

I EXECUTIVE SUMMARY

IMPROVEMENT OF *Poa annua* FOR GOLF TURF #8, 1991 UNIVERSITY OF MINNESOTA

The seed production field established in 1990 at Pickseed West in Tangent, Oregon was harvested in July of 1991. This seed was allocated so that a breeder's seed planting in Oregon and seeded golf course management plantings in Minnesota, at the San Diego CC, and on a golf course near Boston might be established. The Minnesota golf course management plantings were seeded in 5 X 10 foot plots on September 16. The breeder's seed planting was seeded on October 12. This included only the 5 best *Poa annua* selections. Seed from this planting will be harvested and used in 1992 to increase the breeder's seed supply and for more widespread evaluation of the selections. Emphasis is being placed on *Poa annuas* #42 (3A), #117 (10C), #184 (16B), #208 (18D), and #234 (21) for further evaluation, naming & introduction. 150 collections from 23 locations were added to the program. Selections #117 (10C) & #208 (18D) received the highest laboratory cold tolerance ratings while all 5 performed well over winter in the field. Several new selections and F1 hybrids continue to exhibit qualities that identify them for the next cycle of introductions. 2 selections exhibit truly exceptional qualities of color, density, and ruggedness. The ES-ME (excised stem - mist emasculation) technique has been fully employed in the planned crossing program and has enabled completion of many very difficult crosses this year. Seedling plants show more spreading vigor than vegetatively established plants. When progeny from the same parents were evaluated, stolons spread, on average, 1/3 more on first-year plants compared to those on vegetatively re-established second-year plants. This explains some of the success *P. annua* has in becoming established in opportunistic situations. Over 2000 offspring were evaluated. 50 different crosses were performed to develop F₁ hybrids. Several superior hybrids were created that are uniform between plants. Several F₂ hybrids also show promise with improved vigor, density, color, texture, uniformity. Selections #184 (16B) & #117 (10C) are superior parents for use in the production of desirable F1 hybrids. Interspecific crosses with *P. annua* (2n=28) & *P. supina* (2n=14) resulted, surprisingly, in fertile and promising progeny. Three distinct flowering types, day neutral, cold + short day (spring), and short day were identified. Some crosses segregated to a 3:1 ratio of seasonal to day neutral habit which may indicate a simple single gene, 2 allele system of inheritance for spring flowering types. This information will be extremely valuable for future breeding efforts. 14 year old *P. annua* seed germinated at 50%. This gives new dimension to the current poa seed bank in the soil. Cytogenetic analysis of our selections has substantiated that *P. supina* carries 14 chromosomes & *P. annua* normally carries 28. However, 4 of our poas, all of which are dense, dwarfish, dark green, desirable plant types have only 14 chromosomes (1/2 the expected number). This could open the door to much easier inheritance research with *Poa annua* & *Poa supina*. The project is on schedule for evaluation, seed production and introduction of 5 of the original 8 superior lines in the 1996-97 time frame. It is also on track for evaluation of 2nd cycle materials exhibiting superior qualities.

II INTRODUCTION

The following is a report of the research conducted under the project: "Improvement of *Poa annua* for Golf Turf" during the 1990 -1991 year. The activities pursued during the year are summarized and are offered in outline form. More detail is available upon request.

III EVALUATIONS

A. NEW COLLECTIONS

Approximately 150 new collections from 23 locations were integrated into the germ plasm pool over the year. We continued, in 1991, to shift and increase emphasis toward seed production along with evaluation of the initial 8 selections identified for possible introduction as varieties. Some of these materials have also been selected for use as parents.

B. INITIAL SEED PRODUCTION TRIAL (1990)

A seed production field trial was planted on 9 and 10 November 1990, near Tangent, Oregon, under Pickseed West's supervision. The planting consisted of 500 rooted stolons of each of the 8 clones (4,000 total) under consideration for introduction. Seed was harvested on June 28 and 29, 1991 and dried for processing at Pickseed. The following table summarizes the total yield for each of the selections:

<u>Selection Name</u>	<u>Selection Number</u>	<u>Yield (g)</u>
3A	42	345.65
10C	117	188.56
16B	184	329.43
18D	208	124.2
21	234	767.15
29F	391	15.16
55D	417	6.62
NY12	493	216.43

Additional seed was collected from a much smaller seed trial in Minnesota resulting in similar yield patterns.

Seed germination tests indicated that dormancy was an issue in some of the selections, particularly in 10C and 16B. However preliminary experiments indicated that the dormancy was mitigated by a cold treatment. In addition, 200 ppm gibberellic acid was not beneficial.

C. BREEDER'S SEED TRIAL (1991)

Seed (above) harvested this summer from the November 1990 planting (see figs 15 & 16) was allocated 1) to establishing a breeder's seed planting at Pickseed West and 2) to establishing seeded plots for evaluation under golf turf conditions.

A breeder's seed production trial from seed was planted at Pickseed West on October 12, 1991. Seeded plots varied in size according to the amount of available seed. Seed was sown in 100' rows, with rows 6" apart. 16 rows of #234 (21); #117 (10C); and #208 (18D) were planted while 32 rows of #42 (3A) and #184 (16B) were planted. In addition crossing blocks of #42 (3A) x 16B; 3A x 10C; 18D x 10C; and 18D x 16B were also seeded at the same time. We anticipate harvesting breeder's seed from this planting in July of 1992. It is anticipated that one more cycle of seed trials will be needed to produce enough seed for management trials and for establishing foundation seed plantings.

D. SEEDED GOLF COURSE PLOTS

Seed was also allocated to establish seeded plantings that are to be maintained under golf course conditions. Each of the *Poa annua* selections was seeded in 5 by 10 foot plots on the University of Minnesota research area on September 16th. To minimize the possibility of contamination from background poa, the surface layer of soil was steam-sterilized just prior to seeding. Initial germination was observed on September 23rd. See fig. 12.

In addition, arrangements have been made to establish evaluation plantings at the San Diego Country Club (California) in March of 1992. This is the preferred time according to Mr. Gary Dalton, the Superintendent. It is anticipated that another set of plots will be established in the Boston, Massachusetts area early in the Spring of 1992.

E. VEGETATIVELY ESTABLISHED PLOTS

Preliminary observations indicate that plantings of the selections established from seed are more vigorous and productive than plantings established from sod. This includes observations of check plots of bent and poa established as sod. In all cases the turf appeared to be penalized by the process. It may be related to the need for more attention during the establishment period than is currently applied or just the nature of severe root pruning. Symptoms include slower growth and somewhat dwarfed plants for periods that extend far beyond the normal establishment period.

Differences in growth habit of seed propagated versus vegetatively propagated (by stolons) plants were also observed in the performance of selections in the spaced plantings. Plugs grown directly from seed spread far more vigorously than those propagated vegetatively. A rough statistical comparison indicated that the vegetatively established plugs grew at only 2/3 the growth rate of the seed propagated plugs when measured several weeks after transplanting to the field.

A comparison of the performance of progeny resulting from a cross of #117 (10C) with #1832 illustrated this point. These plants grew with exceptional spreading vigor as seedling-plugs in 1990. In 1991, vegetatively derived plugs of these same selections failed to show the same spreading vigor. Yet, newly propagated seedling-plugs from the same cross once again exhibited this strong spreading characteristic. (see fig 17 & 18) It is important to note that, while not strongly spreading, the vegetatively propagated plugs appear healthy and otherwise vigorous.

This fits with the ecological concept of competition for space demonstrated by opportunistic species such as *Poa annua*. Seedling vigor, ie. rapid establishment, favors the species. Later, growth patterns related to consolidation and reproduction become more important.

F. COLD TOLERANCE

We continued to develop and refine our bimodal programmable freezer block (BPFB) technology. Using this equipment, the eight selections currently under a consideration for introduction were evaluated.

Distinct differences in cold tolerance between the selections were observed throughout the winter months. *Poa annuas* #117 (10C) an #234 (PA21) and both *Poa supinas* #391 (29F) and #417 (55D)) were clearly the most cold tolerant. Subjective ratings for the verdure and root recovery were found to correlate well with dry weight measurements of the new root and verdure growth produced. Selection #184 (16B) appeared to be the least cold tolerant while the other poa selections fell into an intermediate level.

All were hardy to the levels normally experienced in the field and none of the materials were damaged in either the mowed or spaced plantings in the field. In addition to the strong laboratory cold tolerance, #117 had exceptional, dark green fall color in the field.

By April, all had started to de-acclimate, however #417 was slower to de-acclimate than the other materials. It was also the first one to initiate growth in the Spring.

G. LONG TERM VIABILITY OF *Poa annua* SEED

A preliminary investigation into the longevity of seed viability was conducted. There is a two fold purpose to this effort: 1) to investigate long term seed viability; and 2) to incorporate diverse, possibly older, germplasm into the program, increasing the genetic base in the program in another way.

Fifteen seed samples were collected from the University of Minnesota Herbarium collection of *Poa annuas*. Samples ranged in age from 7 to 68 years. The seeds were first surface sterilized in dilute sodium hypochlorite. Seed were then placed in Murishige and Skoog (MS) agar medium in petri dishes and held at room temperature. A control of 2 year old seed was used.

The control seed germinated at 90%. Fifty percent of the fourteen year old seed germinated. No germination was observed with seed 25 to 68 years old. However, it was learned that, prior to 25 years ago the whole herbarium was fumigated which could account for these results.

IV BREEDING AND GENETICS

A. EXCISED STEM - MIST EMASCULATION BREEDING TECHNIQUE (ES-ME)

Work on the adaptation and application of a fog (mist) technique (after Burton & others) to enable control of pollen shed and easy emasculation has been completed. The technique results in the expression of the anthers so that they are accessible to emasculation, while at the same time inhibiting pollen shed. ES-ME has solved many of the problems and impediments we have been facing in trying to accomplish controlled pollinations with both *Poa annua* and *Poa supina*. We were also able to gain valuable evidence of carbohydrate partitioning within the excised flowering stems. For more detail on this see our interim report dated May 1991.

B. CROSSING PROGRAM

The ES-ME technique has enabled the planned crossing program to proceed much more efficiently. Efforts continue to combine characteristics such as dark green color, disease resistance, compact habit of growth, fine texture, vigor and other desirable traits found in our selections. Over 2,000 offspring are under field evaluation for improved characteristics. In addition, valuable information is expected on the amount of variation that is expressed in the F₂ generation of certain selections that appear to be naturally selfing. It has also enabled the evaluation of developing F₁ hybrid type varieties that are superior in vigor or some other characteristic.

Approximately 50 different crosses resulting in hybrid F₁ progeny are under evaluation. Twenty different groups of plants in the F₂ generation are also being evaluated. A backcrossing program was initiated to emphasize and stabilize desirable characteristics.

Excellent seedling vigor was observed in almost all of the hybrid seedlings. Many of the plants started from seed in May reached diameters in excess of 12" by September. The extremely prolific growth habit of the most vigorous types may be an exhibition of hybrid vigor. It is certainly a characteristic that needs to be pursued in developing future varieties.

C. F₁ HYBRID PROGENY

F₁ plants, for the most part, are quite uniform. This would be expected in the F₁ generation of highly selfing materials. Examples of this uniformity include seedlings from 42 x 184 (fig 2) and 184 x 117 (fig. 3). This uniformity in the F₁ from these crosses supports the indication that many of the genotypes we are working with are homozygous for many alleles. It is consistent with isozyme work by Darmency and Gasquez (1981)¹ that estimated that there was at most 15% outcrossing occurring in *Poa annua*. However, previous work on this project found both highly selfed and highly outcrossing genotypes. Data from this year supports those findings; indicating that every cross must be approached and planned on an individual basis.

¹ Darmency, H. and J. Gasquez, 1981; Inheritance of triazine resistance in *Poa annua* : Consequences for population dynamics; New Phytol. 89 487-493.

Some of the F₁ crosses exhibiting substantial variation in progeny include 661 x 184 (Fig. 4) and 42 x 1434 (Fig. 5).

Of the F₁s observed this year, 42 x 184, 42 x 493, 184 x 117, and 117 x 1832 show great promise as potential hybrid varieties. It is anticipated that each cross would be developed by each selection being planted in every other row in the field. We will continue to follow seed production, and observation of these materials in field generated crosses.

D. F₂ HYBRID PROGENY

F₂ plant materials resulting from crosses of some of the highly selfed materials exhibited substantial uniformity of type indicating that these materials may lend themselves to further development for introduction. All of these materials exhibited seasonal (Spring) flowering and excellent vigor under space planting conditions. The best of these materials were propagated vegetatively and planted in seed block rows to obtain seed for further evaluation. Examples of these materials include #2386 (117 x 1832) x Self (fig 6).

Some of the F₂ hybrid sets exhibited considerable variation, as expected. Where numbers allowed, ratios of the segregating characteristics are under study. In keeping with the modified pedigree breeding system, selection criteria for these materials include seasonal flowering, fine texture, dark color, and vigor. Seed will be harvested from these plants in the spring to evaluate seed potential. Examples of segregating materials include (184 x 661) x Self (fig 7).

E. BACKCROSSES

This is the first year that substantial backcrossing has been practiced. The primary objective is to stabilize and accentuate desirable characteristics found in progeny resulting from crosses involving advanced selections. The best materials resulted from 208 x 117 progeny backcrossed to 117. This backcross resulted in vigorous plants with desirable flowering characteristics (fig 8). Plans call for expansion of the backcrossing effort in the future.

F. CROSSING BLOCK

A small crossing block was established on the University of Minnesota research area using 4 of the advanced selections: 42, 117, 184, and 208 (fig 9). Seed will be harvested in the summer of '92 and progeny will be evaluated for uniformity and possible use as a synthetic variety. This is similar to the Oregon crossing blocks described earlier.

G. INTERSPECIFIC CROSSES

Work continues with interspecific hybrid crosses between *Poa annua* and *Poa supina* to ascertain if desirable characteristics and new types will be the result.

One very important finding from this research is that the progeny resulting from the interspecific cross are, unexpectedly, fertile. Reports in the literature note that the few hybrids between *Poa annua* and *Poa supina* that have been observed were triploid and sterile.

In addition, in most cases, the F₁ progeny of these crosses tend to flower throughout the growing year. Yet *Poa supina* is a seasonal flowering type and some of the *Poa annua* selections are also seasonal in flowering habit. However, the F₂ progeny segregate for this (flowering) characteristic. In one case [(PS79 x PA19F) x SELF] the ratio to seasonal is 20:1 while in another [(PS93 x PA19F) x SELF] the ratio is 2:1. These ratios are not common Mendelian segregation ratios and one would not expect that to be the case but the information is a valuable addition to our basic knowledge of flowering.

Some very desirable plant types have resulted from these crosses (figs 10 & 11).

There will be an increased emphasis on making interspecific crosses during the next cycle. Backcrossing, an integral part of such breeding, will be included.

H. OBSERVATIONS ON FLOWERING

Observations on flowering over the last 2 years have defined 3 distinct flowering habits in *P. annua*. The 3 types include 1) day neutral types which tend to flower throughout the year; 2) short day plus cold requiring types; and 3) a short day type which flowers under short days with no cold treatment. Type 3 is intermediate in that cold, although not necessary, appears to enhance the flowering response. This discovery is extremely important to the project as well as the general turf and genetics communities.

Examination of flowering data over the last 2 years reveals that F₁ crosses between 234 and 117 or 184 all resulted in day neutral flowering types. Crosses between 117 and 184 resulted in seasonal flowering types, with 1 exception. In the F₂ however, the progeny from crosses of 234 x 117 and 234 x 184 and 184 x 234 resulted in a distinct 3:1 ratio, day neutral to seasonal. If we apply Mendelian genetics this is the ratio expected from a single gene, 2 allele system with complete dominance (day neutral = dominant in this case). It is unlikely that it could be this simple for all genotypes, but this is truly an exciting place to start. Murfet (1977)² reported a simply inherited, completely dominant system in several grasses. We will continue work on this and apply the information in the continuing breeding program. A preliminary experiment is currently being conducted to help ascertain cold requirements for flower induction. Other experiments are in the planning stage for controlled environment

² Murfet, I. C., 1977; Environmental interaction and the genetics of flowering; Ann. Rev. Plant Physiol. 28: 253-278.

chambers. This kind of information will be extremely valuable in allowing us to bring materials into flowering at appropriate times for breeding purposes as well as selecting for seasonal flowering.

V ENVIRONMENTAL IMPACTS 1990-91

Snow cover came early, around Thanksgiving in 1990 after a relatively mild Fall. However prior to Christmas severe cold damaged substantial amounts of turf. Observations with the *Poa annua* selections were that the plots maintained at collar or green height were not damaged and tolerated this severe weather very well.

CYTOGENETICS - CHROMOSOME COUNTS

Chromosome analysis work continues with the advanced selections in preparation for registering them as varieties and to further the production of interspecific crosses. Observations to date generally support the expected; i.e., *Poa supina* materials contain 14 chromosomes and the *Poa annua* materials contain 28 chromosomes. This corresponds to the literature where $2n = 4x = 28$ chromosomes for *Poa annua*. The *Poa supina* selections have 14 chromosomes, which corresponds to the literature which report $2n = 2x = 14$.

Interestingly, three of our *Poa annua* selections have been found to have only 14 chromosomes. All three, #1955, #1976 and #1383 are dwarf types with very fine textured, dense growth habits. These are second cycle selections (under consideration for future introduction). See figs 13 & 14. These should be especially useful in investigating the value of developing inter-specific hybrids with *Poa supina* and providing basic genetic information about *Poa annua*. The existence of these diploids could have far reaching affects in the continual improvement of materials for golf turf.

Table of Chromosome Numbers

<u>Selection Name</u>	<u>Chromosome Number</u>	<u>Comments</u>
1955	14	Dwarf type collected in New Jersey
1959	14	Dwarf type collected in Minnesota
1976	14	Semi-dwarf type collected in Minnesota
1383	14	Dwarf type
42 (3A)	28	Advanced Selection
184 (16B)	28	Advanced Selection
117 (10C)	28	Advanced Selection
208 (18D)	28	Advanced Selection
1930	28	Curly-leaved type collected in Ohio
391 (29F)	14	<i>Poa supina</i>

PLANS FOR 1992

- o Establish seeded field plantings for management under golf course conditions at the San Diego Country Club and at a golf course near Boston, Massachusetts.
- o In June-July, harvest seed from the October 1991 seed production planting in Oregon. Test, analyze, and allocate for production of breeder's seed and establishment of golf course evaluation plantings.
- o In the fall, establish a new breeder's seed production planting in Oregon from the newly harvested seed in preparation for the follow-up planting for foundation seed more widespread golf course and university evaluation.
- o In the fall, seed golf course and university management plantings of the prime selections from the newly harvested seed.
- o Continue evaluation of superior selections in the program. This will include, not only the 5 *Poa annuas* slated for introduction, but also other superior selections in the program.
- o Continue to investigate the possibility of developing F₁ hybrid type varieties.
- o Continue to evaluate the feasibility of producing synthetic varieties from different groups of materials in the program.
- o Continue to investigate the potential from interspecific crosses of *Poa annua* and *Poa supina*. This will include the 14 chromosome *Poa annuas* that have been recently identified in the program.
- o Continue progeny testing - heritability studies with seeds generated for specific self, sib, and hybrid crosses that were accomplished this year.
- o Continue the selective breeding that focusses on seeding habit(s) that are related directly to harvesting seed.
- o Continue to investigate the flowering requirements and the inheritance of flowering habit. This work will eventually be directed toward the control of flowering in the field and on the golf course.
- o Continue to investigate and document the chromosome numbers of selections in the program in preparation for registering materials for varietal status and introduction.
- o Continue to evaluate cold tolerance and winter hardiness of the advanced selections in the program.

- o Continue the backcrossing program to accumulate and stabilize desirable characteristics in the selections.
- o Continue the collection program and incorporation of new germplasm into the program. Hopefully, we have located some help in obtaining *Poa supina* seed from the wild that will offer a broader genetic base for our program.
- o Efforts in 1992 will continue to focus on seed production and field evaluation of the 5 *Poa annuas* selected for introduction as varieties.

Respectfully submitted,

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Fig. 1. Overall view of 1991 space planting at the University of Minnesota Research field.



Fig. 2. 42 X 184 F₁ hybrid exhibiting uniformity of progeny. This hybrid shows great promise for introduction as a variety in the future.



Fig. 3. 184 X 117 F₁ hybrid which exhibits uniformity of type and promise for varietal status.



Fig. 4. 661 X 184 F_1 hybrid exhibiting variation in progeny. Note one very vigorous plant selected to remain in the program.



Fig. 5. 42 X 1434 F_1 hybrid exhibiting substantial vigor with variation in progeny types. Note some very nice plant types.



Fig. 6. F_2 progeny resulting from selfing 2209 (a hybrid between 117 and 1832) exhibiting excellent uniformity, vigor and other qualities.



Fig. 7. F_2 progeny resulting from selfing 2317 (a hybrid between 184 and 661) exhibiting segregation of types. Note the extremes in color and vigor.



Fig. 8. B_1 progeny resulting from a backcross of 2171 (a hybrid between 208 and 117) with 117) showing stability of types and vigor.



Fig. 9. Stolonized crossing block showing alternating rows of 42, 117, 184, and 208.



Fig. 10. Interspecific hybrid progeny of *Poa supina* #343 (Ps 79) and *Poa annua* 223 (Pa 19F). Note vigorous plant.



Fig. 11. Interspecific hybrid of #357 (Ps 93) and #223 (Pa 19F) showing variation in offspring and one plant exhibiting many excellent characteristics.



Fig. 12. Two of the 5 by 10 foot seeded golf course plots on the University of Minnesota campus. Picture was taken 13 days after seeding.

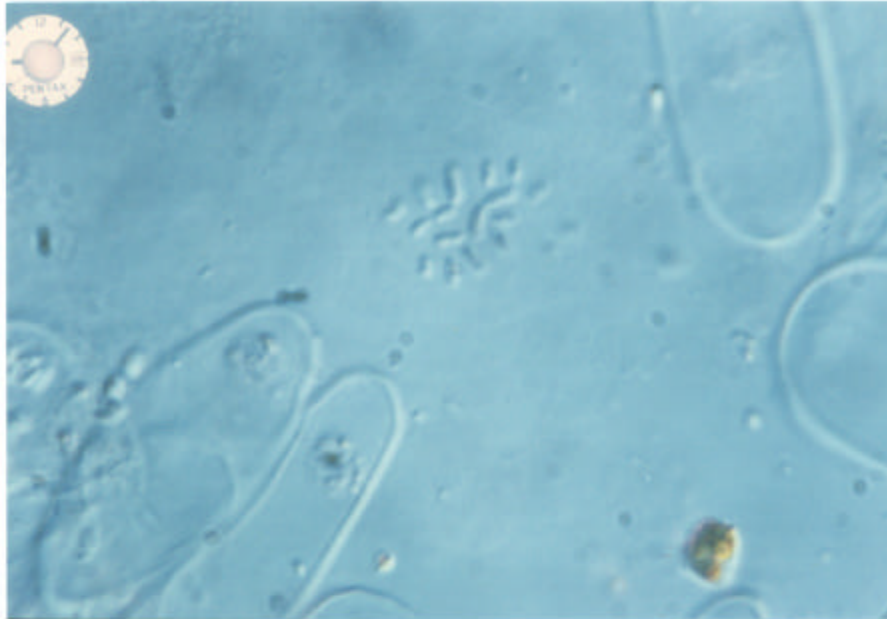


Fig. 13. Photomicrograph of #1976 root-tip cell showing 14 chromosomes.

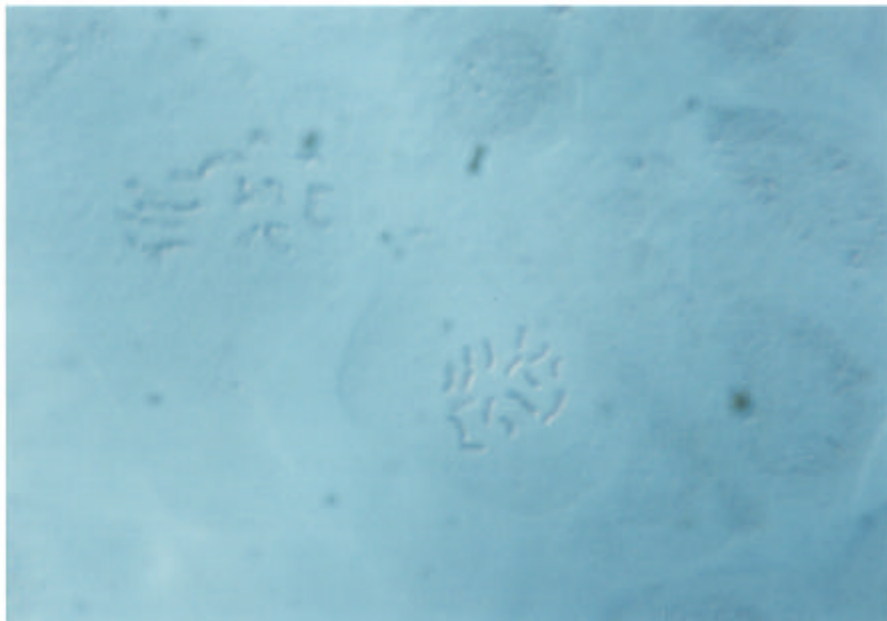


Fig. 14. Photomicrograph of #391 (Ps 29F) root-tip cell showing 14 chromosomes.



Fig. 15. Over-all view of 1990 seed production field at Pickseed West, Tangent Oregon.



Fig. 16. Close-up of #234 (Pa 21) at Pickseed. Pictures were taken at harvest, July 1991.



Fig. 17. 1990 progeny of #117 X #1832 re-established vegetatively in 1991 space planting.



Fig. 18. 1991 progeny of #117 X #1832 in 1991 space planting. Note difference in spreading vigor of the more recently seed-generated plants.



Fig. 19. Selection #42 (Pa 3A) at green height.



Fig. 20. Selection #117 (Pa 10C) at green height. Both pictures were taken on University of Minnesota campus plots.