Forage crop species response to varying soil moisture stress during germination

by William Vogel

A THESIS Submitted to the Graduate Faculty in partial fulfillment of the requirements for the degree of Master of Science in Agronomy
Montana State University

Abstract:
A study was made to investigate the ability of 22 grasses and 11 legume species to germinate against a moisture stress in Manhattan fine sandy loam. The soil was adjusted to various moisture contents corresponding to 5, 9, 10-2/3, 12-1/2 and 15 atmospheres. A check was conducted at field capacity.

When moisture content decreased, germination decreased. In a few species, germination was higher at 9 than at 5 atmospheres soil moisture tension.

At the highest tension the order of decreasing ability to germinate against tension was found to be: Fairway crested wheatgrass, Intermediate wheatgrass, Standard crested wheatgrass, Nordan crested wheatgrass, Russian wildrye, Tall wheatgrass, Lincoln smooth bromegrass, Pubescent wheatgrass, Slender wheatgrass, Tualatin oatgrass, Alta fescue, Switch-grass, and Meadow foxtail. Other species did not germinate.

It was observed that germination of legumes, in general, was depressed to a greater degree than the grasses. This was attributed to the fact that the cotelydons of the legumes pushing through the soil, caused a cracking of the soil surface, thus exposing the seed to the atmosphere.

When the soil, in which species were subjected to moisture tension, was restored to field capacity, it was found that some species exhibited increased germination, some were not affected, and some showed a decrease in germination. Species which showed increased germination were: Standard crested wheatgrass, Russian wildrye, Strawberry clover, Western wheatgrass, Broadleaf birdsfoot trefoil, and Ruby Valley milkvetch.

Species which were not affected were: Fairway crested wheatgrass, Tall wheatgrass, Intermediate wheatgrass, Pubescent wheatgrass, Lincoln smooth bromegrass, Timothy, Ranier red fescue, Whitmar beardless wheatgrass, Ladino clover, and Mandan wildrye. The remainder all showed decrease in germation. It was concluded that subsequent seed viability may be influenced when seed are exposed to conditions where moisture was not present in sufficient quantity to complete the germination phase.

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ABSTRACT

A study was made to investigate the ability of 22 grasses and 11 legume species to germinate against a moisture stress in Manhattan fine sandy loam. The soil was adjusted to various moisture contents corresponding to 5, 9, 10-2/3, 12-1/2 and 15 atmospheres. A check was conducted at field capacity.

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INTRODUCTION

Water plays an important role in field crop production. When moisture conditions are considered in connection with stand establishment, two factors pertaining to the growth process become important; one, germination, and two, subsequent growth.

When a plant emerges, it encounters the above- and below- ground "climate", and the success this plant will have is dependent on many variables such as light, temperature, wind velocity, topography, etc. Most of these factors have been studied and knowledge has advanced to a stage where their effects can be predicted with relative accuracy.

However, less is known about that phase of plant growth which is termed "germination" and factors affecting the phase when a seed is in the soil. It has been found that studies of "What happens to a seed in the soil?" are difficult, even though considerable is known about germination of seed "out of the soil". The soil is a dynamic system which responds to mechanical, physical, and chemical treatments. It is common knowledge that stands can be established by one farmer but not by his neighbor, even when moisture received by their land is equal and management practices are comparable. Salts occur in soils and these salts may exert an effect upon germination or stand establishment. Species and varieties respond in different fashion to factors which limit their germination as is evidenced by grass nurseries planted throughout the west by agricultural experiment stations. Other causes of differential stand establishment include factors of seedbed preparation such as working up land when it is either too wet or too dry, and planting varieties and species which, due to their physiological requirements, have a chance to fail or succeed at critical ranges of conditions.

Another factor which affects germination is temperature. Here again, stand establishment is jeopardized. It would go beyond the scope of this paper to go into detail on this particular point. It will suffice to mention that varietal response varies with temperature. The use and breeding of species which are best adapted to the conditions under which they are to be exposed during the early phase of their life cycle will result in a reduction of failure in establishing stands.

The results of research concerning germination of forage crop seeds in soil at different moisture stresses are given in this paper. It has been the objective to arrive at a measure by which a certain group of species can be separated according to their ability to germinate under limiting soil moisture conditions.

As will be seen from the Literature Review, a considerable amount of research work has been conducted with crops other than forages before this study was undertaken. The bulk of this previous work employed techniques which did not take into account the effect of soil moisture tension which develops in a soil. Soil moisture tension is only one of the two parts which make up the total soil moisture stress. The other part is the osmotic force which is due to the dissolved materials in the soil solution. It has been the latter effect which in the past has been studied and has been published under the general titles of "osmotic pressures" and their effect on seed germination.

LITERATURE REVIEW

Uhvits (12) studied the germination of alfalfa under osmotic stress using lots of 50 seeds. She compared germination using NaCl and Mannitol solutions of equal osmotic pressures to saturate the substrate which consisted of filter paper or sand. She found that the higher the osmotic concentration of the solutions, the lower the percent germination.

There were no differences between results obtained from tap water (check) and solutions with an osmotic pressure of one atmosphere. However, at equal osmotic pressures, the NaCl depressed the germination more than Mannitol. This suggested a toxic effect of the NaCl ions.

Because of the possibility that the initial water is taken up by imbibition, e.g., not conditioned by activity of live tissue, an experiment (12) was carried out with dead seeds. These seeds were killed at 130° C with either infrared heat for thirty minutes or oven heat for one hour. These seeds were given the same treatment as the viable seeds and tested at all concentrations of NaCl and Mannitol. Seeds would swell, and splitting of seeds was often observed. The absorption values were uniform for all concentrations of NaCl. On Mannitol, absorption varied more than on NaCl. The conclusion drawn from this was that imbibition may be important in the first stages of seed absorption and that it is the most important factor in absorption from NaCl solutions of high concentrations. The latter was supported by an experiment in which the recovery of seeds was measured after they had been on substrates of 12 and 15 atmospheres of osmotic tension. The seeds were transferred to plain water substrates, and the result was that recovery was greater after Mannitol than after

NaCl treatment.

At 15 atmospheres, recovery was particularly striking, although the percent of deformed seedlings was smaller when lower tensions were used.

Owen (9) of the Rothamsted Experiment Station studied the relation of a moisture potential to the germination of wheat. He carried out an experiment in which water was supplied to wheat seeds in the vapor phase.

An elaborate system was used by which the temperature could be controlled within .001° C. This gave precise humidity control equivalent to about one-tenth atmosphere of tension. The seeds were suspended in silver wire cradles and were not in actual contact with the solutions used. The water potential was controlled by use of solutions of sodium chloride. These solutions affected the vapor pressure, which was expressed in meters of water. The reason for using this method was that the toxicity factors would be excluded. This may be compared to Uhvit's work, which could be criticized because of the chances the seeds had to absorb the solute as well as the water.

Absorption of solute was studied by Ayers and Hayward (1). They used a fine sandy loam and adjusted it to various degrees of salinity by use of NaCl. The species tested were alfalfa, sugar beets, barley, corn, and kidney beans. Their conclusion was that salinity affects germination in two ways; one, by decreasing the rate of water entry with a corresponding decrease of ease with which seeds will take up water, and two, by facilitating the intake of ions in amounts which become toxic. They also concluded that salt concentration has a different effect on seed germination and seedling growth.

Owen's results with wheat indicated that water potentials corresponding to the permanent wilting point did not greatly reduce germination of wheat. An interesting finding was the fact that there seemed to be an apparent deterioration in seed viability. He found indications which warranted the conclusion that, when seeds were subjected for some time to conditions where germination is slow, the germinating capacity of the seed was reduced. Danielsen and Russell (3) studied the influence of moisture and aeration on the ion uptake by corn roots. Osmotic solutions and soil of known moisture tension were used. Their results indicated that osmotic pressures of solutions remained fairly uniform. However, the ability of the soil to conduct moisture to the place of utilization was not found to be comparable to the solutions. They indicated that significant moisture stress gradients occur in the soil which surrounds plant roots. It could be concluded that their results indicate that when seeds germinate, similar stress gradients would operate.

Davis (4) investigated the comparative salt tolerance of seeds of grass species during germination. He used two substrates, soil and filter paper. To these he added NaCl solutions of increasing concentrations. Eleven species were tested, and the order of salt tolerance at the highest concentrations was found to be: Tall wheatgrass, Russian wildrye, Standard crested wheatgrass, Intermediate wheatgrass, Fairway crested wheatgrass, Smooth bromegrass (Lincoln), Western wheatgrass, Meadow fescue, Alta fescue, Orchardgrass, and Timothy. He also found that the rank species obtain at a certain concentration, will not be identical when a different concentration is used. In his discussion of the problem, he mentions that, in general, Standard crested wheatgrass appeared to be more tolerant to salt than the other species. Western wheatgrass exhibited a marked susceptibility to salt injury during the germination period.

Harris and Tibbman (7) found that, in the presence of soluble salts, a considerable lengthening of the germination period occurred. Also, out of 70 different species of plants, Kentucky bluegrass seemed to be the least tolerant, followed by Orchardgrass, Timothy, and Meadow fescue.

Crocker and Barton (2), in their review of the physiology of seeds, mention many factors which influence the germination of seeds. The coverage they give the present knowledge on seed germination can be used as a guide in subsequent work which pertains to seed germination and will help the experimenter in recognizing the many variables which affect his data.

EXPERIMENTAL PROCEDURE

After 10 Montana soils were evaluated, surface soil of Manhattan fine sandy loam was chosen as the medium in which the germination of 33 species of forage crop seeds were to be tested. This soil belongs to the Chestnut great soil group. It is uniform in texture and high in very fine sand. There is very little evidence of aggregation in the surface soil. This soil is easily handled, has a pH of 7.8, and exhibits a relatively low electrical conductivity (.35 x 10^{-3} at saturation).

A moisture tension curve (figure 1) was prepared by use of the pressure membrance according to the procedures of Richards (10 and 11). All measurements were obtained on soil which was screened through a 2mm sieve.

From the moisture tension curve, the soil moisture content of soil samples to be used during the experiment was determined. This soil was stored in a large can at a moisture content of approximately 12 percent.

Various methods were tried in attempting to obtain soil of a uniform, desired moisture content for use in the germination tests; these included use of the pressure membrane apparatus, mixing of snow with a screened, cold soil of known moisture content, and others. Various techniques for obtaining uniform compaction, germination without condensation, minimizing evaporation, and soil moisture determinations were tried. The generally satisfactory method developed is described in detail.

Sufficient soil was removed to conduct a test and dried at room temperature in a closed room to the desired moisture content. The soil was spread one and one-half inches thick on brown wrapping paper. At short intervals the soil was "rolled over" by folding the paper to

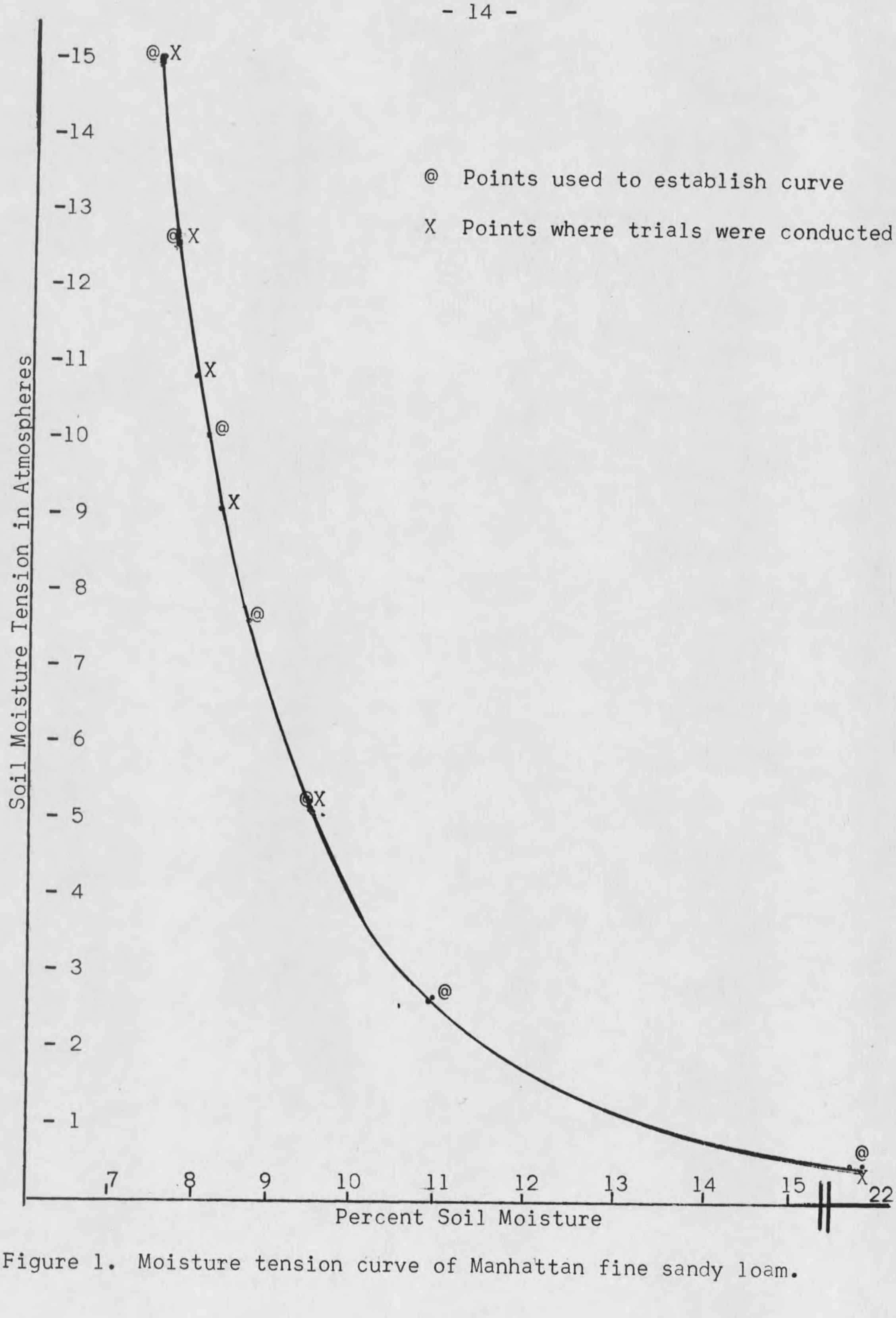

Figure 1. Moisture tension curve of Manhattan fine sandy loam.

facilitate drying. After the moisture content approached a desired level, it was stored in an air-tight can (milk can). This can was shaken and rolled violently, after which a soil sample was taken. When it was found that more drying was needed, the soil was spread out again and dried.

When the desired moisture content was obtained, the soil was passed through a "Bourner divider", and equal portions placed in glass dishes. Subsequent handling of the soil was reduced to a minimum of time to reduce loss of moisture while exposed to the room temperature. After each division, the soil was placed in air-tight cans and when placed in the glass dishes "Saran-Wrap" was used to cover the dish. The dishes used were "fire-King" casserole dishes and had the following dimensions: 12" x 7-1/2" x 1-3/4". All materials used were kept in the same room to diminish vapor flow between, and condensation on, surfaces of different temperatures. The temperature of the room was kept at about 68° F.

To test three lots of 100 seeds per species of the 33 species used, (see page 16) 25 glass dishes were employed per trial. Of these 25 dishes, 24 received four rows each of 100 seeds and the last dish three rows, so that each specie was represented by three lots of 100 seeds. The rows were randomly distributed. Approximately 700 grams of soil were used per dish.

A marker and planter was constructed so that the seeds could be placed in rows at an even depth and of uniform row spacing. After the seeds were planted, they were covered and a nest of weights was placed on the soil in order to compact the soil uniformly. A sheet of "Saran-Wrap" was used to cover the dish. The wrap was held down by a rubber

List of Species Tested

Agropyron cristatum	Fairway crested wheatgrass
Agropyron desertorum	Nordan crested wheatgrass
Agropyron desertorum	Standard crested wheatgrass
Agropyron elongatum	Tall wheatgrass
Agropyron inerme	Beardless wheatgrass
Agropyron intermedium	Intermediate wheatgrass
Agropyron smithii	Western wheatgrass
Agropyron trachycaulum	Slender wheatgrass
Agropyron trichophorum	Pubescent wheatgrass
Alopecurus pratensis	Meadow foxtail
Arrhenatherum elatius	Tualatin oatgrass
Astragalus cicer	Cicer milkvetch
Astragalus falcatus	Sickle milkvetch
Astragalus rubyi	Ruby Valley milkvetch
Bromus inermis	Lincoln smooth bromegrass
Bromus marginatus	Mountain bromegrass
Dactylis glomerata	Potomac orchardgrass
Elymus canadensis	Mandan wildrye
Elymus junceus	Russian wildrye
Festuca arundinaceae	Alta fescue
Festuca rubra	Ranier red fescue
Lotus corniculatus, arvensis	Broadleaf birdsfoot trefoil
Lotus corniculatus, teniufolius	New York narrowleaf birdsfoot trefoil
Medicago sativa	Ladak alfalfa
Melilotus alba	Spanish white sweetclover
Panicum virgatum	Switchgrass
Phleum pratense	Timothy
Poa ampla	Sherman big bluegrass
Poa pratensis	Kentucky bluegrass
Trifolium fragiferum	Strawberry clover
Trifolium hybridium	Alsike clover
Trifolium pratense	Kenland red clover
Trifolium repens	Ladino clover

band. This type of container and covering provided ample oxygen for germination. Then the weight of dish, soil, and seed was recorded for soil moisture determination.

After preparation, each dish was placed in a germinating cabinet constructed from a discarded refrigerator. The germinator was in the same room where all other manipulations occurred, and the temperature was held at 68° F plus or minus one degree. This again helped to reduce vapor movement in the soil. The relative humidity in the cabinet was held at a high level by placing a pan of water at the bottom of the cabinet. Condensation was not observed to occur.

After 14 days, the dishes were removed from the cabinet, the dishes were weighed, the "Saran-Wrap" was removed, and the number of seedlings which had emerged were counted. Sufficient water was added to the soil to bring it to approximate field capacity. The dishes were replaced in the cabinet without the "Saran-Wrap". The "Saran-Wrap" was omitted to reduce damping off and the development of fungal growth at this higher moisture level.

After the dishes were left in the cabinet for another week, a final count was made. The dishes were than placed in an oven, and oven-dry weights were recorded. From this data, soil moisture percentages were calculated.

Five moisture levels were tested according to these procedures. A check was conducted on soil which had 22 percent moisture, which was considered as field capacity. The dishes were left three weeks in order to be comparable to the three weeks' total at which the other moisture levels were conducted. The number of seedlings which emerged after three weeks' field capacity was used as the basis to which all calculations were adjusted, this basis being 100 percent for any given specie.

Moisture levels, corresponding to 5, 9, 10-2/3, 12-1/2, and 15 atmospheres tension were used in this study. The first test was conducted at a moisture level equal to 5 atmospheres tension. The soil used was prepared in the pressure membrane. Since it required a long time to prepare enough soil to test 300 seeds per specie, a start was made with 200 seeds. After another amount of soil became available, the other 100 seeds were tested.

Since only small amounts of soil could be prepared at a time, the soil was taken out of the membrane after it had attained equilibrium and stored in a milk can until enough soil was prepared to run a test. Since this was an elaborate and time-consuming practice, it was abandoned in favor of the drying method. However, at 5 atmospheres tension, the test was run in two parts on soil which had been stored in the milk cans for two different time periods.

All the data obtained were statistically treated (8). In order to obtain a grouping for the species tested, the shortest significant range method (5) was used as a guide for grouping the species for ability to germinate against soil moisture tension. Each test was analyzed separately but all tests considered in arriving at the groupings. At the higher moisture tensions, the species which did not germinate were omitted from the analysis.

The effect of holding seeds at high soil moisture tensions on subsequent germination was also determined. The number of seeds germinating at the end of three weeks, where during the first two weeks the seed was held under a moisture stress, was divided by the number of seeds which germinated after three weeks of field capacity. The data obtained from these calculations were analyzed by a complex analysis of variance. The moisture stress by species interaction least significant difference at the 5 percent level was used to test the hypothesis that a given species at a given moisture stress was significantly different from 100 percent.

RESULTS

Twenty-two grass species and 11 legume species were tested for their ability to germinate under limited moisture conditions.

A considerable variation was observed between species (Table I) and their ability to germinate when moisture becomes limiting. Table I also shows that between field capacity and 5 atmospheres tension germination, in general, was satisfactory. It was found that the species studied could be placed in six groups (Table II). This grouping is based on the over-all consideration of the five moisture levels. In general, the species are listed in order of decreasing tolerance in each group, and the most tolerant group first. It was observed that a specie does not necessarily hold its identical place as moisture becomes more limiting but may change place with other species within or between adjacent groups. Further tests would be necessary to place all the species precisely in a table of this type.

At the highest tension the order of decreasing ability to germinate against tension was found to be: Fairway crested wheatgrass, Intermediate wheatgrass, Standard crested wheatgrass, Nordan crested wheatgrass, Russian wildrye, Tall wheatgrass, Lincoln smooth bromegrass, Pubescent wheatgrass, Slender wheatgrass, Tualatin oatgrass, Alta fescue, Switch-grass, and Meadow foxtail. Other species did not germinate.

In Table III data is presented indicating that significant species differences were obtained in each test. It should be noted that the data of Table I for the five moisture levels is freed of the differences in germinating capacity of the seed used.

The grasses and the legumes exhibited a different germination behavior. Grasses, when not emerging at higher moisture tensions, did not seem to affect the surrounding soil, whereas legumes exhibited the ability to crack the soil due to swelling or partial germination. As a result of this, they became exposed to the atmosphere. The escape of moisture from the soil due to this cracking could not be measured. However, at the higher tensions this was associated with decrease in germination.

It should be mentioned here that seed size could play an important part. It was noted that the milkvetches seemed to exhibit this characteristic to a greater degree than the other legumes.

The grasses in some cases produced faint cracking in the soil over the row. In this case, the cracking was not found to be consistent for specific species, and not to be pronounced enough to be of consequence.

Fairway crested wheatgrass appeared to have more turgid seedlings than other species when germinating under stress. Grass seedlings did not appear to be attacked by any fungus, nor did they exhibit tendencies toward damping off. Mandan wildrye was an exception, in that it appeared to be associated with the development of a particular mold. It was observed that in each trial the soil over a row of Mandan wildrye became infected with this mold. It was concluded that for these trials, Mandan wildrye may not have given a representative response. Further tests would be necessary to verify the results with Mandan wildrye.

At the end of each test, e.g., after three weeks, which included one week at field capacity, final counts were made. The values obtained are expressed in percent of the check (Table IV).

From these values it was observed that, when higher tensions plus succeeding field capacity conditions were imposed on the seeds, some species showed increased germination, others exhibited decreased germination. When the five trials were analyzed, a highly significant interaction was obtained between moisture levels and species (Table V). By use of the interaction least significant difference, three groupings were established (Table VI). Division one contains species which were significantly higher in germination than 100 percent after having been exposed to limited moisture conditions. The second division contains those species which were not significantly affected. The third division contains species which exhibited a decrease in germination.

Table I. Percent germination at 5, 9, 10-2/3, 12-1/2, and 15 atmospheres soil moisture tension expressed as percent of check trial conducted at field capacity.

List of Species	Atmospheres Tension	14-Day Observation 5 Ave.	9 Ave.	10.67 Ave.	12.5 Ave.	15 Ave.	Check .33 100 % Basis
		Percent					
Crested wheatgrass (F)		100	103	96	79	22	91
Crested wheatgrass (N)		89	89	88	49	9	86
Crested wheatgrass (S)		109	108	97	48	11	78
Tall wheatgrass		97	90	86	45	7	90
Intermediate wheatgrass		94	94	85	45	14	72
Russian wildrye		104	89	81	26	8	83
Pubescent wheatgrass		101	101	64	26	4	82
Lincoln smooth bromegrass		92	100	78	26	5	84
Meadow foxtail		82	92	79	22	0	73
Slender wheatgrass		78	94	58	19	2	88
Tualatin oatgrass		74	84	57	15	1	89
Sherman big bluegrass		85	88	76	11	0	52
Hopkins timothy		100	98	68	9	0	90
Ranier red fescue		75	85	58	6	0	88
Mountain brome		93	89	43	0	2	87
Alta fescue		48	54	24	5	1	88
Strawberry clover		91	73	40	1	0	56
Western wheatgrass		61	52	32	1	0	64
Broadleaf birdsfoot		81	70	27	1	0	67
Ladak alfalfa		77	56	32	1	0	65
White sweetclover		70	83	22	2	0	58
Alsike clover		69	63	2	0	0	78
Beardless wheatgrass		41	31	12	1	0	75
Switchgrass		56	50	15	1	1	56
Ladino clover		77	48	1	0	0	66
Kenland red clover		67	48	8	0	0	66
Ruby Valley milkvetch		57	48	17	0	0	29
Kentucky bluegrass		22	24	16	0	0	76
Sickle milkvetch		20	12	34	0	0	49
Narrowleaf birdsfoot		50	27	6	0	0	56
Cicer milkvetch		27	1	3	0	0	54
Mandan wildrye		14	8	3	0	0	39
Potomac orchardgrass		21	14	1	0	0	78
Standard Error of Mean		9.4	4.7	7.4	4.8	2.4	4.5
% Soil Moisture		9.3	8.5	8.2	7.9	7.6	22