that occurred.
7. A general trend indicates lower invasion in the introduced and combination native and introduced mixture as compared with the native seed mixtures.

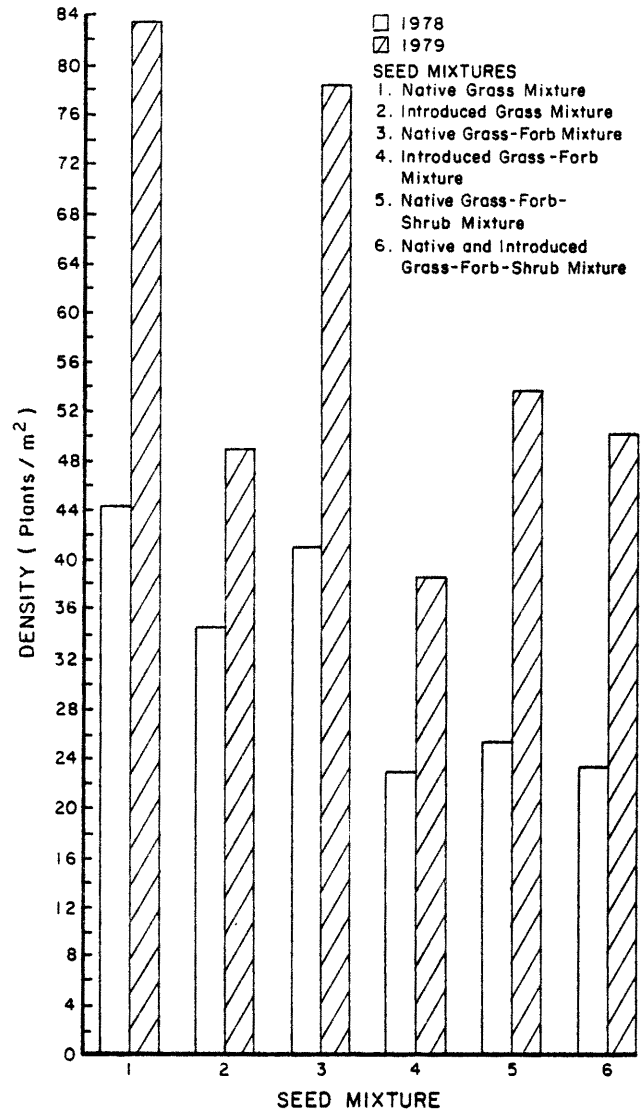


Figure 11. Total density of seeded species for 1978 and 1979 on the Shallowly Disturbed Successional Plot.

8. The greatest biomass for invading species was produced under high fertilization.
9. Percent cover for seeded and invading species has remained relatively stable between 1978 and 1979.
10. Regeneration at the end of the third year was greater for native species than for introduced species.

Annual Disturbance Plot Studies

Each year during 1976 and 1977 disturbance plots were constructed at the Intensive Study Site to examine the effect of soil disturbance on natural invasion and succession. Four treatments were applied simulating various levels of soil disturbance which may accompany oil shale extraction.

The four treatments during each of the above years are as follows:

- Treatment 1: Scrape vegetation off while leaving as much topsoil as possible.
- Treatment 2: Scrape vegetation off and rip the soil to a depth of 30 cm.
- Treatment 3: Remove topsoil and subsoil to a depth of 1 m. These soils were then mixed and returned to the excavated area.
- Treatment 4: Remove top 1 m of soil material and stockpile. Remove an additional 1 m of soil and stockpile. Soil materials were then replaced in reverse order with the material removed from 2 m being placed at the surface.

Results and Discussion

Significant differences were observed between the various treatments on the 1976 plots. Grass cover was significantly greater on Treatment 1 than on Treatments 3 and 4. Grass production was found to be significantly greater on Treatment 1 than on any of the other treatments. Both grass cover and biomass were observed to decrease with increasing levels of disturbance. Prairie junegrass (*Koeleria cristata*), western wheatgrass, and needle-and-thread (*Stipa comata*) comprised, on the average, greater than 80 percent of the grass cover and biomass across all treatments. The primary reason for the reduction in the grass component with increasing soil disturbance was because of the decrease in topsoil quantity and perhaps soil fertility. The increasing levels of disturbance resulted in a reduction and dilution of seeds and vegetative plant parts present in the surface soil material. In related work, Howard and Samuel (1979) observed that freshly stripped topsoil was a good source of seed but, more importantly in arid regions, a source of rhizomes and other vegetative plant parts which allowed plant survival (e.g., western wheatgrass) even under drought conditions. This implies that proper conservation and handling of topsoil in reclamation work may be vital for rapid establishment of desirable species.

Vegetation responses observed on the 1976 plots differed among various surface configurations resulting from the treatments. Therefore, following construction of the 1977 plots a tract-type dozer was driven across all treatments providing a uniform surface configuration. Results from the 1977 plots, which differ somewhat from those of 1976, can be partially explained by conditions resulting from this supplemental treatment of the reconstructed soil surface.

Results from the 1977 plots show significant differences between treatments which are almost entirely the result of differences in Russian thistle invasion across treatments. At least 95 percent of all cover and biomass values for forbs are accounted for by Russian thistle. For the most part, forb cover and biomass were observed to increase with increasing levels of disturbance

while grass cover and biomass generally decreased (Table 12).

Table 12. Cover (%) and biomass (kg/ha) of grasses and forbs across disturbance treatments on 1977 Annual Disturbance Study.

Life Form	Treatment			
	1	2	3	4
<u>Cover</u>				
Grasses	2.27	0.35	0.32	0.04
Forbs	10.28	27.77	34.69	31.00
<u>Biomass</u>				
Grasses	240.8	17.2	20.4	1.2
Forbs	424.4	950.4	1649.2	1397.2

Reeves (in another section of this report) found that mycorrhizal infection potential of the soil decreases with increasing disturbance. This is thought to be the primary reason that Russian thistle (which is non-mycorrhizal) enjoyed a competitive advantage over perennial plants, most of which are mycorrhizal, on the more highly disturbed treatments. Most genera of the climax sagebrush-grass community have been found to be mycorrhizal, and thus without the presence of inoculum in the surface soil the more desirable mycorrhizal plants cannot colonize these disturbances (Reeves et al. 1979). Therefore, topsoil is necessary if rapid invasion and growth of desirable, perennial plants on disturbed lands is desired. To insure that topsoil will serve as a source of seed, vegetative plant parts, and mycorrhizal fungi it should be placed at the surface and mixing with subsoil material should be limited.

The large growth of Russian thistle found on all 1977 treatments is thought not only to be a result of mycorrhizal-plant interactions but also tractor compaction which formed a great number of small depressions on the surface of all treatments (Figure 12). Evans and Young (1972) noted that roughened surface microtopography (similar to that produced by the tractor) aids in Russian thistle seed germination. Hyder et al. (1955) also noted that heavy compaction reduced germination and seedling emergence of perennial grasses. Thus, conditions present on the 1977 plots favored Russian thistle growth by reducing competition and providing suitable microsites.

A comparison between annual and perennial plants was made across treatments on both 1976 and 1977 plots. With increasing intensity of disturbance, annuals were observed to account for greater percentages of total biomass and cover values (Figure 13). Russian thistle was the dominant annual which accounted for the observed trend (Figure 14). In similar work, Terwilliger et al. (1974), Piemeisel (1951), and Costello (1944) also found that Russian thistle dominated early stages of secondary succession following disturbance on semiarid land in the western United States.

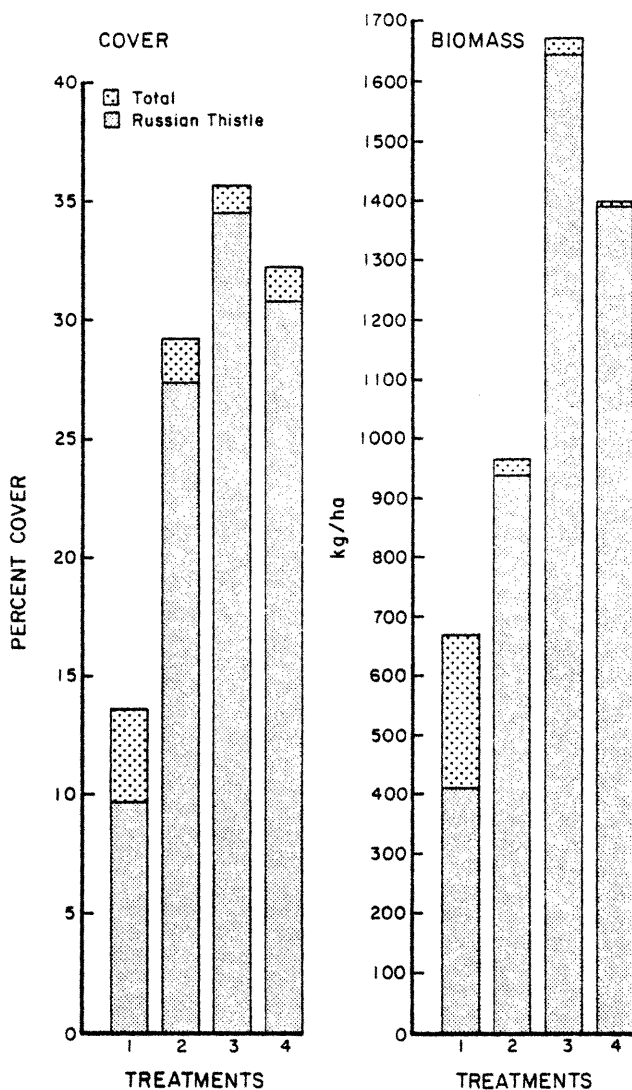


Figure 12. Total cover and biomass of all life forms as compared with cover and biomass of Russian thistle across all disturbance treatments on the Annual Disturbance Study. (See text for treatment descriptions.)

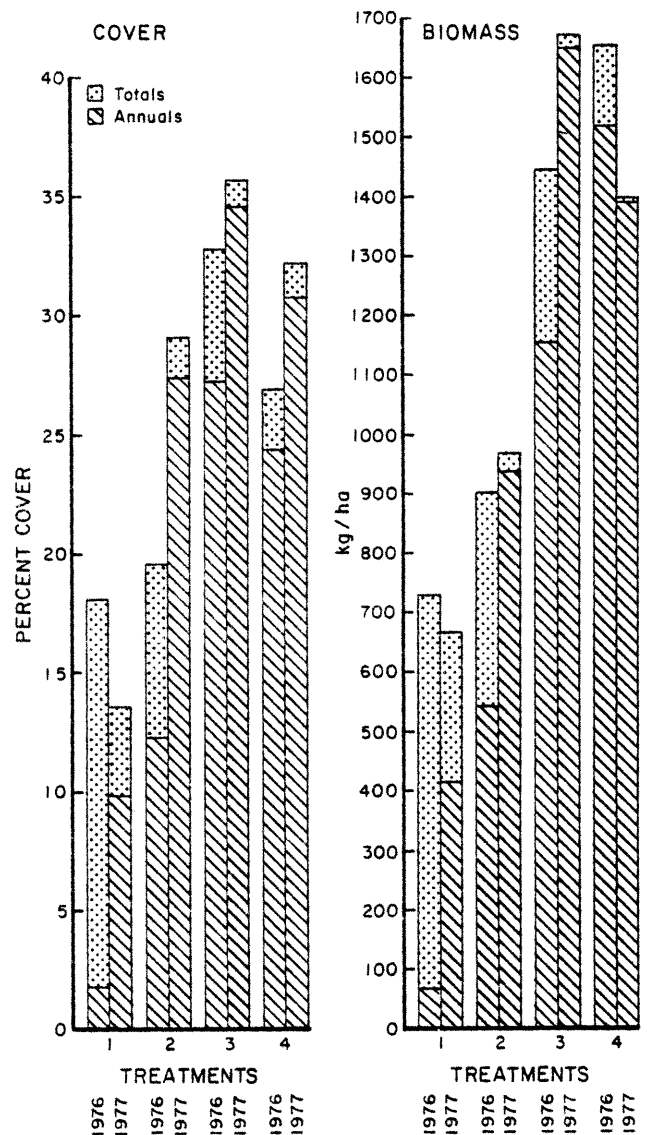


Figure 13. Total cover and biomass of all life forms as compared with cover and biomass of annuals across all disturbance treatments on the Annual Disturbance Study. (See text for treatment descriptions.)

A comparison of species diversity between the various soil disturbances and the native undisturbed vegetation indicated that diversity declined with increasing levels of disturbance (Figure 15). Odum (1971) states that communities with low species diversity are less resistant to outside perturbations (e.g., annual climatic changes), therefore are less stable. Second and third year data from the Annual Disturbance Plots indicate that as intensity of soil disturbance increases, a less diverse and stable community may result if the soil is left to natural invasion.

Conclusions

Preliminary results from the Annual Disturbance Study indicate that to insure the rapid reestablishment of productive and self-sustaining

communities on disturbed lands, reclamation work should take into account the following recommendations:

1. Minimize the level of disturbance on surface disturbed areas.
2. Avoid or minimize mixing of topsoil and subsoil during mining and reclamation.
3. Save adequate amounts of topsoil for surface application.

By following these recommendations a greater amount of mycorrhizal inoculum, vegetative plant parts, and native seed will be provided, thus enhancing the chances for successful reclamation.

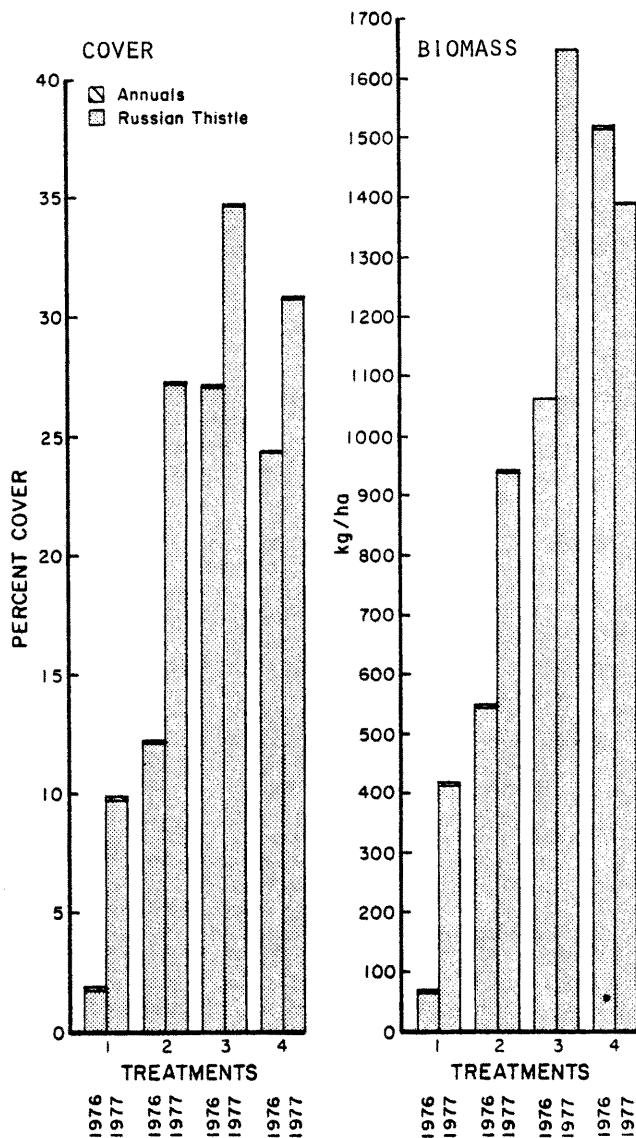


Figure 14. Cover and biomass of annuals as compared with cover and biomass of Russian thistle across all disturbance treatments on the Annual Disturbance Study. (See text for description of treatments.)

SUMMARY

The successional studies at the DOE Intensive Study Site are beginning to show responses that can lead to much improvement in reclamation practices on lands disturbed by oil shale development. The most important step in reestablishing a functional ecosystem on disturbed lands is the conservation and proper storage of the soil material. Soil material which is suitable for a plant growth medium needs to be saved in an adequate amount to provide a soil covering of at least 61 cm over retorted shale material. A soil cover of this depth will provide a sufficient rooting zone for vegetation and allow a functional decomposer community to become established. In addition, the coverage by

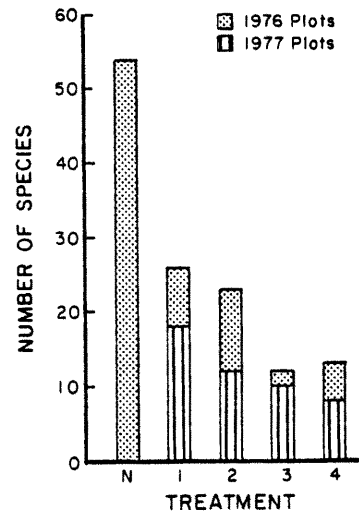


Figure 15. Number of species found in native, undisturbed vegetation and across all disturbance treatments on the Annual Disturbance Study. (See text for description of treatments.)

soil minimizes the problem of uptake of toxic elements by the vegetation from the retorted shale material. A topsoil cover of this depth will store winter storm moisture as a reserve for the summer drought. Maximum benefit from the soil material can be achieved if topsoil and subsoil materials are handled and applied separately.

Different seeding mixtures are showing rather pronounced differences in the diversity of plant species and abundance of life forms that ultimately occupy the revegetated area. Across all successional studies similar total biomass values are being observed for mixtures composed of native, introduced, or a combination of native and introduced species. Cover values, however, are generally greater for the native seed mixtures because of the excellent growth of the native shrub species. Native seed mixtures are generally allowing the greatest amount of invasion, but this may be only an early successional expression. The quantity of each life form in the seeded mixture profoundly affects the density of species within each life form during early successional patterns.

Fertilization with nitrogen and phosphorus generally increases seedling establishment and overall production primarily because of an increase in grass biomass. Forb species, mostly nitrogen-fixing legumes, have shown a slight decrease in production with increasing amounts of fertilizer. Fertilization in most cases has had little effect upon shrub establishment and growth. Like fertilization, irrigation increases production as a result of increased grass biomass. Irrigation also increases plant density but has had little effect on overall vegetation cover.

All reseeded vegetation on the successional studies is exhibiting higher production than the native, undisturbed plant communities. Percent herbage cover is similar for both natural and seeded stands of vegetation, and density values for the

seeded areas are increasing and beginning to approach those of the native vegetation. However, the density of the native species mixtures is increasing at a more rapid rate than the introduced seed mixtures. It should be noted that while the native vegetation is dominated by sagebrush, the reseeded areas are composed predominately of grass species with secondary forb and shrub components.

LITERATURE CITED

- Bernstein, L. 1962. Salt-affected soil and plants. Pages 139-174 in UNESCO arid zone research. XVIII. The problems of the arid zone.
- Black, A. L. 1968a. Nitrogen and phosphorus fertilization for production of crested wheatgrass and native grass in northwestern Montana. *Agron. J.* 60:213-216.
- Black, C. A. 1968b. Soil-plant relationships. John Wiley and Sons, Inc., New York.
- Bower, C. A., and C. H. Wadleigh. 1949. Growth and cationic accumulation by four species of plants as influenced by various levels of exchangeable sodium. *Soil Sci. Soc. Am. Proc.* (1948) 13:218-223.
- Buchman, Harry O., and Nyle C. Brady. 1972. The nature and properties of soils. The MacMillan Co., New York.
- California Fertilizer Association. 1975. The western fertilizer handbook. Interstate Printers and Publishers, Inc., Danville, Illinois.
- Cooke, D. A., S. E. Beacom, and N. K. Dawley. 1968. Response of six year old grass-alfalfa pastures to nitrogen fertilizer in northeastern Saskatchewan. *Can. J. Plant Sci.* 48:167-173.
- Cosper, H. R., J. R. Thomas, and A. Y. Alsayegk. 1967. Fertilization and its effect on range improvement in the Northern Great Plains. *J. Range Manage.* 20:216-222.
- Costello, D. F. 1944. Natural revegetation of abandoned plowed land in the mixed prairie association of northeastern Colorado. *Ecology* 25:312-326.
- Evans, R. A., and J. A. Young. 1972. Germination of *Salsola* in relation to seedbed environment. II. Seed distribution, germination, and seedling growth of *Salsola* and microenvironmental monitoring of the seedbed. *Agron. J.* 64:219-224.
- Harper, J. L., W. T. Williams, and G. R. Sogar. 1965. The behavior of seeds in soil. Part I. The heterogeneity of soil surface and its role in determining the establishment of plants from seed. *J. Ecol.* 53:273-286.
- Hedrick, D. W. 1964. Response of an orchardgrass-subclover mixture in western Oregon to different clipping and fertilizing practices. *J. Range Manage.* 11:221-227.
- Hersman, L. E., and D. A. Klein. 1979. Retorted oil shale effects on soil microbiological characteristics. *J. Environ. Qual.* 8:Oct-Dec 1979.
- Hitchcock, A. S. 1971. Manual of the grasses of the United States, revised ed. General Publishing Co., Ltd., Toronto, Canada.
- Hodder, R. L. 1976. Planting methods and equipment development. Pages 69-82 in K. C. Vories, ed. Reclamation of western surface mined lands. Workshop proceedings, Ecology Consultants, Inc.
- Hodder, R. L., Program Leader. 1978. Research on revegetation of surface mined lands at Colstrip, Montana: Progress report 1975-1977. Research Report 127. *Mont. Agric. Exp. Stn.*, Mont. State Univ.
- Howard, G. S., and M. J. Samuel. 1979. The value of fresh-stripped topsoil as a source of useful plants for surface mine rehabilitation. *J. Range Manage.* 32:76-77.
- Humphrey, R. R. 1959. Forage and water. *J. Range Manage.* 12:164-170.
- Hyder, D. N., F. A. Sneva, and W. A. Sawyer. 1955. Soil firming may improve range seeding operations. *J. Range Manage.* 8:159-163.
- Justice, O. L., and M. H. Reece. 1954. A review of literature and investigation on the effects of hydrogen-ion concentration on the germination of seeds. *Proc. Assoc. Off. Seed Anal.* 44:144-149.
- Lindsay, W. L. 1979. Professor, Department of Agronomy, Colorado State University, Fort Collins. Personal communication, 2 November.
- Lutwick, L. E., and A. D. Smith. 1979. Yield and N uptake by seven perennial grass species as affected by high rates of N fertilizer. *J. Range Manage.* 32:433-436.
- McDonough, W. T. 1977. Seed physiology. Pages 156-184 in Society for Range Management. Rangeland plant physiology.
- McGinnies, W. G., and J. F. Arnold. 1939. Relative water requirements of Arizona range plants. *Ariz. Agric. Exp. Stn. Tech. Bull.* 80.
- McGinnies, W. J. 1960. Effects of moisture stress and temperature on germination of six range grasses. *Agron. J.* 52:159-162.
- Odum, E. P. 1971. Fundamentals of ecology, 3rd ed. W. B. Saunders Co., Philadelphia, Pa.
- Piemeisel, R. L. 1951. Causes affecting change and rate of change in a vegetation of annuals in Idaho. *Ecology* 32:53-72.
- Reeves, F. B., D. Wagner, T. Moorman, and J. Kiel. 1979. The role of endomycorrhizae in revegetation practices in the semi-arid West. I. A comparison of incidence of mycorrhizae in severely disturbed vs. natural environments. *Am. J. Bot.* 66:6-13.

- Richards, L. A., ed. 1954. Diagnosis and improvement of saline and alkali soils. U.S. Dep. Agric. Agric. Res. Serv. Agric. Handb. No. 60.
- Robertson, J. H., and C. K. Pearse. 1945. Artificial reseeding and the closed community. Northwest. Sci. 19:58-66.
- Terwilliger, C., Jr., C. W. Cook, and P. L. Sims. 1974. Ecosystems and their natural and artificial rehabilitation. Pages 67-97 in C. W. Cook, coordinator. Surface rehabilitation and land disturbances resulting from oil shale development. Colo. State Univ. Environ. Res. Center Tech. Rep. Ser. No. 1.
- Viets, F. G., Jr. 1962. Fertilizer and the efficient use of water. Adv. Agron. 14:223-264.
- Wight, J. Ross, and A. L. Black. 1978. Soil water use and recharge in a fertilized mixed prairie plant community. J. Range Manage. 31:280-282.

LONG-TERM FERTILITY STUDY ON LAND DRASTICALLY DISTURBED BY OIL SHALE DEVELOPMENT

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OBJECTIVES

The general objective of this subproject is to determine the long-term fertility requirements and methods of meeting these requirements of N- and P-deficient soil materials disturbed by oil shale development in northwestern Colorado. More specifically the objective involves comparing the usefulness of adding low to moderate annual applications of inorganic N fertilizer with that of adding high rates of inorganic N fertilizer only once at the initiation of the study on plant establishment and growth. At the end of four years, equal amounts of N will have been added to both sets of plots. Additionally, comparison of the effect on plant establishment and growth on the high rates of N fertilizer added initially with and without wood wastes is of interest. Other comparisons showing the effect of fertility treatments on plants are the addition of sewage sludge with and without wood wastes and four combinations of nitrogen and phosphorus fertilizer. Lastly, we desired to determine the effect of adding N to the soil through nitrogen fixation by seeded legumes in combination with grasses.

PROGRESS TO DATE

Long-Term Fertility Plot studies were developed in the summer and fall of 1977. One set of plots was constructed on disturbed topsoil placed over 61 cm of Paraho retorted shale while the second set of plots was established on disturbed subsoil. The two sets were prepared to compare nitrogen availability to plants in an N-deficient subsoil with a topsoil that has considerably more organic matter and thus more nitrogen available for plant growth. Table 13 indicates the treatments applied to the Topsoil Over Shale and Subsoil Plots.

The legume-grass plots were prepared in the summer of 1978. The plants were seeded in the fall on both topsoil and subsoil that were fertilized and unfertilized. Plant growth has been established, and one sampling for plant biomass has been done in 1979.

These studies examine the addition of nitrogen to the soil systems from both external inorganic

sources and internally through N-fixation by legumes. As these methods are evaluated in light of the overall project's objectives, other aspects of maintaining adequate nitrogen in the soil will come under consideration.

Subsoil Plot

Second year soil samples were taken from these plots in May 1979. Ten cores 13 cm deep were taken from each plot using an Oakfield probe 2 cm in diameter. These cores were composited for each plot, quickly air-dried, and prepared for further analysis. They were later analyzed for pH, EC, ammonium nitrogen, nitrate nitrogen, total nitrogen, and extractable phosphorus.

Plant density, aboveground biomass, and cover were measured by species for the second growing season during July 1979. These measurements were taken from permanently marked quadrats in each subplot.

In addition to the above plant measurements the entire plot was sampled for plant invasion. Each plot was walked in 1 m strips by two researchers who recorded surface area occupied by invading species.

Topsoil Over Retorted Shale Plot

Second year soil samples were taken from these plots in May 1979. Cores from ten locations in each plot were sampled to a depth of 6.5 cm and from 6.5 to 13 cm using an Oakfield probe 2 cm in diameter. These cores were composited by depths for each plot, quickly air-dried, and prepared for further analysis. They were analyzed for pH, EC, ammonium nitrogen, nitrate nitrogen, total nitrogen, and extractable phosphorus.

Because of heavy rodent damage during the winter of 1978-1979, these plots were reseeded in May 1979. They were broadcast seeded using the native grass-forb-shrub mixture used previously (see Progress Report 1978-1979 for details). In order to establish a stand at this time of year, an irrigation system was established and a total of

Table 13. Fertilizer treatments applied to the Topsoil Over Retorted Shale and Subsoil Plots seeded with a native grass-forb-shrub mixture.

Fertilizer Application	Treatment Number	Rate of Application
Annual nitrogen applications ^{1,2,3}	1	56 kg N/ha
	2	112 kg N/ha
	3	224 kg N/ha
	4	448 kg N/ha
Initial nitrogen applications ²	5	224 kg N/ha
	6	448 kg N/ha
	7	896 kg N/ha
	8	1792 kg N/ha
Initial nitrogen application plus wood waste ²	9	224 kg N/ha + 11.1 MT/ha wood waste
	10	448 kg N/ha + 22.4 MT/ha wood waste
	11	896 kg N/ha + 44.8 MT/ha wood waste
	12	1792 kg N/ha + 89.7 MT/ha wood waste
Sewage sludge plus wood waste	13	56 MT/ha sewage sludge + 0.0 MT/ha wood waste
	14	112 MT/ha sewage sludge + 22.4 MT/ha wood waste
	15	224 MT/ha sewage sludge + 44.8 MT/ha wood waste
Nitrogen with phosphorus at two rates	16	112 kg N/ha + 56 kg P/ha
	17	896 kg N/ha + 56 kg P/ha
	18	112 kg N/ha + 192 kg P/ha
	19	896 kg N/ha + 192 kg P/ha
Control ²	20	0 kg N/ha

¹N applied as ammonium nitrate.

²Plus overall application of 130 kg/ha phosphorus (applied as triple superphosphate).

³Annual applications 1-4 after four years equal respective treatments of initially applied nitrogen.

13 cm of irrigation water was applied over a six-week period until seedlings appeared to be established.

Due to the immature stage of most of the plant species at the end of the summer, only plant density by life form was measured during August 1979.

Legume Plots

These plots were established to study the effect of legumes on the nitrogen status of soil as well as on nitrogen status and growth of native grasses grown in association with the legumes with and without N fertilizer added.

The following parameters were measured in 1979: (1) plant yield (aboveground dry weight), (2) nitrogen content of native grasses, (3) nitrogen content of legumes, and (4) N-fixation.

During the 1979 field season the legumes were sampled for plant yield when one-third of the plants were in bloom. Sainfoin (*Onobrychis viciaefolia*), Ladak alfalfa (*Medicago sativa*), and cicer milk-vetch (*Astragalus cicer*) bloomed in 1979 and were therefore sampled. The other legume species were well established during this first season but did not bloom and will therefore be sampled in 1980.

Soil samples were obtained prior to planting to establish the initial N content of the plots. The next sampling will be made in the spring of 1980. Grass samples will be obtained in 1980, and N analyses will be run to determine quantity.

The length of time before significant changes in soil nitrogen content or grass nitrogen content occur will also be evaluated in this study. This time factor is important in the revegetation of disturbed sites because, while rapid return to pre-disturbed conditions is optimum, the fertility of the site should be at a level where the plant productivity can be maintained.

RESULTS AND DISCUSSION

Soil Data for Subsoil Plot

Table 14 contains the mean data from the chemical analyses of the soil samples taken in May 1979 from the Subsoil Plots. The pH is typical of the subsoils of the area ranging from 8.0 up to 8.4. Treatments had little, if any, effect on the subsoil pH.

Electrical conductivity (EC) varied from 3.2 to 6.6 mmhos/cm. The treatments that resulted in

Table 14. Influence of fertility treatments on the soil parameters determined on the Disturbed Subsoil Plots. (See Table 13 for a description of fertilizer treatments.

Fertilizer Treatment Number	Parameter ¹					Extractable Phosphorus ² (ppm)
	pH	EC (umhos cm ⁻¹)	NH ₄ ⁺ -N (ppm)	NO ₃ ⁻ -N (ppm)	Total N (ppm)	
1	8.4 ^{ab}	4.2 ^{ab}	3.5 ^{ab}	3.7 ^{abcd}	520 ^{ab}	27.4 ^{abc}
2	8.2 ^{ab}	3.7 ^{abc}	11.3 ^{abc}	2.6 ^{ab}	620 ^{ab}	29.5 ^{abc}
3	8.3 ^{ab}	4.8 ^{abc}	12.7 ^{bc}	7.5 ^{de}	580 ^{ab}	21.0 ^a
4	8.3 ^{ab}	5.3 ^{bc}	12.2 ^{bc}	8.5 ^{de}	542 ^{ab}	24.4 ^a
5	8.3 ^{ab}	6.4 ^{bc}	5.1 ^{abc}	3.6 ^{abc}	273 ^a	21.7 ^a
6	8.1 ^{ab}	3.2 ^a	6.5 ^{abc}	4.7 ^{abcde}	407 ^a	21.4 ^a
7	8.1 ^{ab}	3.5 ^a	14.0 ^c	2.7 ^{ab}	430 ^a	28.6 ^{abc}
8	8.3 ^{ab}	6.6 ^{bc}	13.2 ^{bc}	3.9 ^{abcde}	587 ^{ab}	21.8 ^a
9	8.3 ^{ab}	3.8 ^{abc}	3.6 ^{ab}	2.4 ^{ab}	587 ^{ab}	30.2 ^{abc}
10	8.3 ^{ab}	3.9 ^{abc}	4.9 ^{abc}	1.8 ^a	587 ^{ab}	31.5 ^{abc}
11	8.2 ^{ab}	3.9 ^{abc}	8.9 ^{abc}	3.0 ^{abc}	417 ^a	18.1 ^a
12	8.2 ^{ab}	4.1 ^{abc}	12.7 ^{bc}	6.1 ^{abcde}	417 ^a	22.1 ^a
13	8.3 ^{ab}	4.9 ^{abc}	3.9 ^{abc}	2.6 ^{ab}	293	32.2 ^{abc}
14	8.1 ^{ab}	4.8 ^{abc}	9.7 ^{abc}	7.0 ^{bcde}	920 ^{ab}	48.0 ^{bc}
15	8.0 ^a	6.6 ^{bc}	9.6 ^{abc}	7.0 ^{de}	1333 ^b	49.3 ^c
16	8.4 ^{ab}	4.0 ^{abc}	3.6 ^{ab}	2.5 ^{ab}	383 ^{ab}	18.9 ^a
17	8.4 ^{ab}	3.9 ^{abc}	11.1 ^{abc}	2.8 ^{ab}	500 ^{ab}	12.8 ^a
18	8.4 ^b	4.0 ^{abc}	1.9 ^a	1.6 ^a	470 ^{ab}	30.7 ^{abc}
19	8.3 ^{ab}	3.7 ^{abc}	5.0 ^{abc}	3.7 ^{abcd}	613 ^{ab}	26.4 ^{ab}
20	8.4 ^{ab}	4.4 ^{abc}	4.3 ^{abc}	2.5 ^{ab}	510 ^{ab}	25.0 ^a

¹Means followed by the same letter are not statistically significant at the .05 level.

²Phosphorus determined by the Olsen's sodium bicarbonate extractable method (Watanabe and Olsen 1965).

the highest EC were Treatment 8 (1792 kg N/ha added initially) and Treatment 15 (224 MT/ha sewage sludge + 44.8 MT/ha wood waste). These values were significantly different ($p=.05$) than the lowest EC values but not greatly different from the control. These EC values are high enough to adversely affect growth of many agronomic crops and possibly some native species as well. However, species native to the Piceance Basin do not appear to be affected by the naturally occurring EC values of 4.4 or less.

The highest NH₄⁺-N value was 14 ppm with the lowest 3.5 ppm. The NH₄⁺-N content of the control was 4.3 ppm. Most treatments were not significantly different from the control. The only two treatments that caused a significantly different NH₄⁺-N content in the soil were Treatment 7 (896 kg N/ha) with 14 ppm NH₄⁺-N and Treatment 18 (112 kg N/ha + 192 kg P/ha) with 1.9 ppm NH₄⁺-N. These differences are of little consequence, however, when you consider that extremely large amounts of NH₄⁺-N fertilizer were added and there was little remaining in the soil at the time of sampling.

The NO₃⁻-N content of the subsoil was relatively low, ranging from 1.6 ppm up to 8.5 ppm. Although there were some statistically significant differences due to treatment, these amounts were so low compared to the total amount of NO₃⁻-N added originally that the practical significance of the differences is questionable. Most of the 800 ppm

of NO₃⁻-N added originally in the highest initial nitrogen treatment (Treatment 8) has been lost from the sampled zone (top 15 cm) of the soil. The NH₄⁺-N may have been lost by NH₃⁰ volatilization before nitrification took place or some NH₄⁺-N fixation may have occurred in the soil, but the loss of the NO₃⁻-N is not so easily explained. Nitrate can be lost by leaching; but, the normal rainfall is low in the Piceance Basin so it is doubtful that leaching removed appreciable amounts of NO₃⁻-N. Denitrification could occur if the soil were saturated or nearly so for extended time periods. Again, there were no extensive time periods wherein the soils had a high water level. Additionally, denitrification requires a plentiful organic energy supply. Since these subsoil plots had little organic matter, energy from organic oxidations was severely limited--so, denitrification was not a feasible explanation for the NO₃⁻ loss. Obviously, the plants absorbed some of the NO₃⁻ and NH₄⁺-N. This leaves no logical explanation for the nitrogen losses at this particular time.

The total nitrogen picture appears equally perplexing. The control plots had about 500 ppm nitrogen when sampled. Most of the other plots have similar nitrogen contents. Even the 1,333 ppm nitrogen of the highest sludge application plot (Treatment 15) was not significantly different ($p=.05$) from the control plots. Total nitrogen analyses verifies the loss of most of the NH₄⁺ and NO₃⁻ nitrogen from the soil.

Only two treatments caused a significant increase in the extractable phosphorus. These included the two highest sewage sludge applications (Treatments 14 and 15). With the overall addition of 130 kg/ha of phosphorus to each plot, any differences in extractable phosphorus due to the treatments have been masked except for the two treatments mentioned above.

Vegetation Data for Subsoil Plot

Annual nitrogen treatments at 56, 112, 224, and 448 kg/ha have been applied for two growing seasons. When compared with the control where no nitrogen fertilizer was applied, 224 kg N/ha resulted in significantly lower density of grasses than the control. Similar results were seen in the total seeded species density on the 224 and 448 kg N/ha plots (Tables 15 and 16). The density of forbs and shrubs was not significantly affected by the annual N applications.

The 56 kg N/ha treatment significantly increased the total dry weight of seeded species ($p=.05$) above the control while the three higher rates of annual nitrogen decreased dry weight when compared with the control (Table 16). The two highest N treatments significantly lowered above-ground biomass of total seeded and invading species when compared with the control.

In summary, the second year of annual nitrogen applications did not increase plant density or cover; but, the lowest rate did significantly increase dry weight. At rates above annual applications of 56 kg N/ha plant responses generally declined, which may be due to a salt effect during

Table 15. Density, dry weight, and percent cover of grasses, forbs, and shrubs during the second growing season on the Subsoil Plot in response to various fertilizer treatments.¹

Fertilizer Treatment Number	Density ² (plants/m ²)			Dry Weight ² (kg/ha)			Cover ² (%)		
	Seeded Grasses	Seeded Forbs	Seeded Shrubs	Seeded Grasses	Seeded Forbs	Seeded Shrubs	Seeded Grasses	Seeded Forbs	Seeded Shrubs
1	31.6 ^{bcdef}	4.4 ^a	8.8 ^b	1456 ^{ab}	64 ^a	792 ^a	7.7 ^{abc}	0.7 ^a	21.7 ^a
2	26.8 ^{abcde}	2.0 ^a	5.6 ^{ab}	1308 ^{ab}	28 ^a	688 ^a	7.0 ^{abc}	0.2 ^a	17.0 ^a
3	14.0 ^{abc}	0.8 ^a	3.6 ^{ab}	788 ^{ab}	4 ^a	352 ^a	4.2 ^{ab}	0.1 ^a	12.7 ^a
4	17.6 ^{abcd}	1.6 ^a	4.4 ^{ab}	828 ^{ab}	60 ^a	528 ^a	4.5 ^{abc}	0.9 ^a	11.6 ^a
5	29.2 ^{abcdef}	4.8 ^a	6.0 ^{ab}	1000 ^{ab}	32 ^a	744 ^a	4.9 ^{abc}	0.6 ^a	16.6 ^a
6	20.4 ^{abcd}	2.4 ^a	3.6 ^{ab}	924 ^{ab}	4 ^a	1040 ^a	5.0 ^{abc}	0.1 ^a	21.4 ^a
7	19.6 ^{abcd}	2.4 ^a	4.8 ^{ab}	656 ^{ab}	16 ^a	236 ^a	5.3 ^{abc}	0.3 ^a	5.6 ^a
8	11.2 ^{ab}	2.8 ^a	2.8 ^{ab}	528 ^a	24 ^a	504 ^a	3.1 ^a	0.4 ^a	12.3 ^a
9	27.2 ^{abcde}	3.2 ^a	3.6 ^{ab}	1144 ^{ab}	48 ^a	968 ^a	4.6 ^{abc}	0.7 ^a	23.2 ^a
10	27.2 ^{abcde}	1.6 ^a	8.4 ^{ab}	1244 ^{ab}	8 ^a	400 ^a	5.1 ^{abc}	0.1 ^a	10.0 ^a
11	22.8 ^{abcde}	1.6 ^a	3.2 ^{ab}	748 ^{ab}	16 ^a	536 ^a	4.8 ^{abc}	0.2 ^a	13.5 ^a
12	9.6 ^a	0.8 ^a	0.8 ^a	520 ^a	36 ^a	116 ^a	2.9 ^a	0.7 ^a	3.4 ^a
13	42.8 ^{ef}	1.2 ^a	3.2 ^{ab}	1816 ^{ab}	44 ^a	1068 ^a	8.6 ^{abc}	0.9 ^a	22.5 ^a
14	36.8 ^{def}	2.4 ^a	3.6 ^{ab}	2012 ^b	20 ^a	340 ^a	9.9 ^{bc}	0.2 ^a	8.8 ^a
15	50.0 ^f	2.4 ^a	5.2 ^{ab}	1952 ^b	148 ^a	144 ^a	11.0 ^c	1.3 ^a	4.5 ^a
16	24.0 ^{abcde}	2.4 ^a	5.2 ^{ab}	1276 ^{ab}	56 ^a	704 ^a	7.8 ^{abc}	1.0 ^a	15.1 ^a
17	9.2 ^a	3.2 ^a	1.6 ^{ab}	548 ^a	80 ^a	188 ^a	3.3 ^a	1.2 ^a	4.9 ^a
18	37.2 ^{def}	4.8 ^a	6.4 ^{ab}	1548 ^{ab}	40 ^a	1056 ^a	5.8 ^{abc}	1.1 ^a	22.2 ^a
19	8.8 ^a	2.0 ^a	1.6 ^{ab}	548 ^a	36 ^a	308 ^a	2.4 ^a	1.0 ^a	10.7 ^a
20	35.2 ^{cdef}	5.2 ^a	8.4 ^{ab}	1376 ^{ab}	52 ^a	716 ^a	7.2 ^{abc}	0.9 ^a	18.7 ^a

¹Refer to Table 13 for a description of the fertility treatments.

²Means followed by the same letter are not statistically significant at the .05 level.

the growing season. Nitrogen applied annually at 448 kg/ha significantly increased the salt content (EC) in the soil above the control as early as May 1979 when soil samples were taken (Table 14). This may have caused the negative plant responses at high nitrogen application levels.

Initially Applied N

Nitrogen initially applied in the fall of 1977 at 448, 896, and 1792 kg/ha but not applied thereafter (Treatments 6, 7, and 8; Table 15) significantly decreased seeded grass density below the control, but total seeded density for all life forms was decreased at only the highest rate of N (Treatment 8; Table 16). In general, these applications have resulted in a decrease in density of all life forms.

Dry weight for all seeded and invading species was not significantly reduced by the high initial applications, although in grasses and forbs there was a decreasing trend in dry weight as the amount of nitrogen application increased and in shrubs there was an increasing trend in dry weight up to 896 kg/ha before dry weight declined (Table 15).

Cover of forbs was decreased below the control at all levels of initially applied nitrogen; however, none of the decreases in cover as a response to high rates of initial nitrogen application were significant (Table 15). Again, the decreases in density, cover, and biomass with higher N applications may have been a result of increased salt levels in the soil.

Table 16. Density, dry weight, and percent cover of total seeded species (grasses, forbs, and shrubs), total invaders, and total seeded plus invaders during the second growing season on the Subsoil Plot in response to various fertilizer treatments.¹

Fertilizer Treatment Number	Density ² (plants/ m ²)			Dry Weight ² (kg/ha)			Cover ² (%)		
	Total Seeded	Total Invader	Total	Total Seeded	Total Invader	Total	Total Seeded	Total Invader	Total
1	44.8 def	4.0 ^a	48.8 bcdef	2308 ^{ab}	32 ^a	2340 ^{ab}	30.1 ^a	0.3 ^a	30.4 ^a
2	34.8 ^{abcdef}	11.6 ^a	46.4 ^{abcde}	2024 ^{ab}	100 ^a	2124 ^{ab}	24.2 ^a	1.0 ^a	21.2 ^a
3	18.4 ^{abcd}	4.8 ^a	23.2 ^{abc}	1144 ^{ab}	80 ^a	1228 ^{ab}	17.1 ^a	1.0 ^a	18.1 ^a
4	24.0 ^{abcde}	3.6 ^a	27.6 ^{abcd}	1412 ^{ab}	56 ^a	1472 ^{ab}	17.0 ^a	1.7 ^a	18.7 ^a
5	39.6 ^{bcdef}	19.6 ^a	59.2 ^{de}	1776 ^{ab}	60 ^a	1836 ^{ab}	22.1 ^a	1.4 ^a	23.5 ^a
6	26.4 ^{abcde}	8.4 ^a	34.8 ^{abcd}	1972 ^{ab}	164 ^a	2136 ^{ab}	26.4 ^a	1.6 ^a	28.0 ^a
7	26.8 ^{abcde}	8.0 ^a	34.8 ^{abcd}	908 ^{ab}	160 ^a	1068 ^{ab}	11.2 ^a	1.9 ^a	13.1 ^a
8	16.8 ^{abc}	3.6 ^a	20.8 ^{ab}	1060 ^{ab}	176 ^a	1236 ^{ab}	15.8 ^a	5.7 ^a	21.5 ^a
9	34.4 ^{abcdef}	5.6 ^a	40.0 ^{abcd}	2164 ^{ab}	52 ^a	2216 ^{ab}	28.4 ^a	0.9 ^a	29.4 ^a
10	36.8 ^{abcdef}	6.8 ^a	43.6 ^{abcd}	1656 ^{ab}	52 ^a	1708 ^{ab}	15.2 ^a	0.7 ^a	15.9 ^a
11	27.6 ^{abcde}	7.2 ^a	34.8 ^{abcd}	1300 ^{ab}	96 ^a	1392 ^{ab}	18.5 ^a	3.1 ^a	21.6 ^a
12	11.2 ^a	1.6 ^a	12.8 ^a	672 ^a	172 ^a	844 ^a	7.2 ^a	4.5 ^a	11.6 ^a
13	47.2 ^{ef}	5.6 ^a	52.8 ^{bcde}	2928 ^b	76 ^a	3004 ^b	31.9 ^a	0.6 ^a	32.5 ^a
14	42.4 ^{cdef}	12.4 ^a	54.8 ^{bcde}	2376 ^{ab}	160 ^a	2536 ^{ab}	18.9 ^a	1.8 ^a	20.7 ^a
15	57.6 ^f	22.4 ^a	79.6 ^e	2248 ^{ab}	204 ^a	2452 ^{ab}	16.8 ^a	3.1 ^a	19.9 ^a
16	31.6 ^{abcdef}	6.4 ^a	38.4 ^{abcd}	2036 ^{ab}	24 ^a	2064 ^{ab}	24.0 ^a	1.0 ^a	25.0 ^a
17	14.0 ^{ab}	6.8 ^a	20.8 ^{ab}	816 ^{ab}	336 ^a	1152 ^{ab}	9.4 ^a	8.1 ^a	17.5 ^a
18	48.4 ^{ef}	6.4 ^a	54.8 ^{bcde}	2644 ^{ab}	80 ^a	2724 ^{ab}	29.1 ^a	0.8 ^a	29.9 ^a
19	12.8 ^{ab}	11.2 ^a	24.0 ^{abcd}	896 ^{ab}	160 ^a	1052 ^{ab}	14.0 ^a	3.0 ^a	17.1 ^a
20	48.8 ^{ef}	8.4 ^a	57.2 ^{cde}	2144 ^{ab}	84 ^a	2228 ^{ab}	26.7 ^a	1.0 ^a	27.7 ^a

¹Refer to Table 13 for a description of the fertility treatments.

²Means followed by the same letter are not statistically significant at the .05 level.

Initial N Plus Wood Wastes

The application of 1,792 kg/ha N plus 90 MT/ha of wood wastes caused a significant decrease in the density of seeded grasses and total seeded species as well as in the sum of seeded and invader species. This significant decrease in density was not apparent with forb, shrub, or invading species when considered separately (Tables 15 and 16).

None of the nitrogen and wood waste treatments had a significant effect ($p < .05$) on the dry weight of seeded or invading species (Tables 15 and 16).

The cover of seeded grasses was significantly decreased by the highest application of nitrogen

along with wood wastes (Treatment 12; Table 15) when compared to the control. The other treatments (Treatments 9, 10, and 11) caused the grass cover to be lower than that of the control plot, but these differences were not statistically significant ($p < .05$).

In almost all cases, the addition of nitrogen fertilizer and wood wastes to the subsoil plots caused a decrease in density, dry weight, and cover of all life forms compared to the control. This may be due to the relatively low requirements of the native seeded species used in this study for nitrogen and the relatively high nitrogen application rates. Certainly the highest nitrogen treatments were excessively high for most plant species and would be expected to produce a salt effect on the plants.

It should be noted that the dry weight and cover values of invading species were higher for the 896 and 1,792 kg/ha N with wood waste (Treatments 11 and 12; Table 16) than for the control treatments, although not significant at the .05 probability level. This may be a result of the detrimental effect of the high nitrogen rates on the seeded species. They provided less competition for the invading species, therefore allowing more invasion to occur.

Sewage Sludge With and Without Wood Waste

Although there appeared to be less detrimental effect on plant density due to the sewage sludge plus wood wastes than high nitrogen with wood wastes, there was not a significant increase in density with the sludge and wood waste treatments (Table 15). The lowest sewage sludge application rate (Treatment 13, 56 MT/ha) produced the greatest dry weight and cover values for total seeded and total seeded and invader species (Table 16).

In general, the 1979 measurements indicate that there were more beneficial effects and less detrimental effects with sludge applications to the subsoil plots than most of the other treatments used in the study. This could have resulted from a steady supply of mineralized nitrogen and phosphorus and/or from an enhancement of the physical properties of the soil that could affect moisture, temperature, aeration, and nutrient supply. Differences in microbial numbers or activity could also account for the beneficial effects. Microbial studies have shown a relatively high dehydrogenase activity and phosphatase activity for these sludge treatments so there appears to be a correlation between these microbial activities and plant growth and development.

N Plus P

The results from the high N treatments (Treatments 17 and 19) were similar to the results from Treatments 7 and 8 shown in Tables 15 and 16. It appears that initial high levels of N depressed plant growth of seeded native species. Again, this reduction in growth of seeded species may have allowed the apparent increase in invader establishment and growth shown in Table 16. However, the increase in invasion on Treatments 17 and 19 was not significantly greater than the control at the .05 probability level.

The data suggests that, in general, an N level of less than 112 kg/ha would be optimum for growth of native plant species since this rate or lower applications have resulted in greater plant growth than the control. Also, the 130 kg P/ha base application was an adequate P application since the 192 kg P/ha did not increase plant growth beyond that of the control plots that received the base application of 130 kg P/ha.

Soil Data for Topsoil Over Retorted Shale Plot

Table 17 and 18 show the variation in chemical parameters determined on soil taken at two depths from the Topsoil Over Retorted Shale Plot.

Only a few differences were significant. The sludge treated plots (Treatments 13-15) had a lower pH in the surface soil because of the biological decomposition of the added organic matter and production of acids (Table 17). The highest rate of sludge plus wood waste (Treatment 15) also had the highest salt content. Although there were significant differences in $\text{NH}_4^+\text{-N}$ and $\text{NO}_3^-\text{-N}$ content due to fertilizer treatments, they were not meaningful because of the rather low N levels remaining in the soil compared to the large amounts of N added. It is noteworthy that the two highest sludge plus wood waste treatments (Treatments 14 and 15) contained the highest total N content in the topsoil. Those same treatments also had the greatest extractable P content in the soil samples.

Table 17. Influence of fertility treatments on the soil parameters determined on the Topsoil Over Retorted Shale Plot at Depth 1 (0-6.5 cm). (See Table 13 for a description of fertilizer treatments.)

Fertilizer Treatment Number	Parameter ¹						
	pH	EC (mmhos cm^{-1})	$\text{NH}_4^+\text{-N}$ (ppm)	$\text{NO}_3^-\text{-N}$ (ppm)	Total N (ppm)	Extractable Phosphorus ² (ppm)	
1	8.4	cd	3.5 ^{ab}	4.0 ^{ab}	2.7 ^{ab}	687 ^a	30.3 ^{ab}
2	8.1	cd	2.9 ^a	6.2 ^{abc}	3.9 ^{ab}	857 ^a	23.6 ^{ab}
3	8.1	cd	4.3 ^{ab}	13.0	8.0	760 ^a	19.4 ^{ab}
4	8.0	bcd	4.9 ^{ab}	14.2	12.1	830 ^a	24.5 ^{ab}
5	8.0	bcd	3.6 ^{ab}	2.3 ^{ab}	2.2 ^{ab}	827 ^a	25.8 ^{ab}
6	8.1	cd	3.7 ^{ab}	3.8 ^{ab}	5.5 ^{ab}	743 ^a	24.4 ^{ab}
7	8.1	cd	3.2 ^{ab}	4.5 ^{ab}	2.6 ^{ab}	643 ^a	22.5 ^{ab}
8	8.1	cd	3.3 ^{ab}	9.3	5.1 ^{ab}	1003 ^{ab}	14.5 ^a
9	8.1	cd	3.6 ^{ab}	5.2 ^{ab}	1.9 ^a	703 ^a	18.9 ^{ab}
10	7.9	abcd	3.7 ^{ab}	3.2 ^{ab}	1.6 ^a	930 ^a	20.2 ^{ab}
11	8.0	bcd	4.4 ^{ab}	5.4 ^{ab}	1.8 ^a	817 ^a	20.7 ^{ab}
12	8.0	bcd	5.0 ^{ab}	6.3 ^{abc}	7.0 ^{abc}	807 ^a	15.9 ^a
13	7.9	abcd	3.7 ^{ab}	5.7 ^{ab}	3.9 ^{ab}	913 ^a	32.0 ^{ab}
14	7.8	ab	3.8 ^{ab}	2.9 ^{ab}	4.6 ^{ab}	1987 ^b	40.9 ^b
15	7.7	a	5.4	4.9 ^{ab}	6.0 ^{ab}	1207 ^{ab}	40.6 ^b
16	8.2	d	3.5 ^{ab}	3.5 ^{ab}	2.8 ^{ab}	1153 ^{ab}	18.5 ^{ab}
17	8.0	bcd	3.2 ^{ab}	4.7 ^{ab}	2.8 ^{ab}	907 ^a	13.3 ^a
18	8.1	cd	3.5 ^{ab}	4.2 ^{ab}	2.9 ^{ab}	977 ^a	26.0 ^{ab}
19	7.9	abcd	3.9 ^{ab}	5.1 ^{ab}	3.3 ^{ab}	927 ^a	27.1 ^{ab}
20	8.1	cd	3.7 ^{ab}	2.2 ^a	1.6 ^a	957 ^a	30.1 ^{ab}

¹Means followed by the same letter are not statistically significant at the .05 level.

²Phosphorus determined by the Olsen's sodium bicarbonate extractable method (Watanabe and Olsen 1965).

Many of the significant differences in the soil parameters measured in the surface soil (Table 17) because of fertility treatments were not apparent in the deeper sampling depth (Table 18).

Vegetation Data for Topsoil Over Retorted Shale Plot

Because of heavy rodent damage during the winter of 1978-1979 this plot was reseeded in May

1979 in order to reestablish a new stand of native vegetation. The test plot was sampled in August 1979 for density by life form. However, because of the very preliminary nature of this data and the lack of any significant differences at this time, the vegetation data has not been included in this progress report. More complete vegetation data will be collected during the 1980 growing season after plants have matured and treatment effects have become more pronounced.

Table 18. Influence of fertility treatments on the soil parameters determined on the Topsoil Over Retorted Shale Plot at Depth 2 (6.5-13 cm). (See Table 13 for a description of fertilizer treatments.)

Fertilizer Treatment Number	Parameter ¹					
	pH	EC (mmhos cm ⁻¹)	NH ₄ ⁺ -N (ppm)	NO ₃ ⁻ -N (ppm)	Total N (ppm)	Extractable Phosphorus ² (ppm)
1	8.2 ^b	4.0 ^a	2.3 ^a	2.9 ^a	830 ^a	10.5 ^{abc}
2	8.0 ^{ab}	3.0 ^a	5.3 ^a	3.9 ^a	1450 ^a	11.0 ^{abc}
3	8.2 ^b	4.4 ^a	8.8 ^a	6.3 ^a	850 ^a	9.3 ^{abc}
4	8.0 ^{ab}	3.8 ^a	9.9 ^a	10.6 ^a	770 ^a	8.7 ^{abc}
5	7.9 ^{ab}	2.8 ^a	4.3 ^a	3.8 ^a	820 ^a	11.9 ^{abc}
6	8.2 ^b	4.7 ^a	5.9 ^a	3.9 ^a	865 ^a	8.7 ^{abc}
7	8.2 ^b	5.2 ^a	2.9 ^a	2.7 ^a	885 ^a	7.8 ^{ab}
8	8.0 ^{ab}	4.7 ^a	7.0 ^a	5.0 ^a	820 ^a	5.2 ^a
9	8.2 ^b	4.8 ^a	2.2 ^a	2.6 ^a	820 ^a	8.7 ^{bc}
10	8.2 ^b	4.0 ^a	4.8 ^a	5.9 ^a	860 ^a	10.7 ^{abc}
11	8.0 ^{ab}	4.9 ^a	2.8 ^a	3.3 ^a	975 ^a	8.8 ^{abc}
12	8.1 ^b	4.5 ^a	5.5 ^a	6.0 ^a	710 ^a	11.3 ^{abc}
13	8.1 ^b	4.1 ^a	4.9 ^a	4.5 ^a	960 ^a	23.3 ^{abc}
14	8.0 ^{ab}	4.3 ^a	2.8 ^a	2.6 ^a	1300 ^a	25.2 ^{bc}
15	8.0 ^{ab}	6.7 ^a	1.8 ^a	6.5 ^a	2015 ^a	27.0 ^c
16	8.2 ^b	2.8 ^a	2.4 ^a	6.3 ^a	825 ^a	7.7 ^{ab}
17	8.2 ^b	3.1 ^a	6.0 ^a	4.8 ^a	905 ^a	8.1 ^{abc}
18	8.2 ^b	3.9 ^a	4.3 ^a	3.2 ^a	635 ^a	9.0 ^{abc}
19	7.6 ^a	3.2 ^a	6.2 ^a	4.7 ^a	870 ^a	11.7 ^{abc}
20	8.1 ^b	4.0 ^a	1.9 ^a	2.2 ^a	860 ^a	21.2 ^{abc}

¹Means followed by the same letter are not statistically significant at the .05 level.

²Phosphorus determined by the Olsen's sodium bicarbonate extractable method (Watanabe and Olsen 1965).

Legume Plot

Statistical analysis of the total nitrogen content of grasses grown in association with legumes on subsoil and topsoil with nitrogen added at 9, 50, and 100 kg/ha as ammonium nitrate shows that in the first growing season soil type, fertilizer N application level, and species of legume had no significant effect upon nitrogen content in the grasses. Sainfoin and alfalfa were sampled on July 3 and July 16, 1979, respectively, which were considered to be comparable stages of growth. During this period of time the average total nitrogen content of the grasses decreased from

2.4 percent to 2.1 percent. These results suggest that forage plants should be sampled on several dates for comparisons of nitrogen content. In the 1980 growing season sampling dates will begin as early as May (depending upon the weather) and continue weekly or biweekly through July.

Two legume species out of a possible five matured and were harvested on both subsoil and topsoil plots in the 1979 growing season. Statistical analysis of yields showed no response to level of fertilizer nitrogen. Legumes growing on topsoil had a significantly higher yield ($p=0.9$) than did legumes growing on subsoil, yielding 1.2 and 0.77 MT/ha, respectively. Species of legume also had a significant effect upon yield with sainfoin and alfalfa yielding an average of 1.1 and 0.85 MT/ha, respectively, when grown in pure stands.

Johnson et al. (1979) have shown that crested wheatgrass grown in association with alfalfa and milkvetch (*Astragalus falcatus*) on a semiarid rangeland site has significantly higher protein content and greater herbage yield than crested wheatgrass grown alone. These data were taken in May of 1979 after the stand had been established for eight years. Their work did not include yearly monitoring to determine how long it may take to see this effect. Successive growing seasons will be necessary to evaluate the usefulness of these legumes in improving the nitrogen status of the soil and the quality and quantity of grasses grown in combination with them.

CONCLUSIONS

1. The soil and vegetation data indicate that a large part of the inorganic nitrogen applied to the subsoil plots is not being recovered either in the soil or in the aboveground vegetation. It is probable that appreciable amounts of nitrogen are being lost through volatilization and perhaps leaching. Losses through denitrification are also possible although there is not sufficient soil moisture and organic matter to account for appreciable losses through denitrification.

2. Sewage sludge at the lowest rate (56 MT/ha) had the most beneficial overall effect of any of the fertilizer treatments in the subsoil plots.

3. Apparently the level of phosphorus applied in the control (130 kg P/ha) and several of the other treatments results in sufficient phosphorus in the soil for good establishment and growth of native species grown on the subsoil plots.

4. Inorganic nitrogen fertilizer applied annually at a rate of 56 kg/ha had some beneficial effect after two years upon the growth and establishment of the native species planted, while higher rates of inorganic fertilizer generally had detrimental effects.

5. After one year the legume species studied did not affect the nitrogen content of native grasses growing in association with the legumes.

LITERATURE CITED

- Johnson, D. A., M. D. Rumbaugh, and K. H. Asay. 1979. Physiological criteria for forage plant breeding. In Western Experiment Station Regional Project W-126 Annual Report.
- Watanabe, F. S., and S. R. Olsen. 1965. Test of an ascorbic acid method for determining phosphorus in water and NaHCO_3 extracts from the soil. Soil Sci. Soc. Am. Proc. 29:677-678.

ROLE OF SOIL MICROORGANISMS AS INDICATORS AND POSSIBLE CONTROLLING FACTORS IN PLANT SUCCESSION PROCESSES ON RETORTED SHALE AND DISTURBED SOILS

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OBJECTIVES

Phase I: To evaluate microbial responses and activity during reestablishment of plant communities on disturbed soils.

- (1-1) To monitor soil microbiological processes on test plots at the Intensive Study Site in the Piceance Basin.
- (1-2) To study the effects of topsoil storage on microbiological populations and on microorganism-related nutrient cycling.
- (1-3) To determine the effects of retorted shale on soil microbiological characteristics in laboratory studies.

Phase II: To develop approaches for controlling or modifying plant-microbe interactions.

- (2-1) To evaluate the role of symbiotic nitrogen-fixing plants in community establishment.

ANALYTICAL PROCEDURES

Field Plots Sampled

During the 1979 field season samples were collected from the following plots: Long-Term Fertility Plot (May), Revegetation Technique Plot (May and July), Retorted Shale Succession Plot (June), and Topsoil Storage Plot (May). Also, reference samples were collected adjacent to the Revegetation Technique Plot during the May sampling period to provide controls for evaluating soil organic matter responses to the various planting mixtures and management techniques evaluated in this experiment.

Basic Sampling Procedures

The procedures for analysis of soil pH, soil organic matter, microbial populations, nitrogen fixation, dehydrogenase, phosphatase, ATP, and ^{14}C glucose mineralization have been described in previous progress reports.

Effects of Added Ammonium Ion on Nitrogen Fixation in Surface Soil-Shale Mixtures

Soil from the Revegetation Technique Plot (unfertilized subplots sampled on May 5, 1979) were composited for use in this study. This composited soil was mixed with retorted shale to obtain mixtures of 90 percent soil-10 percent shale and 75 percent soil-25 percent shale. In addition, the shale was used alone. All materials were passed through 32-mesh sieves before use. Ammonium ion was added as ammonium chloride.

The soil water holding capacity was determined using the procedure of Peters (1965) as modified by Sherwood (1979). For most experiments, if the soils were at an appropriate soil water level, the soils were simply amended with a standard 0.5 percent w/v glucose solution. The solutions of glucose at 0.5 percent w/v were also prepared to give final ammonium ion nitrogen levels of 0, 50, and 200 $\mu\text{g/g}$ of soil in the first test systems. In later experiments 0, 5, 10, 15, 35, and 50 μg of $\text{NH}_4^+\text{-N/g}$ soil were added.

The acetylene reduction values were determined as described in previous reports.

Effects of Retorted Oil Shale Water on Acetylene Reduction by a *Rhizobium* Species

Sieved (32 mesh) Paraho retorted oil shale in an amount of 60 g was combined with 540 ml of deionized, distilled water and mixed for one week. Following mixing the supernatant was decanted and

autoclaved. *Rhizobium* 32H1 (Nitragin Company, Milwaukee, Wisconsin) was incubated at 25°C for 96 hours in the following growth medium: sodium gluconate, 5.0 g; mannitol, 5.0 g; K₂HPO₄, 1.28 g; yeast extract, 1.0 g; MgSO₄·7H₂O, 0.1 g; casamino acids, 0.1 g; CaCl₂, 0.06g; FeSO₄, 7.3 mg; NaMoO₄·2H₂O, 2.3 mg; deionized H₂O, 1000 ml; pH 7.2. After incubation the cultures were centrifuged at 17,000 x gravity for 10 minutes; the supernatant was discarded. The cells were washed and resuspended in the following assay medium: sodium gluconate, 3.0 g; K₂HPO₄, 8.7 g; sodium glutamate, 1.0 g; MgSO₄·7H₂O, 48 mg; iron citrate, 33.5 mg; NaMoO₄·2H₂O, 18 mg; deionized H₂O, 1000 ml; pH 7.2. Cell suspensions were adjusted to 35 Klett units (Klett-Summerson Colorimeter, New York), and 10 ml of the cell suspension were aseptically transferred to a 72 ml serum bottle. To each bottle either 0.025, 0.1, 1.0, 2.0, or 3.0 ml of retorted oil shale water were added. The bottles were sealed with rubber stoppers; and, using a sterile syringe 5 ml of atmosphere were replaced with 5 ml of acetylene. After incubation at 25°C in the dark for 14 days acetylene reduction values were determined.

RESULTS

Revegetation Technique Plot

Strong correlations between irrigation and dehydrogenase activity, N₂ fixation, phosphatase, and organic matter were observed (Table 19). Other interactions of importance included species used with phosphatase and soil organic matter and a species x irrigation interaction. (The Revegetation Technique Plot is described by Redente et al. in this report.)

The effects of different plant species mixtures on soil organic matter levels in the irrigated

and non-irrigated plots are shown in Figure 16. With irrigation higher relative soil organic matter levels were observed with the native and introduced

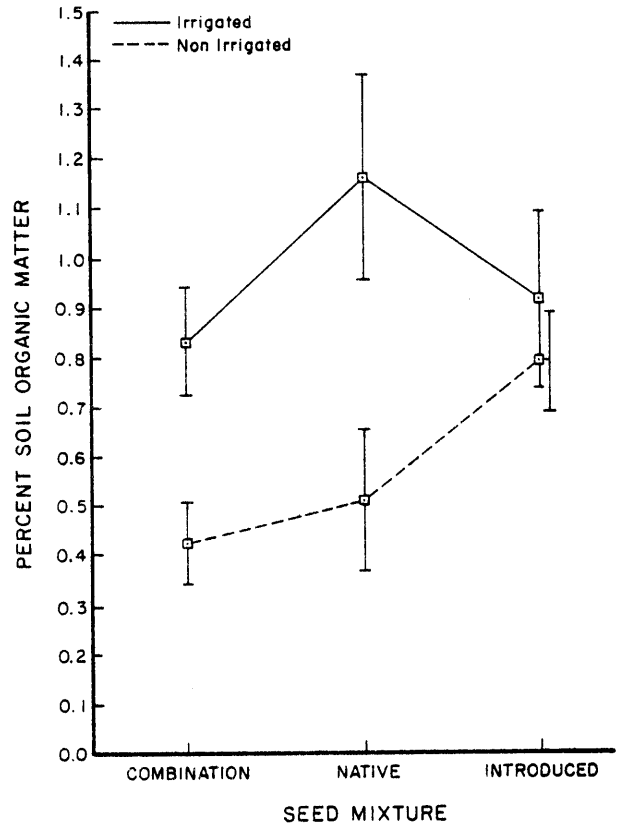


Figure 16. Effects of irrigation and species mixtures on the soil organic matter content for the July 1979 sampling. Means and standard deviation are shown.

Table 19. Significant analysis of variance results for 1979 samplings on the Revegetation Technique Plot.

Variables	Measured Response Parameters					
	Moisture	pH	Dehydrogenase	N ₂ -Fixation	Phosphatase	Organic Matter
<u>Management variables</u>						
Irrigation			**	**	**	**
Fertilization						
Species	**				**	**
Seeding technique		**				
Sampling date	**	**	**	*	**	**
<u>Interactions of variables</u>						
Irrigation x Fertilization		*				
Irrigation x Species	**	**	**		**	
Fertilization x Species		*				
Species x Seeding technique		**				
Irrigation x Species x Seeding technique		*				

* = Significant at .05 level.

** = Significant at .01 level.

mixtures; without irrigation the subplots seeded to introduced species had higher soil organic matter levels. This same trend was noted for all sampling in 1978 and 1979. Similar relative effects of irrigation and species on dehydrogenase, N_2 fixation, and phosphatase activities were observed. It is important to note that irrigation was only applied during the 1977 and 1978 seasons.

Significant effects of irrigation and fertilizer on nitrogen fixation potential were indicated when the combined 1978 and 1979 data for the Revegetation Technique Plot were analyzed. The fertilizer effect was at a 2 percent probability level, and the irrigation effect on N_2 fixation occurred at a 1 percent probability level. On a similar basis, when both years were considered, a fertilizer effect on phosphatase activity occurred at a 1.1 percent probability level.

With different plant species varied nitrogen fixation potentials were noted, independent of irrigation and fertilizer variables. For the combined 1978 and 1979 years the plots where introduced species were seeded had a higher N_2 fixation potential. This was significant at the 7.4 percent probability level and indicates a trend which may become more distinct in the coming seasons.

In a similar manner, the phosphatase activities of the soils were strongly influenced by the plant species seeded at a 1 percent probability level with the introduced species showing the highest activities. Higher relative responses were also shown with irrigation, and fertilization caused slight decreases in phosphatase activity.

These results suggest that different species mixtures will have distinct effects on the below-ground processes at the test site and that fertilization may tend to lead to decreased nitrogen fixation and phosphatase activities.

Controls which were taken across all test plots outside of the zones where plant growth had taken place did not indicate that significant differences in soil organic matter levels existed prior to test plot construction and the application of revegetation techniques.

Surface Disturbed Succession Plot

This plot, which was also sampled two times during the 1979 field season, yielded additional information on the effects of the test management variables on the development of the belowground ecosystem. (The Surface Disturbed Succession Plot is described by Redente et al. in this report.)

As noted in the 1978 year, the major interaction for this plot was that of species with soil organic matter levels (Table 20). The nitrogen fixation potential was significantly reduced for the 1979 sampling ($p < .06$) where mulch was used. When the data for all species were averaged on the non-fertilized subplots, the mulched plots had nitrogen fixation activities of 2.60 nanomoles of ethylene produced per gram of soil while the non-mulched soils had a value of 3.24 nanomoles. As was noted for the 1978 sampling, higher soil organic

matter levels were noted for the introduced grass and the native grass-forb-shrub mixtures.

Table 20. Soil organic matter levels in soils with and without mulch treatments from the Surface Disturbed Succession Plot for May 1979 sampling.¹

Species Mixture	Mulched	Not Mulched
Native grass	0.97 ± 0.33	1.20 ± 0.35
Introduced grass	1.55 ± 0.27	1.42 ± 0.34
Native grass-forb	0.85 ± 0.28	1.14 ± 0.04
Introduced grass-forb	1.27 ± 0.36	1.35 ± 0.24
Native grass-forb-shrub	1.43 ± 0.27	1.46 ± 0.20
Native and Introduced grass-forb-shrub	1.34 ± 0.78	1.37 ± 0.49

¹Percent organic matter by weight, means and standard deviation are shown.

When the data for the Surface Disturbed Succession Plot were averaged over the 1978 and 1979 years, a species effect on dehydrogenase activity was noted at an 8.1 percent probability level and a mulch effect on nitrogen fixation was significant at an 8.5 percent probability level. From the individual analyses for the 1978 and 1979 years, the combined data indicated a strong effect on surface soil organic matter content. No significant interactions with phosphatase activity were noted.

Retorted Shale Succession Plot

The Retorted Shale Succession Plot which was sampled for the first time during the 1979 field season provided the first direct information concerning possible effects of retorted oil shale on the microbiological characteristics of revegetated soils placed over this material. (The Retorted Shale Succession Plot is described by Redente et al. in this report.)

The analyses of variance for this experiment showed significant panel effects across the various soil-shale profile treatments. The pH values were in the range of 8.6 to 8.8 for all the panels including the shale-to-surface treatment while the control soil had a pH of approximately 8.4. For the specific biological parameters the control and capillary barrier panels had generally more similar activity levels while the soils placed directly over shale material had lower values. The dehydrogenase results are shown in Figure 17. The soils placed directly over retorted shale had lower activity values while the capillary barrier soils

had higher activity values and the control soil had the highest dehydrogenase activity. In comparison the shale cover without a soil cover had negligible dehydrogenase activity.

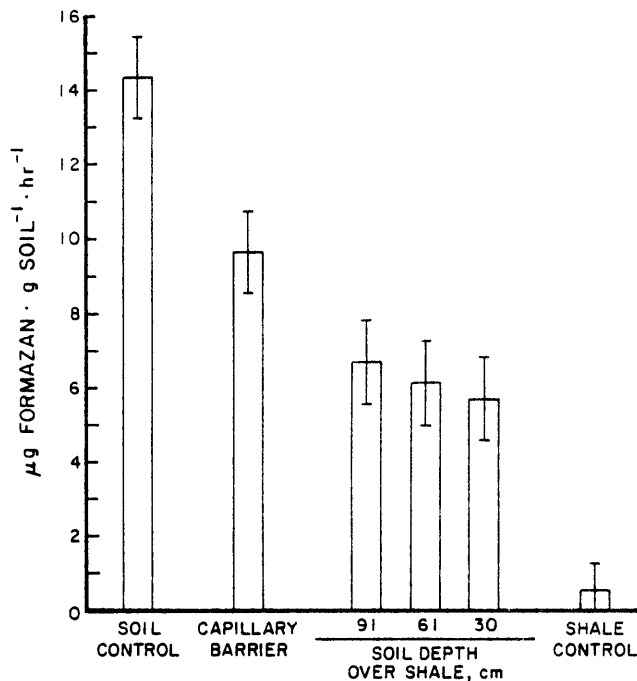


Figure 17. Effects of soil depth and a capillary barrier on the dehydrogenase activity of revegetated soils located over retorted oil shale. The least significant difference range ($p < .05$) is given.

The surface soil organic matter values are shown in Figure 18. The main trend is for the shallower soils placed directly over the shales to have lower organic matter levels which were significantly lower than found in the control, capillary barrier, and 91 cm topsoil treatments. For the phosphatase and nitrogen fixation activities a more distinct set of trends was evident (Figures 19 and 20). For both of these assays the control and capillary barrier treatments had similar values; the surface soils placed directly over the shale materials had distinctly lower values. This was most evident in the case of the nitrogen fixation potential where the soils placed over the shale materials showed approximately one-half the activity of the control and capillary barrier treatments.

When the shale-to-surface panel was not included in the analyses of variance, a species-fertilizer interactive effect on acetylene reduction potential of the soils was suggested at a 4.7 percent probability level. These data indicate that with the native plant mixture a typical repression of nitrogen fixation potential had occurred with added fertilizer nitrogen (Figure 21). In contrast, in the test plots with introduced plants increased nitrogen fixation potentials were observed in the presence of added mineral nitrogen and phosphorus in comparison with the non-fertilized plots. This could be related to possible increased growth of

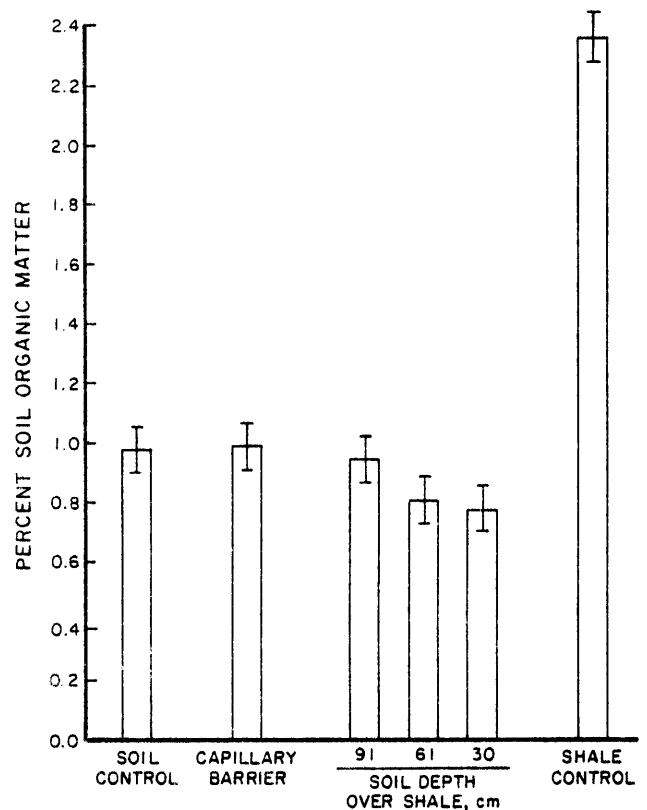


Figure 18. Effects of soil depth and a capillary barrier on the percent organic matter of revegetated soils located over retorted oil shale. The least significant difference range ($p < .05$) is given.

the introduced species which might lead to greater releases of organic matter to the surface soil in the plant root zone. However, no evidence of this type of relationship was suggested by the overall surface soil organic matter assays carried out during 1979.

These results suggest that a capillary barrier may play a vital role in maintaining the integrity of surface soil to allow the development of a plant-microbial system and shale materials may be influencing microbial processes when a capillary barrier is not present. These responses may also be due to the differences in plant growth or soil water contents which may be having secondary effects on microbiological processes.

Stored Topsoil Experiment

The analysis of variance results for the stored topsoil experiment in which four cores at six depths were used are summarized in Table 21. For the majority of the parameters no bore or soil depth effects were evident after 1.5 years of storage. For several of the parameters (including soil moisture, pH, glucose mineralization, mycorrhizal infection, fungi, dehydrogenase, and pH 6.5

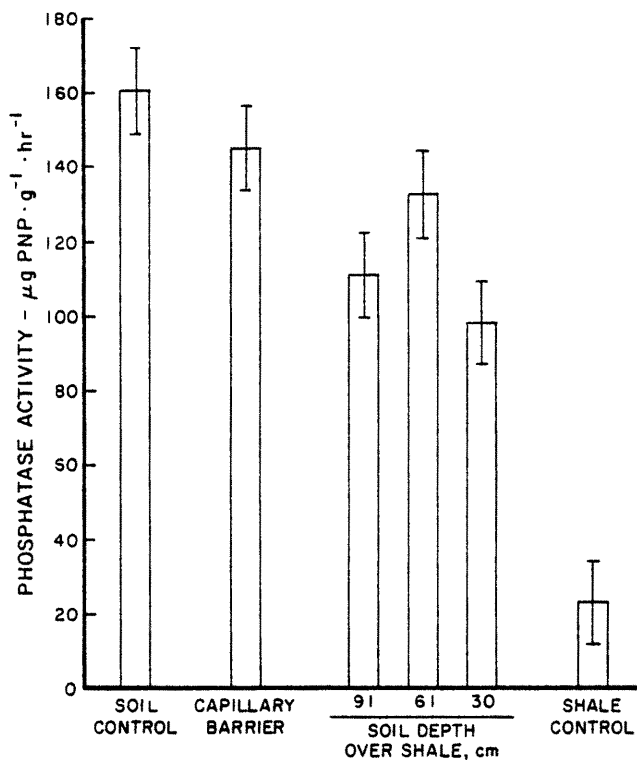


Figure 19. Effects of soil depth and a capillary barrier on the phosphatase activity of revegetated soils located over retorted oil shale. The least significant difference range ($p \leq 0.05$) is given.

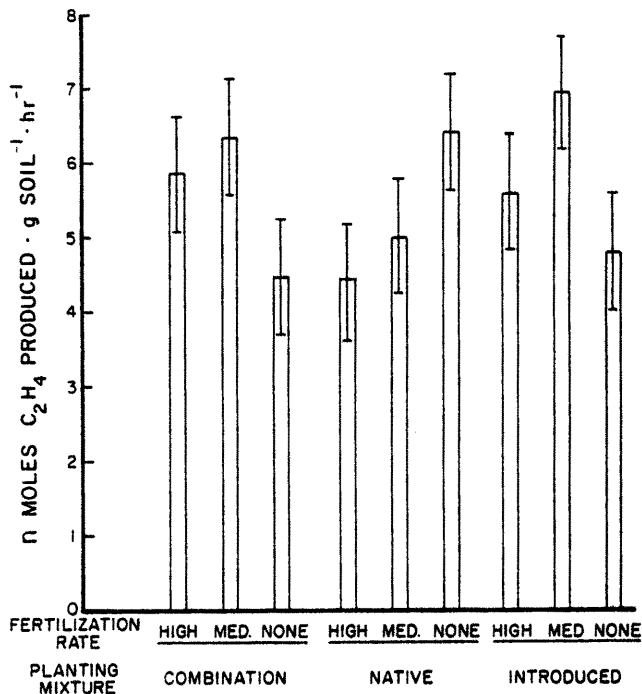


Figure 21. Seeding mixture-fertilizer rate interactive effects on acetylene reduction on the Retorted Shale Succession Plot for 1979. The high fertilization rate is 112 N and 56 P (kg/ha); the medium (med.) rate is 56 N and 28 P (kg/ha).

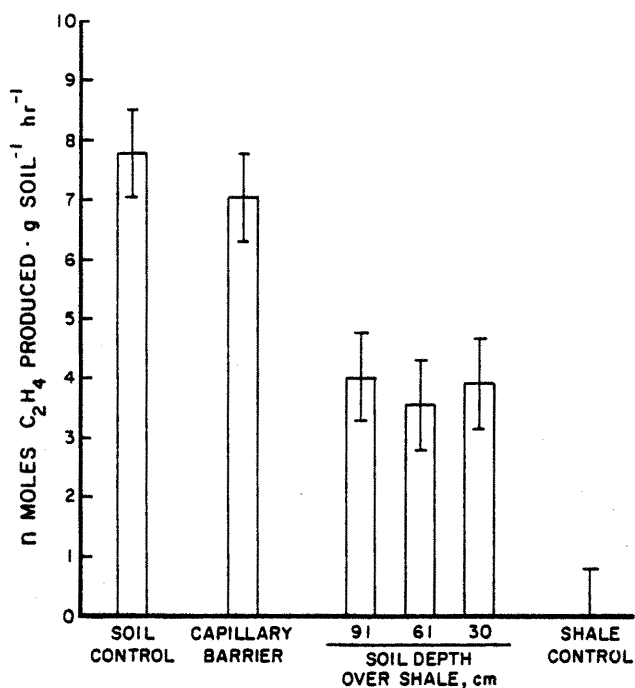


Figure 20. Effects of soil depth and a capillary barrier on the nitrogen fixation potential of revegetated soil located over retorted oil shale. The least significant difference range ($p \leq 0.05$) is given.

Table 21. Analysis of variance results for 1979 for the Stored Topsoil Plot.

Parameter Monitored	Significance	
	Bore	Depth
Soil moisture	*	**
Soil pH	**	
Glucose mineralization	**	
Mycorrhizal infection potential	**	*
Fungal viable counts	*	
Actinomycete viable counts		
Bacterial viable counts		
Dehydrogenase	*	
N ₂ fixation		
Phosphatase pH 5.5		
Phosphatase pH 6.5	**	
Phosphatase pH 8.5		
Soil organic matter		
ATP content of soils		

* = Significant at the .05 level.
 ** = Significant at the .01 level.

phosphatase activity) significant differences were observed between the bores. Only the soil moisture ($p \leq 0.01$) and the mycorrhizal infection potential ($p \leq 0.05$) were significant at various depths. Based on comparisons between the 1978 and 1979 analyses of mycorrhizal infection from the soil, the average percent infection for 1978 was in the range of 19 to 25 percent at all depths while for 1979 this level of infection was only maintained at the lowest depth (for more information refer to the following report on mycorrhizae).

The analysis of variance data for the combined 1978 and 1979 samplings suggests that the major effects which were observed over the 1.5 year period of topsoil storage were due to bore variations and that only with the soil moisture, N_2 fixation potential, soil organic matter, and bacteria were depth or year by depth interactions significant. (Mycorrhizal infection data were not included in this two-year analysis.)

Shale and Added Ammonium Ion Effects on Soil Nitrogen Fixation Potential

Studies carried out by Hersman and Klein (1979) have shown that retorted shale when mixed with surface soil will cause distinct decreases in microbial nitrogen fixation potential. Data from the Revegetation Technique and Surface Disturbed Succession Plots, as noted in this report, suggest that fertilization can lead to a suppression of the natural nitrogen fixation potential of these soils.

In this experiment the combined effects of retorted oil shale and low levels of ammonium ion on nitrogen fixation were examined. This study was conducted based on measured nitrogen levels in surface soil, shale, and shale-soil mixtures (Table 22). These data indicate that the surface soil has a lower level of total nitrogen but higher levels of nitrate and ammonium in comparison with the retorted shale. The retorted shale has a very low level of either of these ions which might be related to the poor microbial growth that has been observed when this material has been tested for its suitability as a nitrogen source.

Table 22. Total nitrogen and extractable nitrate and ammonium ion in surface soil, retorted shale, and soil-shale mixtures from the Intensive Study Site.¹

Sample (%)		% Total N	$\mu\text{g/g}^2$	
Surface Soil	Shale		$\text{NO}_3\text{-N}$	$\text{NH}_4\text{-N}$
100	0	0.979	10	49
95	5	0.086	15	41
90	10	0.078	18	24
75	25	0.086	16	21
0	100	0.145	2	3

¹Data from the soil testing laboratory, Colorado State University.

² NH_4^+ was extracted using 2M KCl.

The effects of retorted shale and added ammonium ion were tested in a series of experiments where nitrogen as ammonium ion was added in the range of 50 to 100 $\mu\text{g/g}$. In all of these initial experiments it was found that essentially no nitrogen fixation activity could be observed with ammonium ion present. In later experiments a range of added ammonium ion from 0 to 30 $\mu\text{g NH}_4^+\text{-N/g}$ was used, and it was possible to observe a suitable response gradient (Figure 22).

Under these test conditions (when 10 or 20 $\mu\text{g/g}$ of ammonium nitrogen were present with 3 percent of retorted shale) an essentially complete inhibition of nitrogen fixation was observed. Similar effects have been observed with 5 $\mu\text{g/g}$ of added ammonium ion nitrogen. The effects of added

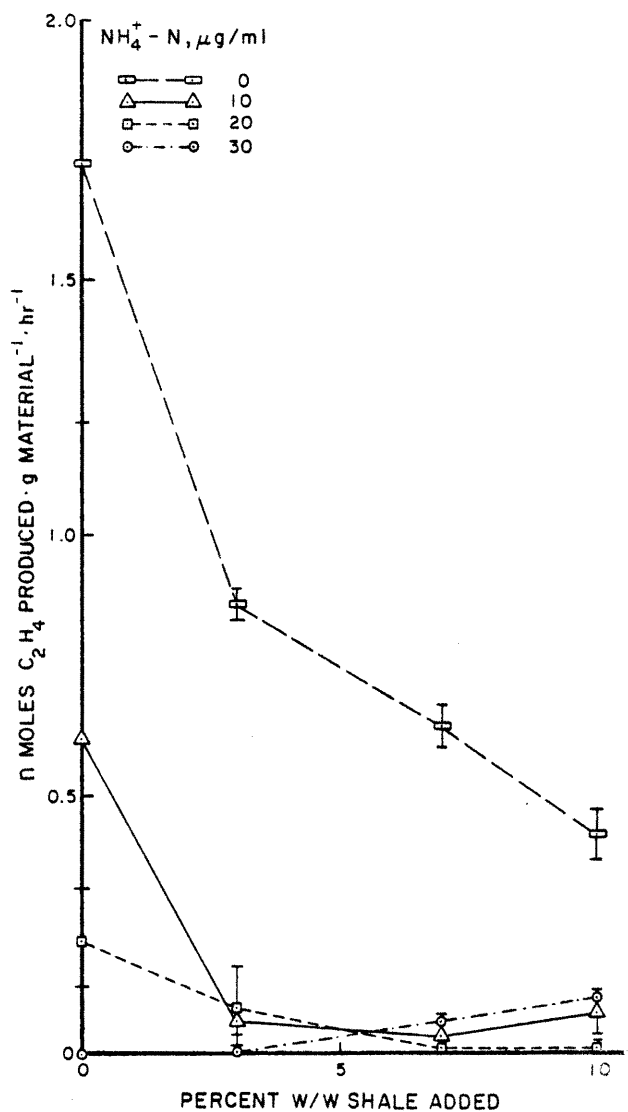


Figure 22. Effects of varied ammonium ion and retorted oil shale addition on the nitrogen fixation potential of surface soil from the Intensive Study Site. Means and standard deviations are shown.

nitrogen fertilizer on nitrogen fixation observed in the fertilized subplots of the Revegetation Technique Plot and the possible effects of retorted shale on nitrogen fixation observed on the Retorted Shale Succession Plot suggest that an interaction of these two factors may lead to more distinct inhibition of surface soil nitrogen fixation potential. It was of interest to note that the surface soil, with a nominal ammonium ion nitrogen level of 49 $\mu\text{g/g}$, did respond to nitrogen additions in the range of 10 to 20 $\mu\text{g/g}$. This suggests that the 2M KCl extractant used in the routine analysis procedure to estimate plant-available nitrogen may be measuring nitrogen which is not capable of influencing the nitrogen fixation capability of soil microorganisms.

Long-Term Fertility Plot Experiment

The Long-Term Fertility Plots were also analyzed for the first time during the 1979 sampling season. These plots and their various treatments are described by Sabey et al. in this report. Samples were taken on May 25, 1979. A single sample was taken from the 5-10 cm depth of each subplot. Treatments are replicated three times (triplicate subplots) for the samples from the Topsoil Over Retorted Shale and Subsoil Plots. Soil moisture, pH, and organic matter were analyzed in each sample together with dehydrogenase, nitrogen fixation (acetylene reduction), and phosphatase activities.

In general, the topsoil samples had lower pH values with an average of 8.54 in comparison with a pH value of 9.01 for the subsoil samples. The lowest pH values appear to have resulted from the sludge plus sawdust treatments; and, in most cases the treated plots had lower pH values than the controls. With increased nitrogen additions, at least in the Topsoil Over Retorted Shale Plot, there appears to be a trend to decrease pH values.

As would be expected major increases in soil organic matter were observed on plots with added sewage sludge and sewage sludge plus sawdust. The microbiological activity parameters which were monitored on this plot included dehydrogenase, phosphatase, and nitrogen fixation. In all cases general increases in activity were observed with the addition of sewage sludge and/or sawdust.

Retorted Shale Leachate Effects on the Nitrogen Fixation Activity of *Rhizobium*

Based on initial results which were obtained during the 1978 field season, additional effort was given to further evaluate the possible use of *Rhizobium* growing in pure culture as an indicator of potential effects of oil shale components on nitrogen fixation by seeded legumes.

Using a 1-10 retorted shale-water leachate and 20 ml of the *Rhizobium* culture per 70 ml test bottle, stimulation and inhibition of *Rhizobium* nitrogen fixation potential were observed with increased addition of the shale extract (Figure 23).

In comparison with the control, only when 3.0 ml of the 1-10 retorted shale leachate were added was a significant decrease in nitrogen fixation potential observed. With 1-7 leachates immediate inhibition of nitrogen fixation potential was observed without stimulation occurring as had resulted from the use of more dilute leachates.

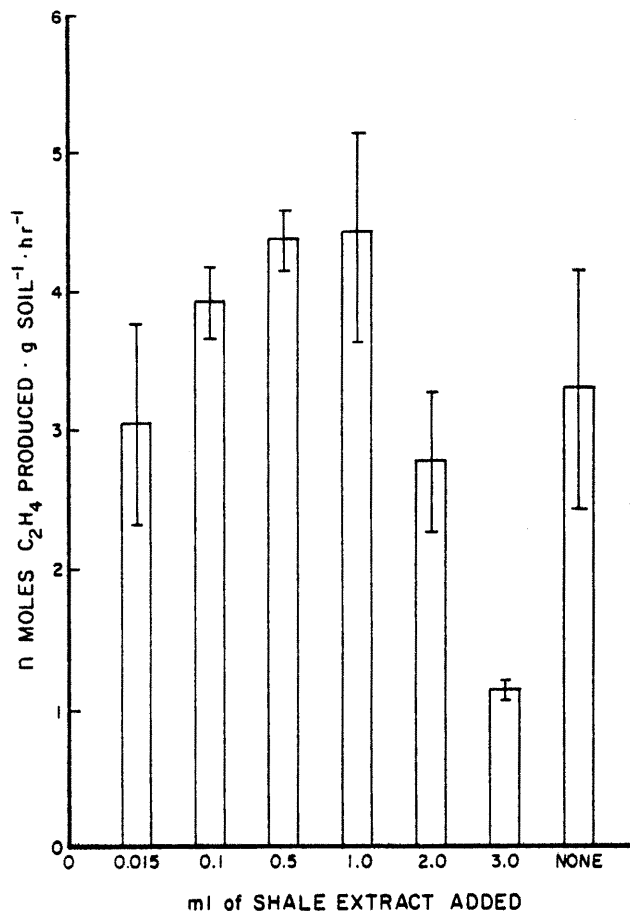


Figure 23. Effects of varied amounts of a 1-10 retorted oil shale leachate on the nitrogen fixation activity of pure cultures of *Rhizobium*. Means and standard deviations are shown.

Nitrogen Fixation Activity of Legume Nodules

Initial studies of nitrogen fixation activity of excised nodules from five of the legumes being evaluated at the Intensive Site were also completed using materials grown on topsoil and subsoil (Figure 24). The legumes which were tested included sweetvetch (*Hedysarum boreale*), crownvetch (*Coronilla varia*), milkvetch, sainfoin, and alfalfa. Based on the use of uniform numbers of excised nodules per analysis flask (other than with the milkvetch) higher nitrogen fixation activity was observed using plants which had been grown in topsoil, and the alfalfa showed markedly higher levels of activity in both topsoil and subsoil. These

should only be considered as relative potential activity values as the number of nodules per plant was not determined and field studies to confirm these potential rates of nitrogen fixation have not been completed. These results do indicate, however, that plants grown in topsoil will have nodules with higher relative rates of nitrogen fixation than those grown in subsoil materials. This may be related to changes in the overall vigor of the plants in these varied growth media.

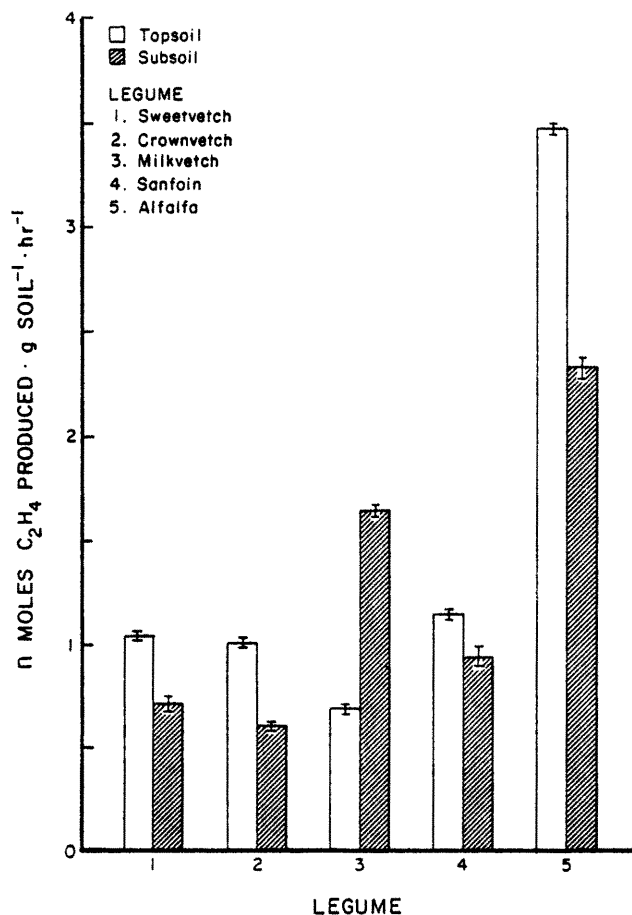


Figure 24. Nitrogen fixation potential of nodules from five legumes being grown in topsoil and subsoil at the Intensive Study Site. Forty nodules were used per sample analysis flask with a volume of 70 ml. Means and standard deviations are shown.

SUMMARY

After the second year of intensive analysis of the study area plots several general concepts appear to be emerging which may be of direct interest for the management of reclamation programs. The results from the Revegetation Technique Plot indicate that irrigation during the first two years of establishment can have long-lasting effects on belowground ecosystem development. As an additional

aspect of microbiological responses to management it appears that fertilizer use can lead to significant decreases in nitrogen fixation and phosphatase activity.

As noted in the 1978 report it appears that specific planting mixtures may be better suited for use under irrigated or non-irrigated conditions and that the use of introduced species may lead to higher dehydrogenase and phosphatase activities, independent of nitrogen and irrigation treatments.

The studies of topsoil storage have not indicated that there are significant changes in general microbial activities over a 1.5 year period. It appears that sampling will have to be much more intensive in the future as the bore effect variations have, in some cases, been greater than the depth effects. The only possible exceptions to this would be in relation to the mycorrhizal infection potential of soils which appears to have decreased in the surface soil layers. This information is discussed in greater detail in the mycorrhizal studies section which was carried out under the direction of Dr. F. Brent Reeves.

The Retorted Shale Succession Plot results were of extreme interest as they indicate that surface soil depth and a capillary barrier may have significant effects on the reestablishment of microbiological processes. Generally, for the parameters examined the plots with capillary barrier soils had activities more similar to those found for the controls while soils placed directly over retorted shale had lower levels of activity. This was most evident for the measurements of nitrogen fixation potential. Although no isolated effects of seeding mixture or fertilizer on microbiological processes were observed at this sampling, the presence of an interactive effect of these variables on the soil nitrogen fixation potential suggests that clearer effects may occur with further development of these plant communities, as was noted for the Revegetation Technique and Surface Disturbed Succession Plots where two years were required to find significant effects of fertilizer additions.

With the possibility that retorted shale plus fertilizer nitrogen may have specific interactive effects upon nitrogen fixation, fertilizer nitrogen use may have to be managed carefully especially if longer-term revegetation management is a major consideration in a particular reclamation program.

LITERATURE CITED

- Hersman, L. E., and D. A. Klein. 1979. Retorted oil shale effects on soil microbiological characteristics. *J. Environ. Qual.* 8:520-524.
- Peters, D. B. 1965. Water availability. Pages 279-285 in C. A. Black, ed. *Methods of soil analysis.* American Society of Agronomy, Inc., Madison, Wisconsin.
- Sherwood, J. E. 1979. The rhizosphere effect on antibiotic-resistant *Arthrobacter* and *Pseudomonas*. M.S. Thesis. Colorado State University, Fort Collins. 90 pp.

IMPORTANCE OF MYCORRHIZAL FUNGI IN REVEGETATING DISTURBED SOILS AND RETORTED SHALE

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OBJECTIVES

The overall goal of this subproject is to determine the changes in the population of mycorrhizal fungi in soils which have been disturbed or stored under conditions simulating those associated with oil shale development. The population changes of mycorrhizal fungi are correlated with other microbiological activities in the soil and with the relative success of establishment and succession of the aboveground vegetation at the Intensive Study Site. The specific objectives for the past year were as follows:

1. To determine the relationships of various fertilizer treatments to mycorrhizal formation in the experimental field plots
2. To test inoculated (mycorrhizal) versus non-mycorrhizal transplants under field conditions
3. To continue to monitor rather drastically disturbed plots for changes in the population of mycorrhizal fungi over time
4. To refine our bioassay methods for greater reproducibility
5. To continue to monitor the Topsoil Storage Plot for changes in the mycorrhizal inoculum potential (MIP)
6. To monitor seasonal variation in the population of mycorrhizal fungi in undisturbed soils at the Intensive Study Site
7. To determine the effect on the population of mycorrhizal fungi of mixing retorted shale with topsoil
8. To determine the species relationships among mycorrhizal fungi and host plants.

PROGRESS TO DATE

Our original hypothesis regarding the role of endomycorrhizae in revegetation practices (Reeves

et al. 1979) in semiarid environments has been corroborated and extended in Wyoming (Miller 1979) and briefly discussed with reference to plant competition (Moore 1979). Our laboratory bioassay (Moorman and Reeves 1979) which measures the mycorrhizal inoculum potential (MIP) of the soil is now used by several investigators (Rives et al. 1980) and is being refined in our laboratory (Bishop 1979) so that it might better evaluate subtle changes in the population of mycorrhizal fungi.

Of our original eight objectives seven are complete or data are being analyzed and one is not yet begun. Additionally, two other important results have been obtained, viz., certain strains of the same species of mycorrhizal fungi are more effective in stimulating growth under controlled conditions in the lab and the vegetation on topsoil stored under greenhouse conditions significantly influences the MIP of the soil after 20 months of storage

Effect of Fertilizer on MIP

Previous research by several investigators indicates that vesicular-arbuscular (VA) mycorrhizal infections are more common in low fertility soils and that additions of P, N, or complete fertilizers reduce the amount of infection (Gerdemann 1975). Indeed, high P additions have been reported to render plants resistant to infection (Mosse 1973). We suspected a similar case might be true on the experimental plots at the Intensive Study Site; if such were the case, then the mycorrhizal microbial component would be substantially reduced or eliminated by additions of fertilizer.

Analyses of data for 1978 and 1979 from the Retorted Shale Succession Plot with 112 kg N/ha + 56 kg P/ha vs 0 kg N/ha + 0 kg P/ha indicates that there are no significant differences in the MIP of profile Treatments 2-6 (soil placed over processed shale--see Redente et al. in this report). Profile 1 (retorted shale without any soil cover) had a 0 MIP value for both fertilizer treatments. This was expected since the processed shale is effectively sterilized. Table 23 gives the MIP values for the retorted shale-soil profiles.

Table 23. Retorted shale panels showing MIP values for June 1979.

Fertilizer Treatment	Soil-Shale Profile Treatments						Mean
	1	2	3	4	5	6	
112 kg N/ha + 56 kg P/ha	0.0 ^a	35.0 ^b	30.7 ^b	29.7 ^b	29.7 ^b	20.0 ^b	29.0
0 kg N/ha + 0 kg P/ha	0.0 ^a	43.7 ^b	26.0 ^b	24.0 ^b	31.7 ^b	31.7 ^b	32.0

^{a,b,c}MIP values in each column with the same letter are not statistically different.

We have not completed the analyses of the 1979 data on the Revegetation Technique Plot nor the Surface Disturbed Succession Plot, but preliminary results support our 1978 data (1978-1979 Progress Report) which indicated that there were no significant changes in the MIP on the fertilized vs unfertilized subplots.

At this time it is concluded that up to 112 kg N/ha and 56 kg P/ha does not significantly alter the mycorrhizal component of the soil, and thus such fertilizer treatments are not detrimental to these organisms. The current study is especially interested in investigating the effects of large additions of fertilizer to soils and their effects on MIP in semiarid environments. This research will begin this year using Drs. Berg and Sabey's Long-Term Fertility Plots at the Intensive Study Site.

Field Test M+ (inoculated) and M- (non-inoculated) Plants

This objective was not accomplished during this year due to a lack of pot culture of the mycorrhizal fungi and to a failure to propagate the experimental plants vegetatively.

Monitor the Annual Disturbance Plot

These plots provided information on the rate of natural recovery of disturbed big sagebrush communities. Analysis of data has begun to correlate the MIP of the four disturbance treatments with the natural succession of the vegetation (see Redente et al. in this report for test plot design).

As indicated in previous research the more severe the disturbance the less the MIP of the soil. This year we have plotted the MIP as a function of the grass density on the four treatments (Figure 25). The relationship of the aboveground vegetation to the belowground MIP exhibits an excellent semilog correlation (0.833). Treatment 1 on the 1976 Annual Disturbance Plot was the least disturbed that year and shows, as would be expected, the greatest recovery. The recovery is reflected in both the high MIP and the highest grass density when compared to all the other plots. Next in recovery is Treatment 2 (1976) then Treatment 1 (1977). This sequence reflects the increasing severity of disturbance. The 1977 Treatment 2 plots

have not recovered as yet. This lack of recovery is also found in the MIP of the soil. Treatments 3 and 4 on both the 1976 and 1977 plots were so severe that little difference between the treatments or the years is apparent. It was expected that the 1977 Treatment 2 plots would recover more quickly, but the MIP may have been reduced to the point where recovery may require substantially more time. There is a potentially interesting inverse relationship between the forb cover and the MIP on these plots. The forbs are mostly the non-mycorrhizal Russian thistle and, thus, do not require mycorrhizal fungi nor do they contribute to increasing the population of mycorrhizal fungi. The low MIP values on Treatment 4 (1976 and 1977) reflect this same relationship with vegetation cover.

The data given in Table 24 is for both April and June (1979) and illustrates the seasonal variation in MIP on these disturbed soils. In Objective 6 we have shown that the MIP values change with the seasons in undisturbed soils at the Intensive Study Site. Table 24 gives the data for the MIPs for April and June on these plots.

Refine the MIP Bioassay

As originally published (Moorman and Reeves 1979) the technique was to measure the MIP of the

Table 24. MIP values for 1979 for the Annual Disturbance Plots constructed during 1976 and 1977.

	MIP Value			Grass Density
	April	June	\bar{X}	
<u>1976</u>				
Treatment 1	38.5	55.0	46.8	20.65
2	41.0	44.5	42.8	7.35
3	17.8	13.0	15.4	0.60
4	11.0	11.5	11.3	0.30
<u>1977</u>				
Treatment 1	29.0	49.5	39.3	5.15
2	17.3	21.0	19.1	0.55
3	10.8	16.5	13.6	0.45
4	7.8	10.0	8.9	0.25

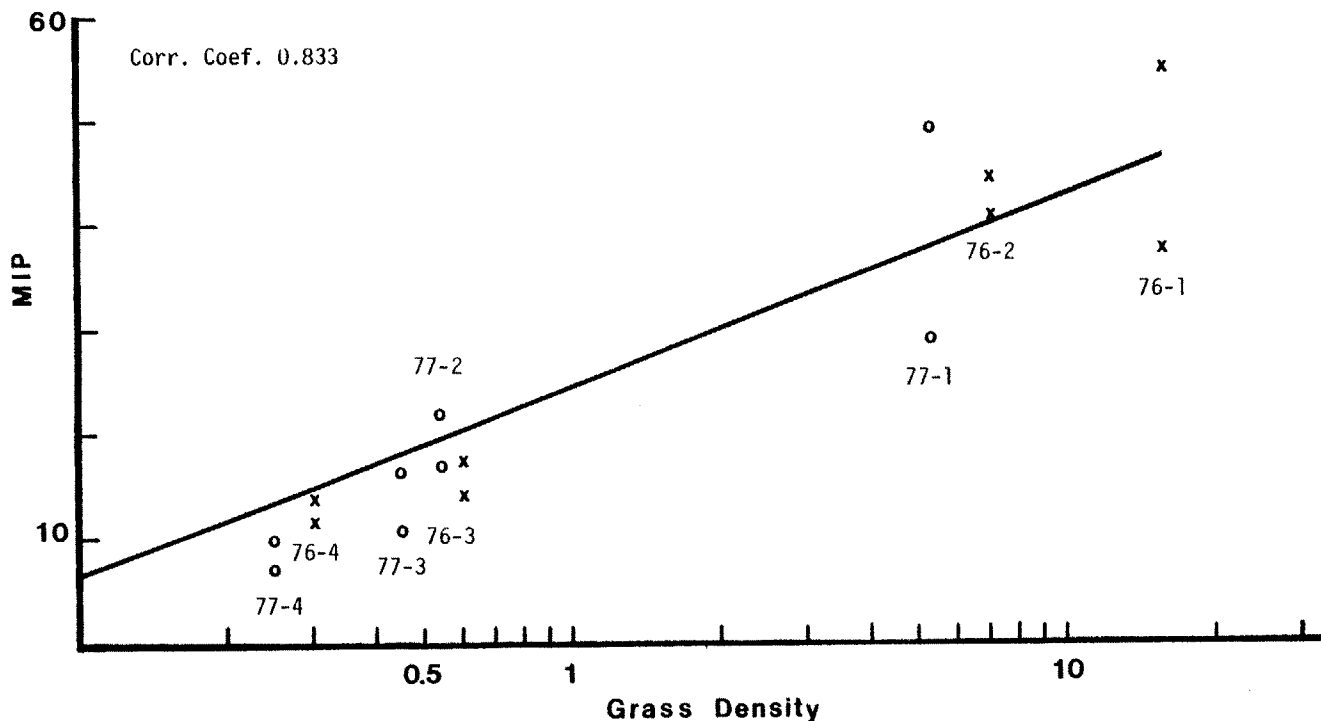


Figure 25. Correlation of grass density with mycorrhizal inoculum potential (MIP) on the Annual Disturbance Plot, 1976 and 1977. Number 76-1 represents 1976 plots, Treatment 1 (least severe); Number 77-4 represents 1977 plots, Treatment 4 (most severe).

soil by growing corn under greenhouse conditions for 30 days. These conditions vary greatly with the season (cool and low light intensity in winter, hot and high light intensity in summer). Previous research (Furlan and Fortin 1977) has indicated that light may have a significant effect on mycorrhiza formation in onion (*Allium cepa*). It is suspected that the results of the bioassay used herein may vary with light intensity. Further, it is desirable to reduce the growth time required for making the analyses. Results of the initial experiments are given in Figure 26 and published in an abstract by Bishop (1979). Figure 26 shows that a MIP plateau is reached at 14 days at 800 and 1600 foot candles (30°C, 14 h day; 21°C, 10 h night); however, at 400 fc there is a progressive increase in MIP values up to 24 days. These results emphasize the necessity of conducting the MIP analyses in growth chambers under standardized conditions.

Although the information on the effects of temperature variation on MIP values are not available, it is expected that similar changes in values would be found. Studies are now utilizing simple growth chambers for determining the MIP values of the different soils at the Intensive Study Site.

Monitor Topsoil Storage Plot

The topsoil initially present was rather heterogeneous (MIP values 10-35). The 1979 data exhibits this same heterogeneity (MIP values 7-30), but for each sample the value is down from 1978. Because of this heterogeneity there are no signifi-

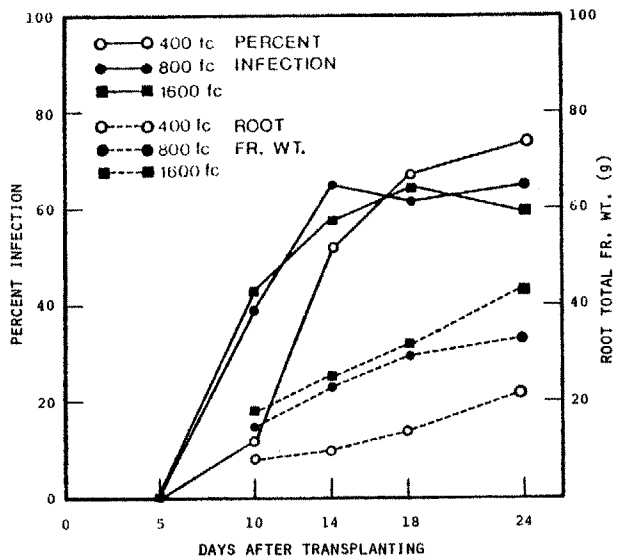


Figure 26. Percent infection, root fresh weight, and light intensity relationships in the bioassay. After 14 days percent infection remains level at 800 and 1600 foot candles.

cant differences between depths; but, when the mean values of each depth are plotted (Figure 27), certain expected trends are obvious. In the upper layers of the Topsoil Storage Plot (15, 30, and 60 cm) the MIP shows a decrease. We tentatively

Table 25. Data for MIP values from the Topsoil Storage Plot taken during 1978 and 1979.

Sample	MIP Values											
	15 cm Depth		30 cm Depth		60 cm Depth		90 cm Depth		120 cm Depth		150 cm Depth	
	1978	1979	1978	1979	1978	1979	1978	1979	1978	1979	1978	1979
Bore 1	16	14	33	27	25	21	26	24	26	22	33	27
Bore 2	10	13	35	22	18	17	22	14	21	12	25	23
Bore 3	30	9	18	7	30	11	15	8	13	15	22	30
Bore 4	19	11	14	7	13	8	12	10	10	13	20	17
Mean	19	12	25	16	22	14	19	14	18	16	25	24

interpret this decrease to be a reflection of the effects of harsh external environmental conditions reducing the numbers of viable mycorrhizal propagules (Table 25). At the greater depths (120 and 150 cm) little decrease in MIP is found. These greater depths are well insulated from external conditions.

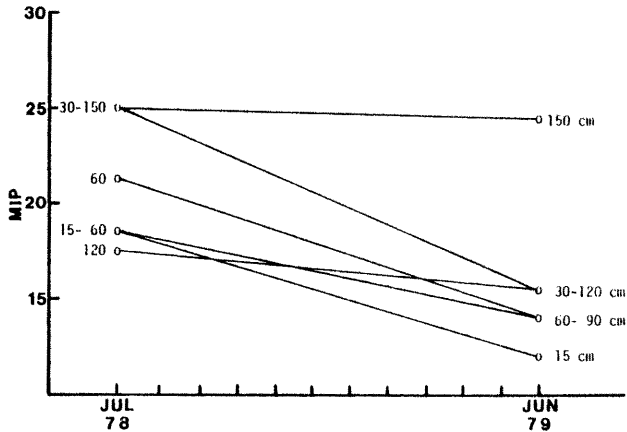


Figure 27. Decrease in mycorrhizal inoculum potential (MIP) in Topsoil Storage Plot with time. MIP decreases in the upper depths (15, 30, 60, and 90 cm) but not at the lower depths (120 and 150 cm).

Monitor Seasonal Variation in MIP of Undisturbed Soils

The experimental plots at the Intensive Study Site were established at different times during the year. In order to determine if significant MIP variation between plots might be a function of seasonal variation in the population of mycorrhizal fungi rather than soil treatment, a program to measure the MIP during different seasons was undertaken. The preliminary results of this work are given in Figure 28 and Table 26. The trends so far appear to indicate that the MIP at most sites increases in the early spring but begins to fall somewhat later in the spring. These results are consistent with the laboratory observations which

indicate that the numbers of spores of mycorrhizal fungi which will germinate usually is greater after a storage period of several months in the cold than without a cold treatment. If data continues to record these trends, indications are that the optimal time for disturbance in order to maintain a high MIP in the soil is during the winter months. Such disturbances would least affect the population of mycorrhizal fungi since at this time the fungi are in a dormant state.

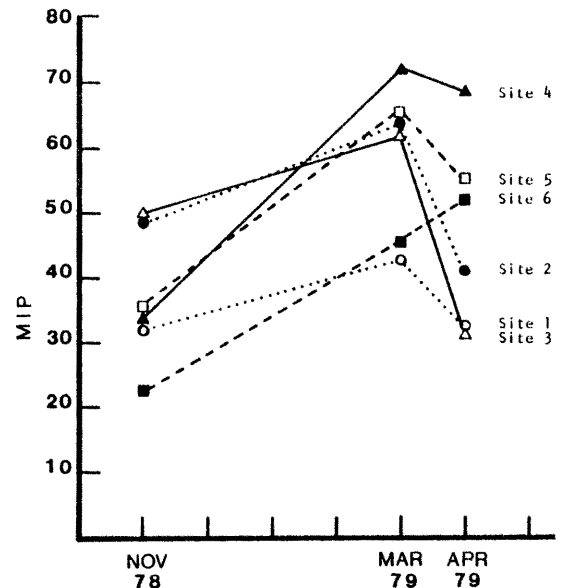


Figure 28. Seasonal changes in mycorrhizal inoculum potential (MIP) of undisturbed soil. Site 1, adjacent to sagebrush; Site 2, between sagebrush plants; Sites 3 and 4, under sagebrush; Sites 5 and 6, bare ground.

The great range of MIP of the soil beneath, between, and at a distance from big sagebrush plants does not indicate that any microhabitat offers a particularly favorable site for maintaining MIP of the soil. Indeed, the apparently bare ground had MIP values nearly as high as beneath the sagebrush plants; and, in one case, Site 3, the MIP beneath the sagebrush was less than the bare ground value.

Table 26. Seasonal variation in MIP of undisturbed soils in the Piceance Basin.

	MIP Values		
	Nov 1978	Mar 1979	Apr 1979
Site 1 Adjacent to sagebrush plants	31	43	33
Site 2 Between sagebrush plants	49	64	41
Site 3 Under sagebrush plants	50	62	32
Site 4 Under sagebrush plants	34	71	69
Site 5 Bare ground	35	63	55
Site 6 Bare ground	22	45	53
Mean	37	58	47

Determine the Effects of Mixing Retorted
Oil Shale with Topsoil

The results of this study are in press (Schwab and Reeves 1980). Our experiments were designed to determine the amount of processed oil shale which when mixed with topsoil would affect the MIP of the topsoil. Two interplaying factors had to be considered: (1) the dilution effect and (2) the direct (toxic) effect of retorted shale. In these experiments sterile sand or processed shale was added in equal amounts (0, 10, 25, 50, 75 percent) to topsoil. Sterile sand in amounts equal to the addition of retorted shale added to topsoil served as controls for the dilution effect. The results are given in Figure 29. Significant differences between sand and topsoil and retorted shale and topsoil do not appear until the concentration of retorted shale reaches 50 percent. Under normal conditions processed oil shale mixed with topsoil should not pose a problem to the population of mycorrhizal fungi until it approaches the 1:1 ratio.

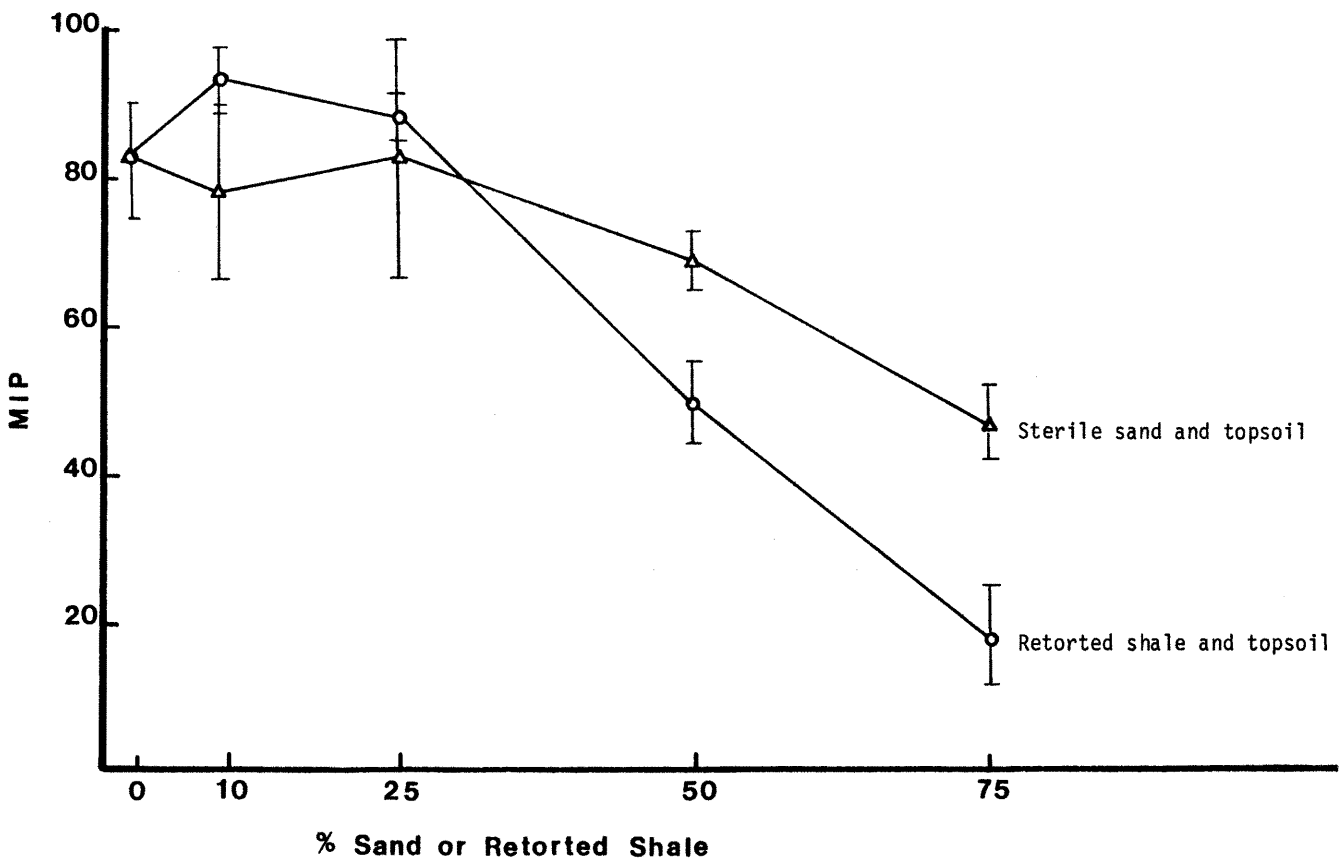


Figure 29. Effects of mycorrhizal inoculum potential (MIP) of topsoil by the addition of processed oil shale or sterile sand. Processed shale decreases the MIP more than sand at 50 and 75 percent shale added to topsoil.

Determine Species Relationships Among
Mycorrhizal Fungi and Host Plants

The results of this research are represented in papers presented or scheduled to be presented this year (Reeves et al. 1979, Schwab 1979, Schmidt et al. 1980, Schwab et al. 1980). Our experiments have been designed to elucidate the normal mycorrhizal relationships of native species in undisturbed soils and in species on disturbed soils. In surveys we have found that in undisturbed rangeland areas 90 percent or more of the species are mycorrhizal, but in severely disturbed areas most of the species are non-mycorrhizal (Reeves et al. 1979, Schwab 1980). In Objective 3 it was pointed out that pioneer species on severely disturbed soils are usually non-mycorrhizal. Schwab (1979) found that the time required for newly seeded species to reach the maximum level of infection varied with the species and that non-mycorrhizal species and species which became only moderately mycorrhizal were the best invaders on disturbed soils. This suggests there is a continuum for non-mycorrhizal to heavily mycorrhizal species and pioneers are non-mycorrhizal or only weakly mycorrhizal.

In addition to monitoring the Topsoil Storage Plot we have data on topsoil storage which complements those reported in Objective 5 (Schmidt 1980). Topsoil collected in May 1978 was placed in wooden flats (30x30x90 cm), planted with different species of native and agronomic plants, and left in the greenhouse. The MIP of the soil after 20 months is given in Table 27. This experiment represents topsoil stored under greenhouse conditions and the effects different plant species have on maintaining the MIP of the soil. Table 27 also indicates that typically non-mycorrhizal species (fourwing saltbush and winterfat) lower the MIP of the soil when compared to mycorrhizal species or corn (*Zea mays*) or the control soil (no plants are grown in this

Table 27. Average MIP values of topsoil storage after 20 months.

Host Plant	MIP Value
Corn <i>Zea mays</i>	87
Globemallow <i>Sphaeralcea coccinea</i>	81
Western wheatgrass <i>Agropyron smithii</i>	67
Indian ricegrass <i>Oryzopsis hymenoides</i>	47
Big rabbitbrush <i>Chrysothamnus nauseosus</i>	44
Big sagebrush <i>Artemisia tridentata</i>	39
Fourwing saltbush ¹ <i>Atriplex canescens</i>	15
Winterfat ¹ <i>Ceratoides lanata</i>	12
Control	28

¹Non-mycorrhizal species.

soil). This suggests that the non-mycorrhizal species may in fact decrease the MIP by acting as allelopathic species towards normally mycorrhizal species. This is tentative data and must be repeated before any final conclusions can be made regarding the interrelationships of MIP and plant species.

In preliminary experiments designed to test the effects of various species of mycorrhizal fungi on the growth of plants, pot cultures of *Glomus* spp. isolated from Colorado (Piceance Basin) were chosen for comparison with other species. The results are shown in Figure 30. The Colorado strain of mycorrhizal fungi which we have in culture is not as effective as other species of mycorrhizal fungi, especially *Glomus mosseae*, when used to stimulate growth of tomatoes. Current problems involve quantifying the inoculum so that the results are more nearly comparable. Therefore, the data should be considered only preliminary. These results will be extended to native species (grasses) to see if the stimulation in tomatoes can be duplicated in native grass species. It may well be the case that one or more elite strains of mycorrhizal fungi not present in the Piceance Basin can be chosen to enhance growth in native species.

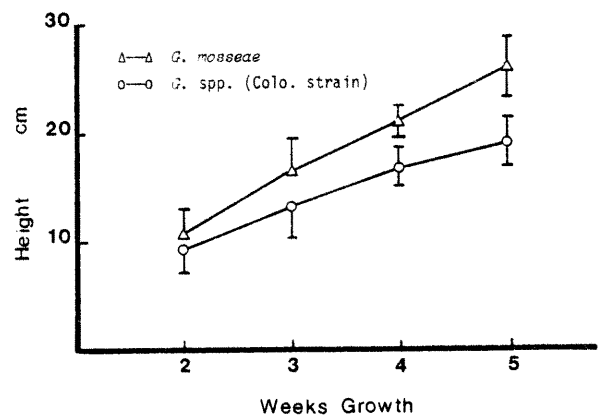
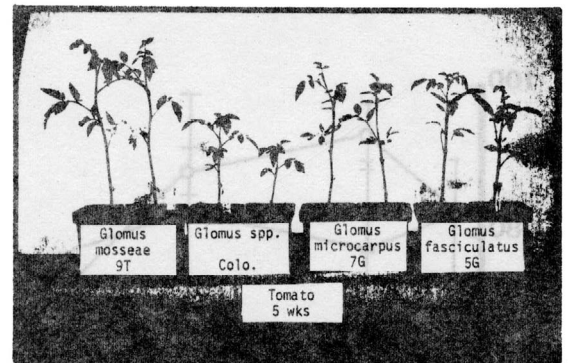


Figure 30. Comparison of growth stimulation in tomatoes by two different strains of mycorrhizal fungi. *Glomus mosseae* (9T) enhances growth when compared to the native Colorado strain after five weeks. When grown with two other strains (7G and 5G) tomatoes show stimulation similar to strain 9T.

CONCLUSIONS

Our research on the relationship of populations of mycorrhizal fungi to the success of revegetation techniques has demonstrated that in undisturbed soils most plants are mycorrhizal, that disturbance of the topsoil substantially reduces the population of mycorrhizal fungi, and that on heavily disturbed soils most of the invading plants are non-mycorrhizal species. Our objectives have been to determine which factors of disturbance substantially alter (increase or decrease) the viable population of mycorrhizal fungi. To date research has shown that: (1) the addition of fertilizers (N and P) to soil do not affect the fungi and is a useful technique if plant growth is stimulated; (2) if heavily disturbed areas are not fertilized and seeded the population of mycorrhizal fungi decreases and the plant cover develops very slowly; (3) our methods by which we measure the population of mycorrhizal fungi are affected by light intensity; (4) after 14 months in the field stored topsoil exhibits a decrease in the population of mycorrhizal fungi at depths up to 90 cm but not at depths of 120 nor 150 cm; (5) there are great seasonal variations (in undisturbed soil) in the population of mycorrhizal fungi; (6) when processed shale is mixed with topsoil no detectable decrease in the population of mycorrhizal fungi is found until the shale exceeds 50 percent of the volume of the soil; and (7) certain species of mycorrhizal fungi are more effective in stimulating growth than are other species.

LITERATURE CITED

- Bishop, C. L. 1979. Effects of light intensity on vesicular-arbuscular mycorrhizal infection in *Zea mays*. J. Colo.-Wyo. Acad. Sci. 11:28. (Abstract)
- Furlan, A., and A. Fortin. 1977. Effects of light intensity on the formation of VA endomycorrhizas in *Allium cepa* by *Gigaspora calospora*. New Phytol. 79:335-340.
- Gerdemann, J. W. 1975. Vesicular-arbuscular mycorrhizae. Pages 575-591 in J. G. Torrey and D. T. Clarkson, eds. The development and function of roots. Academic Press, New York.
- Miller, R. M. 1979. Some occurrences of vesicular-arbuscular mycorrhiza in natural and disturbed ecosystems of the Red Desert. Can. J. Bot. 57:619-623.
- Moore, P. D. 1980. Mycorrhizal associations. Nature 282:780.
- Moorman, T., and F. B. Reeves. 1979. The role of endomycorrhizae in revegetation practices in the semi-arid west. II. A bioassay to determine the effect of land disturbance on endomycorrhizal populations. Am. J. Bot. 66:14-18.
- Mosse, B. 1973. Plant growth responses to vesicular-arbuscular mycorrhiza. IV. In soil given additional phosphate. New Phytol. 72:127-136.
- Reeves, F. B., D. Wagner, T. Moorman, and J. Kiel. 1979. The role of endomycorrhizae in revegetation practices in the semi-arid west. I. A comparison of incidence of mycorrhizae in severely disturbed vs. natural environments. Am. J. Bot. 66:6-13.
- Rives, C. S., M. I. Bajwa, A. E. Liberta, and R. M. Miller. 1980. Effects of topsoil storage during surface mining on the viability of VA mycorrhiza. Soil Sci. [In press.]
- Schwab, S., F. B. Reeves, R. L. Dix, R. T. Ward, and W. Slauson. 1980. Relationships among aboveground vegetation types, soil chemistry, and populations of VA mycorrhizal fungi in the Great Basin Deserts of Colorado. To be presented at AAAS Meeting, 9-12 April, Las Vegas.
- Schwab, S. 1979. Rate of formation of VA mycorrhizae in seedlings of seven species native to the mid-elevation sage community of northwestern Colorado. J. Colo.-Wyo. Acad. Sci. 11:28. (Abstract)
- Schwab, S., and F. B. Reeves. 1980. The effect of retorted oil shale on VA mycorrhizae formation in soil from the Piceance Basin of Northwestern Colorado. To be published in the Oil Shale Symposium Volume, Denver, Colorado.

ECOGENETIC VARIABILITY IN NATURAL POPULATIONS AND THE STRUCTURE OF NATURAL COMMUNITIES AS RELATED TO THE REESTABLISHMENT OF VEGETATION ON DISTURBED ARID SHRUBLANDS

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OBJECTIVES

This study has two parts: (1) the population or ecotype phase--which is to evaluate the natural variation within species (especially shrubs) native to the Piceance Basin and to provide recommendations regarding source materials which can be expected to give long-term, natural success on particular disturbed sites and (2) the plant community or vegetation phase--which is to provide basic information on the present vegetation as it varies over the landscape of the Piceance Basin and to structure this information so that it is relevant to other reclamation research in this project, to the design of revegetation programs, and to the measurement of success of plant community reestablishment.

The second part was given primary emphasis in the first years of the study when the experimental garden for the first phase was being established. The plant community studies have now reached a degree of completion permitting the final vegetation model to be given in this report. A "User's Guide" is presented in outline form. The Guide will provide simplified use of the vegetation/environment model for reclamation design.

The first part, dealing with inherent variability in ecological adaptations among populations of individual species, has data from the ecotype garden sufficient to show initial patterns of response for a few selected species.

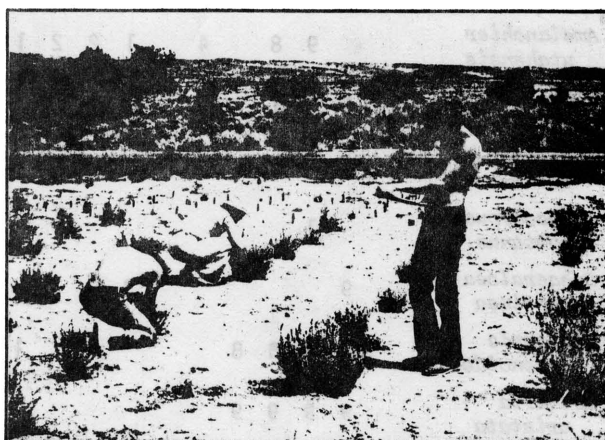
PROGRESS TO DATE

Phase One--Ecotype Studies

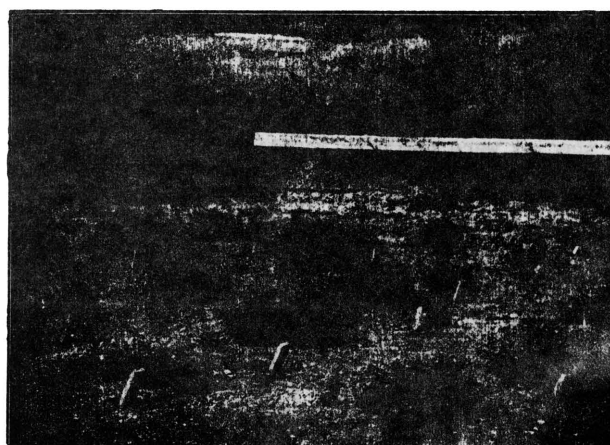
Measurements

Representatives of nine species from 102 populations completed their growing season in the ecotype garden at the Intensive Study Site (Figure 31). The measurements taken are given in Table 28. The number of populations and individuals for each species was reported in an earlier progress report (1978-1979). The measurements of the 1979 field

season along with some from 1978 gave the first substantial evidence for ecotypic variation in this study. The data for winterfat have been analyzed and the results are reported herein.



Shrub section of the garden



Forb, *Sphaeralcea coccinea*, section of the garden

Figure 31. Ecotype garden at the Intensive Study Site.

Table 28. Characteristics measured for each individual of each species in the ecotype garden at the Intensive Study Site during the 1979 growing season. Indicated are the number of times measurements were made during the season.

Species	Characteristic										
	Phenology	Plant Height	Vegetative Stem Length	Reproductive Stem Length	Stem Diameter	Nodes/Stem	Leaf Size	Leaf/Stem Color	Life Form	Dormancy Break	Dormancy Onset
<i>Symphoricarpos oreophilus</i>	9	9	3	2	9	1	1	1			
<i>Ceratooides lanata</i>	8	4	4		4						
<i>Amelanchier utahensis</i>	9	8	4		1	2	2	1	1		
<i>Atriplex canescens</i>	5	1	1								
<i>Purshia tridentata</i>	9				2			1	1		
<i>Cercocarpus montanus</i>		1									1
<i>Sphaeralcea coccinea</i>	9					3	1	1			1
<i>Oryzopsis hymenoides</i>	8	8	8	8					1		
<i>Koeleria cristata</i>	9	9	9	9					1		

Satellite Garden

Populations of three of the species have been established in a satellite garden at a higher elevation. Individuals of each population of a species were randomly positioned, planted, and hand watered in late August 1979. The numbers of populations for these species are given in Table 29.

The garden (at 2,440 m) is located north of the C-b Federal Oil Shale Lease between Steward and Scandard Gulches. It is in a fenced area, used previously for a species adaptability study, and has known environmental parameters. It receives approximately 15 cm more precipitation per year than the Intensive Site and has a shorter growing season. The purpose of this garden is to compare population responses in an environment different from the main garden on the Intensive Site and, more importantly, to test predictions of the relative response and success of different populations based on information gathered at the main garden.

Table 29. Species and populations in the satellite garden.

Species	Number of Populations	Number of Plants/Population
<i>Amelanchier utahensis</i>	9	6
<i>Symphoricarpos oreophilus</i>	5	6
<i>Ceratooides lanata</i>	2+2 ¹	6

¹Yet to be collected

Analysis

One of the most important aspects of ecotypic differentiation as it relates to reclamation has to do with differences in life cycle. Successful reclamation materials must not only be able to live for a period after initial establishment but must also be able to perpetuate themselves. Most populations of winterfat reached the flowering and fruiting stages in the Intensive Site garden, but at different times; this permits an evaluation of the genetically-based differences in life history characteristics and their relationships to probable reproductive success or failure on different reclamation sites.

Seven populations of winterfat growing at the Intensive Study Site have been measured for vegetative and reproductive growth (Table 28). Six populations were collected from various environments in the Piceance Basin, the seventh from the Huerfano River Valley in southern Colorado. (Site descriptions were presented in the progress report for 1977-1978.) Each plant was scored on a scale of one to four during the growing season for phenological advance from "vegetative only" through possession of "flower buds" and "flowering" to "seed set". These responses over the season are presented in Figure 32.

The responses fall into three sets. Population 1 (as the first set) contains small plants which flower and fruit earliest. Population 7 has delayed flowering and has the least reproductive success in the ecotype garden. The five other populations are intermediate in phenological timing. The differences among populations are statistically significant ($p < .01$) for each measurement date. If the phenological stages are summed for each population over the growing season, the relative flowering and fruiting success of these populations can be summarized (Figure 33). SNK multiple range tests identified the groups which are significantly different.

The implication of these results for reclamation planning are obvious. Population 7, and others from similar or even longer-season localities, could not be recommended for sites like the Intensive Study Site nor those with a shorter

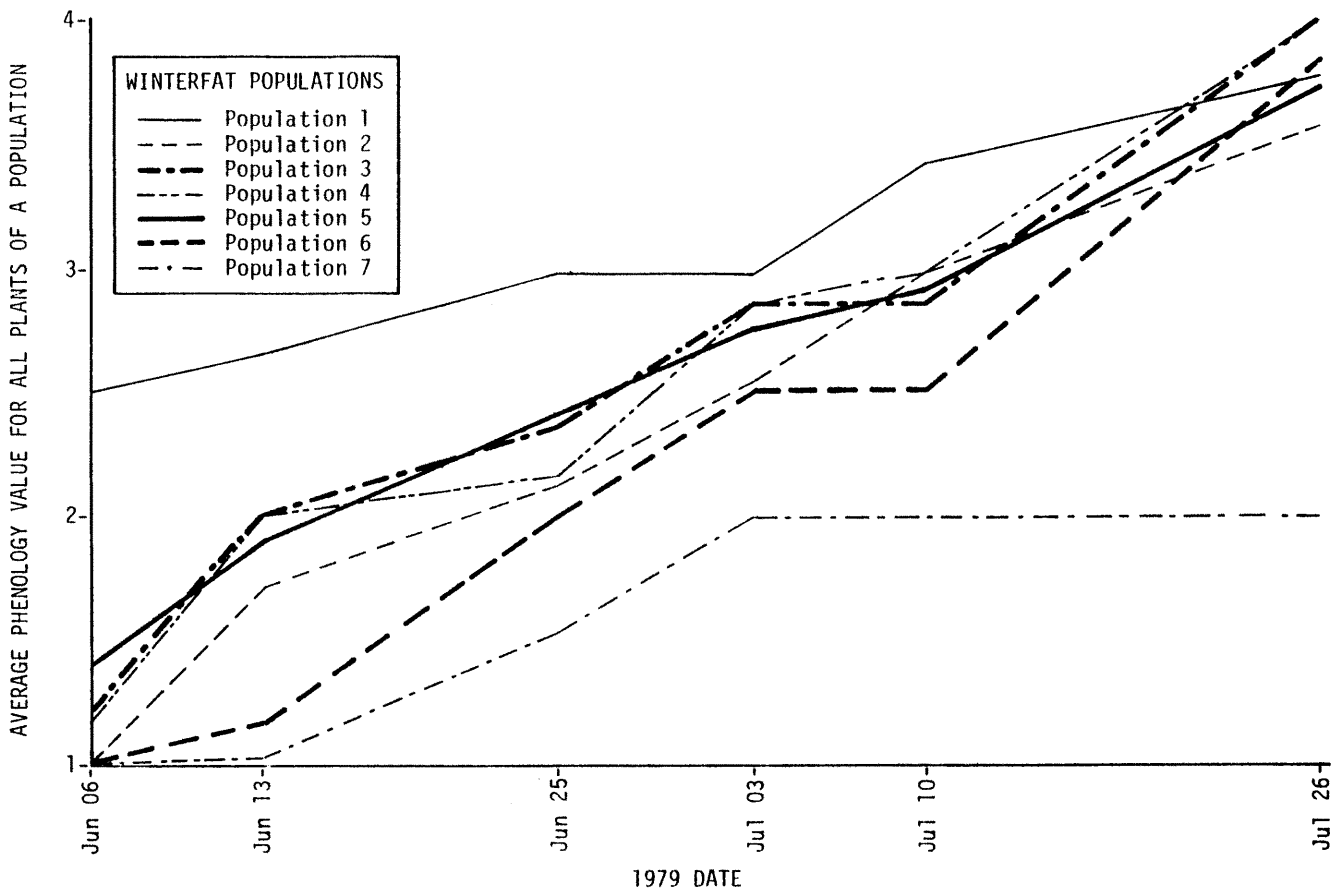


Figure 32. Average phenological development of seven winterfat (*Ceratoides lanata*) populations. Plants were scored 1--for vegetative growth only, 2--for flower buds present, 3--for anthesis, and 4--for fruits present. Separate ANOVA calculated for each date showed significant differences.

growing season. Although it might survive initial establishment and grow for a period, it would be very unlikely to reproduce itself at a level sufficient to maintain itself.

Additional biological justification for this conclusion is given in Figure 34. The vegetative growth of the stems producing flowers and fruits shows that the early-flowering populations develop less vegetative growth prior to flowering. Population 7 (with different adaptations) does not deploy resources to reproduction until substantially more vegetative growth has occurred. These differences, again, are genetically based and are not merely induced by different environments.

The other implications for reclamation are apparent. Population 1 from a high elevation in the Piceance Basin, may not be the best for reclamation in areas at lower elevations even though it sets seed. The reduced allocation to vegetative growth is apt to make this population less competitive than those populations which invest more in vegetative growth yet reproduce satisfactorily.

Other data, including relative amount of seed dispersal and onset and breaking of dormancy, show a similar pattern of statistically significant differences among populations of winterfat.

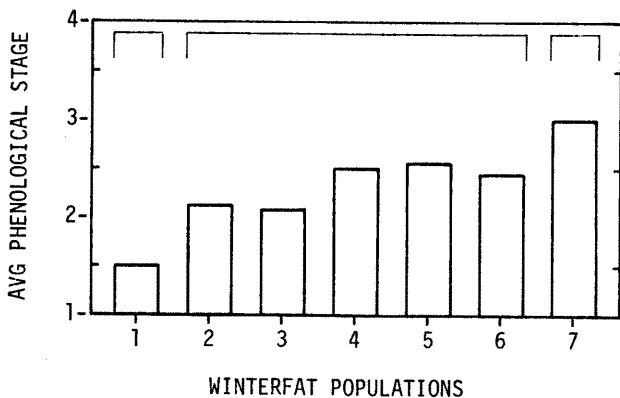


Figure 33. Total average phenological advance of seven winterfat (*Ceratoides lanata*) populations measured in 1979. Brackets show grouping by SNK multiple range test. Phenological stages include: 1--vegetation only, 2--flowering buds present, 3--anthesis, and 4--fruits present.

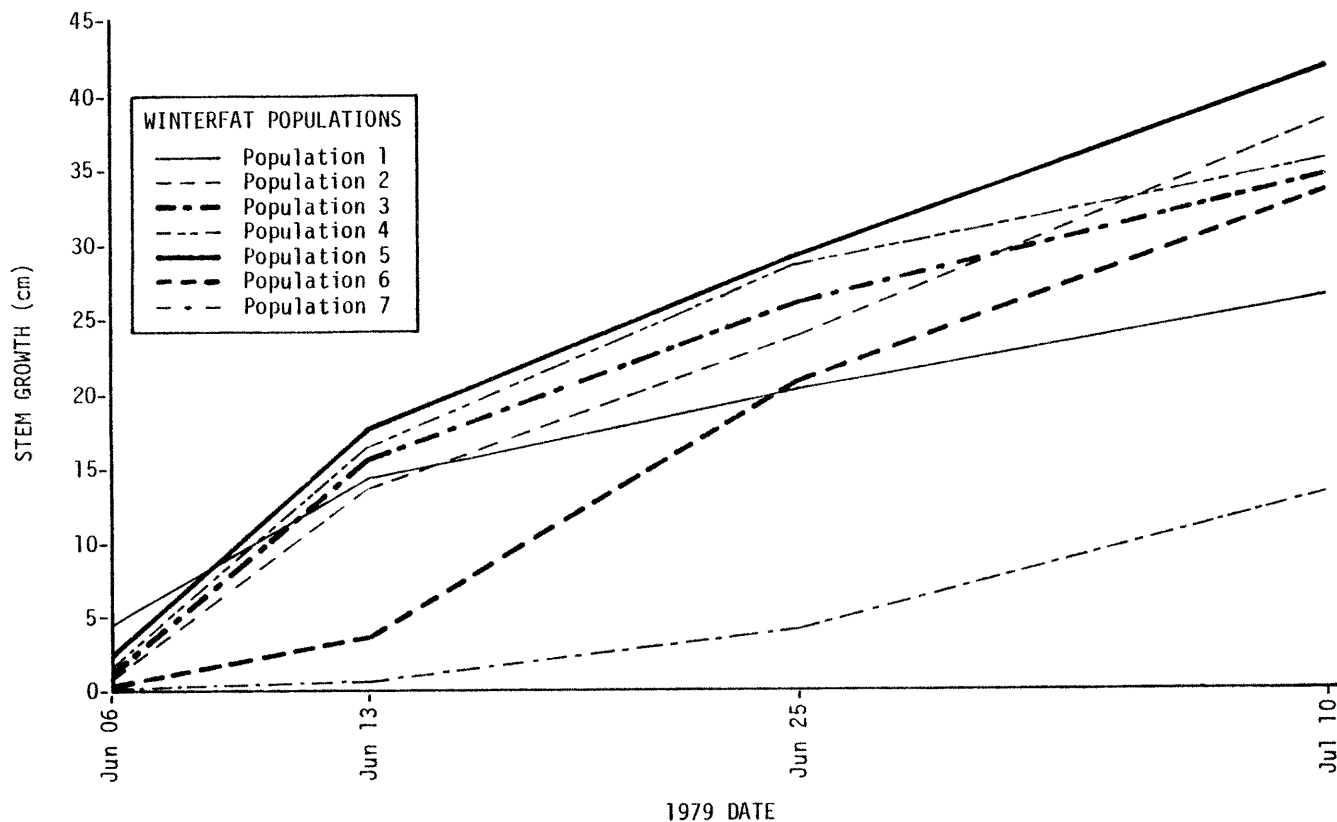


Figure 34. Progress of vegetative stem growth of seven populations of winterfat (*Ceratooides lanata*). ANOVA shows significant ($p < .005$) differences for size at each date and for total growth.

It is obviously important to understand the relationship of the different responses in the garden to the differences in the original environments of these populations. The length of the growing season has been found to relate closely to response differences. The early flowering population (Population 1) shows adaptation to short seasons, while the late flowering population (Population 7) evidently needs a considerably longer season to complete development (Table 30). The statistical correlation (r) of phenology with length of growing season is -0.94 and is highly significant ($p < .001$). The proportion of variance in phenology accounted for by growing season length is 0.88 or 88 percent. The relation of growing season to the relative amount of dormancy on October 14, 1979, is similar ($r = 0.97$). The corresponding regressions are given in Table 31. Other environmental factors may also be important; the correlation (r) of dormancy with annual precipitation is 0.81 .

Table 30. Site characterization for the seven populations of winterfat (*Ceratooides lanata*).

Population	Elevation (m)	Growing Season (days)	Annual Precipitation (cm)
1	2,615	<60	58
2	2,301	60-100	47
3	1,993	60-100	36
4	1,945	60-100	35
5	2,103	60-100	40
6	2,027	60-100	38
7	2,377	>100	30

Table 31. Relation between measured responses in ecotype garden of winterfat (*Ceratooides lanata*) and their original environments.

Response	Environment	Correlation	Intercept	Slope	Significance
Dormancy, Oct 14	Growing season length	-0.97	3.6	-0.51	$p < .005$
Phenological Index	Growing season length	-0.94	4.9	-0.85	$p < .005$
Dormancy, Oct 14	Annual precipitation	0.81	1.0	$.03$	$p = .01$

Since there are only seven populations involved for winterfat, multiple factor involvement cannot be ascertained. For example, length of season and annual precipitation are highly correlated in an inverse way ($r=-0.87$), and results of multiple regression with these as well as other combinations of factors are not significant beyond the first step. Multiple factor analyses are being pursued for other species of this study where population representation is greater.

To summarize, significant biological differences among ecotypes of winterfat have been identified and are likely to exist to varying degrees in the other species. The evidence and interpretation of these differences should provide part of the basis for better decisions on the selection of plant materials for reclamation.

Phase Two--Phytosociology

The results of the vegetation sampling reported earlier (see progress reports of 1978 and 1979 for sampling regime and types of stands sampled) have been used to form a model of the major vegetation types of the Piceance Basin. To fulfill the two primary aims of this phase of the study two concrete applications of the model are presented herein. First, a "User's Guide" is outlined. This guide will enable the reclamationist to predict (by use of simple environmental measurements and reference to the vegetation model) the plant species combinations best adapted to the specific site to be reclaimed. Second, the relationships of the native vegetation of the Intensive Study Site to the other kinds of communities in the Piceance Basin are presented in a way which emphasizes the controlling environment. The reasoning behind these aspects of the study are in the Progress Report of 1979, pp. 164-181.

Predicting Vegetation from the Environment

The use of the vegetation model for reclamation is outlined in Figure 35. Principal component analysis of 136 sampled stands of vegetation on the basis of their species composition resulted in four independent components of variation (Figure 35, Step 1). These give the four most important dimensions of variation in vegetation across the landscape. Reclamation concerns can be focused on this reduced set of dimensions of variation.

Accordingly, the stands were classified into community types by their position in the four dimensional principal component space (Figure 35, Step 2). The classification method used was the sum of squares clustering technique of Orloci (1975). Each community occupies a separate region of this space. This is an effective way to divide the essentially continuous vegetation into units relevant to reclamation. The distribution of the nine community types on the first three principal components is given in Figure 36. Hierarchical arrangement of the communities is given in Figure 37. The details of this vegetation model, presented generally here, will be published elsewhere. The application to the objectives at hand continues here.

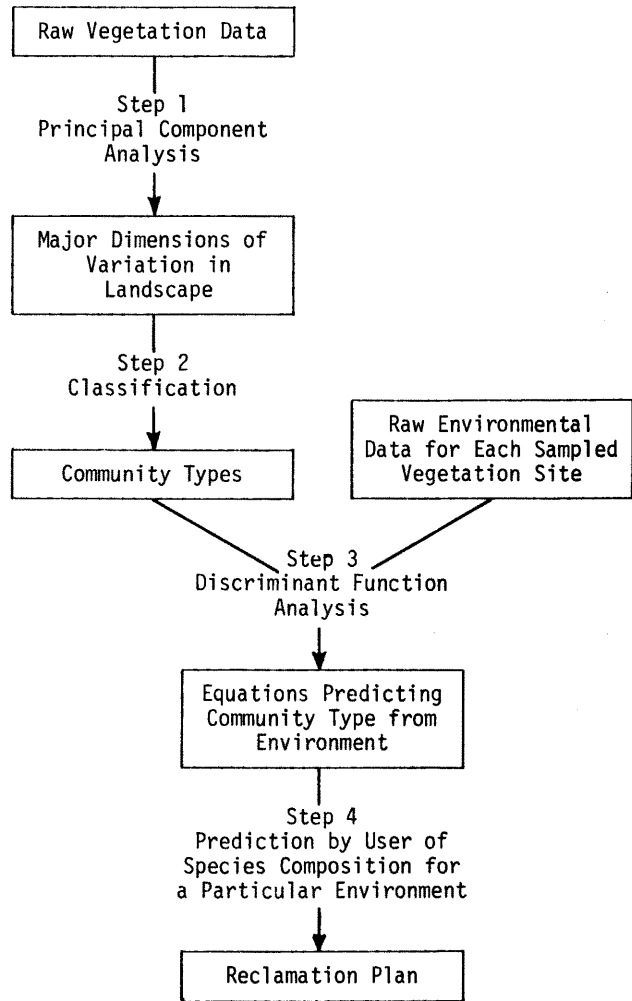


Figure 35. Portrayal of the way the vegetation model is constructed and then related to the environment allowing a user to predict from the environment the communities best adapted to a particular site. Steps 1-3 (already carried out) enable a user to complete Step 4, illustrating the way in which this research can be used as input to an overall reclamation plan.

The communities defined only by species composition are related to their environments by using stepwise discriminant function analysis (Figure 35, Step 3). This technique uses the measured environmental variables for each stand in each community type as predictors of the stand's community type. As in multiple regression, equations are constructed which predict group membership. Given the specific environment of a site to be reclaimed, both the community type (which has the most similar environment) and the probability of membership in that type can be calculated (Figure 35, Step 4).

Two complications in the use of this technique are noted. First, some of the kinds of vegetation included in this model are less likely than others

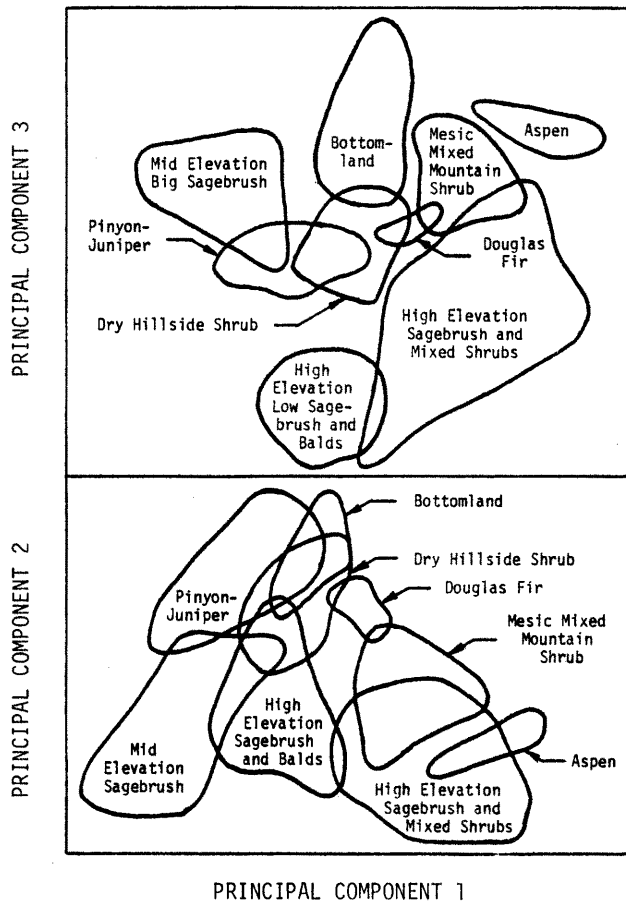


Figure 36. Display of nine community types on the first three components of stand ordination. Although types overlap when any two dimensions are viewed, there is essentially no overlap in the three dimensional component space and no overlap if the fourth dimension is added.

to be included in a reclamation plan. For example, aspen and Douglas fir stands (and also stands which are on very steep slopes) are present in the complete model but will rarely be targeted for reclamation. Accordingly, different discriminant functions have been produced which include and exclude these stands. Second, as in multiple regression different combinations of predictor variables, including power terms and the interactions of two or more variables, can be used. Even through better prediction within the original data is possible, there is increasing danger of producing spurious results arising from the specific data used the closer a saturated model is approached. To avoid this, only interactions of the basic terms which are thought to have biological significance were used (e.g., the way elevation, slope, and exposure are thought to combine to reflect potential evapotranspiration).

The discriminant functions derived will be presented in a form tailored for easy use by a reclamation user. The form will be a self-contained handbook or user's guide which will

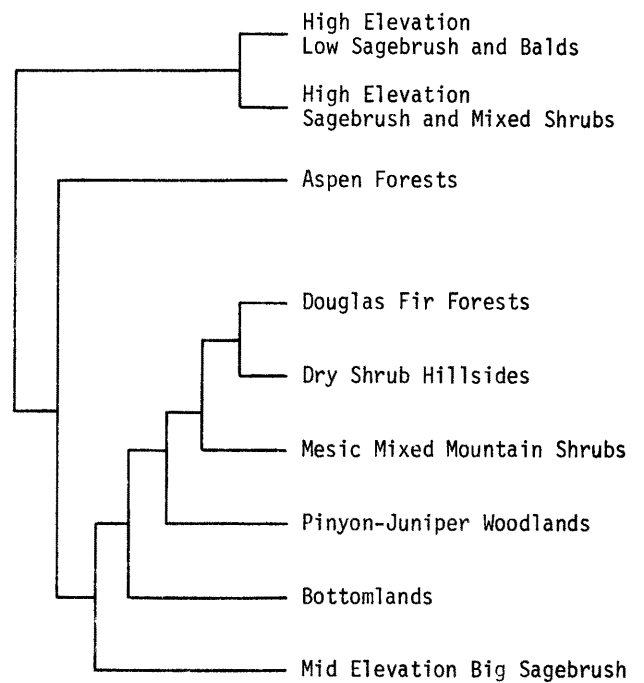


Figure 37. Hierarchical arrangement of community types based on position in four-dimensional principal component space.

present the step-by-step process of predicting the likely species combinations to effect long-term, low maintenance, natural success for specific sites within the Piceance Basin. The following are the contents (in outline) of the "User's Guide".

- I. Introduction: the use and limitations of the guide.
- II. Measurement of the environment of a site to be reclaimed: slope, exposure, elevation, topographic context, soil texture, etc.
- III. Calculation of derived environmental scalars: potential evapotranspiration, temperature, water stress, radiation index, etc.
- IV. Combining environmental scalars to predict the most likely community type and the probability of membership.
- V. Description of species composition of predicted groups.

Section III will include all the necessary tables and equations for combining the basic environmental measures into the form necessary for the prediction equations. No other outside sources are needed. Section IV will give an equation for each discriminant function. Solving these equations will give the community to which the measured environment is closest. Another set of equations, given the results of the first, will allow the calculation of the probability that the site is within that community.

When these calculations are performed on the original data, over 92 percent of the stands are correctly classified. Inspection of misclassified stands reveals that they are intermediate between their actual and predicted communities. This is explained by the slightly different ways the vegetation continuum is divided up by the vegetation classification routine, as opposed to discriminant function analysis. No stands were misclassified into unrelated (more distant) community types. To accommodate the artifact of classifying essentially continuous variation, the community descriptions given in Section V will include discussion of intra-community variation. This will enable the user to select modified species compositions for sites which have nearly equal probabilities of belonging to different community types.

Extrapolation from the Intensive Study Site

The second main objective of this research is to indicate how other reclamation research results obtained on the Intensive Study Site can be extrapolated to environments and communities outside of the site itself. One question we can answer regarding such extrapolation is, "What are the most important environmental differences between the mid elevation sagebrush communities of the Intensive Site and other communities; what are the relevant environmental differences between the on-site and off-site vegetation?" Relevant environmental differences are those to which the vegetation itself is responding as it differs in landscape position.

Since actual observations of the environmental causes of vegetation variation are impossible in this context, the strategy is to interpret extant vegetation variation through correlation with the environment. The results of community ordination and classification followed by discriminant function analysis as outlined above is just this.

Some of the results of the analysis of the six communities in the Piceance Basin that were most closely related to the vegetation of the Intensive Study Site and which compose the predominant part of the landscape are given in Table 32. The entries in the table are the standardized discriminant function coefficients for the first two functions which separate the six communities. Only the first two are given since they account for 92 percent of the variance of the community classification. There is a coefficient for each environmental variable for each function, and they are interpreted as regression coefficients. The higher the absolute value of its coefficient, the more the factor contributes to the separation of groups. Only the most important factors are given in Table 32. For example, elevation and growing season water stress are most important on the first dimension while growing season water stress and percent slope are weighted most on the second. (The environmental variables are defined in the Progress Report 1979, Table 27.)

This means that of all the environmental variables tested the ones with the greatest weights give the widest and most consistent community separation. Since they also reflect environmental differences which are biologically relevant (e.g., water and insolation), it is plausible to conclude that these are the factors to which the vegetation

Table 32. Standardized discriminant function coefficients for selected environmental variables. Functions are significant at .005. The canonical correlation coefficients are 0.97 and 0.83 and account for 80 and 12 percent of the variance in group membership, respectively.

Environmental Factor	Discriminant Function	
	One	Two
Elevation	12.6	-3.4
Growing season water stress	9.8	6.1
Growing season potential evapotranspiration	7.7	4.0
Slope	0.5	3.9
Radiation index	-4.1	1.0

is responding as it varies over the landscape. Notice that this is an argument based on correlation, not experimental manipulation, but it is strengthened by the biological truisms that the distribution of water and radiation on the landscape are biologically important and that they control the distribution of communities on the landscape.

The pertinence of this to the question at hand--"What are the important, relevant environmental differences among these communities?"--is made clear in Figure 38. Here each stand was evaluated on the basis of its environment by the discriminant function. The average position of stands for each community is then plotted against the two discriminant functions. The territory (or region) in this environmental space for each community is also indicated. The figure is an environmental ordination of the six community types constructed from those factors most important as described above. The difference in scale of the horizontal and vertical axis results from their being scaled to the proportion of the total variance for which they account (80 and 12 percent, respectively).

Identifying the environmental differences among communities from this ordination is facilitated by including for each community its average value for the individual environmental variables contributing to each axis (= discriminant function). Growing season, potential evapotranspiration, and slope are given here. The environment of mid elevation sagebrush can be seen to differ from that of high elevation sagebrush in having more potential growing season evapotranspiration, while it differs from mesic mixed mountain shrub in evapotranspiration as well as slope steepness.

Some of the relevant environmental differences that could be plotted on the ordination in this way are instead given in Table 33. Similar tables, including all the significant (two dozen or so) environmental measurements and scalars which discriminate among these community types, are being prepared. They will be made available, by request, as an appendix to this report. This will enable other reclamation researchers to compare

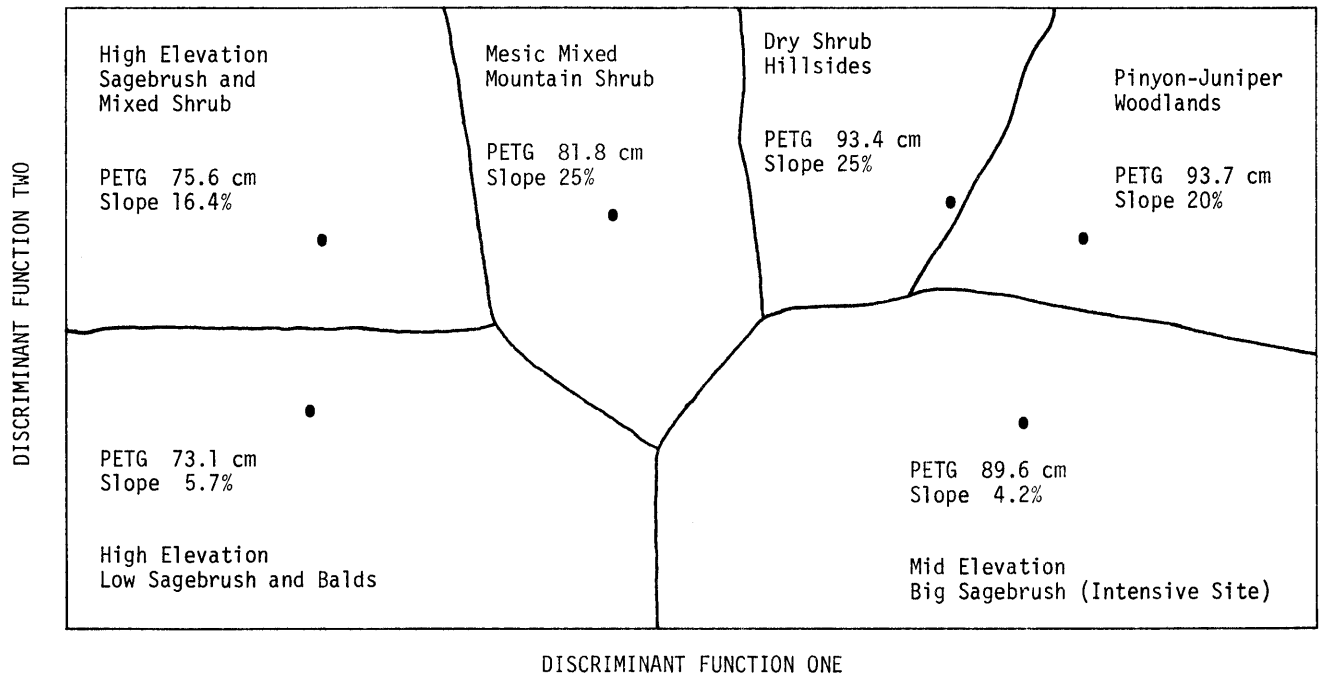


Figure 38. Environmental ordination of communities by first two discriminant functions (see Table 32). The points represent mean community position. Partitions represent the range of territory of each community on these two functions. Potential evapotranspiration for the growing season (PETG) and slope are given. See text for explanation.

Table 33. Average environmental values for communities. The ranges given are one standard deviation. The signed numbers express the differences from the mid elevation big sagebrush community typical of the Intensive Study Site.

Community	Elevation (m)	Potential Growing Season Evapotranspiration (cm)	Percent Slope	Radiation Index
Mid Elevation Big Sagebrush	2062 ± 98.4	89.6 ± 4.8	4.2 ± 2.5	.478 ± .010
High Elevation Big Sagebrush and Balds	2600 ± 21.7 +538	73.1 ± 5.4 -16.5	5.7 ± 2.7 +1.5	.485 ± .026 +.007
High Elevation Sagebrush and Mixed Shrub	2571 ± 45.1 +509	75.6 ± 5.0 -14	16.4 ± 8.2 +12.2	.500 ± .039 +.022
Mesic Mixed Mountain Shrub	2432 ± 126.2 +370	81.8 ± 8.8 -7.8	25.6 ± 12.9 +21.4	.506 ± .046 +.028
Dry Shrub Hillsides	2044 ± 247.4 -18	93.4 ± 11.8 +3.8	25.3 ± 12.6 +21.1	.502 ± .061 +.024
Pinyon-Juniper Woodlands	2013 ± 98.7 -49	93.7 ± 8.4 +4.1	20.6 ± 11.8 +16.4	.498 ± .054 +.020

factors they find experimentally to be important for reclamation with the factors we found to be important for controlling community distribution on the landscape.

SUMMARY

Data for the population phase of the study further define the ecogenetic differences within certain species and permit initial interpretations regarding the probability of success of different source materials on various sites. For example, mid elevation populations of winterfat from the Piceance Basin appear to be suitable for several sites in the Basin, while a high elevation population and one from outside the Basin would have restricted site success due to their special phenological habits.

Data for the second part on community variation in the Piceance Basin form the basis for a multivariate model of vegetation/environment relationships. The model is outlined in the form of a "User's Guide" which upon completion will enable the reclamationist to predict (by use of simple environmental measurements) the plant species combinations best adapted to the specific sites to be reclaimed. The model may also be employed for use in extending other research findings of the Intensive Study Site to the different kinds of communities found in the Basin.

LITERATURE CITED

Orloci, Lazlo. 1975. Multivariable analysis in vegetation research. Dr. W. Junk, The Hague.

SELECTION OF NATIVE GRASSES AND LEGUMES FOR IMPROVED REHABILITATION

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OBJECTIVES

This research involves the development of improved plant material for the oil shale region by testing and selective breeding of a small number of important species. The hypothesis is that a few improved strains of each species could be developed from genotypes of local or regional origin which would be adaptable in the rehabilitation of diverse, stable (self-sustaining), and functional ecosystems. The improvements sought are those traits important to early growth and land stabilization, such as seed size and lowered dormancy which contribute to rapid establishment and better seedling vigor, as well as the capacity to produce useful quantities of seed. These must be achieved without losing the ecological adaptation or altering the role played by these grasses and legumes in advanced successional stages.

Research and selection and breeding of plant materials is summarized in the following four objectives.

1. Collect and assemble germplasm sources.
2. Evaluate spaced plants in two nurseries and clonal propagules or progenies under controlled stress for various seed and plant characters; produce test seed.
3. Evaluate seed progenies at the Intensive Study Site and satellite field plantings for survival, vigor, and adaptation.
4. Use suitable breeding procedures for recombination of parents selected by progeny testing to develop improved strains.

PROGRESS TO DATE

Research on the various species has proceeded at different rates. For grasses we are at Objective 3 and at Objective 4 for the development of improved strains. The legume work has proceeded at a somewhat slower pace with the research at Objectives 2 and 3. Work was done mainly at the Intensive Study Site with some procedures in the breeding nurseries. Progress in 1979 and early 1980 is discussed under the four headings which correspond to the objectives.

Collection of Germplasm

The native western wheatgrass (*Agropyron smithii* Rydb.) nursery planted in the Environmental Plant Center (EPC), Meeker, became well established and was harvested in late summer 1979. This nursery was planted to provide a permanent seed source from cloned native plants. Collection of Indian ricegrass has been accomplished except for seed from the ecotype garden at the Intensive Study Site. That seed may need to be grown in increase nurseries before an adequate supply can be collected for testing and study.

No new collections of either lupine (*Lupinus argenteus* Pursh and related taxa) or Utah sweetvetch (*Hedysarum boreale* Nutt.) were made in 1979; however, a nursery with the same goal as the native western wheatgrass nursery mentioned previously was planted for both lupine and Utah sweetvetch at the Environmental Plant Center, Meeker. This planting will be used to study the native collections and will act as a seed increase nursery for each.

Evaluation of the Sources in Nurseries

Spaced nurseries for western wheatgrass and Indian ricegrass presently serve as sources of parent plant material of selected elite plants. Both the lupine and Utah sweetvetch seed increase nurseries at Meeker (mentioned previously) had become established by June 1979 but remained in the vegetative stage for the rest of the year. The first seed harvests for these nurseries are expected in 1980. Data for these grasses and legumes are detailed below.

Western Wheatgrass

A second seed harvest at the western wheatgrass nursery in Meeker was made to give two years of data for both the Fort Collins and Meeker nurseries. Data were also taken on culm height and spike length at Meeker to see if these traits can be used (as in many grasses) for ecotype recognition and as components of seed yield and harvestability. Net seed yields are not available at this time. Harvest in 1979 at Meeker was made on individual plants in each plot, and the analysis of variance of plot means in three replications showed highly significant ($p < .01$) differences among the sources for culm height and spike length (Table 34).

Table 34. Plant and seed characters of western wheatgrass at Meeker, 1978 and 1979.

Source	Culm Height (cm)		Spike Length (cm)		100-Seed Weight (mg)
	1978	1979	1978	1979	1978
<u>Varieties</u>					
Arriba	76.6	66.1	13.6	12.9	505
Barton	60.6	68.1	13.0	14.3	456
Flintlock	69.9	58.3	13.4	13.6	527
Mandan 456	81.3	64.2	13.4	13.1	473
Rosana	66.3	61.7	13.3	13.8	553
<u>Accessions</u>					
A-13981	73.4	71.7	13.3	13.8	421
A-16592	62.8	67.5	11.6	14.1	382
A-16634	71.4	63.2	12.9	13.5	367
A-16931	67.4	57.2	1.27	11.9	450
BN19823	66.2	57.9	14.1	15.6	424
BN19824	86.8	69.0	16.4	16.8	517
C-27	70.2	63.9	13.4	14.0	583
NM-429	80.1	76.8	15.8	15.6	476
P-727	68.4	66.0	14.6	14.8	402
E6-37	58.4	54.4	13.0	14.5	444
<u>Collections</u>					
Colo City	68.9	57.8	13.7	14.0	416
Tincup	68.7	66.0	12.0	12.0	466
TS-11-11/15	69.9	64.7	12.1	13.2	426
TS-11-16/20	70.4	69.0	12.6	15.7	448
360	66.3	56.6	11.3	13.9	415
361	63.0	59.5	12.0	11.9	442
370	80.5	61.6	16.4	14.4	470
372	62.5	55.1	12.0	13.1	493
378	71.0	59.3	11.0	12.1	432
385	68.1	52.5	12.2	11.5	278
387	61.3	54.0	10.2	11.6	401
390	62.3	52.0	11.6	12.1	388
399	52.6	51.7	9.2	12.3	357
440	62.3	54.3	12.5	12.0	314
525	64.5	54.6	13.2	14.3	462
Mean	68.9	61.2	12.9	13.5	442
S.E. (mean)	4.5 ¹	3.0 ¹	0.9 ¹	0.8 ¹	35 ¹
Correlation: 1978-1979	0.68 ¹		0.70 ¹		
Correlation: Culm height and Spike length	0.76 ¹ (1978)		0.55 ¹ (1979)		

¹Differences among entries and correlations are highly significant.

The correlations between the two years of data at Meeker show that there was no appreciable change in seed or plant characters between the two years. This indicated that the differences between sources are mainly due to genetic, not environmental, causes.

Selection of the best individuals for progeny testing in the Piceance Basin is based on

spread, vigor, and 100-seed weight. As last year's report showed (p. 195-197), there is a range of values available for selection. The opportunity was taken for genetic improvement with these traits, individually and collectively. Testing of 96 selected progenies is in progress at the Intensive Study Site; first year performance results for 49 of them are reported herein.

Indian Ricegrass

The nurseries established at the Agronomy Research Center (CSU, Fort Collins) and the Meeker EPC in 1977 each contain 16 sources represented by 15 plants. The 480 plants were individually harvested for seed in 1979. Additional plants grown from seed collections of Dr. Lang (University of Wyoming) and native collections made by this sub-project and planted in 1978 were harvested for the first time in 1979.

The second year comparisons for leaf traits and panicle height (related to ecotypic differences in herbage yield per plant in Indian ricegrass) were analyzed statistically with highly significant variation found among sources for many traits (Table 35). Data on seed yield, seed weight, and their relationships await analysis. Within and between nursery collections, correlations have also been calculated for some of these traits.

Table 35. Indian ricegrass source information and plant characteristics.

Source	Origin	Panicle Height (cm)	Leaf Width (mm)	Leaf Length (cm)
Breeders	North Dakota	46	3.0	52.9
Paloma	Pueblo, CO	54	2.2	52.7
Sharps	Kansas ¹	49	2.3	51.9
Warner	Wiggins, CO	39	---	---
M-700	Montana	37	1.8	27.7
NM-15	New Mexico	36	2.0	54.9
NM-168	Los Lunas, NM	44	2.2	56.0
P-2575 ²	White Bird, ID	54	3.2	59.0
P-15597	Bridger, MT	46	3.0	44.9
P-15598	Bridger, MT	39	1.9	39.2
P-15650	Bridger, MT	41	3.3	51.7
P-15657	Bridger, MT	47	2.1	31.6
16503-65	Holbrook, AZ	---	1.8	51.3
525-36	Utah	---	3.1	46.3
E-1	Eagle, CO	46	---	---
PB-1	Piceance Basin	---	3.5	55.0
PB-2	Piceance Basin	36	2.3	34.9
PB-4	Piceance Basin	47	2.7	42.0
SC-1	Colorado	41	---	---
Mean		44	2.5	46.3
LSD (0.05)		---	0.3	3.0
Correlation:				
Leaf length x leaf width			0.41	
Panicle height x leaf length			0.46	

¹State where strain was developed.

²Released by SCS as Nezpar.

Single-plant selections of 34 Indian ricegrass plants have been made for progeny testing based on high seed yield, heavy 100-seed weight, and vigor in the belief that these characters are important for the most effective rehabilitation technology. Seed progenies of these elite plants have been planted in progeny tests in the Piceance Basin.

Indian ricegrass seed germination, seed covering thickness, seed shape, and 100-seed weight were studied to determine the variability among sources and the relationship between the traits. These data appear in Table 36. There were significant differences in germination, seed covering thickness, and seed weight for 15 sources from both nurseries. There was no difference in location response for these characters--indicating that they are influenced more by genotype than by environment. Seed covering thickness was positively correlated to seed weight (+0.48) and negatively correlated to germination (-0.47). This indicates that care in selection for moderate seed covering thickness could result in increased germination (i.e., decreased dormancy). The lowering of dormancy by direct selection among progeny of nondormant seeds is also a goal in breeding of Indian ricegrass.

Until this goal is reached, the use of scarification to break dormancy of seed lots is

necessary. The scarification method (rubbing machine) developed by this subproject (see 1978-1979 progress report) was used in combination with November planting for the progeny tests in 1978 and 1979. Establishment in 1979 was excellent indicating that the scarification method was successful in increasing germination up to 40 to 50 percent. This improvement is necessary for getting maximum use of Indian ricegrass for rehabilitation. The scarification method is mechanical and could be used by industry with very little training or expertise.

Lupines

Thirty-two collections, seeded at the Meeker EPC in October 1978, were established as small seedlings by June 1979 but did not produce enough harvestable seed to be used for seed data analysis.

N₂ fixation was assayed in seedlings grown from some of these collections. They were planted in sand in greenhouse flats, inoculated with rhizobial cultures, and watered with mineral nutrients other than N. The acetylene-reduction assay was used as an estimate of comparative N₂-fixation capability. The results are not available at this time.

Table 36. Seed data for Indian ricegrass harvested from the Fort Collins and Meeker nurseries.

Strain	Fort Collins			Meeker			Average		
	Germination (%)	100-Seed Weight (mg)	Seed Covering Thickness (μ)	Germination (%)	100-Seed Weight (mg)	Seed Covering Thickness (μ)	Seed Width	Seed Length	Shape Index
Breeders	5.4	345	55	5.4	331	56	1.8	3.4	1.9
Paloma	1.5	444	70	4.5	436	73	2.0	3.8	1.9
Sharps	0.2	460	71	1.8	408	78	2.0	4.0	2.0
M-700	0.3	272	76	0.0	241	74	1.7	3.0	1.8
NM-15	0.9	385	77	1.8	309	75	2.0	4.0	2.0
NM-168	2.2	469	71	5.2	422	73	2.0	3.9	1.9
P-2575 ¹	8.2	335	42	7.7	336	44	1.6	4.2	2.6
P-15597	0.2	323	50	2.6	307	51	1.7	3.5	2.0
P-15598	0.6	338	72	1.0	298	77	1.9	3.2	1.7
P-15650	0.3	271	50	0.3	275	51	1.8	3.2	1.8
P-15657	0.6	290	67	1.9	318	62	1.6	3.6	2.3
PB-2	1.0	287	69	3.9	341	64	1.9	3.2	1.7
Warners ²	0.6	357	67	---	---	---	1.9	3.4	1.8
16503-65 ³	---	---	---	1.5	399	78	2.0	3.7	1.9
525-36 ³	---	---	---	6.7	275	56	1.9	3.1	1.6
Mean	1.7	352	64	3.2	343	65	1.9	3.4	1.9
LSD (.05)	1.2	72	8	1.2	60	8			

Correlations:

Seed width x seed length (0.16)
 Seed width x leaf width (0.39)
 Seed length x leaf length (0.38)

¹Released by SCS as Nezpar.

²Not planted in the Meeker nursery.

³Not planted in the Fort Collins nursery.

Utah Sweetvetch

Four collections including one commercial source from Utah were directly seeded (eight replications) at the EPC in Meeker in 1978. Like the lupine, they became established in 1979 but did not produce any flowers or seed. There was good flowering in Fort Collins on the 102 plants of the Utah collection in the 1977-established nursery. The 11 best seed producers were harvested individually and will be multiplied.

Testing Source Material and Progenies in Critical Environments

The testing of elite material in critical environments is the most essential step in proving which commercial varieties, strains, accessions, ecotypes, or single-plant progenies can succeed in the rehabilitation process. Moreover, by identifying the best mother plants with this criterion the breeder can develop the synthetic strains which are needed for that specific task (i.e., to perform well on disturbed areas in the Piceance Basin). Progeny tests for both the grasses and a legume evaluation planting were seeded at the Intensive Study Site. All 1978 plantings showed good growth in 1979 and will be discussed subsequently in separate sections.

Western Wheatgrass

Progeny tests planted at the Intensive Study Site in 1978 and 1979 used selections based on data from the two evaluation nurseries and the direct seeding strain trial planted in 1976. The 1978 progeny test was planted in a randomized complete block design with three replications. The data are presented in Table 37. While most selections did well, there were some whose performance could be considered excellent; those entries with ratings of 3.7 or over are strong candidates for recombination. Spring (1980) data based on winter hardiness and spring growth need to be taken before final selection can be made. The second progeny test which was planted in fall 1979 will not produce usable data until summer 1980.

Indian Ricegrass

Replicated progeny tests were established at the Intensive Study Site in fall 1978, spring 1979, and fall 1979. Data from fall 1978 and spring 1979 establishments are presented in Table 38. The spring test differed in time of planting in order to test two types of scarification procedures and to insure a successful progeny test. The seeds planted in fall 1978 were treated by mechanical scarification (rubbing machine), and the seeds planted in spring 1979 were scarified with sulfuric acid. As can be seen in Table 38, there were significant differences ($p < .01$) between selections for both tests with some proving to be excellent. These selections are the 16 progenies with a vigor score of 3.7 or more in October 1979. The spring 1979 progeny test also showed some excellent entries, and many of the same selections proved superior in both tests. One thing to note is the generally

Table 37. Fall vigor scores (October 9, 1979) for 49 progenies of elite western wheatgrass plants seeded at the Intensive Study Site in November 1978.

Elite Parent ¹			
Row	Strain	Plant	Vigor ^{2,3}
<u>Variety</u>			
FC- 4	Arriba	1	3.3
FC- 4	Arriba	2	4.3
M -12	Arriba	4	4.3
FC- 3	Barton	3	3.7
FC- 3	Barton	4	3.7
FC- 3	Barton	5	4.0
FC- 4	Barton	5	3.7
M -19	Barton	1	3.7
M -19	Barton	3	2.7
M -19	Barton	4	3.7
M - 5	Flintlock	5	3.3
M -11	Flintlock	2	3.7
M -15	Flintlock	3	2.7
M -15	Flintlock	5	3.0
M - 1	Mandan 456	3	3.3
M -19	Mandan 456	1	3.7
M -19	Mandan 456	3	3.7
FC- 2	Mandan 456 (006)	2	4.0
FC- 3	Mandan 456 (006)	5	4.3
M -11	Rosana	1	3.7
FC- 2	Rosana (002)	1	4.3
FC- 3	Rosana (002)	4	4.0
<u>Accession</u>			
FC- 1	A-13081	3	3.7
M - 9	BN-19824	2	3.3
M - 9	BN-19824	3	2.6
M -17	BN-19824	2	4.0
M -17	BN-19824	3	3.0
M - 2	C-27	1	3.7
M -17	C-27	1	3.7
FC- 2	NM-429	2	3.3
M -18	NM-429	3	2.3
FC- 3	P-727	4	3.7
<u>Collection</u>			
FC- 1	Colo City	5	4.0
M - 6	Colo City	1	4.0
M - 8	Colo City	1	3.3
M -18	Tincup	3	3.0
G - 2	010	5	3.3
G - 2	031	5	2.7
G - 3	071	3	3.0
G - 3	101	3	3.3
G - 1	221	2	3.0
FC- 2	TS-11/16-20	2	4.0
FC- 3	360	2	3.0
M - 6	370	4	3.3
M - 8	387	5	3.7
FC- 4	411	4	4.0
FC- 1	525	5	3.3
FC- 2	525	4	3.7
FC- 2	525	2	3.7

¹28 parents out of 49 tested are expected to be retained.

²Mean = 3.5±0.2

³Selection cut-off 3.7.

lower scores of the spring 1979 progeny test. This shows the importance of fall planting to take advantage of available early spring soil moisture and utilize the entire growing season. Both these tests will be evaluated in spring and summer of 1980 to determine winter survival and regrowth. The fall 1979 progeny test was planted with mechanically-scarified seed of eight selections from both the Lang Wyoming collections and this project's native collection. This test will start producing data in spring 1980. The value of the

Table 38. Fall evaluation (October 9, 1979) of 34 progenies of elite Indian ricegrass plants seeded in the Intensive Study Site in fall 1978 and spring 1979.

Elite Parent			Vigor Score	
Row	Strain	Plant	Fall Planting	Spring Planting
<u>Varieties</u>				
8	Breeders	4	4.0	2.3
3	Breeders	M ¹	4.7	2.0
4	Paloma	3	3.2	--- ²
5	Paloma	2	4.0	3.8
5	Paloma	3	4.2	---
5	Paloma	5	3.7	3.5
1	Paloma	M	3.3	---
3	Paloma	M	3.0	---
1	Sharps	2	4.0	2.0
1	Sharps	5	3.3	2.7
6	Sharps I	2	3.3	2.3
6	Sharps I	3	3.7	0.7 ³
6	Sharps II	3	2.3	2.7
5	Sharps	M	2.8	2.7
3	Warners	2	4.8	---
3	Warners	5	3.7	---
6	M-700	3	2.3	2.7 ³
6	M-700	4	2.0	3.0 ³
7	M-700	3	2.3	2.3 ³
7	M-700	4	3.0	3.0 ³
<u>Accession</u>				
1	NM-15	3	4.2	---
1	NM-168	4	3.5	2.0
6	NM-168	1	4.0	2.7
6	NM-168	3	4.3	3.3
7	NM-168	3	4.2	3.7
7	NM-168	4	4.8	3.3
7	NM-168	5	4.5	3.3
5	NM-168	M	2.7	---
6	NM-168	M	3.3	2.7
	P-15597	M	4.0	---
3	16503-65	M	3.0	---
<u>Collection</u>				
3	E-1	5	1.2 ³	
5	PB-2	3	2.3 ³	
1	SC-1	2	2.3 ³	
Mean			3.4±0.5	2.7±0.2

¹M = source harvested by row in Meeker.

²Not included in spring.

³These rows did not produce seed.

scarification is in enhancing the uniformity as well as the speed of establishment so that quicker and more precise selection can be done.

Lupines, Utah Sweetvetch, and Other Legumes for Nitrogen Fixation

The legume study plot (which was planted in conjunction with Drs. Berg and Sabey) was established to evaluate native and introduced legume species that have high N-fixing abilities. The types of legumes planted were alfalfa, lupines, Utah sweetvetch, cicer milkvetch, crown vetch, sainfoin, purple prairieclover (*Petalostemon purpureum*), and Swainson's pea (*Sphaerophysa salsula*). They were planted in October 1978 on both topsoil and subsoil disturbed plots. Ratings were made in June and October 1979, and the October data (which closely matched the June data) are given in Table 39). Response to the soil types varied from legume to legume with Utah sweetvetch being definitely better on the more clayey subsoil and the lupines being better on the siltier topsoil. Species and strains performing well were: all sainfoin accessions, all cicer milkvetch strains, several sources of Utah sweetvetch, L-031 and L-142 lupine collections, and Penngift crownvetch. The alfalfa, purple prairieclover, and Swainson's pea were represented by only one strain, but all became established in the first year.

Spring 1980 evaluations for winter hardiness, survival, and spread are crucial for the legumes, especially the non-natives, because of their luxuriant growth in fall 1979. On-site investigation of nitrogen-fixation is already in progress under Dr. Klein's subproject, and we will cooperatively determine variation in N-fixation between and within species.

Recombination of Breeding Material

Five recombination blocks have been or will be established by spring 1980. A 66-elite-clone western wheatgrass recombination block was set out in spring 1978 and harvested for seed in 1979. A 96-elite-clone recombination nursery was planted in spring 1979 with elite plants identified by data from Fort Collins, Meeker, and the Intensive Study Site as heavy seed-weight plant with good seed yields from vigorous sources. All 96 are represented in progeny tests in the Piceance Basin. Less desirable plants based on progeny test results can be cut back before flowering to remove their pollen contribution. This recombination block will be harvested in summer 1980, the seed produced representing the first generation of an adapted synthetic. This seed will be tested in the Piceance Basin starting in fall 1980.

Indian ricegrass has two established recombination blocks and one planned for spring 1980 transplanting. The two established blocks, low-dormancy and five-strain recombination, both yielded a first-year seed harvest in 1979. From the low-dormancy block the seeds will be tested without scarification, and those seeds which germinate

Table 39. Fall evaluations (vigor scores, October 9, 1979) of the legume adaptation study at the Intensive Study Site seeded October 1978.

Species and Strain	Vigor Scores ¹	
	Topsoil	Subsoil
<u>Utah sweetvetch</u>		
ISS ² (Piceance Basin)	3.2	3.5
E/ISS ³ (Piceance Basin)	2.8	3.2
Pipeline (Piceance Basin)	2.0	3.2
Stewart (Utah)	3.8	4.2
Native Plants Inc. (Utah)	4.5	4.2
Mean	3.3	3.7
<u>Lupine</u>		
L-031	4.5	2.0
L-142	4.0	3.0
L-225	3.5	2.5
L-232	1.5	1.0
Mean	3.4	2.1
<u>Cicer milkvetch</u>		
Lutana	3.0	4.3
C-4	4.0	4.3
20-15	4.0	5.0
Dotzenko	4.0	5.0
Strohman	4.0	4.3
Sugarbeet 2	4.3	5.0
Wellington	4.5	4.3
Mean	4.0	4.7
<u>Crownvetch</u>		
Penggift	3.2	3.5
Emerald	2.5	2.8
Chemung	3.2	2.5
Mean	3.0	2.9
<u>Sainfoin</u>		
Eski	5.0	5.0
Melrose	5.0	4.8
Remont	4.8	5.2
Mean	4.9	5.0
<u>Purple prairieclover</u>		
Kaneb	2.8	2.0
<u>Alfalfa</u>		
Ladak	5.0	4.0
<u>Swainson's pea</u>		
San Luis Valley	4.0	3.5

¹Scored 0-5 from poor to excellent (a few ratings of 6 were given to indicate superior establishment).

²Collected inside the fence at the Intensive Study Site, Piceance Basin.

³Collected east of the Intensive Study Site, Piceance Basin.

promptly will be planted in spring 1980 to make up the second-cycle selection of low dormancy plants. The five-strain intervarietal recombination will also have its seed planted in spring 1980 to produce a second cycle. The new recombination block will be based on the results of the Indian ricegrass progeny tests in the Piceance Basin with elite parent clones to be transplanted in spring 1980.

The lupine seed increase nursery in Meeker will be used as a recombination block allowing natural selection to operate against inferior phenotypes with seed harvested from the more fruitful and superior plants. A similar procedure will be used with Utah sweetvetch at Meeker as well as multiplying the genotypes of the 11 high seed-producing plants identified in Fort Collins in the Stewart strain.

SUMMARY

Progeny tests for western wheatgrass and Indian ricegrass based on nursery data identifying desirable elite plants have been planted and partially evaluated at the Intensive Study Site in the Piceance Basin. From these results synthetic strains will be developed using the recombination blocks established or planned for spring 1980.

Evaluation tests of legumes in the Piceance Basin showed good growth in 1979. Future observations will indicate how useful native and introduced perennial legumes will be for rehabilitation, including their nitrogen contribution. Among the non-natives, sainfoin and cicer milkvetch strains appeared to perform best in the first year of growth at the Intensive Study Site.