I EXECUTIVE SUMMARY

IMPROVEMENT OF *Poa annua reptans* FOR GOLF TURF #9, 1992
UNIVERSITY OF MINNESOTA

The seed production field planting seeded in late October 1991 at Pickseed West in Tangent, Oregon was harvested in late June of 1992. Seed yields varied from almost 15 pounds for *Poa annua* #208 to a low of almost 5 pounds for #234. However, #234 was accidently subjected to an herbicide application that severely damaged the planting. Projected estimates of seed production potential varied from 246 pounds per acre for *Poa annua* #42 to 800 pounds per acre for *Poa annua* # 208. This seed was cleaned and will be allocated to selected golf course plantings and storage for use during the next growing season for seed production. Some of the seed was planted, in late October, in a new increase planting in Tangent, Oregon in preparation to accumulate enough seed for a breeder's seed planting in 1993. 315 New collections, including to foreign materials were introduced into the project during 1992. Several of the new materials carry stress resistance and a surprising number were found to be 14 chromosome types. Several male sterile plants were also identified during the evaluations. *Poa annua* #117, #184, #208, and PS#417 continue to exhibit cold tolerance. One new material that holds promise for a future introduction was found in the 3rd selfed generation of *Poa supina* # 56-S3. Another resulted from an interspecific cross between *Poa annua* and *Poa supina* (PS79XPA19F). Observations continue to confirm that seeded plantings perform better than sod established plantings. Seeded plantings grew at a rate 1/3 faster than sodded plantings. Seed germination tests indicated a dormancy problem in some of the prime selections, particularly #117 and #184. Preliminary research showed that moist vernalization at 4C for as little as 5 days can overcome the after ripening requirement in relatively new seed. Under normal seed storage conditions, the requirement was serviced after 7 months. Continuous flowering in *Poa annua* is a major problem on the golf course. Some off season flowering has been observed in some very desirable plants. Research with parental material, F1, F2, and F3 populations suggest that the flowering habit in *Poa annua* may be controlled by a single gene. Examination of segregating F2 and F3 progeny indicate a 3:1 ratio of continuous flowering to seasonal. Almost all of the perennial types in our breeding program exhibit a more seasonal flowering habit, flowering for a short time in the spring. We have found that most of the perennial materials possess the normal number of chromosomes(2N=28). However, a substantial proportion of our materials contain only the diploid number (2N=14). The 14 chromome plants are fine textured, dense, with dark green coloration. Flow cytometry research has been very successful and has enabled us to conduct the large number of chromosome evaluations that have allowed identification of the 14 chromosome materials. Golf course seeded plantings were established at the University of Minnesota turf research site; at the San Diego Country Club at Chula Vista, CA; and at "The Country Club" in Brookline, MA. Evaluations at both locations are showing differences, but all materials appear to be performing in excellent fashion. Efforts have been initiated to investigate appropriate steps to be followed for introduction of several of the prime selections as varieties by 1996 or 97.

II INTRODUCTION

The following is a report of the research conducted under the project: "Improvement of *Poa annua* for Golf Turf" during the 1991 -1992 year. The activities pursued during the year are summarized and are offered in outline form. More detail is available upon request.

We continued, in 1992, to shift and increase emphasis toward seed production along with evaluation of the 5 prime selections identified for possible introduction as varieties. Some of these materials have also been selected for use as parents.

III EVALUATIONS

A. NEW COLLECTIONS

Approximately 315 new collections, including some from Canadian and Afghanistan sources, were integrated into the germ plasm pool over the year. Some of the materials from Afghanistan exhibit a unique habit of growth with a rugged, rough appearance. The Afghanistan material was received from the USDA seed storage facility in Pullman, Washington.

Several of the golf course collections turned out to be examples of diploid *Poa annua* types (14 chromosomes verses the normal 28 in tetraploid *Poa annua*). These plants exhibit considerable variation in flowering habit, color, and vigor, however most are characterized by very dwarf growth habit and fine texture. For example, growth of four of these accessions in a turf where other poas and bentgrass was maintained at 1/2", never reached the mowing height . Leaf width measurements indicate the fine texture these exhibit. Cold tolerance appears to be reduced in these materials, but more evaluation is required.

Propagation of these diploid plants has been difficult due to the lack of seed production. Male sterility was observed in at least two accessions, and appears to be due to failure of normal pollen formation; the anthers appear empty. Evaluation of other diploid types is underway.

The origin of some of these diploid *Poa annua* is not known, however one type originated from one of our crosses of two tetraploid types. This poses many cytological questions that we hope to work on in the future.

B. SEEDED GOLF COURSE EVALUATIONS

Seed was also allocated to establish seeded plantings that are to be maintained under golf course conditions. Each of the five prime *Poa annua* selections was seeded in 5 by 10 foot plots on the University of Minnesota research area on September 16th 1991. 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 23 (Fig. 1, 2, 3).

Additional evaluation plantings were established at the San Diego Country Club (California), Mr. Gary Dalton, Superintendent. in late April of 1992 (Fig 4, 5, 6). Another planting was seeded at The Country Club, Brookline, Massachusetts, Mr. Bill Spence, Superintendent, on 18 June 1992.

Evaluations of all of the plantings indicated excellent performance and quality of most of the selections. At one location the seeding performed well even after being subjected to severe algae infestation and an accidental scalping.

Seasonal observations, in Minnesota, of the selections indicate that during the midsummer, 42,117, 184, and 234 performed very well, presenting excellent texture, color and general appearance. Color in the fall showed more differences. 184 presented the best color at this time along with fine texture and excellent density. 42, 208, and 234 ranked next and 117 turned off color to light green. This indicates the need for more thorough evaluation of 117 before releasing it. Otherwise, performance, to date, of all of the materials at all sites, indicates work toward introduction should continue as a very high priority.

New evaluation plantings of the five elite selections along with some of the second cycle materials were seeded for future evaluation under both collar and greens height of cut at the University of Minnesota turf field research area.

C. COLD TOLERANCE

Cold tolerance evaluations continue to indicate that selections #117, #184, #208, and PS #417 develop the greatest cold tolerance.

D. NEW SELECTIONS

Several new materials exhibit characteristics that may make them candidates for the next cycle of breeding. One is an S3 (third generation self of one of our poa supinas (PS56-S3). Another is the result of an interspecific cross between *Poa annua* and *Poa supina* , PS79XPA19F. Both of these materials present interesting opportunities and indicate the potential value of interspecific crosses to the project.

IV SEED AND SEEDLING OBSERVATIONS

Preliminary observations continue to indicate that plantings of the selections established from seed are more vigorous and productive than plantings established from sod. This seedling vigor may be one of the most important characteristics in the success of *poa*

annua at becoming established as an invader. However, wide genotypic differences have been observed within the population, ranging from dwarf to vigorous spreading types. A light seeding of 1.3 pounds per 1,000 square feet sown in late september resulted in dense turf by April in Minnesota of several of the selections

Variation was also noted in germination of many of the materials currently in the advanced selection phase of the project. germination tests of seed indicated that an after ripening treatment might be required to overcome seed dormancy in several of the selections. Our research indicated that several of the selections benefit greatly from a preplanting vernalization treatment. This was particularly true with #184, # 493, and #117. Dormancy was overcome by imbibing the seed for eight hours and subjecting it to 4 degrees C vernalization exposure for a minimum of 5 days (Fig 8 & 9). Otherwise, the dormancy or after ripening requirement was fulfilled after 7 months in storage. More research into this phenomena is indicated.

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 about 2/3 the rate of the seed propagated plugs when measured several weeks after transplanting to the field.

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

V SEED INCREASE

A. SEED INCREASE TRIAL (1991-92)

A seed increase field trial was planted, from seed produced in the initial increase last year (1991), near Tangent, Oregon, under Pickseed West's supervision. The planting consisted of separate blocks of 3A, 10C, 16B, 18D and #21, (Fig.18 & 19). These are the 5 selections that produced seed in sufficient quantities in the 1991 planting to warrant continuation of the evaluations leading to naming and introduction.

Seed was harvested in June of 1992, dried, and processed to the point where it required one more step in the process by Pickseed personnel. The seed was further processed through an air separator to remove extraneous material at the University of Minnesota.

The following table summarizes the total yield for each of the selections in 1992.

Table 1. 1992 Seed Yield Estimates from First Increase for Breeder's Seed at Pickseed USA, Tangent, Oregon, Harvested 1992

Selection #	42	117	184	208	234*
Seed Wt. (lb)	9.02	9.24	15.27	14.83	4.90
Area (sq. ft.)	1600	800	1600	800	800
Yield (lb/acre)	246	503	416	807	267

*The small amount of seed associated with #21 is not indicative of the seed production potential of this selection. The reduction in yield is directly related to an mistake in preemergent herbicide application to a substantial amount of the seed production block.

As expected, more than one cycle of seed trials will be needed to produce enough seed for management trials and for completing "Breeder's" seed needs in preparation for production of foundation seed.

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.

A new crossing block was established with #42, #117, #184, and #208. In addition a planting of one of the second cycle selections (1930) which possesses a thick, dark green, curly leaf was established. This plant may require a cold induction to initiate flowering. Inheritance of the "curly" characteristic is also of interest. This material could be a productive and interesting parent once flowering requirements are unravelled.

B. BREEDER'S SEED TRIAL (1992)

A breeder's seed production trial from increase was planted at Pickseed West in late October of 1992. Because of limitations on the space available, one pound of seed each of numbers 42, 184, 208 and 234 were planted. It was determined that selection # 117 seed needed further evaluation for variation in flowering characteristics in the seedling population before increasing further.

VI BREEDING AND GENETICS

A. FLOWERING

Poa annua is known for continuous flowering during the growing season. However genotypes exhibiting seasonal flowering characteristics (spring only) offer great potential for improvement of the species. Evaluations of parental material, F1, F2, and F3 populations suggest that the flowering habit is controlled by one simply inherited gene. Examination of segregating F2 and F3 progeny reveal a ratio of 3:1, continuous to seasonal flowering habit. Small amounts of late season flowering also exist within the seasonal groups, but the mechanism, or cause of this is unknown at this time. Incidently, the seasonal flowering trait is also associated with perennial types only.

Evidence suggests that these flowering habits are correlated with the physiological requirements, particularly cold and to a lesser extent, photoperiod. The genetic system described fits well with the high heritability and rapid evolution that has been reported for plant type in *Poa annua*.

Materials & Methods:

F1, F2, F3, and S1 generations of plants, resulting from most reciprocal combinations of three *Poa annua* genotypes, were studied for flowering habit and pre-reproductive period, or days-to-flowering, during 1992. The parental genotypes (117, 184, & 234) represent typical types of flowering habit in *Poa annua* (Fig. 7 & 10). Seeds were germinated in April and May, then transplanted to the field on May 20 and June 23. Flowering data, consisting of whether or not a plant was in flower, was then collected at approximately two week intervals from June 29 to October 1. On August 1, flowering habit classification data was taken, based on representative types of *Poa annua* flowering habit (Fig. 9 & 10). This categorical data was analyzed using Chi-square.

Discussion - Flowering Requirements:

Attempts at relating the inheritance data to flowering requirements have been difficult, because artificial induction of the seasonal type *Poa annua* plants (such as 117 and 184) has not been successful. Continual types (like 234) flower prolifically under all conditions, indicating a lack of, or minimal flowering requirement. Seasonal *Poa annua* plants typically do not flower when overwintered in a warm greenhouse. However, under natural, or outdoor conditions, the flowering requirements of these plants appear to be satisfied by early winter (mid December), since the transfer to a long day photoperiod, warm greenhouse results in flowering after a short period of time. (data not presented).

Cold seems to be the major flowering requirement, with photoperiod secondary. Because this is based on observations made under various growing conditions, further study is required. However it appears likely that gene(s) for cold requirement are involved.

Discussion - Inheritance:

The segregation data of the F2 and selected F3 generations fits well with a single locus model having two alleles. The significant 3:1 ratios appearing in the F2 and F3 families with putative heterozygous parents are the best evidence for this genetic model. However patterns for the late flowering characteristic are not evident. Comparison to previous year's flowering data on F2 populations of crosses between 184 and 234 indicate no similar patterns. Therefore GxE is a possible cause of this late summer flowering habit. Due to the lower numbers of individuals in the previous year, further study should determine if the late flowering characteristic is genetic or environmental in nature.

The habits of flowering described here correlate with two subspecies commonly used for *Poa annua. P. annua* var. *annua* exhibits the continuous flowering habit, while *P. annua* var. *reptans* (perennial type) typically is of the seasonal type. Some differences between these subspecies have been described previously to be genetic in nature (Law, 1977; McNeilly, 1981). In addition, Till-Bottraud (1990) calculated high estimates of heritability and a rapid evolution for these subspecies types. A single locus system, as proposed here for flowering habit, could play an important role in this rapid evolution.

Understanding the breeding behavior of this flowering habit will provide better understanding of the ecology and competitiveness of *Poa annua* in the golf course environment, and, of course, would be of tremendous value to the breeding program.

VII CYTOLOGY - CYTOGENETICS

A. CHROMOSOME COUNTS OF INTERSPECIFIC HYBRIDS

Cytological examination and chromosome counts will be necessary for proper description and introduction of any selections or varieties. The information is also crucial to the conduct of any breeding program where differences in ploidy exist.

P. supina (2n=2x=14), is reported to be one of the evolutionary parents of *P. annua* (Koshy, 1968 and ref. cited therein). On this basis, this species was introduced into the breeding program for the improvement of *P. annua*.

During the last three years, some interspecific hybrids between *Poa annua* and *Poa supina* have been synthesized. Plants resulting from these crosses have exhibited good to excellent color and average vigor and other desirable turf qualities. Interestingly, all appeared to exhibit a continuous flowering habit, regardless of parents. This was interpreted as an indication that there was a high probability that these were actually hybrids. However, verification required cytological examination and chromosome evaluation.

In addition, crosses of this type have been reported in the literature, but resulted in sterile plants. Contrary to that, several of our crosses exhibited some level of fertility resulting some selfs and a limited number of backcross progeny. Cytological examination of the progeny revealed that the PS39 X PA184 appeared to possess 28 chromosomes. The reciprocal cross revealed 21 chromosomes with some possible pairing of some of the chromosomes.

In the case of one of the backcrosses to PA 184, counts of 21, 14 and 15 were made. All of these findings indicate that continued research with interspecific crosses could be productive both on the scientific as well as practical basis of incorporating desirable characteristics from each of the species.

Through out this work we were constantly impressed that with these grasses, chromosome counts are difficult to make, expensive, time consuming, and tedious to conduct, but they are necessary. Clearly we needed to find a better way of ascertaining the ploidy levels in the plant materials that we are working with.

B. EXAMINATION OF PLOIDY LEVELS THROUGH FLOW CYTOMETRY

Nannfeldt (1937) noted that *P. annua* is tetraploid (2n=4x=28) and the chromosome counts obtained matched that of three previous researchers. However, more recently there have been 2 reports of 14 chromosomes in this species (Ellis, Calder, and Lee, 1970; Hovin, 1958) indicating some polyploid variation in the species. This should not be too surprising because ploidy level varies in other members of the genus. *P. sphondylodes - viridula* in Japan (Koba and Tateoka, 1991) are known with 2n= 28, 35, 42, 49, and 56. In *P. pratensis*, chromosome counts ranged from 2n=33 to 92, with the majority having 56, 63, 70, or 77 chromosomes (Speckmann and van Dijk, 1972).

DNA measurements through flow cytometry of appropriately stained isolated nuclei can provide a comparatively rapid and easy assay of ploidy level compared to chromosome counts. Keeler, et al. (1987) noted a high correlation between nuclear DNA content measured by flow cytometry and chromosome number from root-tip squashes in *gerardii* allowing rapid determination of ploidy level.

On the bases indicated above, research was initiated to: 1) Determine if variation in ploidy level exists in accessions of *P. annua* through analysis of chromosome counts of root-tips and analysis of DNA content; and 2) Determine chromosome number in *P. supina* accessions and in hybrids between *P. supina* and *P. annua*.

Root-tips from individual *Poa* plants grown under greenhouse conditions were stained according to the HCl-Giemsa method (Guerra Filho, 1983).

Plant material was prepared in sample buffer according to Galbraith (1983). 35 mg of distal portions of leaf blades of *Poa* unknown and *Pisum* cv. 'Little Marvel' as an internal

standard were placed in 200 microliters of chopping buffer (45 mM $MgCl_2$, 15 mM sodium citrate, 20 mM MOPS, 0.1% triton X-100, pH 7.0) and chopped for 2-3 minutes with a single edged razor blade. The solution was filtered through a 10 micron nylon mesh. Staining solution was prepared containing propidium iodide and boiled Ribonuclease A (Sigma Chemical) made in fresh chopping buffer. Staining was accomplished by mixing equal volumes of filtered sample and stain solution resulting in a 50 ppm stain concentration with 20 micrograms Ribonuclease A. Samples were analyzed with a Becton Dickinson FACStar Plus. Propidium iodide fluorescence was excited at 488 nm from an argon laser and emission read at 625 nm at a 35 nm band width. Plots were generated which allowed presentation of nuclear fluorescence (channel number) versus number of nuclei. Data from 10,000 nuclei were routinely taken for analysis. The coefficient of variation of the peaks representing 2C nuclei ranged from 4.83 to 9.06% in an analysis from a typical experiment. A low coefficient of variation indicates low degradation of nuclei during sample preparation and analysis. The value for the DNA content of *Pisum* cv. 'Little Marvel' is from Michaelson, et al. (1991).

A computer program allowed the selection of peaks representing sample and *Pisum* DNA from histograms at given fluorescence levels (mean channel number). DNA content of samples was calculated by using the *Pisum* standard of 8.55 picograms of DNA per 2C nucleus: (sample mean channel number X *Pisum* DNA content= 8.55) / (*Pisum* mean channel number) = unknown sample DNA content.

Results from root-tip metaphase preparations showed *P. annua* with 28 or 14 chromosomes and *P. supina* with 14 chromosomes (Figure 11, 12, 13); (Table 2). Based on few reports of *P. annua* with 14 chromosomes, we did not expect to find many individuals at this ploidy level. After initial characterization, 14 chromosome types could be recognized by their finer texture and narrower blade width (mean of 1.62 versus 3.04 mm measured at the base of the blade) compared to 28 chromosome types (Fig. 14). We determined DNA content through flow cytometric analysis of leaf nuclei due to difficulties in easily obtaining good root-tip preparations.

Laser flow cytometry output for 1 individual of each cytotype (14 chromosomes, 1.57 pg/ 2C nucleus; 28, 3.75 pg/ 2C nucleus) shows the peak for *Pisum*, the internal standard, and that the peaks for 14 and 28 chromosome cytotypes can easily be distinguished (Fig. 16). The DNA content of each accession for which root-tip chromosome counts were also determined is presented in Table 2.

Examination of the correlation between chromosome count and DNA content for each accession showed that 97% of the variation in DNA content could be explained by chromosome number. Analysis of tissues of two individuals indicated similar DNA content of roots and leaves within each (not shown).

An analysis of sources of variation was accomplished (Table 3). The standard error for variation in samples run through the flow cytometer 3 times (machine error), variation

in different sets of leaves of the same plant (leaf variation), and variation between roots and leaves (root-leaf variation) were calculated. The latter two sources of variation were higher than the machine variation, but still quite low compared to the values of DNA being compared. The results of analysis of sources of variation taken together with the correlation between DNA content and chromosome number supports the conclusion that we may infer ploidy level from analyzing one sample of a plant's leaves for DNA content.

Figure 16 shows flow cytometry data from the accessions on which chromosome counts were done and from additional accessions of *P. annua*, *P. supina* and hybrids between these species. More individuals were found to have 14 chromosomes, in addition to others that had 28 chromosomes, the most commonly reported number for *P. annua*. No individuals were found to have a DNA content consistent with a ploidy level higher than 4x with 28 chromosomes. The DNA content for *P. supina* was consistently lower than the DNA content of *P. annua* with 14 chromosomes.

Also shown in Figure 16 are the DNA values for a number of hybrids between *P. annua* (28 chromosomes) and *P. supina*. All appear to have a DNA content consistent with the *P. annua* ploidy level. Tetraploid (*P. annua*) by diploid (*P. supina*) crosses were made as well as some reciprocal crosses. Diploid by tetraploid crosses providing this result could not be due to pollen contamination producing selfed progeny. However, the production of unreduced gametes is one possible mechanism to explain in the observation of hybrids at a DNA content consistent with the *P. annua* parent of these crosses.

Discussion

DNA content measured by flow cytometry closely matches ploidy level from root-tip squashes and allows rapid determination of ploidy level in *P. annua*. The ease of this analysis improves our ability to exploit ploidy variation. The production of tetraploid hybrids from crosses of diploids and tetraploids has been observed and discussed from the standpoint of plant evolution and breeding (e. g. in *Dactylis*: Carroll and Borrill, 1965; van Santen, 1988). One mechanism through which this variation could occur is by the production of unreduced gametes; future research could examine this possibility for opportunities for breeding and improved under-standing of gene flow with close relatives of *Poa annua*.

We wish to acknowledge Mike Hupke, Immunobio-logy, University of Minnesota, for running our samples on the flow cytometer.

Respectfully submitted,

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Table 2. Nuclear DNA content and chromosome number for Poa annua selections and accessions. The mean DNA content for the 28 chromosome accessions is 4.11 and for the 14 chromosome accessions is 2.20 picograms DNA per 2C nucleus. A correlation coefficient of r=0.986 was found for the known P. annua 28 and 14 chromosome selections. The amount of variation in DNA content that could be explained by chromosome number was 97% (r^2=0.972). The DNA content and chromosome count for two individual P. supina accessions is also noted.

Selection	Chromosome Number	DNA Content (pg)
Poa annua		
42	28	3.96
234	28	4.38
117	28	4.13
1930	28	4.22
208	28	3.79
184	28	4.31
493	28	4.03
1959	14	2.23
1955	14	2.26
1976	14	2.23
1383	14	2.10
Poa supina		
391	14	1.79
417	14	1.82

Table 3. Sources of Variation in Measurements of DNA content of *Poa annua*.

	Standard Error
Machine Variation variation in the same sample run 3 times (5 samples tested)	.003 to .012
Leaf Variation variation in 3 different sets of leaves of the same plant (3 plants tested)	.032 to.036
Root - Leaf Variation variation between root and leaf samples of the same plant (2 plants tested)	.002 to.074

Fig. 1. *Poa annua* # 234, established from seed, mowed at 1/4", July 1992

Fig. 2. *Poa annua* # 208, established from seed, mowed at 1/4', July 1992

Fig. 3. *Poa annua* # 42, established from seed, mowed at 1/4", July 1992

Fig. 4. *Poa annua* #42, seeded 4/26/92, mowed at 1/8", as it looked on 8/27/92

Fig. 5. *Poa annua* # 184, seeded 4/26/92, mowed at 1/8", as it looked at the San Diego Country Club on 8/27/92

Fig. 6. *Poa annua* # 234, seeded 4/26/92, mowed at 1/8", as it looked at the San Diego Country Club on 8/27/92.

Fig. 7. Typical Plant Type of a Continuous Flowering Habit *Poa annua* Plant.

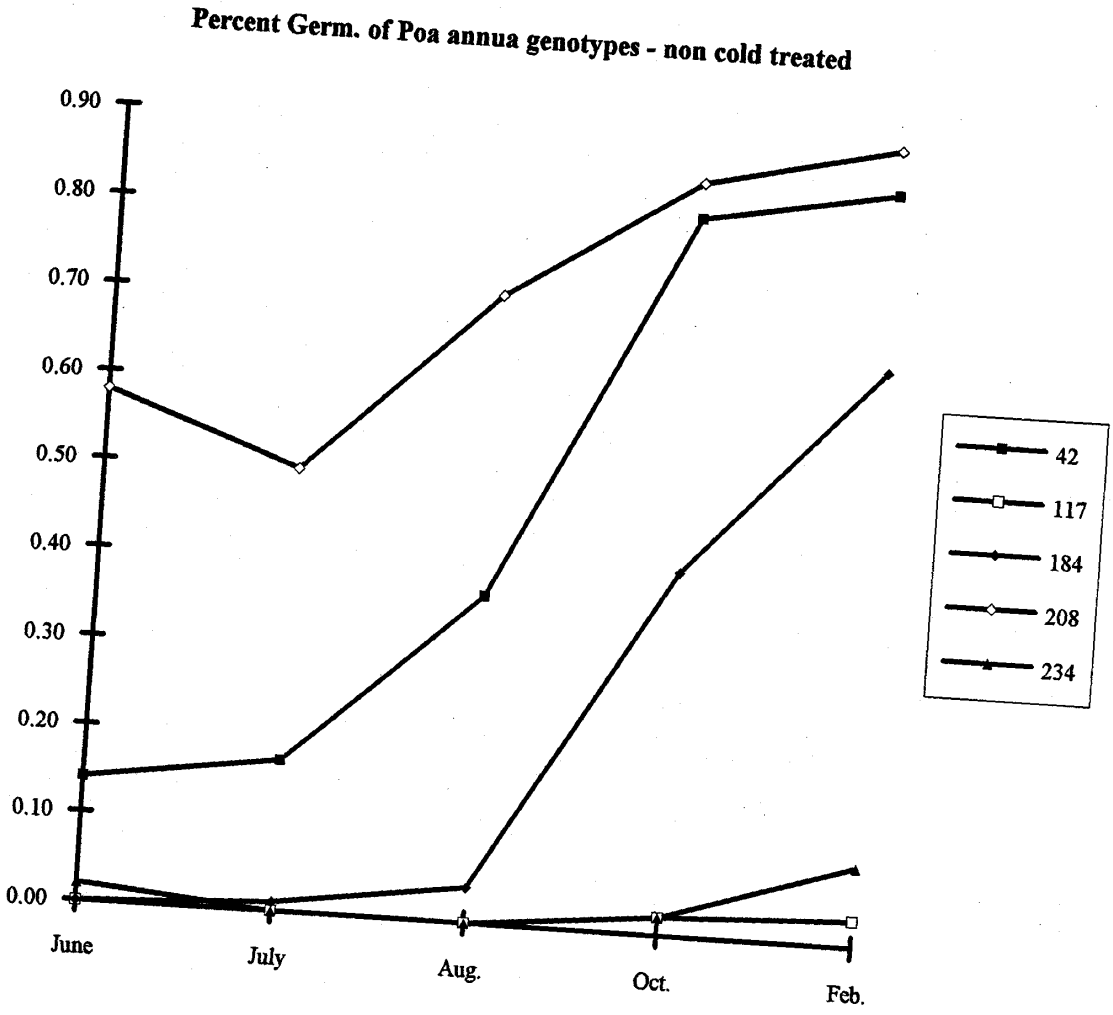

Fig. 8. Germination of non-vernalized *Poa annua* seed

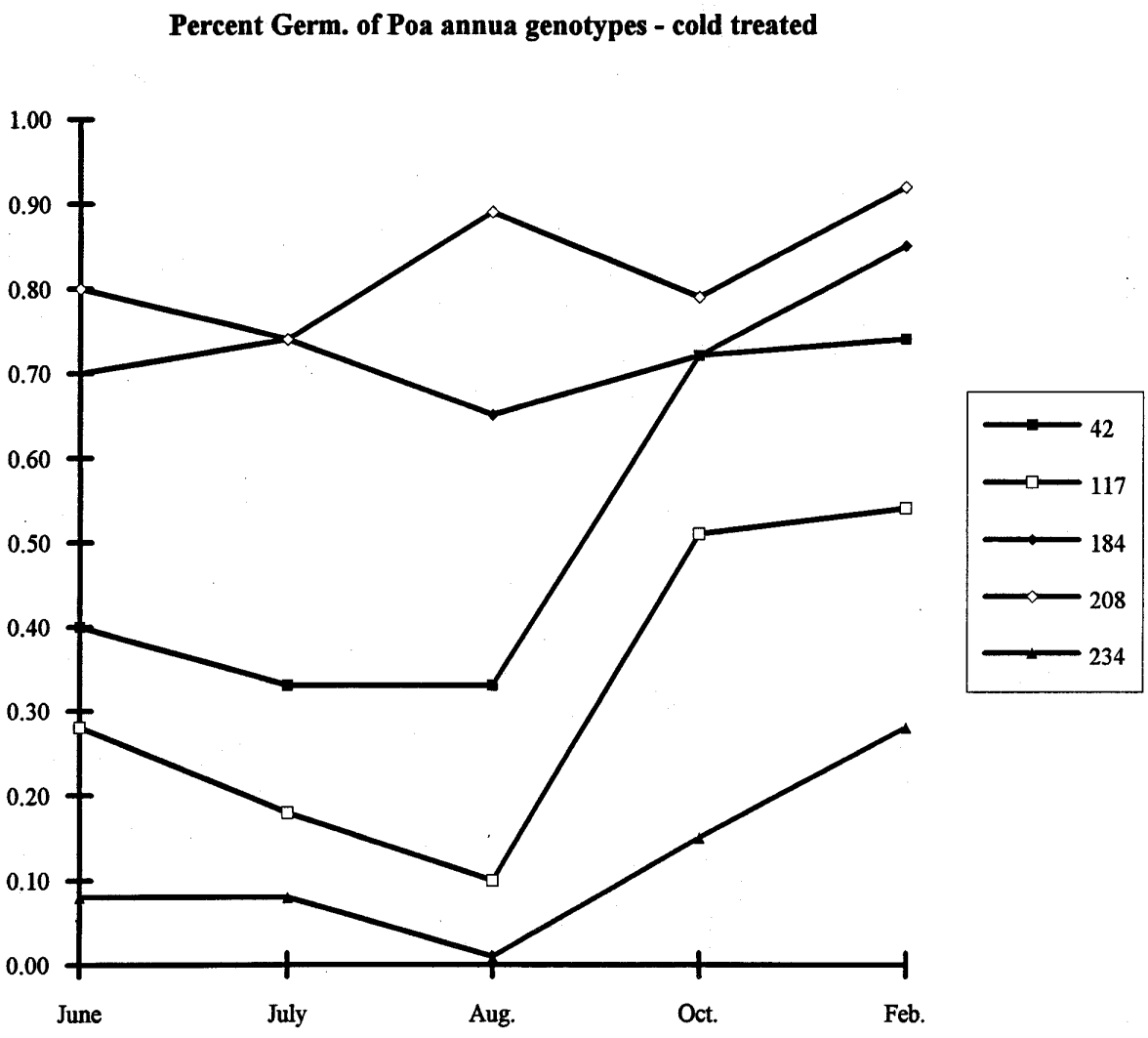

Fig. 9. Germination of vernalized *Poa annua* seed after 14 days of vernalization

Fig. 10. Typical Plant Type of a Seasonal Flowering Habit *Poa annua* Plant.

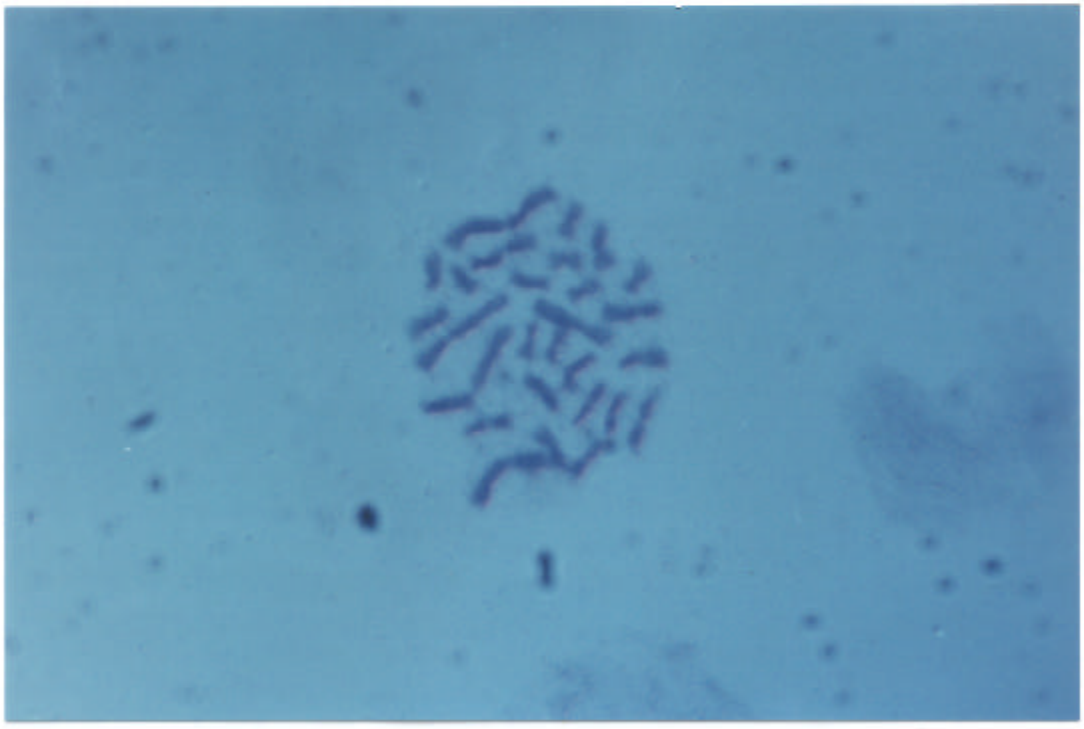

Fig. 11. Typical Chromosme Figure for the 28 Chomosome Type *Poa annua*; # 493

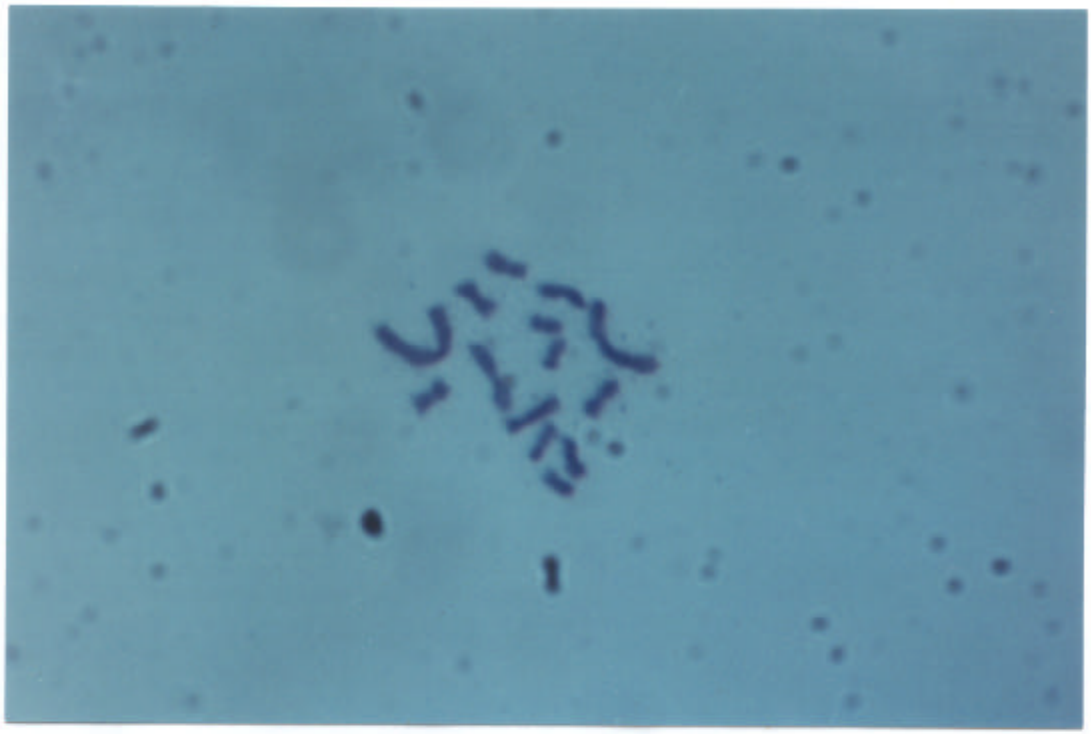

Fig. 12. Typical Chromosome Figure for the 14 Chromosome Type *Poa annua* # 1955

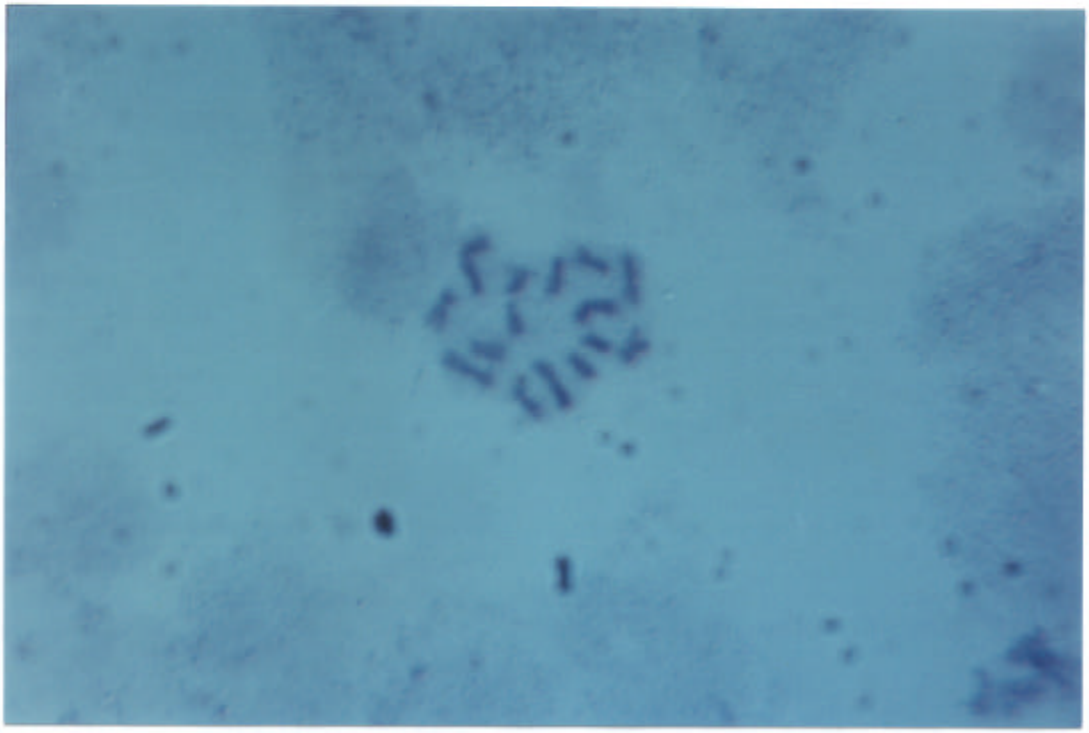

Fig. 13. Typical Chromosome Figure for the 14 Chromosome type *Poa supina* # 276.

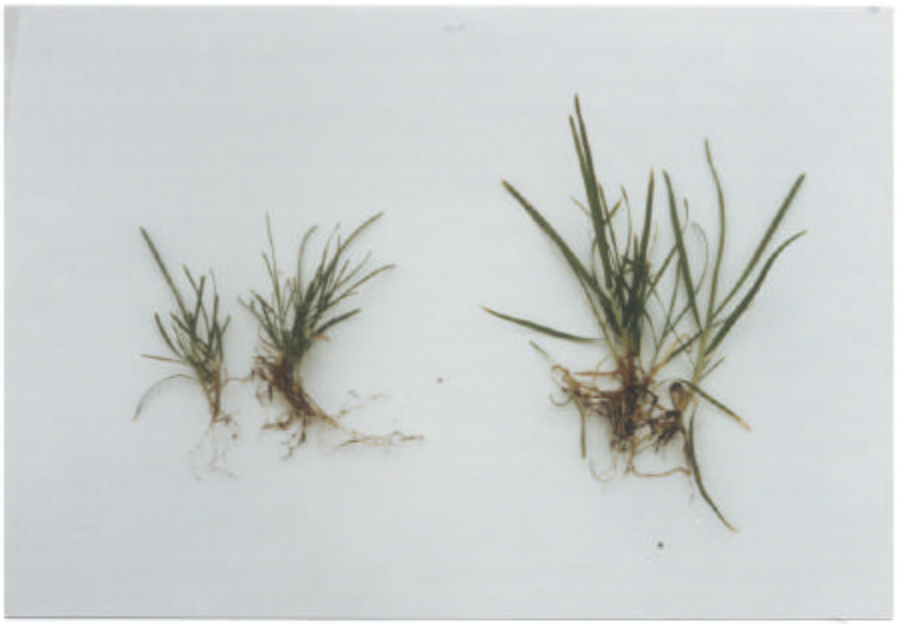

Fig. 14. Comparison of Plant Habit for 14 Chromosome *Poa annua* (left) and 28 Chromosome Type (right). Note short stature and fine texture.

Fig. 15. Close up of flowering in an experimental crossing block (center row is #117).

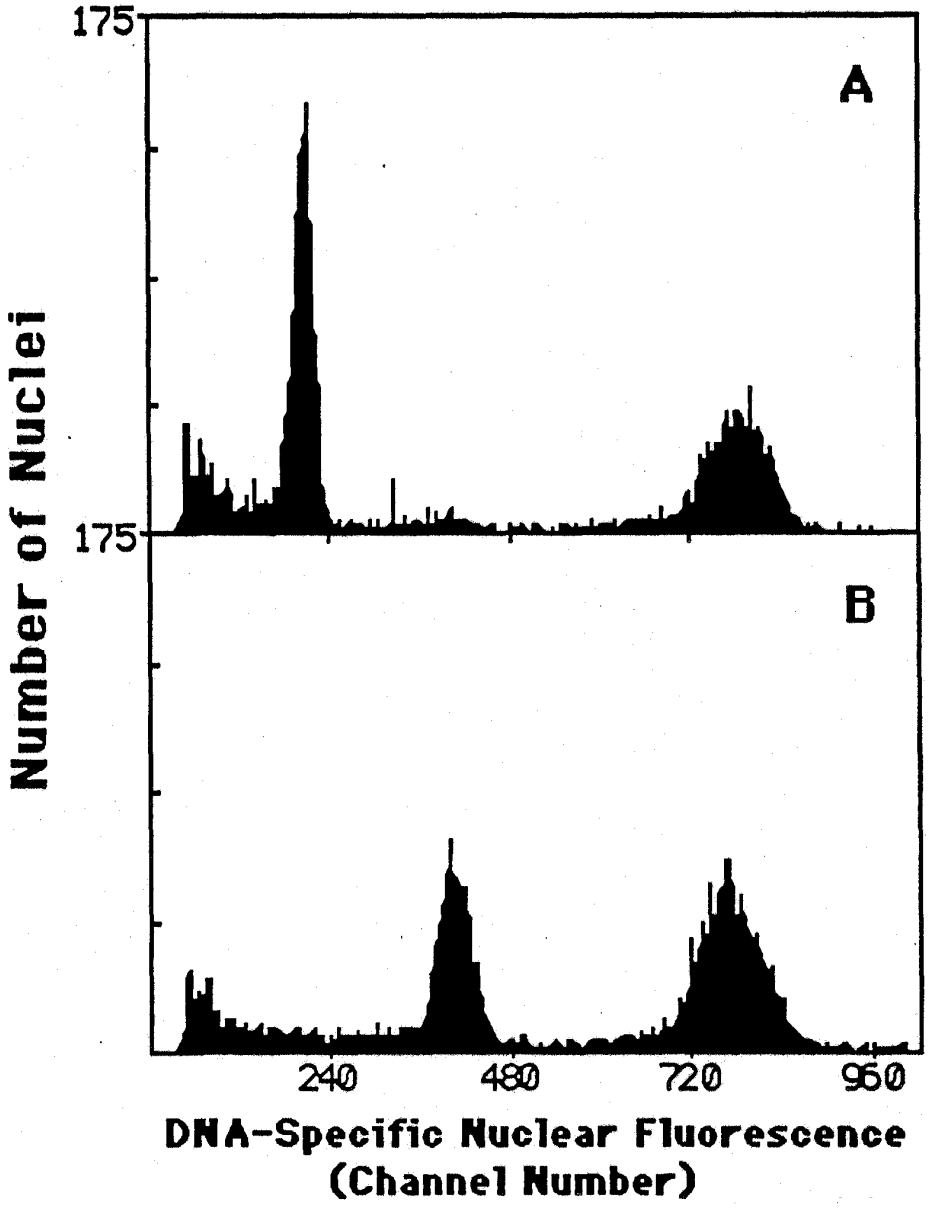

Figure 16. Nuclear DNA content of various accessions of *Poa annua*, *Poa supina*, and hybrids between *Poa annua* and *Poa supina*. Accessions represented by open symbols determined by chromosome counts and flow cytometry; all others determined by flow cytometry only.

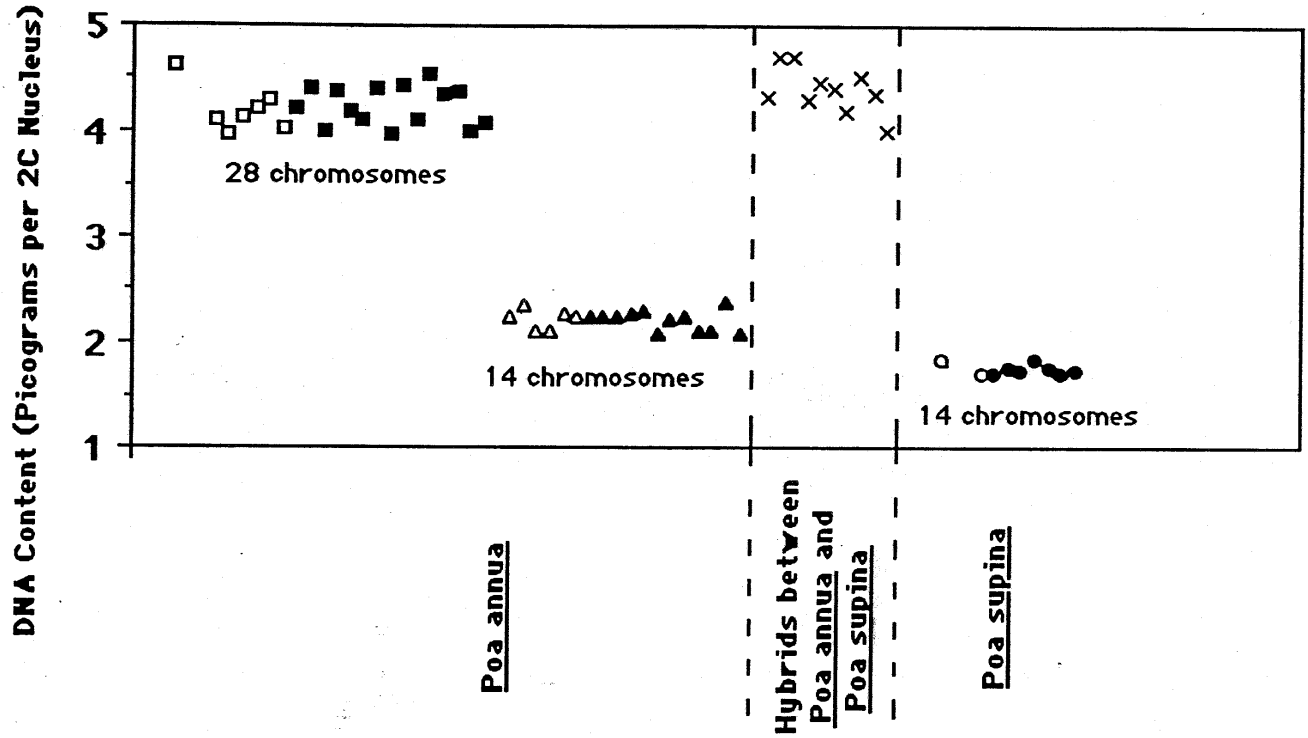

Figure 17. Nuclear DNA content for two *Poa annua* accessions with differing cytotype in relation to the internal standard (*Pisum sativum* cv. 'Little Marvel.' DNA content = 8.55 pg/ 2c nucleus). A) *Poa annua* accession #1383 (14 chromosomes; nuclear DNA content mean = 1.57 pg/ 2C nucleus). B) *Poa annua* accession #234 (28 chromosomes; nuclear DNA content mean = 3.75 pg/ 2C nucleus.) Right-hand peak in each represents *Pisum* nuclei, left hand peaks are sample nuclei.

Fig. 18. Seed Production Block at Tangent, Oregon of *Poa annua* #42 Showing Status of the Plot, Planted Late October 1991, Picture Taken on 4/20/92

Fig. 19. Seed Production Block at Tangent, Oregon of *Poa annua* #184 Just Prior to Harvest, Planted Late October 1991, Picture Taken Late June 1992.