

**Proceedings of
Scotts Turfgrass
Research Conference**

Volume 4—Turfgrass Breeding

January 1973





First row, left to right: William Daniel, John Madison, Eugene Mayer, Joseph Kuc, Lincoln Taylor, Richard Bangs.

Second row, left to right: William R. Kneebone, Allen Dudeck, Leroy Nittler, William Richardson, Ming Yu, Terry Riordan.

Third row, left to right: Joseph Duich, Derwyn Whalley, Jack Fisher, Joseph Higgins, John Long.

Fourth row, left to right: Douglas Taylor, Reed Funk, Virgil Meier, Dale Benedict, James Gabert, Kenyon Payne.

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PREFACE

This volume contains the proceedings of a Turfgrass Research Conference on Turfgrass Breeding sponsored by and held at the O. M. Scott & Sons Company, Marysville, Ohio on June 18-20, 1972. Workers concerned with a wide array of turfgrass species, turfgrass problems, and turfgrass conditions joined with members of the Scott's research staff in fruitful discussions. The formal (and some of the informal) expressions of problems and solutions are included between these covers.

Four such conferences have now been held. It has been my distinct privilege to participate in two of these. I know that I speak for other participants (and for those who have been able to join later by means of published proceedings) in applauding O. M. Scott & Sons for their leadership and farsighted public spirit in organizing and completing this series. I should like also to express my personal appreciation to Dick Bangs and Bill Schwaderer of Scotts and to Ken Payne and Bill Daniel, my associates in the editing process.

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William R. Kneebone
Chief Editor

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BREEDING GRASSES FOR TURF PURPOSES

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Introduction

The development of truly outstanding varieties is one of the greatest needs of the turfgrass industry. The most important attributes of these varieties should be dependability, reduced establishment and maintenance requirements, durability, and attractive appearance (Funk and Pepin, 1971). Each of these attributes can be heightened over what they are now as we achieve improved genetic resistance to diseases, insects, nematodes and weeds, a more desirable growth habit, better turf-forming properties, and greater tolerance of various environmental stresses, such as heat and drought. Adaptation to specialized uses and environments is also important. For example, better turfgrasses are needed for shaded areas, athletic fields, close-cut tees and fairways, and for poor soils. Varieties of unique or striking color would be very useful for special landscaping patterns and effects.

The leading cool-season turfgrass species include Kentucky bluegrass (Poa pratensis L.), the fine fescues (Festuca spp.), the bentgrasses (Agrostis spp.), and perennial ryegrass (Lolium perenne L.). In each of these species considerable genetic improvement can and should be made.

Kentucky Bluegrass

Kentucky bluegrass is a pseudogamous, facultative apomict having complex cytological characteristics (Akerberg, 1942; Grazi et al., 1961; Tinney, 1940). Although the basic chromosome number of Poa is $x = 7$, most plants of P. pratensis possess from 35 to 100 chromosomes. In fact, biotypes ranging from 28 to over 150 chromosomes have been reported.

Apomixis, as it exists in Kentucky bluegrass, is a very advantageous method of maintaining a superior hybrid variety. Elite cultivars such as Merion, Fylking and Nugget most likely originated as chance hybrids in nature. They maintain their superior hybrid genotype because nearly all of the seed is produced asexually through apomixis and is thus genetically identical to the mother plant. Other elite Kentucky bluegrass plants have been collected which are highly sexual. Warren's A-20 and Warren's A-25 are examples. However, they must be propagated vegetatively to maintain their superior qualities and varietal identity.

All currently available bluegrass cultivars originated as individual plant selections from old turf areas or as aberrant plants in seed production fields or nursery rows where no attempt had been made to control parentage. Approximately 40% of the plants collected from pastures, roadsides and other old turf areas of the Northeastern United States have been sufficiently apomictic to maintain varietal identity (Funk and Han, 1967). Such collections will continue to be a source of new varieties and also germplasm for hybridization. Previous studies by Duich and Musser (1959) and Grazi, Umaerus and Akerberg (1961) have demonstrated that bluegrass cultivars such as Merion and Fylking are highly apomictic. Merion produced 3-4% nonmaternal plants. Fylking produced only 2%. Furthermore, most of these aberrant plants were very weak. The majority of the plants studied by Tinney and Aamodt (1940) and Akerberg (1942) were also predominantly apomictic. Clausen, Keck and Hiesey (1944-1945) were unable to obtain hybrids using *P. pratensis* as the female parent in interspecific crosses although reciprocal crosses were often successful. Because of these factors, plus the cytogenetic complexity of Kentucky bluegrasses, an extensive research program has been conducted to develop techniques and procedures required to develop an effective program to improve Kentucky bluegrass by intraspecific hybridization (Funk and Han, 1967; Han, 1969; Pepin, 1971; Pepin and Funk, 1971).

Substantial numbers of bluegrass hybrids have been produced at Rutgers using a greenhouse crossing technique described by Han (1969). Plants growing in field nurseries can be brought into

the greenhouse any time from December to May and forced to flower using cool nights and extended daylength. Seed of Merion and Fylking produced in our greenhouse normally contains over 10% hybrids. Many of these hybrids originate from the fertilization of unreduced eggs (Pepin and Funk, 1971). The egg of apomictic strains of Kentucky bluegrass normally initiates division to form the proembryo before or very soon after the flower opens (Tinney, 1949). Thus, the frequency of hybrids may be enhanced by applying pollen as early as possible. At New Brunswick, anthesis of bluegrass occurs at night, normally between midnight and 5:00 a.m.

Studies by Han (1969) and Pepin (1971) demonstrate that non-hybrid aberrant progeny generally show substantial inbreeding depression in nearly all characteristics relating to vegetative and reproductive vigor. On the other hand, vigorous hybrids can be obtained. Although the average hybrid is somewhat less vigorous than the average vigor of its highly heterozygous selected parents, a workable number of hybrids have excellent vigor and frequently show positive transgressive segregation. Many of the more promising F_1 hybrids are highly apomictic.

Several second cycle hybrids developed by using first cycle hybrids as female parents are being evaluated. A number of these second cycle hybrids show excellent performance in turf evaluation trials in New Jersey. Additional experience with such hybrids in combination with extensive cytological study is needed to assess the value of recurrent intraspecific hybridization. Some of our best current hybrids, such as P-27, P-72 and P-106, have about 95 chromosomes. It remains to be seen whether hybrids with even higher chromosome numbers will be of practical value.

The cytogenetic complexity of Kentucky bluegrass makes it necessary to work with large populations. Also, because of the severe physiological stress placed on a plant by continued close mowing, it is difficult or impossible to select for many characteristics important in turf performance in the spaced-plant nursery. Experience has thus indicated the importance of screening large numbers of hybrids and of selections in either small seeded or small clonal plots receiving turf maintenance at an early stage in

the breeding program (Funk, Engel, and Halisky, 1967). Because of the extreme range of conditions under which turf is grown, considerable improvement needs to be made in our advanced evaluation procedures.

Interspecific hybridization involving the use of four *P. pratensis* varieties as female parents in crosses with *Poa compressa* L. has been studied (Dale, 1971). Crosses involving the highly sexual varieties, Warren's A-20, A-25 and A-26, produced a very high frequency of hybrids. Many of these hybrids were vigorous and produced considerably more panicles than are generally found in Kentucky bluegrass. However, all such plants produced very few or no viable seeds. Cytological studies indicated that these predominantly sterile hybrids originated from the fertilization of reduced eggs by reduced male gametes. The 54 interspecific hybrids produced, using the highly apomictic Belturf Kentucky bluegrass as the female parent, showed moderate to good seed fertility and acceptable seedling vigor. Some of these hybrids produced rather attractive turf. Cytological studies indicated that these fertile hybrids originated from the fertilization of unreduced eggs by reduced male gametes. These triploid F_1 hybrids are moderately to highly apomictic.

Fine Fescues

A number of promising red fescue plants have been collected from old turf areas of the Northeast. They are of two distinct types. Some have the non-creeping or chewings type growth habit similar to Jamestown and Highlight. They produce a fine, dense, low-growing, attractive turf, and the best local selections show better turf performance than any of the commercially available cultivars. Other plants have a strong spreading habit with rhizomes which compare favorably with our better bluegrasses. These strongly spreading types have good seedling vigor, a medium texture and show excellent potential for use on roadsides and in mixtures with Kentucky bluegrass for lawn type turf. Unpublished data from Rutgers University suggests that the strongly spreading type red fescues are reproductively isolated from the non-creeping red fescues. The spreading types shed pollen in mid-afternoon, whereas,

the chewings types shed pollen just before sunrise. These spreading types, (Festuca rubra L. subsp. rubra), are reported to have 56 chromosomes. The chewings types, (F. rubra L. subsp. commutata Gaud.), have 42 chromosomes. Some hard fescue (F. longifolia Thuill). varieties recently developed in Europe also show considerable promise.

Bentgrass

Bentgrasses do best in cool, moist climates. However, where professional management can be provided, it is possible to use these grasses throughout all but the hottest regions of the United States. Many of the elite cultivars of creeping bentgrass are propagated vegetatively for use on putting greens and tees. Penncross creeping bentgrass is grown from Syn 1 seed.

Breeders of future bentgrass varieties should make use of some excellent cytogenetic studies conducted in Great Britain (Jones, 1956).

Table 1. Genome relationships in Agrostis*

Species	Chromosome Number	Genome Constitution
1. <u>Agrostis canina</u> L. subspecies <u>canina</u> velvet bentgrass	14	A ₁ A ₁
2. <u>A. canina</u> subspecies <u>Montana</u> Hartm.	28	A ₁ A ₁ A ₁ A ₁
3. <u>A. tenuis</u> Sibth. colonial bentgrass	28	A ₂ A ₂ A ₂ A ₂
4. <u>A. stolonifera</u> L. creeping bentgrass	28	A ₂ A ₂ A ₃ A ₃
5. <u>A. alba</u> L. Redtop	42	A ₁ A ₁ A ₂ A ₂ A ₃ A ₃

*Adapted from Jones (1956).

Hybrids between A. tenuis and A. stolonifera showed low fertility, whereas hybrids between either A. tunuis and A. alba or A. stolonifera and A. alba demonstrated good fertility.

Perennial Ryegrass

Perennial ryegrass (Lolium perenne L.) is a cross-pollinated species with 14 chromosomes. It is a cool-season bunchgrass best adapted in maritime climates having mild winters and cool, moist summers such as New Zealand, the British Isles and the coastal areas of Washington, Oregon and Northern California. The ryegrasses are not as well adapted in continental climates where winters are cool and summers hot. A number of new synthetic varieties of perennial ryegrass have been specifically developed for turf use. These varieties show excellent performance for sports turf in Europe. In the United States, they produce good turf on athletic fields in the maritime climates of the Pacific Northwest. They appear very promising for winter overseeding of warm season grasses in the South. Throughout much of the Northeast they are being used on sandy coastal plain soils where Kentucky bluegrass is not well adapted. They are also useful on school grounds and similar areas for overseeding turf areas devastated by disease, insects or wear.

Additional improvements needed in our turf-type ryegrass include better mowing qualities, greater disease resistance and better summer and winter performance. A leafy, fine-textured, low-growing type with less persistence would also be very useful for mixtures.

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Discussion Period

Dr. Daniel: The stem rust that has been hitting Nugget, we've seen in space plantings, particularly, a very definite degradation in summertime with almost severe loss in fall and winter. Have you observed this also?

Dr. Funk: We have a very difficult time keeping Nugget alive in our spaced-plant nurseries. There are two factors involved in our situation. First, aphids weaken the plant and, second, stem rust causes additional damage to the weakened plant. If we don't at least control the aphids, we can lose the spaced-plant nursery of Nugget. Varieties, like Anheuser, show this damage to some degree while others, like Belturf, show excellent resistance to this complex of aphids and stem rust. In a space planted row of Nugget adjacent to a space planted row of Belturf, we notice a tremendous difference in performance.

Dr. Payne: In the Poa compressa hybrids, are you looking at them from genetic interest or do you hope to transfer some desirable characteristics from the P. compressa?

Dr. Funk: Our initial interest in making this cross originated from some work that was done in California a number of years ago by Dr. Jens Clausen and his co-workers. They found that it was difficult or impossible to use P. pratensis as a female parent in interspecific hybridization. They grew out rather large progenies where P. pratensis was used as a female parent. They couldn't find any hybrids. Whereas, when they used it as a male parent they found quite a few hybrids. Just by chance we notice a few natural hybrids between P. pratensis and P. compressa, with P. pratensis used as a female parent. We thought we'd see if P. pratensis could be used as a female parent in this particular cross, and we found that obviously it could. This was our initial objective. I think there is some potential from the practical standpoint of bringing in germplasm from P. compressa to improve P. pratensis for use on poor soil. I think there are a lot of characteristics from P. pratensis which could be used to improve P. compressa.

Dr. Fisher: Do you think that there is a possibility that the higher chromosome numbers in the apomictics of Poa pratensis may have occurred from natural hybridization in the past between P. compressa and P. pratensis?

Dr. Funk: This is something that we really haven't the information to answer. It would just be speculation. There is certainly a possibility that this could have occurred. The two seem to be related and we know that the fertilization of unreduced gametes of P. pratensis by reduced gametes of P. compressa can give fertile, apomictic hybrids. These hybrids can be recognized as not true P. pratensis, but I suspect that if we crossed these hybrids with P. pratensis, it would be difficult to distinguish their offspring from some of the things we have been calling P. pratensis.

Dr. Fisher: The reason I bring this up is that we are accumulating some evidence that Norin 10, the ancestor of the Mexican wheats, has rye genes in it. We know that Norin 10 came from the ancient Japanese wheat, Daruma, which I am investigating now. Daruma has rye characteristics in it and may be an intergeneric hybrid of hexaploid wheat and rye.

Dr. Yu: Can a reduced egg of bluegrass develop into a seed without fertilization?

Dr. Funk: We don't have good cytogenetic data on all of our crosses. However, we do know that when we use a highly apomictic plant, such as Belturf as a female parent, that many of the hybrids result from the fertilization of unreduced eggs. Many of these hybrids are highly apomictic. When we use highly sexual plants of Kentucky bluegrass, such as Warren's A-25 as female parents, I suspect that we are fertilizing mostly reduced eggs. Many of our A-25 x Anheuser hybrids are highly apomictic; whereas, most of our A-25 x Nugget hybrids are highly sexual.

Dr. D. Taylor: You showed us pictures of creeping fescues and the different amount of creeping under space conditions. Do you observe the same differences when they're closely mown? Do the creeping types fill in better than the chewings, or not?

Dr. Funk: Yes. The old spreading variety that we made a number of years ago has a lot of leaf spot susceptibility, and so forth, but it has shown better recovery from summer thinning than the chewings types. On our roadside plots, the value of the rhizomes of the spreading types are even more marked than under our closely mowed trials. We've been mowing at three-quarter inch in our fescue evaluations, which really isn't quite fair for these spreading types. They're not quite as tolerant of close mowings as the chewings. Improved turf-type varieties of these spreading types may be quite valuable.

Dr. Payne: You mention the technique for getting the bluegrasses to seed in the greenhouse. Have you tried that with the fine fescues -- the cool nights and long days?

Dr. Funk: Yes. I was visiting Dr. Felix Juska (Agricultural Research Service, USDA, Beltsville, MD.) a few years ago and he mentioned that he was having difficulty getting his fine fescues to flower in the greenhouse. Apparently, it is very easy to cause fescues to revert to the vegetative stage if you move them to a warm greenhouse. We've been successful in bringing fescues into the greenhouse as early as January if we keep our greenhouse temperatures cool, especially our night temperatures, and immediately put the plants under long days. There's quite a difference between varieties of bluegrass and fescues in this regard. We can bring the common types of bluegrasses into a cool greenhouse with long days, around Thanksgiving Day, and they'll flower. In fact, we have some varieties of bluegrass that will flower in the greenhouse, just if it is kept cool. If the night temperatures in the greenhouse get down in the 40⁰s, they'll flower without any outside cold temperature induction. Other varieties, including many of our better turf types, have a much greater winter requirement.

Dr. Long: You mentioned that we have a problem with summer thinning of bluegrasses due to high temperatures. Is this a physiological type response that you are seeing?

Dr. Funk: We know we get much poorer recovery if we fertilize heavily with nitrogen. We know there are disease problems involved, and we know there's a tremendous difference between varieties and their ability to tolerate this summer condition. Summer survival is a complex situation. We have observed large variety x year and variety x location interactions in summer survival.

Dr. Long: What is the major disease of perennial ryegrasses in your region?

Dr. Funk: In New Jersey, I would say that crown rust is probably the most apparent. In some of the cooler areas, such as Rhode Island and the Pacific Northwest, probably *Corticium* red thread. *Helminthosporium* leaf spot is also of some significance.

Dr. Payne: What treatments are you using to keep your nurseries free of volunteers and weeds?

Dr. Funk: We try to fallow our nursery for two or three months before we transplant to kill any perennial grasses and to reduce the weed population somewhat. Then we apply 2,4-D and dicamba for weed control in the fall. It does a good job on everything but annual grasses, which we just let grow to give us a little winter protection from your old stems, and so forth. Then in the spring we apply Dacthal at a moderately high rate. Generally, this will give us pretty good weed control until seed harvest.

Mr. Gabert: What are the main diseases of tall fescue in your area?

Dr. Funk: This is something we haven't looked at. There's certainly a lot of leaf spotting and when we've taken them to the pathologists, they've always found the *Helminthosporium dictyoides*. They've also found many other organisms. I just wouldn't be able to give an answer on that. Obviously, winter kill is also a problem under high nitrogen fertility and close mowing, even under New Jersey conditions.

Dr. Yu: I would like to know if hot water emasculation can be used in tall fescue, and what the proper procedure would be?

Dr. Funk: We haven't any experience with hot water emasculation of tall fescue.

Mr. Mayer: You reported that you thought you saw some tip chlorosis or tip damage on your Belturf and Adelphi bluegrass varieties in Pennsylvania and thought it might be smog injury. And then I believe you thought it might be *Alternaria* or some other disease organism. Have you confirmed what the problem is?

Dr. Funk: I think Dr. Duich could give us better information on that.

Dr. Duich: We've identified it as *Curvularia* sp.

Dr. L. Taylor: Is the reason for using variety blends in bluegrass more (1) that we don't know which variety we need and that one will be dominant, or (2) that blends have intrinsic advantages over single varieties?

Dr. Funk: I could say that both things are important. When we're recommending varieties for the general consumer we don't know what the situation will be four or five years from now on disease complexes. An example is this new *Curvularia* sp. that Dr. Duich mentioned on Belturf and some of the hybrids involving Belturf. We don't know the soils or management conditions. If we have three or four components in a blend, hopefully the component that's best adapted to the situation will predominate and give better long term dependability and performance. I can see some adverse problems with blends if they're not properly formulated, if they have an overly aggressive component that is a very heavy thatch former, or some other problem. I think that we should do much more research on blends. The Southern leaf blight situation on field corn illustrates the importance of genetic diversity. Adequate genetic diversity can be of considerable insurance value in a perennial planting.

Dr. Daniel: When stripe smut gets into a variety, how has it affected seed production as you've observed it?

Dr. Funk: We haven't had any problem with stripe smut in our annual seed nurseries. It's only a disease that's a problem in our older seeded turf plots, usually three years or older.

BIOCHEMICAL BASIS FOR DISEASE RESISTANCE AND ITS POTENTIAL APPLICATION IN PLANT BREEDING

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Introduction

I will discuss biochemical mechanisms for disease resistance in plants. I hope to leave you with three messages. One, that we can now begin to explain disease resistance in plants in biochemical terms--in terms of metabolic pathways, in terms of enzymes, in terms of organic compounds. The second message that I'd like to leave with you is that disease resistance does not depend upon the presence or absence of genes to do something, but whether the genes are rapidly activated so that they produce a response early enough and in sufficient magnitude to control the infectious agent whatever it may be -- fungus, bacteria or virus. I consider this a very important concept. We're not talking about the presence or absence of genes for resistance, but rather whether they are activated early enough and in sufficient magnitude to control the infectious agent. And finally, third, that we can manipulate this genetic information, much as a microorganism does, to induce resistance and to immunize plants against disease, much as animals have been immunized against disease. We will see, in fact, that there are many similarities between disease resistance mechanisms in plants and disease resistance mechanisms in animals.

I'll be spending most of my time discussing two systems. One is a disease of the Irish potato, Late Blight, caused by the fungus Phytophthora infestans. Late Blight made quite a contribution to history; it caused the epidemic of potato in Ireland which had such devastating effects on the population and resulted in mass migrations, especially to the United States. Late Blight is still a very important disease. The other disease that I'm

going to talk about is a disease of green bean. The disease is bean anthracnose, caused by Colletotrichum lindemuthianum.

Why have we chosen these model systems for our studies? Potato tubers are easy to get. We have grown many different lines of potatoes, differential varieties that are resistant to one race and susceptible to another of P. infestans. The same thing holds true for beans. We can easily grow the bean tissue and we have maintained many cultivars that differ in their reactions to different races of the C. lindemuthianum. Both fungi can be easily grown in vitro, and distinct races of both are available.

Testing for Potato Disease Resistance

Potato tubers are sliced, put into petri dishes, inoculated and incubated. Tuber slices of the cultivar Kennebec sprayed with sterile de-ioned water and incubated in petri dishes for 72 hours show some natural discoloration. The same cultivar inoculated with race 124 of P. infestans shows the compatible or susceptible interaction. After 72 hours there is very little difference from the control and you might be tempted to say that this is resistant tissue. If you wait another 24 hours to 48 hours, however, fungal growth is readily evident on the surface and sporulation occurs. The symptoms of the susceptible reaction are not evident for some considerable time, during which the organism is ramifying in the tissue without apparently damaging it.

Now let's look at the resistant interaction. If we place race 4 on the cultivar Kennebec, we induce a hypersensitive reaction. This is an immediate response that is evident as early as 12 hours after inoculation and has probably reached its maximum intensity about 24 hours after inoculation. We've used a heavy inoculum, but we've also used an equal inoculum in inoculating with the other race where we had a compatible interaction. In looking at these you'd be tempted to call the hypersensitive response to race 4, the susceptible interaction, and the response to race 124, the resistant interaction. In fact, hypersensitivity gives high resistance -- an example of great incompatibility. As soon as the organism gets into the plant, we have the death of a few cells and the organism gets no further. The organism is contained.

One of the thoughts that has been very prevalent in disease resistance work is that there are plants that have genes for resistance and there are plants that don't have genes for resistance. It has been suggested that a specific gene product determines resistance to each pathogen. This concept can be misleading. We set up some experiments to explore this concept and to see whether it might be misleading. We made thin layer chromatograms of extracts from tubers of two cultivars of potato, Russet Burbank and Kennebec. The tubers were inoculated with two races of *P. infestans* which can attack the former, but not the latter cultivar. Kennebec is resistant and Russett Burbank is susceptible. We also inoculated with the fungus *Helminthosporium carbonum*, which does not attack the potato. It is a non-pathogen of potato, but it is a pathogen of corn. What did we see? If we inoculate these tissues, extract them, run chromatograms of the extracts, and spray the chromatograms with different reagents which will detect metabolites, we see a pattern of metabolites which is a fingerprint for an interaction. Depending upon the reagent we use to spray with or how we extract tissues, we can see different compounds. Spraying with antimony tri-chloride, for example, detects among other things, terpenoid-type or steroid-type compounds. Each spot on the chromatogram represents at least one compound that has appeared after spraying. Extracts of fresh tuber tissue that hasn't been inoculated have some of these metabolites. If we let the tissue age, indeed we see that a few new metabolites appear and others increase. If we inoculate Russett Burbank with *P. infestans* (susceptible), we see very little increase in the terpenoid-type or steroid-type compounds as compared to the aged uninoculated tubers. If we inoculate the Kennebec, the resistant variety, we see a very marked build-up of metabolites. It's tempting to say then that Kennebec has genetic information which is controlling the synthesis of these compounds and that the Russet Burbank does not. Actually that's a misleading statement. If we inoculate Russet Burbank and Kennebec with the non-pathogen of potato, *H. carbonum*, we find that both cultivars are equally able to synthesize these terpenoids very rapidly after infections. We can not state that Russet Burbank doesn't have the genetic information to produce these compounds. All we can say is that in the presence of a race which can attack

the Russett Burbank (for some reason the genetic information is not being expressed) which would lead to the production of these compounds. It's also possible that the compounds are being degraded in the compatible interaction.

What is the relationship between the terpenoids and resistance? We find that there are a number of terpenoid compounds that consistently arise in incompatible or resistant reactions which we do not find in the susceptible or compatible reactions. These compounds are inhibitory to *P. infestans*. They are also inhibitory to *H. carbonum*, and they accumulate very rapidly after infection to levels which can account for the containment of the organism in the plant. It appears we have a mechanism which resembles a disease resistance mechanism in animals. We don't have complex protein antibodies produced or interferon-type substances, but we do have simpler compounds. They may be terpenoids and/or phenols, depending on the plant infected. These compounds, in the case of plants, may have a very important role in disease resistance. In resistant tissues, they build up very quickly to levels which inhibit further development of the infectious agent.

Mechanisms that Activate and Suppress Disease Resistant Reactions

These compounds, which build up in resistant tissues, are sometimes called phytoalexins. One that we've worked with in potato is the compound rishitin. We have also isolated another compound, phytuberin. Rishitin becomes evident very soon (24 hours) after inoculation and rapidly accumulates. Phytuberin also builds up in the incompatible or resistant reaction. How reliable is this accumulation? We have used eleven cultivars of potatoes and six races of the fungus. We have tested numerous compatible and incompatible interactions. In all these cases, rishitin and phytuberin rapidly accumulate in the incompatible but not the compatible or susceptible interaction. We feel confident indeed that this is a real thing and these two terpenoids are associated with the disease resistance reaction.

What is it that is inducing the synthesis and the accumulation of rishitin in the incompatible interaction? Is it something very specific that arises only in the interaction? Can we induce the plant to produce and accumulate the rishitin without the living fungus? We found that a cell-free sonicate of any race of the fungus can induce the production of rishitin and phytuberin in the potato tuber to levels even greater than those found in an incompatible interaction. Interestingly enough, even cultivars "lacking genes for resistance," accumulate rishitin and phytuberin when treated with sonicates of *P. infestans*. Russet Burbank is a cultivar that is susceptible to all the known races of *P. infestans*; therefore, it would be tempting to say that it does not have the major R genes for resistance to *P. infestans*. However, a sonicate from any race of *P. infestans* induces rapid accumulation of both terpenoids. The accumulation would not have occurred if the potato had been inoculated with a compatible race. How specific is this response of the potato to *P. infestans*? We can induce the accumulation of rishitin and phytuberin with a broad spectrum of fungi including *H. carbonum*. We're not talking about a specific antibody-antigen-type of relationship. We're talking about a broad spectrum of compounds which are rather non-specific but which the fungus will either induce the accumulation of or not, depending upon whether it can or cannot attack that particular host. One wonders then what is it that's happening in the compatible or susceptible interaction. If the potato that is susceptible has the potential for producing the antibiotic compounds, why is it that this potential is not expressed; that this potential is not realized? There are several possibilities. One, is that in the susceptible interaction, these antibiotic compounds are produced but they're rapidly detoxicated. They are broken down into non-fungitoxic compounds. Another possibility is that, for some reason in the susceptible interaction, the fungus is suppressing the synthesis or accumulation of these compounds. It is because of the suppression of a general resistance mechanism, that it is able to attack the potato.

We set up a series of experiments in which we inoculated with the compatible race, waited a period of 12 or 24 hours, and then inoculated with the incompatible race. Inoculation with the incompatible race normally leads to the accumulation of rishitin and phytuberin. What we found was

that when we preceded the incompatible race with the compatible race by 12 or 24 hours, we suppressed the response to the incompatible race. Susceptibility, at least in this system, was based on somehow suppressing a resistance response. Not a lack of a potential for producing the compound, not a lack of potential for inducing its production, but rather an active mechanism for suppression.

If we have inoculation with an incompatible followed by a compatible race, the incompatible race completely protects the potato against subsequent inoculation by the compatible race. If we hit it first with the resistant combination, we get the build up of rishitin and start a series of metabolic events which protect the potato. If we inoculate the potato with the compatible race, then wait a period of 12 hours and inoculate it with the incompatible race, rishitin accumulation is markedly reduced and we also don't have protection. We have, in fact, susceptibility. A 24-hour lag between inoculation with the compatible and incompatible race increases suppression.

Now what happens on the metabolic level when we do this? Metabolically, how do we explain this? We find an interaction, then the accumulation of rishitin is suppressed by a compatible. Another terpenoid accumulates which is not fungitoxic. It appears that by suppressing rishitin accumulation, we've caused the accumulation of another compound in the pathway. This compound is not toxic to the fungus. The fungus can grow.

Inoculation with a compatible race 12 to 24 hours before treatment with a fungal sonicate also suppresses accumulation of rishitin and phytuberin. It's not a matter of one race competing with another, but is in fact that the compatible or susceptible combination interferes with the response to the incompatible race or its products.

Not only has the accumulation of rishitin and phytuberin been suppressed, but the necrosis associated hypersensitive reaction or resistant reaction is also not evident. For some reason, now the potato may recognize the incompatible race as compatible.

This is a bit of the potato story. And indeed we see that compounds are produced in resistant reactions. These compounds are inhibitory. They are produced at levels that can inhibit microorganisms. We can induce their production with chemicals produced by the fungus. In this disease, susceptibility does not appear due to the ability or inability to produce a compound by the plant, but rather whether it happens in an interaction soon enough and in sufficient magnitude.

Disease Susceptible and Resistant Interactions in Green Beans

Let's take a look at another interaction. This is the interaction of green bean with Colletotrichum lindemuthianum. Inoculate seedlings of the susceptible cultivar Red Kidney with the beta and gamma race of the C. lindemuthianum, and a susceptible reaction is apparent. Is it really accurate, however, to say that this is a susceptible plant? We say it is susceptible because there is sufficient damage to cause some economic loss. In the case of an ornamental, it's an aesthetic loss. This is not, however, a biochemical definition. When we look at the plant, we see it's not an accurate definition by any means. Even in the susceptible interaction, we have areas of very real and very marked resistance. As we approach the root zone, the lesions become very restricted. Down in the root area itself we find typical hypersensitive-type reactions. The organism germinates in these areas, penetrates, but you get a hypersensitive reaction -- a very rapid resistant reaction. Even this susceptible plant, susceptible to the beta and gamma races of fungus, has tissues that are expressing hypersensitive resistance.

If we assume the genetic information is the same in all the cells of the plant, it's tempting to say that this plant, indeed, has genetic information for resistance and in one part it's being expressed and another it's not. It's not a mechanical matter because the organism germinates and penetrates. It would be very nice to get to the key for the expression that's so effective in roots and somehow use this key to express the genes in all tissue.

In the bean - C. lindemuthianum interaction, as with potato-P. infestans, we have cultivars resistant to some races and susceptible to others. We can take a cultivar, the genetic information

of the plant being constant, and vary the race of the fungus. Or we can take several cultivars which react differently to a single race. This is a beautiful system to crosscheck all of our work and all the work I describe with seedlings is valid for plants and pods. The same cultivar may change from complete resistance to complete susceptibility depending on the race of fungus used for inoculation. This argues very strongly against the thought that a resistant plant has genes for doing something and a susceptible plant does not. Here we see the same plant reacting one way to one race and another way to another race.

Let's look at these interactions in a little more detail. If we inoculate a susceptible or resistant cultivar with the spores of *C. lindemuthianum*, the spores germinate within 3 to 5 hours and germinate equally well on a susceptible or resistant cultivar. About five hours after germination, a thick walled appresorium forms. This is a structure from which the organism will emerge again and then penetrate into the plant. This doesn't happen immediately. About 36 to 48 hours after inoculation the organism emerges again from the appresorium and penetrates into the plant. Up until this time, the reaction is identical on the susceptible and the resistant plant. Thirty-six to forty-eight hours after inoculation, when the fungus has penetrated into the resistant plant, we see a granulation of the cytoplasm -- a hypersensitive reaction -- and the fungus is contained. The fungus goes no further than one or two cells. At that same time, in the susceptible interaction, the organism has grown considerably more but there has not been a granulation of the cytoplasm, the cells are intact. This is true, even though the fungus has grown considerably more. After 96 hours, the fungus has ramified through the tissue and some cell walls appear disorganized. Ninety-six hours, or perhaps 110 hours after inoculation, lesions appear in the susceptible interaction. Resistance, therefore, represents a very high degree of incompatibility, whereas susceptibility is a high degree of compatibility. In the susceptible interaction the fungus and the plant get along for a long period of time, until the organism has worn out its welcome. Only then does injury occur. What's happening chemically in this tissue? Total phenols accumulate in bean seedlings after inoculation with a

compatible or incompatible race of *C. lindemuthianum* or a non pathogen of bean, *H. carbonum*. In the incompatible reaction of the gamma race of *C. lindemuthianum*, however, the build up of phenolic compounds starts about 48 hours after inoculation and proceeds throughout the time interval we used for our study. In the susceptible or compatible interaction, we don't get this build up early, but only when lesions typical of the susceptible interaction start to appear. The susceptible and resistant interaction appear to be doing the same thing. The resistant interaction does it very quickly; the susceptible does it much more slowly. The total phenol accumulation in both reactions is about the same.

Perhaps an analogy here would be a case of smallpox. A person who has never been immunized against smallpox contacts smallpox and is dying, but is producing, just before death, tremendous quantities of antibodies. Unfortunately, the response may be too late. The person who has been immunized can react immediately to smallpox and the infectious agent never gets to do any damage. If we inoculate with *H. carbonum*, a very marked increase in phenols occurs in 12 to 24 hours, and this high level is maintained throughout the test. The compound that we're talking about particularly in this interaction is the compound phaseollin. We're not limited to this compound and we observe a series of compounds that contribute to the resistance mechanism. Phaseollin, one of the antibiotic compounds produced by the green bean in response to infection, and rishitin, which is produced in the potato interaction, are very different compounds. The pattern of increased accumulation of phenols may also hold true for enzymes controlling the synthesis of these compounds.

A possible key enzyme in the synthesis of phaseollins phenylalanine is ammonia lyase. A very marked increase in the activity of this enzyme occurs in the resistant interaction 36 to 48 hours after inoculation. An increase also occurs in the susceptible interaction, but it comes much later. This follows the pattern of phenolic synthesis and accumulation. Therefore, an enzyme, the catalyst for the synthesis of these compounds, very closely follows the pattern for accumulation of the compounds themselves. A very marked increase in the enzyme's activity is evident 12 hours

after bean seedlings were inoculated with *H. carbonum*. Activity returns to the level in uninoculated tissue 12 to 24 hours later. Several compounds, other than phaseollin, accumulate in tissue inoculated with *H. carbonum*.

Establishment of Disease Resistant Strains

It appears a single compound is not responsible for disease resistance, but rather a spectrum of compounds is involved. They may be related, but they are different. This helps explain why in nature resistance is the rule and susceptibility is the exception. When we use a fungicide to control disease, we very often pick up resistant strains because we're dealing with a single compound. The chances of mutating around a single compound are infinitely greater than the chances of mutating around a series of compounds that are produced in a natural resistance mechanism.

If the things I've said about disease resistance are true, and of course I believe they are, then we should be able to immunize plants. We should be able to protect a bean as we did the potato by inoculating with a race of pathogen which cannot attack that cultivar and protect it against a race that can. We should also be able to use a non pathogen -- inoculate a plant with a non pathogen to protect it against a pathogen.

The bean cultivar, Perry Marrow, is susceptible to the gamma and resistant to the beta race of *C. lindemuthianum*. Seedlings inoculated with the gamma race show large coalesced lesions. Seedlings first inoculated with the beta race and 24 hours later inoculated with the gamma race are absolutely protected. Therefore, by inoculating with the race that cannot attack before we inoculate with the race that can, we can protect the bean. A very high degree of protection against a race of the pathogen that can attack is induced by prior inoculation with *H. carbonum* or an *Alternaria* species, both non pathogens of bean.

We can do the same thing with a reciprocal race differential using a cultivar susceptible to the beta and resistant to the gamma

race. After inoculation with the beta race, we see typical lesions. If we inoculate with the gamma race, wait 24 hours, and inoculate with the beta, we get complete protection. We also see a very high degree of protection using the two non pathogens of bean. This can be repeated using leaf tissue and pods.

Animals are immunized against infectious agents by using the attenuated agent; i.e., the infectious agent is treated so that it is unable to function in causing disease but still is able to induce the immune response in the animal. If the mechanisms for protection in plants and animals are similar, we should be able to do the same thing in our plant system. We've accomplished this by taking advantage of the fact that *C. lindemuthianum* is sensitive to temperatures from 37°C. to 40°C. At these temperatures the bean plant is unharmed but growth of *C. lindemuthianum* is stopped. We inoculated bean plants with a race of *C. lindemuthianum* that can attack and let the fungus develop. The plants were then heat-treated and re-inoculated with the same race. There were no symptoms and the fungus was contained. If we heat treat alone, and then inoculate, we actually increase susceptibility. The protection, therefore, is not due to the heat treatment, per se. It's due to the heat treatment of the fungus in the plant.

How far from the site of induction can we demonstrate protection? Plants discussed earlier were protected by being sprayed with inoculum. To demonstrate distance effects, drops of inducer were placed on hypocotyls at some distance from the challenge inoculum. In some cases, inducer and challenge were applied at the same time; in other cases, inducer preceeded challenge by 12 to 24 hours.

When a time interval of 12 hours separated inducer and challenge, plants were completely protected. It's not that nothing happened, but rather that we observed a typical hypersensitive reaction. A message must have gone from the inducer site to the challenge site -- a distance of 0.5 to 1.0 cm. The message signaled the plant to recognize the compatible race as incompatible.

In the next series of experiments, we put spores of an incompatible race on hypocotyls as little drops. The drops were collected after about 48 hours and fungus removed by filtration through a .22 micron millipore filter. The resulting diffusate was sterile. The diffusate was placed on another hypocotyl and the site inoculated with a race of the fungus that should cause disease. The plants treated this way were completely protected. Diffusates from compatible interactions did not protect. The living organism is no longer needed to induce resistance and we can protect with a chemical factor produced in the incompatible interaction. The chemical inducing resistance is not itself inhibitory to the fungus.

What happens if we inoculate with a non pathogen of bean? Can we do the same thing? The answer is, yes. We can protect with many Colletotrichum species non pathogenic on bean. The picture is not as simple as I've explained it but it becomes even more fascinating when we look at just one other piece of information. Protection with the diffusates depends not only upon the presence of inducer in the diffusate, but also upon the presence of genetic receptor sites in the plant.

Cultivars are protected by diffusates from incompatible reactions only if they are resistant to the race used to prepare the diffusate.

Therefore, diffusate from the incompatible reaction of the beta race on the cultivar, Perry Marrow, resistant to the beta race, will protect Perry Marrow against the gamma race. The diffusate will not protect the cultivar Top Crop, susceptible to the beta race, against the beta race. The reciprocal situation is equally true. Mixed diffusate from the beta race on Perry Marrow and gamma race on Top Crop will protect both cultivars against the gamma and beta races, respectively. Tennessee Green Pod is resistant to the beta and gamma races, but it is susceptible to the delta race of C lindemuthianum. Diffusates from Tennessee Green Pod inoculated with the beta or gamma race protect against the delta race. Diffusate from the beta race on Tennessee Green Pod will also protect cultivars, such as Perry Marrow, resistant to the beta race, against the gamma race. The same diffusate will not protect cultivars, such as Top Crop, against the beta race. The reciprocal situation is equally true.

This is very good evidence that not only a factor for induction is necessary for protection, but a genetically controlled receptor site must be available to get the message. What happens if we inoculate with a non pathogen of bean like Colletotrichum truncatum? All green bean cultivars have the receptor site for resistance to C. truncatum and diffusate from C. truncatum on green bean will protect any green bean cultivar against the beta, gamma or delta race of C. lindemuthianum. In fact, C. truncatum even protects green bean cultivars that are susceptible to all the known races of C. lindemuthianum. The cultivars susceptible to all the known races of C. lindemuthianum have the receptor site for resistance to C. truncatum.

This of course is all very exciting to us because it follows a pattern which follows the genetics of susceptibility and resistance. Once again I emphasize the importance of expressing genetic information for resistance. This doesn't mean that the geneticists were wrong in their thoughts concerning resistance. Indeed, resistance is genetically controlled, as are all things. But the thinking as we see it is that it's the regulatory genes that are controlling the expression of the structural genes that become important in many cases of resistance. In other words, the question is not whether a plant can do something but will it do something. Also, it appears to us that, although there is a great deal of specificity in the genetic make up and response, still the compounds that are produced in response to infection are not highly specific. A different phytoalexin is not needed for each infectious agent. The central factor becomes whether the phytoalexins are produced soon enough and in sufficient magnitude. I hope I've emphasized the similarity between disease resistance mechanisms in plants and animals, how we can induce resistance mechanisms, and the fact that resistance appears based on the expression of genetic information soon enough and in sufficient magnitude.

We are very interested in applying this information to practical disease control. And we've extended these thoughts to other diseases. We've worked with Helminthosporium diseases of corn,

fire blight of pears and apples, apple scab, coffee rust, cucumber scab, and Fusarium wilt of muskmelon. It would be truly exciting to us and most rewarding if we could induce resistance in embryonic tissue and cells in tissue culture. Plants developing from these tissues or cells may have the metabolic switch for resistance permanently turned on. The geneticists of the future may derive their resistant plant stocks from undifferentiated plant cells in a test tube.

Discussion Period

Dr. Duich: If I follow you correctly, can we assume that in any natural plant community, where we have the potential presence of many saprophytes, and obligate and facultative parasites, we might get a natural immunization which could trigger the accumulation of terpenoids. In many situations, therefore, we may get a lack of a disease ephiphototic condition.

Dr. Kuc: I think this is true.

Dr. Duich: I'm pointing at something else. We find that in certain aspects of turf, especially in bentgrass cultures, an observation that seems to tie in very well where we make a high use of fungicides. This involved the use of general type "contact" fungicides to begin with, and particularly where we have concentrated with individual use fungicides and arbitrarily sprayed one strip of bent cultivars with one fungicide and another strip with a second type. Late in the season, particularly with preventative type spraying, we often see a complete breakdown of the effectiveness of the fungicides. We now seem to be seeing this problem even more severely with the use of systemic fungicides. For example, we've recognized for many years that Merion bluegrass has a very high degree of natural resistance to Helminthosporium vagans in the spring. The organism does attack, but it more or less is restricted to a point. We've always considered this to be a good reaction of natural resistance, but when we use the systemic fungicides, we suddenly find that varieties or cultivars which for years and years has been resistant to a pathogen, are all of a sudden very susceptible.

Dr. Kuc: I don't want to mislead you. We've talked about some phenols and terpenoids. Depending on the plants used in our studies, different compounds accumulate. In some cases, there may be terpenoids; in some cases coumarins; in some cases steroid glycoalkyloids; in some cases phenols like chlorogenic, caffeic acid are part of the defense mechanism. In some interactions, several of these compounds may be produced. Not all

organisms will induce resistance, and I don't believe one compound alone explains resistance. Very often, if we're protecting against a Colletotrichum, we use different Colletotrichum spp. that don't attack that variety. With some organisms, we've had very good luck. They've protected against a very broad spectrum of fungi; for example, Alternaria, and different Helminthosporia. It's not unreasonable to think that in the soil immunization against pathogens may occur. When this balance is disrupted, immunization does not occur. However, there are several factors involved with induced protection. One, not all organisms will protect. Saprophytes, in general, are not efficient in protection. There are some organisms that won't germinate on a plant or grow and penetrate into a plant.

Dr. Whalley: Once induced, how long does this resistance last?

Dr. Kuc: The resistance lasts the life of the plant in the area that you've induced it. The drawback, at least with the systems that we've used, is that we induce resistance in the area that we treat. If we treat a section of hypocotyl, that section of hypocotyl will be resistant for the life of the plant. New growth is not protected unless we spray it. We hope that we can get around this problem. We can protect tissue at a distance of about 0.5-1.0 cm from the induction site.

Dr. Whalley: Do you visualize any possibility of spraying crops artificially with an incompatible inoculum to protect them from the development of disease?

Dr. Kuc: Cooperating with the Horticulture Department, we have done this on a practical scale for two years in controlling wilt of watermelons. In Southern Indiana, watermelons are normally transplanted into the field. It's no problem to dip in an inoculum of a non-pathogen of watermelon before transplanting. This technique gave us good results. Survival was increased and the horticulturists were pleased with two years of trials. I think it is very promising to use inoculum which will protect. I think it's even more promising to use some of the chemical factors which will induce resistance.

Dr. Daniel: Earlier you said resistance just moved up the stem a small amount, and now you're saying you take a transplant and dip it, as if it affected the plant for a longer period.

Dr. Kuc: No, the wilt is a root pathogen. This wilt is an organism that gets into the roots. When we dip the root system into the suspension, only the treated roots are protected.

Dr. Daniel: There are new roots?

Dr. Kuc: Yes, new roots are formed. But apparently there is enough of an effect there to protect it for a long enough time to get over a critical period.

Dr. Daniel: Out in nature we often see a disease attack barely start, then the weather changes. Does a slight attack protect against the next attack?

Dr. Kuc: I think under some circumstances it may. Since these are biochemical phenomena, they will be influenced by temperature and other factors that affect metabolic systems. Sometimes, just a stress can induce resistance. We can induce the accumulation of phytoalexins by applying stress. By stress, I mean some fungicides or some antimetabolites, which under higher levels would be toxic, but under low levels will produce a stress and trigger a series of events which will lead to the accumulation of phytoalexins.

Mr. Mayer: Along these same lines, you take a variety and it becomes infected with a disease and maybe 95% of it is wiped out, with a few remaining plants left, then make a selection out of this because you assume these are resistant. But again the disease attacks progeny of these plants. Do you think that maybe we have this triggering effect the first time and it didn't carry over into the progeny?

Dr. Kuc: That's possible. However, the inducing factor and a genetically controlled receptor site are necessary for protection. When crosses are made and a segregating population is

obtained, different genetic complements are inherited and different responses to pathogens are observed.

Dr. Payne: You reported spraying the material onto mature plants. Have you done this and was it effective in immunizing?

Dr. Kuc: Yes. Now again a word of caution. We have not been successful in protecting against a root pathogen by spraying the foliage. We've only protected the tissue that we've treated, but we can spray materials on plants and observe protection.

TECHNIQUES FOR ASSESSING SEEDLING VIGOR AND ESTABLISHMENT AND POTENTIAL APPLICATION IN BREEDING PROGRAMS IN GRASSES

R. Derwyn B. Whalley and Cyrus M. McKell 1/

Introduction

Differences in success of establishment among grass species are well known and have been attributed to differential plant response to such environmental conditions as the permeability of the soil to seedling roots, soil moisture content, low or high temperature, and light intensity. Species that consistently show a rapid germination rate, fast rates of root and top growth, a robust growth habit, or resistance to stress are often referred to as having seedling vigor. Success in seedling establishment may be enhanced, therefore, by either providing favorable environmental conditions or by selecting species that have a high degree of vigor during the seedling stage.

Growth of a new grass plant starts with activation (Mayer and Poljakoff-Mayber, 1963) of the embryo and other seed parts, proceeds through the seedling stage to the period of rapid vegetative growth and, finally, maturity of the established plant (Williams, 1964) (Fig. 1). With the emergence of the radicle, seedling establishment begins and may not be considered a success until the plant has developed an adequate root system and leaf area to sustain a high rate of growth (Fig. 2). Whalley et al. (1966a) consider the seedling stage in three-phases: (1) the heterotrophic stage which occurs from inhibition to the initiation of photosynthesis; (2) the transition stage during which time the seedling obtains complex organic compounds from both photosynthesis

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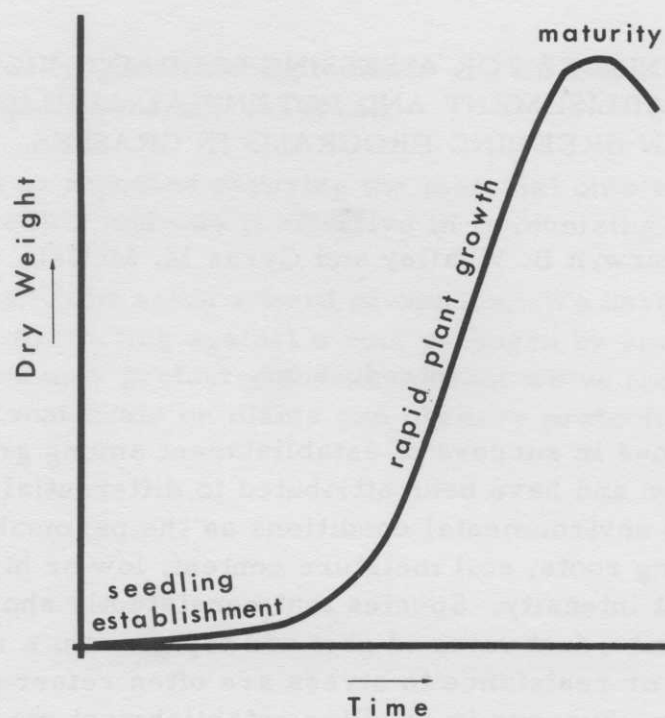


Figure 1. Relative growth curve of barley. Adapted from data of R. F. Williams (1964).

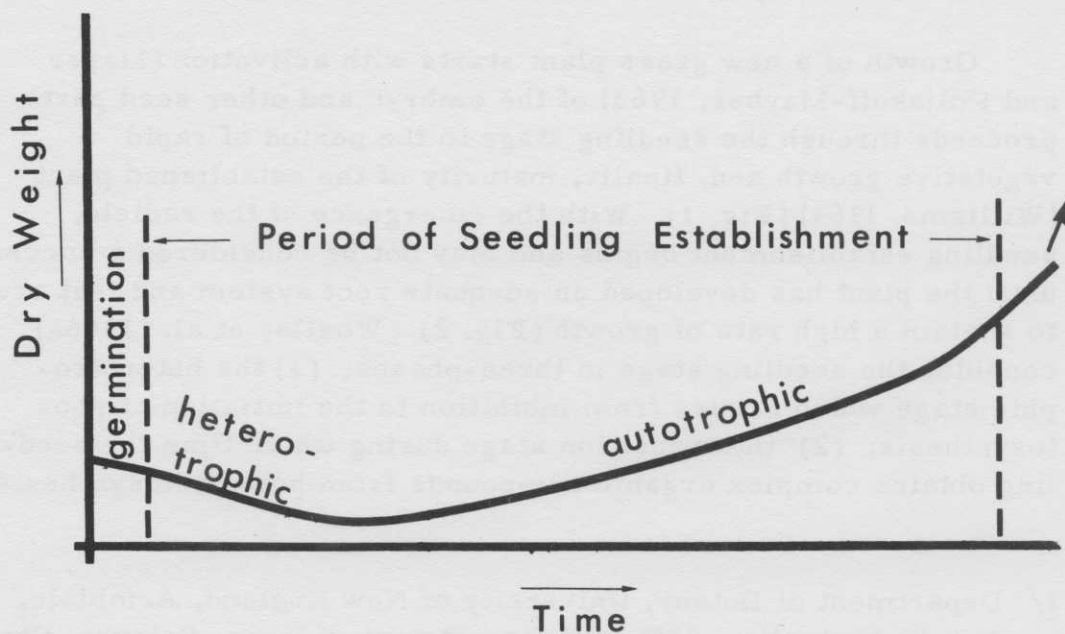


Figure 2. Relative growth curve of barley. Adapted from data of R. F. Williams (1964).

and the remainder of the endosperm, and (3) the autotrophic stage which is completely dependent on its own photosynthetic products. During the establishment stage, intense competition from within the species and from other species may result in the loss of many seedlings, even under favorable environmental conditions. There is no á priori reason why an individual which is efficient and vigorous during the heterotrophic stage should be equally so during the autotrophic stage, apart from the advantages bestowed by a rapid start. With such exceptions as the cereals, members of the Gramineae do not have large seeds with the attendant advantage of extensive food reserves and, as a result, have evolved various seedling characteristics which aid in establishment.

Seedling Vigor

There are two distinct approaches in discussing seedling vigor in the literature. The approach used by seed testers where the objective is to detect and understand reductions in seedling vigor related to adverse conditions of seed harvest, treatment or storage (Heydecker, 1969a). And the agronomic approach where the objective is to understand and improve species and strains with inherently poor seedling vigor.

The general aim in all seedling vigor testing is for quick, convenient tests which have predictive value concerning the field performance of the seed sample tested.

Seed Testing Approach

The necessity for vigor testing, as distinct from germination testing, arises because the field performance of different seed lots may range from excellent to poor despite similar results from standard germination tests (Perry, 1967). The Vigor Test Committee of the International Seed Testing Association (Heydecker, 1969b) attempted to compare a number of vigor tests using three carefully selected lots of pea and six of wheat seeds. A number of cooperating laboratories performed several standard vigor tests. The striking thing about the results was the high correlation between different tests in ranking the quality of the different seed lots.

Agronomic Approach

Most biologists use the term "seedling vigor" in its broadest sense to describe a vigorous growth habit that involves a more rapid size increase than that of competing plants of the same age. Prompt seedling growth, as soon as conditions become favorable, is an obvious advantage toward successful establishment. Seedlings with a fast root and top growth must be supported by efficient enzyme systems with which to mobilize stored food reserves in the endosperm tissue. Rapid transport of soluble sugars to growing points and areas of high metabolic demand is a further necessity for a vigorous seedling.

Seedling vigor is of critical importance in plant competition. The traditional view of Clements (1970) and later of Donald (1963) holds that competition is a physical process of plants drawing from a pool of such factors as air, light, nutrients and water. When the pool is limited or subject to depletion, then the more vigorous seedling that draws rapidly from the pool or can explore a greater volume of the environment will be the successful competitor.

Tests for seedling vigor have been proposed by numerous authors as a means of evaluating or quantifying certain aspects of seedling growth. The predictive value of these tests in terms of later field performance has often not been adequately established. Some of the more recent workers have attacked this problem (Abdullahi and Vanderbilt, 1972; Edje and Burris, 1971; Moutray and Frakes, 1971; Wright and Major, 1971) with variable success.

Characteristics Important in Seedling Vigor

Seed Size and Weight

Seed size and weight are extremely important. Davies (1927) summarized results with commonly used British grasses stating that, "Under normal field conditions, the size of the endosperm is an important factor in determining the potential ability of a species to establish itself." Considerable variation in seed size

exists in seeds produced from the same plant, according to Whalley et al. (1966c). They also reported that commercially harvested seeds of Phalaris tuberosa var. stenoptera may contain a high proportion of immature seeds. Germination percentage and rate were lower in small and immature seeds than in large mature ones. Within species, seed weight is of greater significance to seedling vigor than it is between species or genera. Kittock and Patterson (1962) concluded that, "The closer the genetic background of compared lines, the higher the correlation between seed weight and seedling vigor." This general relationship holds even down to the individual plant (Carleton and Cooper, 1972).

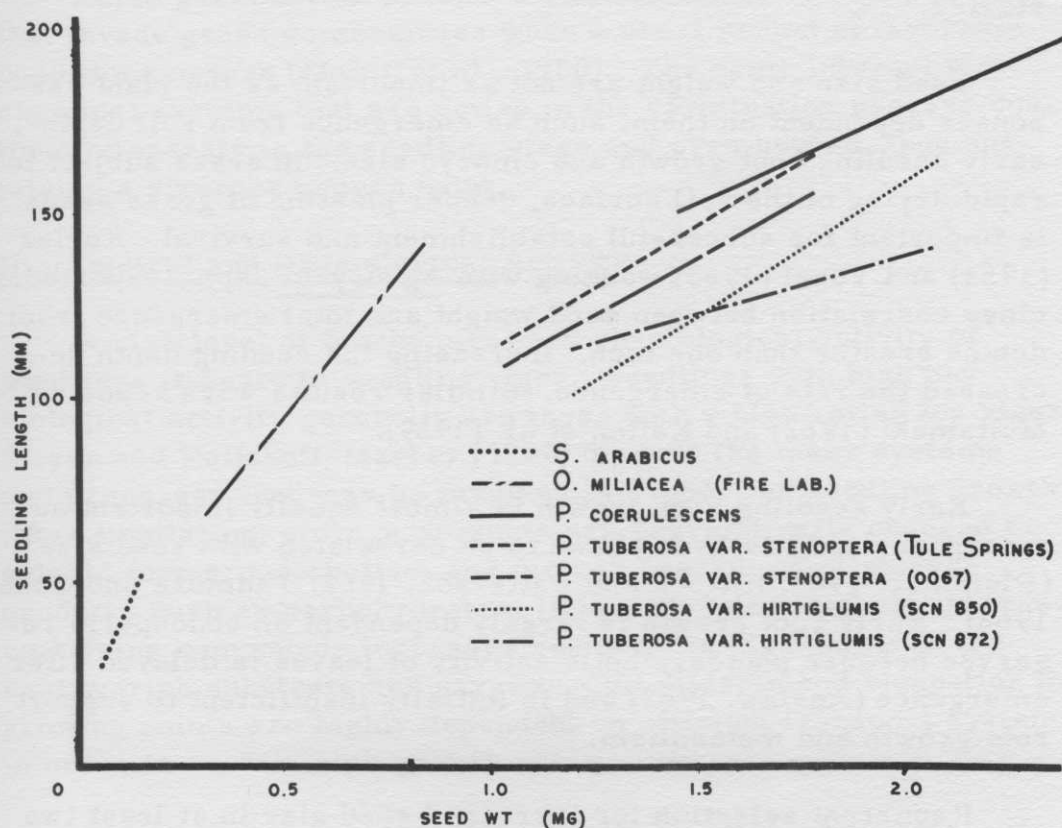


Figure 3. Seed weight in relation to total seedling length. Schismus arabicus and Oryzopsis miliacea show a greater degree of correlation between seed weight and seedling length than do the various Phalaris species and varieties.

In one of the earliest papers published in the Journal of the American Society of Agronomy, Zavitz (1908) reported consistently greater yields from plantings of large well-filled cereal seeds than from small well-filled seeds. Many reports emphasize the importance of a greater volume of endosperm food reserve in relation to subsequent seedling size or rate of growth (Hove and Kleinendorst, 1962; Demivlicakmak et al., 1963; Kneebone and Cremer, 1955). Whalley et al. (1966a) showed that large seeds had longer seedling growth than small seeds of the same species (Fig. 3). One extremely small seeded species, Schismus arabicus, grew at a fast rate but, in the absence of light, was through growing even before radicle emergence of larger-seeded species had started.

Seed size and weight are not as important as the plant responses dependent on them, such as emergence from soil depths, early seedling root growth and embryo size. In areas subject to rapid drying of the soil surface, deeper planting of grass seeds is important for successful establishment and survival. Rogler (1954) and Vogel (1963), working with Agropyron spp., reported a close correlation between seed weight and total emergence from depths greater than one inch. Increasing the seeding depth decreased the rate of emergence. Similar results were reported by Multamaki (1962) and Kalton et al. (1959).

Early seedling root growth is almost equally important as emergence and has been shown to be correlated with seed size (Plummer, 1943; Kittock and Patterson, 1962; Tadmire and Cohen, 1968). Early root growth is largely dependent on endosperm reserves because photosynthetic activity of leaves is delayed after emergence (Anslow, 1962) and is initially insufficient to support root growth and metabolism.

Recurrent selection for increased seed size in at least two perennial grass species, Panicum antidotale (Wright and Major, 1971) and Bromus inermis (Trupp and Carlson, 1971), has lead to increased seedling vigor, as measured by several criteria. There would probably be wide variability in the possible rate of progress in both seed size and seedling vigor with different grass species.

Rapid Germination

Rapid germination is another characteristic that may contribute to a vigorous and successful seedling. Seeds which germinate rapidly have obvious advantages in terms of competition for physical factors of the environment. With a rapidly drying seedbed, this advantage becomes even more important. Horning and Canode (1963) used a vigor score similar to the germination speed index of Maguire (1962) to evaluate smooth bromegrass seed harvested under different conditions. They found a good correlation between the vigor score and other methods of testing seed quality.

Rapid germination is often a characteristic of weedy grasses that invade grass communities when a short period of favorable conditions occurs (Major et al., 1960). The same internal biochemical systems that are active in the germination process continue to operate in the seedling stage and even beyond, thus sustaining a vigorous growth habit.

Biochemical and Physiological Activity

A high level of biochemical and physiological activity of seedlings is basic to seedling vigor. Seedlings with high physiological activity generally are those with a high rating for vigor. Mayer and Poljakoff-Mayber (1963) discuss the many systems and processes that may be involved in supporting seedling growth. After imbibition, storage products are enzymatically changed to soluble forms; metabolism and further breakdown of storage products such as carbohydrates, lipids, proteins, and phosphorus-containing compounds increases resulting in high demands for energy-rich substrate and oxygen. Cell division and elongation in growing points are highly dependent on efficient transport systems in order to sustain high growth rates.

Reports on metabolic rate as an index of physiological activity in vigorous seedlings are conflicting. Mahadevappa (1967) reported that leaf tissues of pearl millet failed to show any differences in the rate of respiration, nor could he find a correlation between respiration capacity and growth rate. Earlier,

Figure 4-A

Conversion of starch to sugars of Phalaris tuberosa var. stenoptera (Tule Springs) which is rated low in seedling vigor.

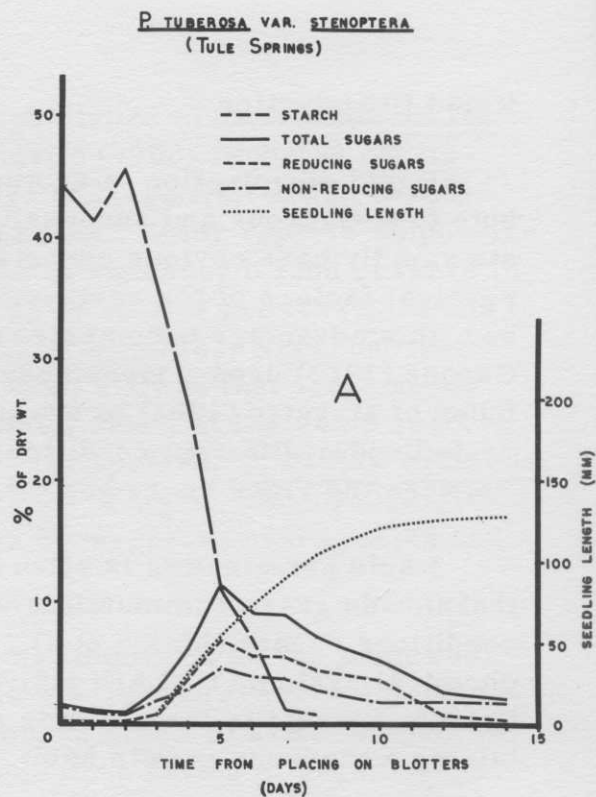
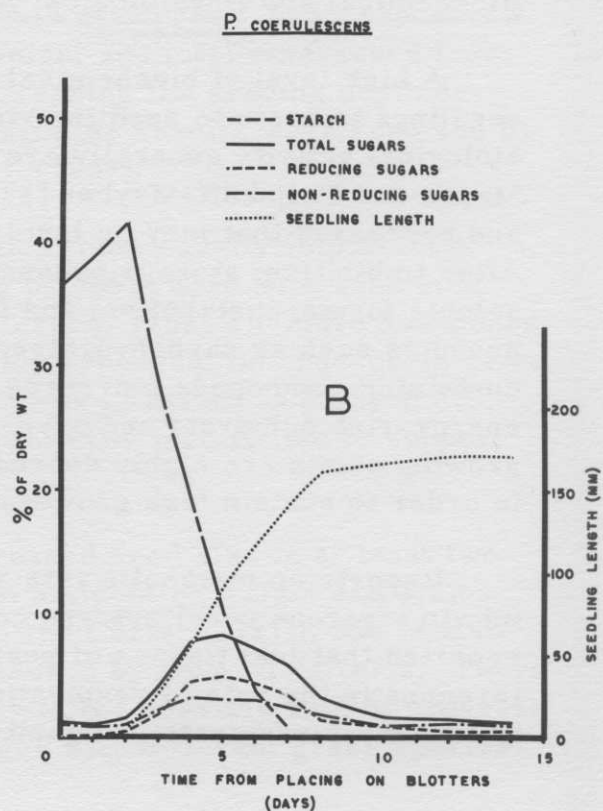


Figure 4-B

Conversion of starch to sugars of P. coerulescens which is rated high in seedling vigor.



Throneberry and Smith (1955) reported essentially the same result with germinating corn seeds. However, Kittock and Law (1968), Woodstock and Feeley (1965) and Cantrell et al. (1972) report close correlations between respiration rates and seedling vigor.

Rapid mobilization of endosperm reserves and transport appears to be another factor in seedling vigor. Whalley et al. (1966b) and Whalley and McKell (1967) show that the degree of availability of sugars or the lack of sufficient transport to the growing seedling may partially account for differences in seedling vigor among several species of Phalaris (Fig. 4). Considerable study is needed before a clear picture can be drawn of the rate of biochemical and physiological activity in vigorous seedlings.

Force of Growth

Force of seedling growth is a valuable plant attribute that may be critical in survival. Where rapidity of emergence depends on a high growth rate of the coleoptile and shoot, the force of emergence may be more dependent on endosperm reserves and a durable epidermis. Kalton et al. (1959) made observations on the relation between seed size and emergence from different depths. Emergence was delayed or reduced with an increase in seedling depth but the difference in seedling emergence between large and small seeds increased. Lawrence (1963) sowed seeds of Elymus junceus at depths of from 0.5 to 1.5 in. and reported a positive correlation between total emergence and seed size. Subsequently, seed yield and forage yield were also correlated.

Rate of Growth

Rate of growth, being one of the most obvious characteristics, is often regarded as evidence of seedling vigor. Both rapid top growth and root growth can be a distinct advantage in seedling establishment. Early achievement of an adequate leaf area supplies the needs of the developing seedling for products of photosynthesis in addition to occupying surface area that would otherwise be used by competing species. Loomis and Williams (1963)

suggest that the time required for attainment of an optimum leaf area and, thus, increased productivity can be shortened by planting more seeds per unit area or by planting seeds that produce a large initial leaf area per plant. Thomas (1966) showed that leaf size is correlated with seed size but that the relationship decreases with time. Shibles and MacDonald (1962) compared two varieties of birdsfoot trefoil which were known for low and high seedling vigor. Even though both varieties were equal in net photosynthetic rate per unit area of cotyledon, the variety known for its high seedling vigor had a higher rate of production of photosynthetic surface, even though seeds of the same size were used.

Root System Development

Rapid development of a root system sufficient to garner water and nutrients for the young seedling is perhaps a higher priority than early top growth, if endosperm reserves in large seeds are still available. Plummer (1943) attributed the success or failure of establishment of twelve range grasses to total root development prior to summer drought. The most successful species, Agropyron cristatum, produced a greater total root length in the seedling stage. Differential response of tops and roots to temperature must be considered. Brouwer (1966) reviewed the literature concerning factors affecting root growth rates and presented information showing that at low temperatures, root growth is favored more than top growth. An increase in temperature results in a burst of top growth giving an increase in shoot root ratio. Other factors such as low nitrogen, moisture stress, and shade also cause low shoot/root ratios.

Cohen and Tadmor (1969) reported large differences among 10 species of grasses and legumes in seedling root growth. Seedling root elongation in the upper soil layer (2 to 12 cm) was greatest at 25°C. Whereas in the deeper layer (12 to 22 cm), 20°C appeared to be the most favorable temperature. Seed size also correlated well with rate of root elongation. Phalaris tuberosa seeds were the smallest in their study and had the slowest elongation rate, whereas the opposite was true for wheat and barley. Low temperatures accentuated these differences. For species

like Phalaris, which is adapted to Mediterranean climates, larger seed varieties must be developed to provide increased vigor of root growth to overcome the adverse effects of low soil temperatures during the moist season. Hyder et al. (1971) also attributed differences in the ease of establishment between blue grama and crested wheat grass to differences in rooting depth.

Resistance to Environment

Resistance to physical and biological environment may be a direct expression of seedling vigor caused by a tough epidermis, an anatomical or physiological makeup that makes a seedling non-susceptible to plant pathogens, or a physical constitution that enables a seedling to withstand mechanical damage of various types. Conversely, resistance as an aspect of seedling vigor may be merely the result of a high rate of growth, the ability to grow at an unusual temperature regime, or the ability to withstand other environmental stresses because of a high level of biochemical or physiological activity. Whatever the case, a resistant seedling may easily be classed as a vigorous seedling if it results in a substantial advantage in establishment. Several studies give examples of the significance of resistance. High temperatures and high humidity prior to germination were shown by Helmers et al. (1962) to eliminate seeds that did not have the potential for high seedling vigor. Disease resistance (Isley, 1957) is another aspect of plant vigor but one which draws on more fundamental seedling characteristics, such as anatomy and physiology. Resistance of reed canary-grass seedlings to flooding or acid conditions was correlated with field performance (Mark and McKee, 1968), thus indicating the importance of this type of vigor in the seedling stage for overall plant performance.

Environmental Conditions and Seedling Growth

Temperature

There is wide variability in the temperature optima of different species and often different requirements in the early and later stages of seedling growth (Cohen and Tadmor, 1969). In

mixed stands, different temperature conditions usually function through competition in that a species favored by the temperature conditions has a competitive advantage over other species. The importance of temperature adaptation should not be overlooked in breeding programs.

Water

Again, species vary widely in their response to water availability during germination and establishment. Easy to establish species, such as annual ryegrass, will germinate at 4-6 bars below the limiting water potential of other more difficult species (McWilliam et al., 1970; McWilliam and Phillips, 1971). Turfgrasses are often established under less than ideal soil moisture conditions and, undoubtedly, selection for germination at low soil water potentials would be valuable. There does not appear to be any relationship between germination at low water potential and drought tolerance of the mature plants (McWilliam and Phillips, 1971).

Fertility

A cultural practice, such as fertilization, is an easy way of increasing the competitive advantage of desirable species, provided the differential responses of the species present are known. Less well understood are the different requirements of species at various stages during seed germination and establishment. Ayeke and McKell (1969) showed that the addition of nitrogen with phosphorus decreased the top and root growth of Oryzopsis miliacea seedlings during the five days from radicle emergence in the dark. With Lolium multiflorum, lower rates of N increased growth rates which then declined at higher rates. Whalley and McKell (unpublished data) have recently shown that crested wheatgrass and Russian wildrye seedlings respond to phosphorus during the early stages of growth, and that a response to nitrogen does not become apparent until two weeks following seedling emergence. There were also some differential species and strain responses.

Table 1. Seeding rates and seedling survival of a dryland perennial grass compared with a common turfgrass (data recalculated to common units and rounded off).

<u>Species</u>	<u>Crested Wheatgrass</u>	<u>Kentucky Bluegrass</u>
Seeding rate	56 gm/100m ² *	1500 gm/100m ² #
No. of viable seeds/gram	346 ⁺	3,570 [#]
No. of viable seeds sown/m ²	200	54,000
No. of seedlings 7 days after sowing/m ²	3*	
No. of seedlings 9 days after sowing/m ²	30*	
No. of seedlings 14 days after sowing/m ²	44*	
No. of plants 12 months after sowing/m ²	25*	11,000 [#]
Percentage establishment	12	20

*Hyder et al. 1955, ⁺Whyte et al. 1959, [#]Musser and Perkins, 1969

It is clear that the responses of grass seedlings to soil nutrient levels during the early stages of establishment are little understood, but are of great importance for the establishment of turfgrasses. Undoubtedly there is scope for breeding work to select strains which can respond to high fertility levels during the establishment phase.

Techniques for Measuring Seedling Establishment

The techniques for measuring seedling establishment have not been studied to the same extent as seedling vigor. However, there are problems involved, particularly with reference to the definition of stand establishment and the time of assessment. The end of seedling establishment could be taken as the end of the lag phase when the seedling has developed a good root system and is commencing its period of rapid growth (Fig. 2).

Few reports are available where repeated seedling counts have been made. Examples of such data are given in Table 1. If seeding rates similar to those used for lawn grasses are

used in a dryland situation, the intraspecific competition is extremely high, and the individual plants develop poor root systems because of intense competition. During a period of stress, the whole stand is likely to succumb. A valid assessment of stand establishment cannot be made until plant numbers are more or less constant with time and should include below as well as above-ground information. There is a need for basic data regarding the shape of the survivorship curves for different species under different situations.

Philosophy of Vigor Testing

In any evaluation of turfgrass breeding material for seedling vigor, the inclusion of species adapted to establishment under range conditions is desirable. Such studies would provide valuable insights to the plant breeder who works with species intended for less demanding environments. In addition, the environment provided by the average amateur lawn grower is usually somewhat less than ideal.

The sequence of events from imbibition to maturity is essentially a continuum. Far too often plant breeders interested in seedling vigor have assessed material at only one or two times. In no case has the predictive value of a single seedling vigor test for seedling performance during the entire continuum been satisfactorily established. Several important tests are summarized below:

Germination Under Water Stress (McWilliam and Phillips, 1970)

The ability to germinate under water stress is undoubtedly important in many situations, and the measurements can be performed on a routine basis.

Rate of Germination (Maguire, 1962)

The speed of germination is easily measured under standard conditions and can be an important characteristic under adverse environmental conditions.

Seed Size (Trupp and Carlson, 1971)

Selection for seed size has been shown to be correlated with early growth rate and is easily measured.

Seed Respiration Studies (Woodstock, 1969)

There appears to be good correlation between early seed respiration rates, R Q values and early seedling growth rates. However, it is essential to measure respiration rates of individual seeds which presents technical difficulties with the small seeded turfgrasses.

Growth Measurements (Heydecker, 1969a p. 206)

The measurement of top and root growth of a mass of seedlings at some point in time after seeding does not give the information required in view of the small percentage of plants which survive to maturity. Such measurements must be on an individual and repeated basis; tedious for plant tops and difficult for plant roots. Further information on survivorship curves in a turfgrass situation are necessary.

Seedling vigor presents considerable problems to the plant breeder because of the many factors involved. Nevertheless, the most promising approach seems to be one based on continual multiassessment of material on an individual basis rather than reliance on any single vigor test.

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Discussion Period

Mr. Gabert: Undoubtedly you've observed many seedling vigor tests in the laboratory. Can you make any comments on the diseases or fungi that are apparently growing on the ungerminated or, in some cases, the germinated seeds? Also, the corresponding events which occur immediately following or preceeding your observations in terms of whether these fungi caused seed deterioration or whether the fungi came as an after effect or saprophyte?

Dr. Whalley: As you know, there are a large number of microflora spores and bacteria on the outside of seeds. Many seeds also carry a heavy endogenous microflora inside the endosperm. I've noticed with crested wheatgrass and also with Russian wild-rye that these endogenous bacteria can become very important. If you measure the respiration rate of one of these seeds, you may find a high respiration rate. If you then put the seed in a petri dish and let it germinate, it may grow fast for a day but then the testa breaks open and a mass of bacteria comes out and growth stops. Surface sterilization does not affect this endogenous material. I feel in certain circumstances some seed lots carry a very heavy endogenous microflora and these are undoubtedly important in the field.

The cold test for corn, which is used as a vigor test, is concerned with resistance to attack by microorganisms. All the test does is to see how resistant a seed lot is to saprophytic and pathogenic microorganisms present in the seed or soil. Undoubtedly, I think the microorganisms are most important, and perhaps one could select the species and strains with a low endogenous endosperm microflora.

Dr. Long: After acid scarification and you go into a standard germination test in petri dish, you notice a great deal more fungal activity. We're wondering whether one alters the endosperm, or change the carbohydrate picture here to where carbon material is in a more available form; thus, encouraging the microorganisms?

Dr. Whalley: Probably. Some of the resistance to saprophytic and facultative parasitic microorganisms is related to the leakage of carbohydrates from the endosperm of the seeds during germination. You can show this very clearly with corn. If seeds are germinated in ordinary petri dishes, the water from the filter pad can be analyzed for sugars during the test. Seed lots which do well in the cold test usually have low sugars in the solution, low leakage from the endosperm and, therefore, less energy substrate for the microorganisms. I would suggest that the acid treatment in some cases increases the leakiness and, as a result, the growth of fungi and bacteria.

Dr. Long: In a turf seeding environment, if a rapid drying seedbed is our major problem, what single test might be most indicative of getting to something that would apply?

Dr. Whalley: If seedbed drying is important, I would screen breeding material for two things -- rapidity of germination and germination under moisture stress. Both of these are relatively easy tests to do. The rapidity of germination is simply a matter of counting the number of seeds germinated each day in a standard germination test. The germination under water stress requires a little more equipment but is possible on a routine basis. If you arrange an appropriate membrane with a thin layer of soil above it (less than a centimeter thick) and adialysed solution of polyethylene glycol (20,000 molecular weight) below, the soil may be kept at say -5, -10 and -15 bars water potential for the duration of a germination test. Breeding material can then be screened for percent germination at different water stresses.

Dr. D. Taylor: Have you any additives which may speed germination of seeds, or have you run across any inhibitors? The reason that I ask is that if you're planting a mixture and one component seems to dominate, can you slow down or speed up the germination and development of any one component?

Dr. Whalley: I don't know of any growth regulators or compounds that will speed up germination or slow it down in the way that you asked. The only ones that I am aware of are those which either break or induce some form of seed dormancy.

Dr. Fisher: With respect to your lack of uniformity in your similar corn seeds, one of our plant breeders brought me some oat seed that he had separated into the primary seed in the spikelet and the secondary, which were screened for identical weights. Yet, the primary germinated and grew more rapidly than the secondary. When these were sectioned, we found that invariably the primary seed had one more foliar ridge in the embryo than the secondary one. This shows up very markedly in oats, which is a panicle grass. I suspect in bluegrass it must be very important.

Dr. Whalley: Yes, I entirely agree with you. There has been some work where people have either by manipulation or selection obtained seeds of similar total size weight, but bigger and smaller embryos, and found exactly the same effect that you mentioned. Those with larger embryos seem to get a better start and grow faster. I don't know how long the advantage continues, or how good the correlation is between embryo size or leaf number and seedling survival or any plant size characteristic at say six or eight weeks after germination. I think there is a poor correlation in crested wheatgrass, but there may be better correlation with other species.

Dr. Yu: For different plant species, how would you define the seedling stage?

Dr. Whalley: The development of a plant from the time of seed imbibition to the time of seed production is essentially a continuum. For convenience one tends to divide this continuum into a number of stages, but I think it is important to remember that these subdivisions are essentially arbitrary. Therefore, the end of the seedling stage is when you define it. Because of my interest in seedling survival, I would perhaps use a definition based on mortality rates.

Dr. Fisher: I would say that the seedling stage is over when the apical meristem makes its first new foliar ridge. But that's arbitrary.

Dr. Whalley: Yes.

Dr. Long: I think it would be interesting to have Dr. Nittler comment on his observations in terms of varieties as to germination rates in relation to nutrients.

Dr. Nittler: I haven't really studied this. I do know that some varieties have much larger seeds than others. I'd be interested if somebody would run a correlation between the seedling establishment and varieties that have different seed size.

Dr. Long: Did you notice any difference in your work when you had minus calcium versus calcium? Did some germinate? Also, did the rate of seedling growth rate vary?

Dr. Nittler: No, I haven't really noticed any difference, though we get poor germination with some solutions. It varies with seed lots sometimes. We tested several samples of Windsor and found that some samples that germinated poorly otherwise will germinate relatively well if potassium nitrate is used. But we haven't correlated this with seedling growth.

Dr. Payne: You had a calcium-nitrogen imbalance there that was causing a differential?

Dr. Nittler: Yes, I think this is part of it.

Dr. Madison: This is not a question, but a commentary. I have looked at Penncross from the standpoint of competition affecting establishment. After 14 phytochrons, my weaker plants under the strong stress of competition had made 14 phytomers, of which 4 were still functioning. At the other extreme, with no competition, I've had vigorous plants with over 1,000 phytomers after 14 phytochrons, and with as many as 1,400 phytomers. In these extremely vigorous plants, the prophyll has a bud which produces a second prophyll and second branch within the first prophyll. This observation shows a characteristic of the extremely vigorous plant; the prophyll has a bud.

PENNFINE PERENNIAL RYEGRASS DESCRIPTION AND PERFORMANCE CHARACTERISTICS

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Introduction

Under the provisions of the 1970 Plant Variety Protection Act, the problem of providing an objective description of a novel variety has become most critical. The absence of refined biochemical and associated techniques still necessitates reliance on traditional morphological and use performance data.

This paper deals with the methods used to describe Pennfine perennial ryegrass (Lolium perenne L.) and is intended as an aid to others facing the problem of providing varietal descriptions.

Origin and Breeding History

Pennfine ryegrass is a nonfluorescent turf-type perennial ryegrass developed as a three-clone synthetic variety at The Pennsylvania Agricultural Experiment Station (Duich, Perkins, and Cole, 1972). All three clones were selected in Southeast Pennsylvania; two are golf course fairway selections, and the third was found on a tennis court.

Original seed sources are unknown and the areas were seeded and reseeded as turf for approximately 40 years. All clones in the ryegrass program required a spread of 12 inches minimum, and the clones in question ranged from 16 to 28 inches. This fact leads one to estimate an age of at least 15 years under close-cut turf selection pressure.

Breeder's selection involved texture, mowability, winter survival, snowmold tolerance, and heading and anthesis dates. Prior to turf quality testing, uniformity of heading and anthesis were also confirmed in two years of testing in Oregon under commercial production conditions.

The Pennfine variety shows a high degree of uniformity for a three-clone synthetic. Uniformity testing involved two generations of space planted and turf trials. Two Foundation growers failed to detect distinctive variants in 40 acres of production in Oregon and Washington. Observations to date indicate Pennfine to be a relatively stable variety on the basis of morphological characters and turf response.

Commercial production will involve three generations: Breeders, Foundation and Certified. Breeders' seed will be produced at University Park, Pa., and others in the Pacific Northwest states. Commercial seed will be in the "certified only" category, with seed failing to meet certified standards not eligible for variety name.

Mature Plant Comparisons

To compare plants of perennial ryegrass, measurements were made in late June 1971 of fifty plants each of six cultivars of perennial ryegrass growing in Western Idaho on the Rathdrum Ranch of the Jacklin Seed Company. The ranch is located approximately 20 miles east of Spokane, Washington. Each plot of ryegrass was approximately 5 m long by 2 m wide. Plots had been fertilized and irrigated as was customary for seed production in this area. All plants measured had headed and anthesis had occurred. Seed would probably have been ripe in 10 to 14 days. Plots had been established in mid-September 1970 to evaluate ryegrass cultivars under environmental conditions for commercial seed production.

Measurements were made of flagleaf size, head length, internode length and head weight. Data were analyzed by analysis of variance and significant differences delineated by Duncan's modified (Bayesian) least significant difference test.

Head weight

To compare head weight, five replications of 15 heads each of six cultivars of perennial ryegrass were weighed to 0.01 g. The heads were air-dried and trimmed, with stems below the lowest floret removed. Heads of Pennfine weighed the least and those of Norlea, the most (Table 1). The coefficient of variability for this measurement was 6.2%.

Head length

To compare head length, 50 heads of each of six cultivars of perennial ryegrass were measured to the nearest millimeter. The heads had been air-dried. Measurements were made from the tip of the head to the internode below the lowest floret. Heads of Pennfine were the shortest and those of Norlea, the longest (Table 2). The coefficient of variability for this measurement was 14.8%.

Flagleaf

To compare size of the flagleaf in six cultivars of perennial ryegrass, measurements of the flagleaf length and width were made on 50 plants each of six cultivars of perennial ryegrass. Flagleaf width to the nearest 0.1 mm was measured at a spot 4 mm from the basal end where the leaf joined the leaf sheath. Measurements were made with a 10X magnifying glass equipped with a reticle calibrated to 0.1 mm. Flagleaf length was measured to the nearest 0.5 mm using a steel metric tape. Length measurements were made from leaf tip to a point where the leaf and leaf sheath joined; i.e., on a line with the ligule. The flagleaf of Pennfine was by far the shortest and narrowest, while that of Norlea was the longest and widest (Table 3). The derived measurement LW accentuated leaf measurement differences so that all six cultivars were significantly different, one from the other. In contrast, differences in the ratio of flagleaf length to width (L/W) were not as diagnostic of cultivar differences as measurements of flagleaf length, width and LW. The coefficients of variability for measurements of flagleaf length and width were 17.6 and 14.2%, respectively. For the derived quantities LW and L/W , the coefficients of variability were 25.9 and 19.2%, respectively.

Internode length

To compare internode length, measurements to the nearest millimeter were made of the first (basal) six internodes of the ryegrass head. Internode length was measured to the nearest millimeter. For each internode, Pennfine was the shortest. Internodes of Linn were either the longest (internodes 2, 3) or not significantly different from the longest (internodes 4, 5). The coefficients of variability for internode length ranged from 21.3 to 25.7%.

The ratios of one internode to another were also compared (Table 5). Some varieties differed significantly ($p = .05$) from others in the ratios of each internode length to internodes 1 and 2. Ratios of the length of each internode to internodes 3, 4, 5 and 6 were generally nonsignificant. The coefficients of variability for the length of each internode to internode 1 varied from 17.3 to 28.6%; for the ratios to internode 2, from 46.6 to 68.0%, and for the ratios to internode 3, from 67.8 to 102.3%.

Ranking of six cultivars from least to greatest for plant characteristics, the relative ranking was: Pennfine, 12; Manhattan, 29; Linn, 43; Pelo, 52; Norlea, 55, and NK-100, 58 (Table 6). Thus, as the above data indicate, Pennfine has smaller flagleaves, shorter and lighter heads, and shorter internodes than the other five cultivars evaluated.

Relative maturity

Observations June 3 and 16, 1971, indicated that with regard to relative maturity and head emergence, Pennfine and Linn would be classified as early, NK-100 as medium, Norlea as medium late, and Pelo and Manhattan as late (Table 7).

Seed characteristics

No discernible characteristics of Pennfine ryegrass seed have been detected to date, but work is continuing along these lines. Seed size, number and weight appear to be appreciably smaller and lighter than Linn, and similar to Manhattan. These data cannot be finally assessed until a representative number of samples are obtained from production areas to determine location and year variables.

Trial Performance

Pennfine ryegrass was developed for turfgrass use. Turfgrass trials initiated in 1965 at University Park, Pa., compared various cultivars under acceptable maintenance procedures. Seedlings were made at 4 pounds per thousand and maintained at 1 1/4-inch cutting height. Fertility was at the moderate level of 3 pounds of nitrogen per thousand. Irrigation was applied at wilt stress levels only. Chemical applications were limited to grub treatment with aldrin and annual applications of phenoxy compounds for broadleaf weed control.

Turfgrass performance data in University Park trials are shown in Tables 1 to 4. All evaluations for density, mowing quality and disease were made by two to four observers. Seedling growth was measured and compared to the quick growing annual as the standard in four mowings following emergence. Leaf width measurements were made on 50 first and second leaves below budleaf per plot with a micrometer. Fluorescence determinations were made at The Pennsylvania Department of Agriculture Seed Laboratory.

Density

Pennfine and Manhattan ryegrasses persisted at the highest density levels among nine cultivars over a 5-year period (Table 8). Utilizing a 1 to 10 rating scale with 10 = best, Pennfine and Manhattan exceeded 8.5 for six evaluating periods, and out-performed all other entries.

Mowing quality

A distinguishing feature of Pennfine is the mowability or relative absence of foliar shredding following mowing with a reel mower (Table 9). This is a well-recognized problem of ryegrasses during high temperature periods. With one exception, Pennfine maintained a mowing quality above 8.0 for 5 years, averaging 8.3. Other cultivars ranged from 2.6 to 7.3.

Disease tolerance

Natural disease infections of epidemic magnitude included snowmold, Fusarium roseum (foliar), leaf rust and Helminthosporium (species unidentifiable by culture) (Table 10). Of nine

cultivars, Pennfine resistance ranked first for snowmold, third for Fusarium, fourth for rust, and first for Helminthosporium.

Additional snowmold and winter survival data were obtained from forage plot seedlings at the U.S. Pasture Laboratory (Table 11).

Fluorescence, vertical growth and texture

All cultivars were analyzed for purity, germination and fluorescence by The Pennsylvania State Seed Laboratory (Table 12). Pennfine, Manhattan, Pelo, Norlea and Combi showed zero fluorescence, whereas other perennials ranged from 3.5 to 5.0%.

Four measurements following seeding in 1965 showed Pennfine had the least vertical growth, averaging 40% of annual ryegrass. Manhattan averaged 50%, and other cultivars ranged from 60 to 80%. Low vertical growth is very desirable, particularly in terms of competition and mowing frequency when ryegrasses are used as companion grasses with slower developing species, such as bluegrass.

Leaf-width measurements during the second growing season (1967) showed Pennfine had the finest texture of the nine cultivars. Pennfine averaged 2.5 mm for the first mature leaf, and other cultivars ranged from 2.7 to 5.6. The variety name was suggested by this textural character.

Overseeding

Varietal distinction is supported by bermudagrass overseeding trials at State College, Miss., and College Station, Tex., as shown in Tables 13 and 14, respectively. Both stations report a unique high temperature persistence during the latter part of the season. Pennfine has also persisted under high temperature conditions in Southern California, as noted in a personal letter from Stan Spaulding, University of California, South Coast Experiment Station, Santa Ana.

References Cited

Duich, J. M., A. T. Perkins, H. Cole. 1972 Registration of Pennfine Perennial Ryegrass. Crop Sci. 12:257.

Table 1. Comparative weight of 15 heads of six cultivars of perennial ryegrass.

Variety	Weight, g
Pennfine	1.50 a*
Manhattan	2.05 b
NK-100	2.19 bc
Linn	2.23 bc
Pelo	2.56 c
Norlea	3.27 d

*Means followed by the same letter do not differ significantly at $p = .01$ by Duncan's modified (Bayesian) least significant difference test.

Table 2. Comparative head length of six cultivars of perennial ryegrass.

Variety	Head length, cm
Pennfine	16.73 a*
Linn	19.72 b
Manhattan	23.44 c
Pelo	25.05 d
NK-100	25.93 d
Norlea	30.22 e

*Means followed by the same letter do not differ significantly at $p = .01$ by Duncan's (Bayesian) least significant difference test.

Table 3. Comparative size of flagleaf of six cultivars of perennial ryegrass (plants headed).

Variety	Length (L), mm	Width (W), mm	LW, mm ²	L/W
Pennfine	138.6 a*	4.29 a	603.58 a	32.66 a
Linn	175.9 b	5.38 b	949.87 b	33.47 a
Manhattan	206.7 c	5.29 b	1102.12 c	39.60 b
Pelo	255.8 d	6.26 c	1610.99 e	41.11 bc
NK-100	262.5 d	5.52 b	1460.18 d	47.96 d
Norlea	282.2 e	6.55 c	1858.21 e	43.72 c

*Reading vertically for each comparison, means followed by the same letter do not differ significantly at $P = .01$ by Duncan's modified (Bayesian) least significant difference test.

Table 4. Comparative internode length of heads of six cultivars or perennial ryegrass.

Variety	Internode number					
	1	2	3	4	5	1
	mm					
Pennfine	24.0 a*	20.2 a	15.6 a	13.6 a	12.1 a	11.1 a
Manhattan	25.0 a	21.6 a	17.1 a	15.4 b	14.1 b	12.4 ab
Norlea	28.2 b	25.2 b	17.5 a	16.9 bc	14.0 b	13.6 b
Pelo	31.8 c	27.5 b	20.3 b	18.6 cd	14.8 bc	13.2 b
NK-100	32.4 c	26.8 b	21.3 b	20.0 d	17.5 d	15.6 c
Linn	34.5 c	27.7 b	21.6 b	17.9 c	16.1 cd	13.2 b

*Means followed by the same letter do not differ significantly at $p = .01$ by Duncan's modified (Bayesian) least significant difference test.

Table 5. Comparative internode ratios, heads of six cultivars of perennial ryegrass.

Variety	Internode ratio				
	2/1	3/1	4/1	5/1	6/1
Linn	.82 a*	.64 a	.54 a	.49 a	.39 a
NK-100	.84 ab	.67 a	.63 b	.56 bc	.50 c
Pennfine	.85 ab	.60 a	.58 ab	.52 abc	.47 bc
Manhattan	.87 ab	.69 a	.62 b	.57 c	.50 c
Pelo	.88 ab	.60 a	.60 b	.49 a	.43 ab
Norlea	.90 b	.63 a	.62 b	.50 ab	.50 c

	Internode ratio				
	1/2	3/2	4/2	5/2	6/2
Linn	1.25 a	.79 a	.66 a	.59 ab	.48 a
NK-100	1.45 a	.96 a	.85 b	.79 b	.68 b
Pennfine	1.20 a	.78 a	.68 a	.61 ab	.56 ab
Manhattan	1.17 a	.80 a	.72 ab	.66 ab	.59 ab
Pelo	1.18 a	.76 a	.70 ab	.56 a	.50 a
Norlea	1.18 a	.73 a	.69 a	.60 ab	.55 ab

*For each ratio comparison, cultivar means followed by the same letter do not differ at $p = .05$ by Duncan's modified (Bayesian) least significant difference test.

Table 6. Comparative rank order for 12 characteristics of six cultivars of perennial ryegrass.

Variety	Head		Flagleaf				Internode length					
	Width	Length	Length	Width	LW	L/W	1	2	3	4	5	6
Pennfine	1*	1	1	1	1	1	1	1	1	1	1	1
Manhattan	2	3	3	2	3	3	2	2	2	2	3	2
Linn	4	2	2	3	2	2	6	6	6	4	5	4
Pelo	5	4	4	5	5	4	4	5	4	5	4	3
Norlea	6	6	6	6	6	6	3	3	3	3	2	5
NK-100	3	5	5	4	4	5	5	4	5	6	6	6

*Ranked in ascending order from least (1) to greatest (6).

Table 7. Comparative time to head, six cultivars of perennial ryegrass.*

Variety	Reproduction status		Relative maturity
	June 3	June 16	
Linn	headed		early
Pennfine	headed		early
NK-100	starting to head		medium
Norlea		headed	medium late
Pelo		starting to head	late
Manhattan		starting to head	late

*Based on observations by Jacklin Seed Company personnel.

Table 8. Ryegrass turf evaluation -- density, University Park, Pa.

Variety	Density rating*					
	7/15/66	9/16/66	7/27/67	8/29/67	9/4/68	8/9/69
Pennfine	8.8	9.0	9.1	8.9	9.0	8.8
Manhattan	8.7	8.6	9.1	9.0	9.0	8.7
NK-100	8.0	7.0	8.3	7.8	7.0	6.5
Pelo	8.2	7.3	8.7	8.7	7.3	7.2
Norlea	7.7	5.8	7.7	6.8	6.2	6.3
Branbantia	8.3	7.3	8.0	8.2	7.2	6.0
Combi	9.0	8.0	8.0	8.3	7.3	6.2
Linn	6.7	6.8	7.7	7.0	5.7	5.3
Annual	1.7	1.0	1.2	---	---	---
LSD .05	1.1	1.3	1.6	1.4	1.7	1.9

*Scale 1 to 10; 10 = best.

Table 9. Ryegrass turf evaluation -- mowing quality, University Park, Pa.

Variety	Mowing quality*					
	7/15/66	8/11/66	8/27/67	7/28/68	8/9/69	Average
Pennfine	9.0	8.0	8.2	8.3	7.8	8.3
Manhattan	8.0	7.0	7.5	7.7	6.2	7.3
NK-100	4.0	3.0	4.5	5.7	4.7	4.4
Pelo	5.3	3.3	4.5	6.3	6.7	5.2
Norlea	8.0	5.7	5.5	7.0	6.7	6.6
Branbantia	6.0	4.3	5.5	6.7	4.7	5.4
Combi	6.0	3.3	5.0	6.3	4.7	5.1
Linn	4.7	2.3	2.0	2.0	2.2	2.6
Annual	2.0	---	---	---	---	---
LSD .05	1.2	1.9	1.3	1.4	1.3	

*Scale 1 to 10; 10 = best.

Table 10. Ryegrass turf evaluation -- disease, University Park, Pa.

Variety	% snowmold		F. <u>roseum</u> *	rust*	<u>Helminthosporium</u> spp.*	
	1966	1970	1966	1967	1968	1969
Pennfine	6	2	3.3	2.0	1.7	2.1
Manhattan	8	8	2.0	1.2	6.0	7.3
NK-100	7	37	6.0	2.0	5.0	6.5
Pelo	8	13	5.0	2.0	5.0	6.2
Norlea	10	20	6.3	7.0	4.0	4.5
Branbantia	12	—	6.0	1.5	3.7	4.3
Combi	10	30	5.3	3.0	4.3	4.0
Linn	10	43	5.0	1.0	2.3	1.5
Annual	55	99	2.0	--	--	--
LSD .05	13	17	1.4	2.1	2.0	2.2

*Scale 1 to 10; 1 = least.

Table 11. Winter survival as influenced by snowmold and winter-kill under forage plot conditions, U. S. Pasture Laboratory, University Park, Pa. 1971-72.

Variety	Nitrogen Kg/Ha	% Survival April 1971
Pennfine	50	100
"	100	100
Norlea	50	90
"	100	90
Tetraploid	50	70
"	100	100
Linn	50	35
"	100	70
		<u>April 1972</u>
Pennfine	0	99
"	50	67
"	100	43
"	200	50
Norlea	0	94
"	50	74
"	100	55
"	200	46
Tetraploid	0	43
"	50	13
"	100	11
"	200	1
Linn	0	54
"	50	9
"	100	4
"	200	3

Table 12. Ryegrass turf evaluation -- fluorescence, vertical growth, and leaf width, University Park, Pa.

Variety	% fluorescence	Seedling vertical growth	Leaf width, mm	
			Leaf 1	Leaf 2
Pennfine	0	40	2.5	2.6
Manhattan	0	50	2.7	2.7
NK-100	5.0	80	2.9	2.9
Pelo	0	80	3.1	3.1
Norlea	0	60	3.1	3.1
Branbantia	3.5	70	3.6	3.6
Combi	0	80	3.3	3.4
Linn	4.0	80	3.8	3.8
Annual	91.0	100	5.6	5.8
LSD .05	--	15	0.5	0.5

Table 13. Ryegrass overseeding evaluation, State College, Mississippi, 1969-70.

Variety	Winter turf quality evaluations*							
	11/12	12/18	1/13	2/11	3/26	4/9	5/5	6/5
Pennfine	6.5	6.7	6.7	5.2	8.7	6.7	4.0	7.0
Manhattan	7.2	7.0	7.2	6.2	7.7	6.7	1.7	6.5
Medalist II	6.6	6.0	6.7	4.0	5.5	4.5	1.7	4.5
Annual	7.7	7.5	7.0	2.7	5.2	6.0	1.5	3.5
Magnolia	8.2	6.5	6.5	3.0	5.5	4.7	1.0	3.0

*Rating scale: 1 to 10; 10 = best, 5 = acceptable turf.

Table 14. Ryegrass overseeding evaluation, College Station, Texas, 1969-70.

	Days to emergence	Color	Avg. putting surface*	Average transition*	<u>Poa annua</u> competition
Pennfine	5-6	med to dark	1.2	2.0	good
Manhattan	7	med	1.1	2.0	fair
NK-100	5	light to med	1.6	3.1	good
Linn	5	light	1.9	3.1	good
Pelo	5	light to med	1.9	2.4	good
Medalist II	5	light to med	1.3	3.0	good
Annual	3	light	2.4	3.3	good

*Scale: 1 to 5; 1 = excellent, 5 = unacceptable.

SEEKING DWARFNESS IN BLUEGRASS

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Introduction

In looking at the bluegrass breeding, some of the interesting questions are: How infrequently could one mow? How dwarf would you like turf to be? And, of course, the most basic question -- how do you get it?

Another question is, of course, spread. We can get spread of 4 to 8 times Merion, the standard in tests. In certain places we have a need for vigor; therefore, we need things that produce a lot of spread, fill-in and recovery for athletics, parks, roadsides, poor nutrition areas and similar places. One way to measure spread is by individual clone spread as inches, meters and ratings.

Seedling Vigor

Another thing we have talked about a great deal is seedling vigor. Merion bluegrass is slower than many. Park claimed seedling vigor. We also know bluegrasses vary in how their leaves angle out along the stems. We have tended to turn our backs on the common types and those that tend to have a narrow leaf angle, as well as initial leaf emergence high above the ground.

In 1962 we began to notice dwarfness in certain plants. We have collected for ten years. There are over 100 variations which came from one plant chosen in 1962. In the greenhouse, leaves unmowed for one year varied from 4 to 20 inches. We were pleased with the spread of some of the plants. Several dwarfs produce less seedheads down very low, quite late in seed production.

Leaf Height

We have kept about 50 of the better dwarfs with leaf height of less than 3 inches. They are usually slow in spread and many of them tended to fail. You want something that has rhizome spread, that does emerge, fill out, keeps coming, and has the ability to rejuvenate itself. Seedheads have been very sparse on most dwarfs, particularly during the first year of production, with low seedhead height - 20 cm., as an example. During the second year, near normal seed was harvested. Among all bluegrasses in test, seedhead heights vary from 30 inches down to 5 inches. Usually seedhead height is about twice leaf length.

Many of the grass varieties now on the market, or to come on the market, are noted for the ability to produce leaves close to the ground. This is one of the targets of dwarf varieties. The plant with the tendency to develop leaves close to the ground has resulted in more tillers -- more leaves compared to more common types.

Summary

Briefly, we have made several selections, and we are interested in dwarfness. Our question is: How dwarf can it be? And, on top of it - how do you serve the seed grower and the seed processor if you go this route? If one goes vegetative, as with Warren's A-20 or A-34, that is simpler than trying to take it all through the seed field of our normal procedure. Disease resistance has always been important whatever the variety.

Does the dwarfness fit into the future? If so, will it be vegetative? How dwarf can we go as we work towards blending will be an interesting question. Certainly it will take a lot of finesse in management.

THE ORIENTATIONS OF THE RHIZOMES OF KENTUCKY BLUEGRASS

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Introduction

The popular concept of the rhizome (a lax underground stem growing through the soil until its tip eventually turns up to become a new plant) implies a particularly disoriented and disorganized structure wandering aimlessly through the soil without real orientations or controls, apart from those imposed by immovable objects in the soil. This misconception is not merely due to lack of knowledge by the layman, but is implied by the description presented in most botanical textbooks and in one of the most prominent manuals of the grasses. However, studies have shown that the rhizomes of grasses are under pronounced orientational control from the time of their initiation as rhizome buds, until their tips ultimately emerge from the soil and become aerial (leafy) shoots.

For the first time, the orientations of the Kentucky bluegrass rhizome during its life history will be described in their chronological and biological sequence. Some of these observations have been presented previously (Fisher, 1964, 1965, 1966a, 1966b, 1972), while others have not.

The Origin and Initial Orientation of Rhizome Buds

Grasses are not truly bilaterally symmetrical. The point of attachment of each leaf deviates slightly from the plane of bilateral symmetry. One leaf may be displaced in one direction, the next in the opposite, or several consecutive leaves may be displaced in the same direction. This is believed to reflect

their phylogenetic relationship to a primitive three ranked progenitor. We have found no means of predicting the deviation of future leaves not yet initiated. However, once the first cell division in the initiation of a new leaf has taken place, the deviation of that leaf from the plane of symmetry has been determined and this deviation can be observed microscopically.

In Kentucky bluegrass this has proved to be simpler than, for example, the cereal grasses. The apical meristem of bluegrass is elliptical in cross section, not circular. Surmounting the apex and extending down both flanks is a pair of cell tiers, one on each side of the plane of symmetry (Fig. 1; F in Fig. 2). When a new leaf is about to be initiated, a cell undergoes a periclinal cell division, increasing the girth of the apex cross section at that point (angle B, Fig 2). Divisions spread around the apex in both directions, meeting at a point displaced from the line of symmetry on the opposite side of the apex (angle A, Fig. 2). Thus, one side of the leaf primordium is of greater partial circumference than the other. This disparity in size is visible in the mature leaf where "half" of the blade is slightly wider than the other. It is this deviation from the line of symmetry of the initiating leaf that determines the deviation from symmetry of the positioning of the axillary bud from which a rhizome (or a tiller) develops.

The bud primordium arises from cell divisions deep within the base of the leaf primordium (Fig. 3). Therefore, the bud develops from tissue giving rise to the leaf above. It is positioned, circumferentially, at the point where the two semi-circles of the developing leaf primordium came together; that is, on the side opposite the future position of the mid-rib of the leaf above (Fig. 2). The concept that a bud arises in the axil of the leaf immediately below the bud is convenient geographically, but biologically the bud is most closely related to the leaf above, whose blade will be on the opposite side to the bud (Sharman, 1942, 1945, 1947; Etter, 1951). This unit of structure, which ultimately consists of the leaf above, a section of stem (the internode), and a bud at the base of the internode on the side opposite the mid-rib of the leaf above, has

Figure 1. Top view of the living apex of Kentucky bluegrass. The lines show the plane of bilateral symmetry and point to the paired tiers of cells, one tier on either side of the plane of symmetry.

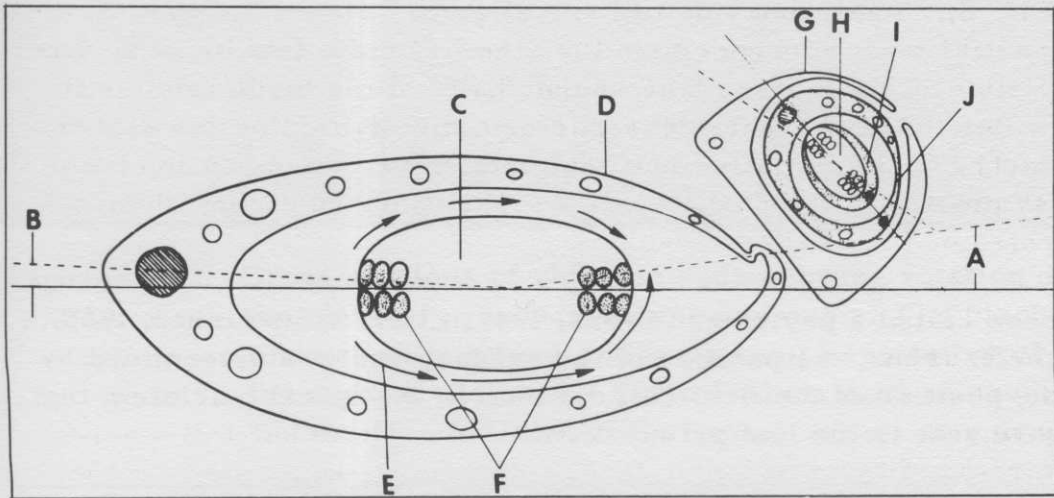
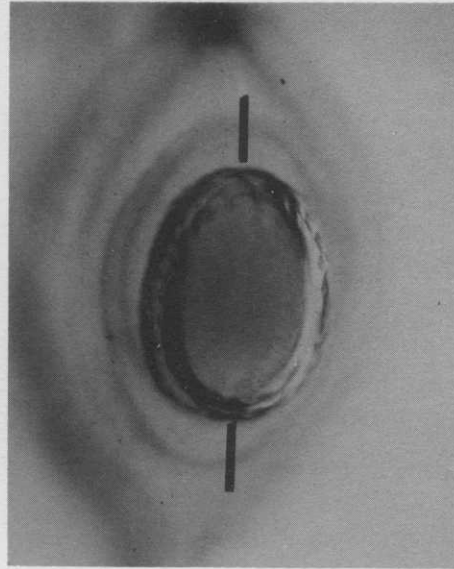


Figure 2. Diagrammatic representation of a bluegrass serial shoot, viewed from above: (A) angular deviation of the leaf sheath from bilateral symmetry; (B) angular deviation of median strand from bilateral symmetry; (C) apical meristem; (D) leaf sheath; (E) arrows represent rates of development of a foliar primordium around the apical meristem; (F) paired tiers of cells straddling the plane of symmetry of the apical meristem; (G) bud prophyll; (H) bud apical meristem; (I) youngest cataphyll, and (J) first true cataphyll of bud. From Fisher, J. E., *Can. J. Botany* 50: 743-750, 1972.

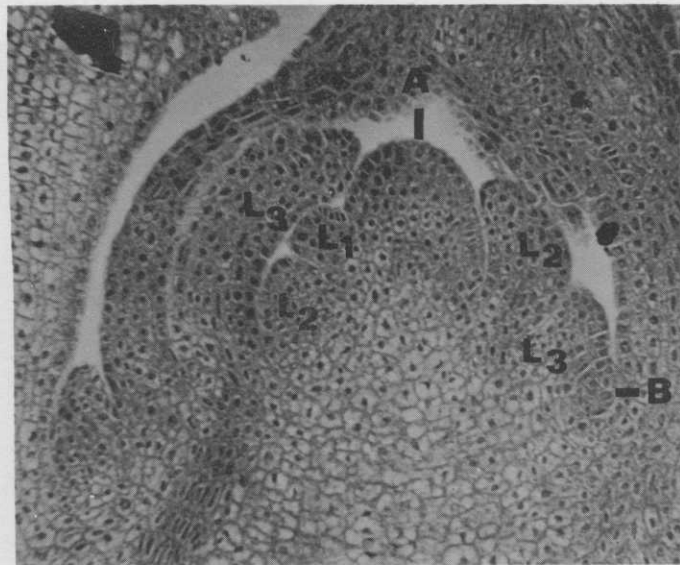
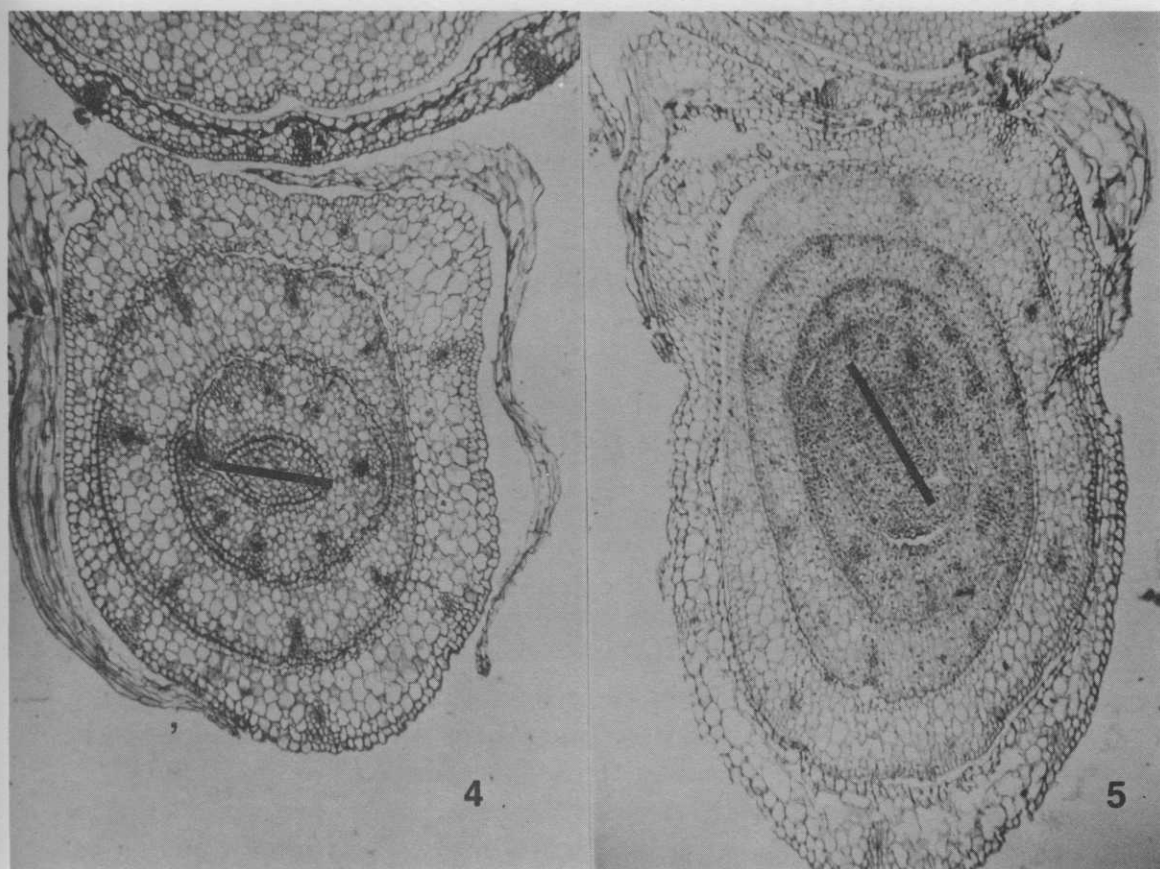


Figure 3. Longitudinal section on the plane of symmetry of a rhizome tip: (A) apical meristem; (B) bud primordium developing from the base of a leaf primordium; (L_1 , L_2 , L_3) leaf primordia in order of age. Each primordium except the youngest, L_1 , consists of two portions when viewed in longitudinal section.

been called a phytomer (Evans, 1949; Etter, 1951; Fisher, 1965, 1972). Thus, the position of the axillary bud was determined by the position of the first cell division in the apical meristem that gave rise to the leaf primordium.

In the development of the bud, the biologically controlled orientation of organs initiated originally by the activity of a single meristem cell is continued. The bud produces foliar organs, the first of which is the two-veined prophyll. It is oriented with its larger side next to the main shoot from which the bud arose (Fig. 2). The bud cataphylls (reduced leaves with a true mid-rib, commonly called bud scales) are oriented with their plane of symmetry approximately at right angles to that of the main shoot (J in Fig. 2).



Figures 4, 5. Cross sections of rhizome buds showing rotation of the plane of symmetry of the bud apex. The black lines are positioned on the planes of symmetry of the apex of each bud. From Fisher, J. E., *Can. J. Botany* 50: 743-750, 1972.

Non-geotropic Orientations During the Initial Growth Phase of Rhizome Buds

After a rhizome bud has formed, it may remain in a dormant condition for long periods of time. In fact, rhizome buds on existing rhizomes often never develop beyond the five or six cataphyll stage (Etter, 1951, 1953). If active growth of a bud commences,

the prophyll and the oldest one or two cataphylls are not usually involved. They are normally ruptured by the expanding cataphylls within and remain as dry tattered scales at the base of the bud (Fig. 4, 5, 6).

The rhizome bud developing from the base of an aerial plant turns downwards preparatory to active elongative growth into the soil. This reaction is apparently a polarity reaction, related to the fore and aft axis of the shoot from which the bud arose, and not caused by susceptibility of the bud to the force of gravity (Fig. 6, 7). Rhizome buds on plants growing on the rotating clinostat rotating at 10 revolutions per hour (gravity stimulation being multidirectional rather than unidirectional) oriented themselves with respect to the fore and aft axis of the stem from which they arose. They did not appear to be affected by gravity. Later, gravitationally sensitive orientations were severely disorganized by growth on the clinostat.

The initial bending appears to be a function of the cataphylls. Cataphyll veins thicken and develop sclerenchymatous cell walls more rapidly on the side away from the stem (outer or abaxial side), hence active growth of the side of the cataphylls next to the stem (adaxial), causes bending of rhizomes. As the cataphylls in succession form a telescoping tube through which the young rhizome bud stem grows, the rhizome stem is therefore directed into a downwards direction by differential cataphyll growth, and not by active bending of its own true stem. Excised stems of young rhizome buds are relatively structureless and very flacid.

The plane of symmetry of the apex of the young bud was originally, approximately at right angles to that of the main shoot (Fig. 2). If this continued, the plane would ultimately be parallel to the soil surface, or more correctly, at right angles to a plumb line. However, the plane of symmetry of the rhizome apex during active elongation is at right angles to the soil surface, hence parallel to a plumb line. This rotation of symmetry of the rhizome apex takes place during the formation of the first 7 to 10 cataphylls, and prior to the most active elongation phase (Fig. 4, 5, 8). This shift is at least partially independent of gravity, since buds

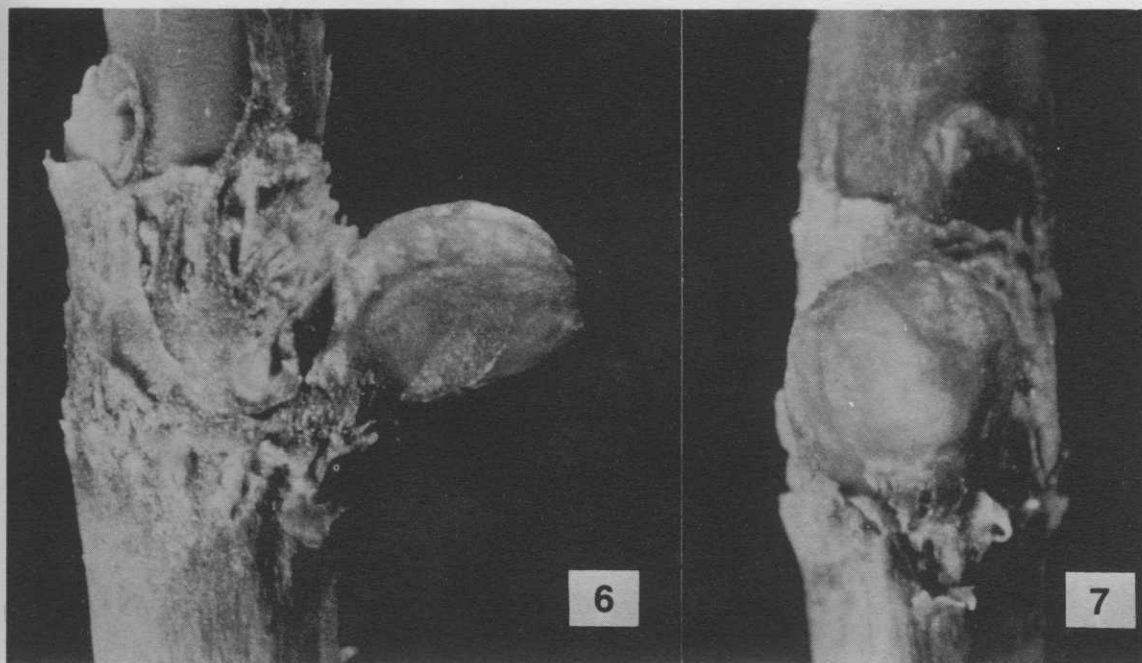


Figure 6. The initial downwards bending of a young rhizome bud.
Note the prominent veins.

Figure 7. Front view of the bud in Figure 6, showing the beginning
of lateral bending towards the right.

on the plants on the clinostat show rotation of the plane of symmetry. However, they do not cease the rotation of the plane of symmetry at a particular angle, either to the soil surface or to the main plant axis and no two seem to cease at the same angle. The gravitation susceptibility seems to occur towards the latter stages of rotation of the plane of symmetry when internode elongation of the young rhizome stem becomes accelerated.

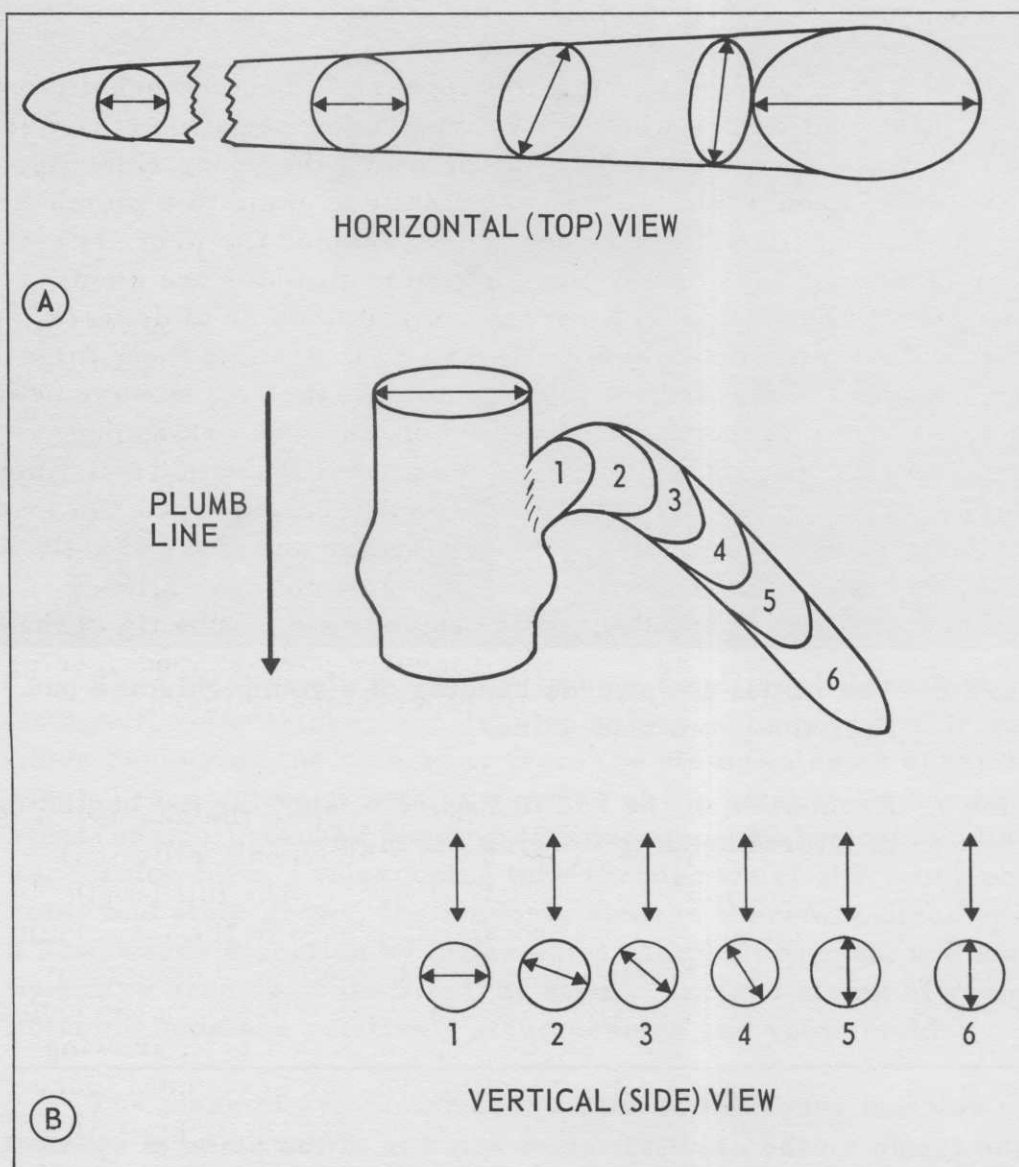


Figure 8. Diagrammatic illustration of the rotation of symmetry in the developing rhizome of Kentucky bluegrass: (A) top view, the arrows represent the plane of symmetry at each location. The main shoot is at the right side: (B) side view of the developing rhizome. Each cataphyll is numbered. The circles beneath represent cross-sections at each of the numbered cataphylls. The arrows above each circle show the plane of symmetry of the main stem from which the rhizome arose.

Geotropic Orientations During the Active Elongative Phases of Rhizome Growth

The young developing rhizome appears to become sensitive to gravitational forces when active internode elongation becomes pronounced. The growth phase during which the young rhizome is extending deeply into the soil at an oblique angle to a plumb line (a plagiogeotropic response), I have termed the primary stage (Fisher, 1965). After the containers in which the plants were grown were tilted by a predetermined number of degrees, the rhizomes reoriented themselves so as to assume their former angle to the plumb line. The contour of the soil surface does not appear to influence the orientation of bluegrass rhizomes. During this plagiogeotropic stage the cataphylls are quite distinctive. Viewed in their entirety, they are distinctly cone-shaped. In cross section, they are elliptical to almost circular and more heavily walled on the side of the main vein. They do not have a longitudinal indent or invagination on the thinner side. The tip of the young cataphyll often has a small triangular pore-shaped opening close to, but slightly beneath its tip. Occasionally there is no opening evident.

The secondary stage begins with a change in the shape and form of the new cataphylls. They become markedly elliptical and have an indentation or invagination running the full length of the cataphyll on the thin-walled side opposite to the main vein. In overall view, they are distinctly sword-shaped. The pore at the tip becomes a definite slit. Then a major change in the geotropic response takes place as the tip, which had been growing downwards, rapidly shifts its direction into the horizontal plane. Under ideal midsummer conditions, the secondary stage is the longest of any.

During the final or tertiary stage a small, incomplete, membranous ligule forms at the base of the apical slit in the first cataphyll. The first signs of the large, thin-walled cells believed to be involved in leaf closure in mature above-ground leaves can just be observed above the ligule on either side of the main vein. Each of the next five successive cataphylls begins to develop

certain leaf-like characteristics. However, the collar region that, in mature leaves, causes the blade to bend obliquely away from the axis of the sheath does not develop in tertiary stage cataphylls. During the final stage in geotropic response, the rhizome tip begins to turn upward, describing a long arc through the soil until it finally emerges and changes to a new aerial shoot. This geotropic response has been called negative orthogeotropism, now frequently abbreviated to apogeotropism.

When the tip of the rhizome penetrates the soil surface the first two or three organs are not true leaves, but merely green cataphylls of the tertiary stage. They originated from the apex before the rhizome had emerged from the soil. Thereafter, normal aerial leaves are produced.

In each of the three stages--primary, secondary, tertiary--morphological form and structure of the cataphylls is intimately linked with a specific physiological response to gravity to such an extent that one can be used as an accurate indicator of the other (Fisher, 1965, 1972).

Circumnutational Growth Movements of Rhizomes

Rhizomes can penetrate objects in the soil, such as wood, asphalt, heavy clay, roots and tubers of other species, and almost anything else in their paths, except stones or metals. They do so by means of circumnutational growth of the individual cataphylls. Circumnutation has been described (Larson, 1962) as the spiral pathway in space described by the tip of a stem during growth. Apparently most stems exhibit some degree of circumnutation. True circumnutation is not a torsional twisting, such as exhibited by morning glory vines; hence, is not analagous with a screw. To demonstrate the movements of a rhizome tip, an apparatus was constructed which consisted of a short, pivoted, balance arm, having a cone at one end into which a rhizome tip was guided. At the other end was an inclined mirror onto which a collimated light beam was directed, the reflection being projected onto the distant wall of a darkened room (Fisher, 1964).

A circular, clockwise, 24-hour rotation of the rhizome tip was observed at 1,000 X magnification. The actual diameter of revolution was between 0.5 and 3.0 mm. A minor, circular, clockwise revolution of one to two hour periods, superimposed on the major revolution, was determined at 10,000 X magnification. The actual diameter of revolution was between 0.001 and 0.010 mm. In soil, the magnitude of these rotations would probably be much less due to the physical damping action of the soil. These rotational movements undoubtedly aid the rhizome in penetration of the soil.

Horizontal Orientation of Rhizome Buds

If a plug of bluegrass sod is planted in a vegetation-free area of soil, new rhizomes, when produced, will grow outward much like the spokes of a wheel. Ultimately they turn up to produce new aerial or leafy shoots. When the daughter plant has emerged, its plane of bilateral symmetry is parallel to the direction in which the rhizome has been growing. Therefore, rhizome buds produced by the daughter plant will be positioned either directly away from the original parent plant, or directly towards it (Fig. 9). Serebryakova (1968) has stated that the bud positioned towards the parent probably never develops since one never sees a rhizome growing back towards the parent plant when the parent is isolated and not in a dense sod.

I have found that these buds do develop but the rhizomes developing from them bend tightly around the stem of the daughter plant until they, like the buds on the other side of the daughter plant, are growing away from the original parent (Fig. 10, 11). This highly oriented bending is accomplished through differential growth responses in the cataphylls of the rhizome. One side of the bud grows at a much higher rate than the other (Fig. 6, 7). This bending around the aerial shoot resembles closely the bending downwards of the young rhizome bud, as outlined in the preceding portion of this report. If the rhizome is severed before the rhizome tip emerges from the soil, this orientation of subsequent rhizomes does not occur. If the rhizome tip has emerged from the soil prior to severing and has formed a new aerial shoot, the orientation remains for more than four weeks. The orientation away

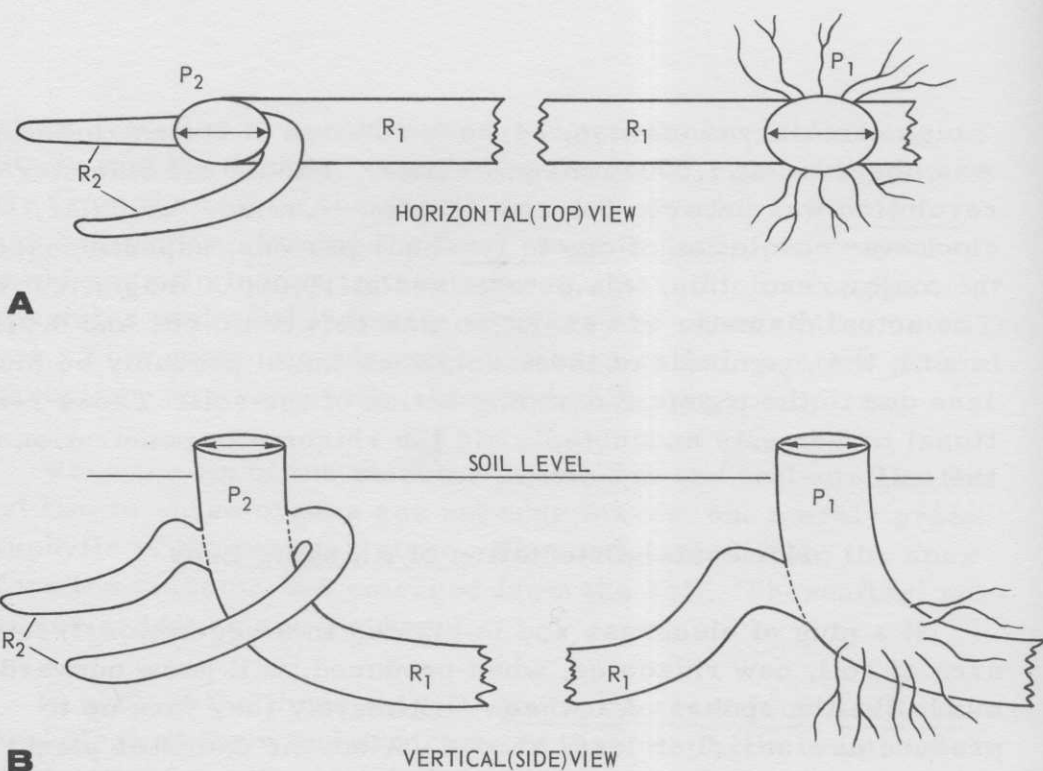


Figure 9. Diagrammatic representation of the movement of rhizomes away from the older plants in a rhizome - aerial shoot series. Rhizome buds that arise facing the rhizome - aerial shoot series bend around the base of the shoot and move outwards into non-inhabited soil. (P_1) Older plant in series; (P_2) younger plant in series; (R) rhizome connecting P_1 and P_2 ; (R_2) young rhizomes arising from the base of P_2 .

from the original plant does not appear to be the result of toxic or growth-regulating substance secreted into the soil by the parent. Newly emerged daughter plants, if transplanted in their same location but rotated by 180° , will produce rhizomes that will grow directly through the parent plant without any apparent inhibition of growth. Apparently the inner buds bend due to some polar type stimulus from the daughter plant and/or from the rhizome from which the daughter arose.

Thus, the rhizomes developed by each succeeding plant orient themselves with respect to the preceding plant in each rhizome train (Fig. 9), making the bluegrass plant a potent invader of new areas of soil. Besides the rhizomes initiated by the parent and by the daughter plants, certain dormant buds along the rhizomes

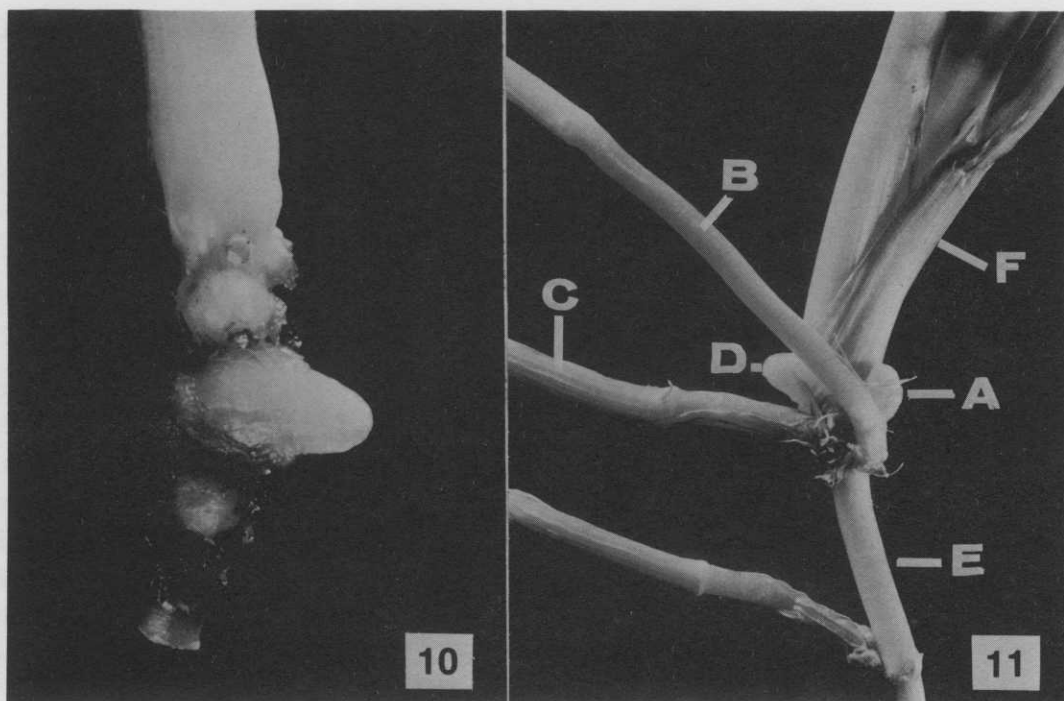


Figure 10. Prominent lateral bending of a rhizome bud.

Figure 11. The bending away from the parent rhizome (E), and around the aerial shoot (F) of a rhizome bud (A) and a developed rhizome (B). Rhizome (C) and rhizome bud (D) have developed from the opposite side of the aerial shoot and show no lateral bending.

suddenly begin active growth and form still more rhizomes. The result is that the area of soil is populated in an ever increasing circle of plants that continually moves outwards from the parent. In the meantime, the older plants at the center of the circle are continually producing new rhizomes. Thus, the soil becomes interlaced with a dense mat of plants and rhizomes which becomes constantly denser at the center until the characteristic bluegrass sod is formed. Few plant species can compete with this domineering growth pattern.

Summary

The rhizome of Kentucky bluegrass is not the lax, aimlessly growing, underground stem of popular misconception. It is a highly organized structure, subject to many orientations with a changing morphology throughout its various growth phases.

It arises as a bud primordium in a predetermined location dictated by the parent shoot apical meristem. It bends downwards into the soil while rotating its plane of symmetry by reason of biological influences derived from its parent stem. Its orientations within the soil are determined (a) vertically, by gravitational stimulus on unknown biological sensors, and (b) horizontally, by strictly biological controls. It bores through the soil using a highly complicated motion in which one helical movement of small amplitude, but rapid rotation, is superimposed on one of large magnitude but slower rotation. Finally, it turns upwards towards the soil surface with its morphology changing in preparation for its ultimate role as an above-ground shoot. It is an exceedingly complex organ.

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TURFGRASS RESEARCH IN BRITISH COLUMBIA

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Introduction

A turfgrass research program was initiated at Agassiz, B. C., in 1969 by the Research Branch, C.D.A. This is a new program for the Agassiz Research Station which is located 70 miles east of Vancouver, B. C., at the eastern end of the Lower Fraser Valley. Agassiz has a maritime climate with an annual precipitation of 64.3 in. which includes 33 in. of snow. Winterkill is rare, but when it occurs it is usually confined to bentgrass and perennial ryegrass. In addition to the Agassiz location, cooperative uniform tests are conducted at the Kamloops Research Station in the interior of the province where conditions are closer to a continental type of climate.

Cool season grasses, fine fescues, bentgrasses and perennial ryegrasses are well adapted to Coastal British Columbia. Although the Kentucky bluegrasses are not as persistent, there is need for this species in home lawns and sports turf because of its ease of management and its wear resistant qualities. However, in the interior of the province, Kentucky bluegrass and fine fescues are both well adapted. Major diseases are leaf spot-melting out complex (Helminthosporium spp.), Fusarium patch (Fusarium nivale), snowmold (Typhula spp. and F. nivali) and red thread (Corticium fuciforme). Less frequent are Ophiobolus patch (Ophiobolus graminis var. avenae), Rhizoctonia brown patch (Rhizoctonia solani), fairy ring, leaf rust and powdery mildew. It is quite evident that our species and variety performances reflect resistance to these diseases.

Variety Trials

Varietal evaluation has been a major part of the Agassiz turf-grass program to date. Seedings made in 1969 and 1970 include a total of 242 entries in replicated trials. Fine fescues, Kentucky bluegrasses and mixtures are evaluated at two heights of cut, 3/4 in. and 1 1/2 in.; perennial ryegrasses at 1 1/2 in., and bentgrasses at 1/4 in..

Fine fescue

Among the creeping red group, Dawson, S59, Rasengold and Leo have approached Pennlawn in performance. SAI-67 from Scotland is particularly promising in a preliminary trial. The chewings fescues, as a group, are relatively more attractive with superior density and resistance in some cases to red thread. In particular, Koket, Highlight, Rolax and Wintergreen are promising. Jamestown is outstanding for its dark green color while Golfrood shows resistance to Fusarium patch. C-26, a dark green hard fescue, has been superior to all fescues tested for its density and red thread resistance.

Kentucky bluegrass

The performance of this species has been directly related to varietal resistance to the leaf spot-melting out complex. The melting out phase of this disease has been so severe that in plots of certain varieties, weeds have taken over 25% of the plot within a three month period. Merion has been most resistant to date, followed by Nugget. Other varieties tested having better than average resistance are A34, Baron, Birka, B101, EVB 391, Fylking, Golf, K412, Newport, Pennstar, Sodco, Sydsport and Windsor. The incidence of Helminthosporium spp. is so great that it would appear to be an ideal screening area for new resistant varieties.

Bentgrass

Among the colonial bentgrasses, Brabantia (Evate), Tracenta and Bardot are promising. Penncross creeping bentgrass and Kingstown velvet bentgrass are both outstanding in their class.

Perennial ryegrass

Two varieties have been promising to date: Norlea for its winter hardiness, and Manhattan for its superior density and attractive dark green color. There is a growing interest in the use of perennial ryegrass for intensively used playing fields. To date, our perennial ryegrass trials have not been subject to wear, but this factor will be an essential part of future trials. Test plots have been established in 1972 of perennial ryegrasses, Kentucky bluegrasses, timothies, and miscellaneous grasses, both in pure stands and in mixtures. Evaluation of their performance and contribution of some of the improved Kentucky bluegrass cultivars using a wear treatment during the period of maximum use is a major goal.

Mixtures

Trials are underway aimed at providing information for home lawn recommendations. Under trial are Pennlawn and Boreal fescue, Merion and Park Kentucky bluegrass and Highland bentgrass in all combinations and at several seeding ratios. The vigor and competition from Highland is most evident and it appears that the contribution of the fescue or Kentucky bluegrass component is dependent upon variety. Within three years, Highland dominates most mixtures. Although well adapted, Highland presents a management problem to the homeowner, who almost invariably cuts his lawn at a height normally recommended for Kentucky bluegrass.

Fertility Trials

A comparison of effects of sources and levels of nitrogen and some minor elements in Penncross bentgrass maintained at 1/4 in. cutting height is now in its third year. A less extensive trial is also underway to investigate the cumulative effects of several levels of phosphorous and potassium on Seaside bentgrass.

High Sand Greens Mixes

An installation has been made to observe the response of six vegetative bentgrass cultivars growing on sand-organic matter mixtures. Variables under observation are two depths of sand, sawdust vs. peat, and three depths of organic matter mixture. We are hoping to obtain information on the irrigation requirements and relate these to evapo-transpiration and other climatological data.

Disease Control

Experiments in snow mold (mostly Typhula spp.) control have been initiated on three golf courses in the interior of the province. Demosan and PCNB are the most promising treatments after one year of trials, but certain treatment x location interactions are apparent.

TURFGRASS IMPROVEMENT AT MICHIGAN STATE

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Introduction

Our climate in Michigan is perhaps different than some of you may realize in that we are frequently subject to drought. Usually every three out of four years we can count on quite a severe drought period during the summer. We get 30 to 35 in. of precipitation annually, which normally would seem to be enough. Dry conditions, however, are the rule rather than the exception in many sandy soil areas during most of our summers. With over 25% of our lawns in shaded situations, we need a good turfgrass with both drought and shade tolerance.

Background

A number of people are doing fine work in bluegrass variety development. This species, of course, is queen of the turfgrasses for cool, humid regions. When the opportunity came to establish a breeding program three or four years ago at Michigan State University, it was decided that perhaps the fine fescues had the most to offer in providing what is needed in terms of shade and drought tolerance, as well as the ability to grow with relatively lower levels of fertilizer and management. Accordingly, when the decision was made to go into a breeding program, it was decided to imphasize the fescues. Dr. Fred Elliott was doing forage breeding work and had included some work with the fescues. As a result, when the present project was initiated in 1968, a field of about 300 single plants of fine fescues existed that Dr. Elliott had acquired through the Plant Introduction. Of this group, there were only two that were strong creepers. One with the

wide leaf, apparently 56 chromosome complex, and the other relatively narrow leafed, creeping-type. From subsequent observations, the narrow leafed, creeping-type is not commonly found.

There are some very acceptable fescue lawns in shaded areas in many of our home conditions, but there are very few excellent fescue lawns in full sunlight. Leaf spot becomes so severe that seeding a red fescue lawn is not recommended. Sometime during the summer, it will become infected. As a result, it was decided to concentrate on leaf spot resistance and spreading habit to provide a variety that sod growers as well as homeowners could use. Michigan has a large sod growing industry. It is estimated to be a \$35-40 million/year crop. The sod producer needs something that will creep relatively quickly in order to provide a more rapid turn over than can be obtained from present red fescue varieties.

Breeding Materials and Procedures

Using the two plants from Dr. Elliott's nursery as basic materials, a crossing nursery was established and through plant accession and other sources, germ plasm was accumulated. A year later, Dr. Joseph Vargas finished his Ph.D. at the University of Minnesota and came to Michigan State as a full-time turfgrass pathologist. A setup was thus available to begin screening and actively working on the disease phases of this particular project. After two or three generations, spreading habits improved considerably. While most of the creeping plants are of the wider leaf type, we are not as concerned about this as the European grass breeders because it is felt that the wide leaf will blend better with bluegrasses as turf.

A screening program was established three years ago and Dr. Vargas had quite a bit of difficulty in the initial phases in finding the right spore load with which to inoculate these fescues. With spore loads which had been used in Minnesota in the bluegrasses, all plants became infected. The next step was to find a concentration of spores sufficient to isolate a few surviving fescue plants. In addition to coming down to a 30,000 spore/cc load,

he found that if he suspended the spores in an agar slurry, which was then sprayed on the plants and placed in a mist chamber for 48 hours, very excellent inoculations would be obtained and a few tolerant plants could be isolated. With this technique, from the materials we were starting with, having no indications or hints as to whether there was any resistance at all in them, we isolated about 1%. These were inoculated a second time and we knocked out about 90% of those. As a result of three winters of screening, 18 clones survived that were inoculated a third time after they had grown in the field the winter of 1971-72. Two clones were from C-26, which is the most tolerant variety and might be expected to provide considerable leaf spot resistance. Surprisingly, it didn't. Three or four survivors are from Dr. Reed Funk's collections -- F-3 material and material that he's collected in the East. One is a chewings type, which is from an accession which appears to be another species, and the rest are of the broad-leafed creeper types. These are being intercrossed in the greenhouse as well as in the field this summer (1972) and, by continued selection pressure, it is hoped that a new plateau of resistance can be obtained. It is felt that with this strong screening and the fact that very few have survived this very severe test, that while we don't have the resistance that Merion brought to the bluegrasses, some progress is being made.

A winter hardy meadow fescue has been developed which has survived very well in contrast to the 50% survival that prevails with Kentucky 31, Alta and other tall fescues in Michigan tests. In another planting made last fall in conjunction with Merion Kentucky bluegrass, it had 100% survival. Alta tall fescue went out 50% this winter (1971-72). Two thousand pounds of seed were produced in Oregon in 1971, and this is now in trial. This could be useful on a large industrial lawn or large site where a very fine highly groomed grass isn't necessarily needed, but where uniformity is desirable. It appears from the performance of the parental material, that it will sustain itself in combination with Merion bluegrass very well. The leaf width made it difficult to distinguish from Merion, although it can be distinguished easily during the winter when the color patterns are somewhat different. It hasn't been tested for wear tolerance.

A series of tests has been conducted in a shade nursery developed in 1960 under non-irrigated conditions. This last summer

(1971), water was piped to the area and a new set of eight or nine experiments are under way. The shade is about 90-95%. The present tests include mixtures and blends, and a comparison of varieties using chemical disease control.

Dr. Vargas is using sprays in succession as the season progresses, and is spraying half of each plot to try to control diseases during the summer. Some very interesting differences are evident.

Other Studies

Dr. James Beard and his graduate students have developed an extraction technique that seems to be working very well in the electrophoretic identification of protein components in grass varieties. Hopefully, this will lead to varietal identification. Good reproductability between the bent varieties has been obtained. Progress is being made with Poa. It seems well established that test plants must be grown in the growth chamber under quite the same conditions. When this is done, very good breakouts of different protein fractions can be obtained.

RESEARCH TO IMPROVE METHODS OF TESTING VARIETAL PURITY

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Introduction

When I started work at Geneva nineteen years ago we were testing varieties in the field, and one assignment I was given was to improve testing methods. Obviously, field plots have some major disadvantages. First, they usually take from six months to a year to conduct. Second, no matter when the seed is received it is necessary to wait until spring to start a test.

Most of my work has dealt with growing seedlings in greenhouses and growth chambers and trying to find ways seedlings of one variety differ from those of another. I started with forage and grain crops, then seven years ago we added turfgrasses. Thirteen years ago our work became part of a Northeast Regional project on testing for varietal purity. Pennsylvania, New York and the USDA are the only agencies involved with it now. Several more states were involved when the project started. At that time it dealt mainly with testing alfalfa varieties in field plots. Dr. Guy McKee of The Pennsylvania State University has done much of his work on chemical tests of seeds and seedlings. Thus, at Geneva we have concentrated our work on seedling growth.

Seedling Growth Research

I have divided my research on seedling growth roughly into six categories. The first approach was to grow seedlings and observe them carefully to detect any characteristics in which those of one variety differ from those of another. The second was use of controlled environmental conditions, mainly temperature, photoperiods and light intensity. We manipulated these to

find conditions that would induce seedlings to express their genetic differences in some observable characteristic. Third, we used chemicals, either as foliar sprays or as soil treatments, to see if seedlings of one variety might respond differently from those of another. Fourth, we defoliated seedlings and observed the regrowth to see if this would result in observable varietal differences. Fifth, we grew seedlings in sand and used various nutrient solutions to determine if varieties would respond differently to a lack of various mineral nutrients or to different levels of them. The sixth approach was acceleration of blooming or heading to make these plant parts available for observation in a short time. This can have some real advantages. For example, about 10% of vernal alfalfa plants should have yellow flowers. If we should test a seed sample labeled Vernal and find no plants with yellow flowers we would know it was mislabeled. These approaches can never be separated completely but they make convenient categories for the purpose of this talk. For instance, in growing seedlings for chemical applications we must consider the temperature, light intensity and photoperiods under which they are grown. This is also true when a person is studying effect of mineral nutrition.

Observation of Seedlings

By careful observation of seedlings during the first three or four weeks of growth, it is often possible to detect characteristics in which varieties differ. Alfalfa varieties were found to differ in serration of the first leaf. Oat varieties differ in length of internodes. Kentucky bluegrass varieties differ in leaf width, growth habit and length of leaf sheaths.

Controlled Environments

In the second approach, one of the first things we did was to develop a test for distinguishing hardy from non-hardy alfalfa. When we grew plants with short photoperiods, non-hardy varieties developed stems twice as long as those of hardy varieties. When we used not only short photoperiods, but also cold dark periods, stems of non-hardy plants were five times as long as

those of hardy plants. When Kentucky bluegrass seedlings were grown with high light intensity and long photoperiods, useful varietal differences developed. Of the 25 varieties we tested, seedlings of 20 developed red lower leaf sheaths. Seedlings of the other five were entirely green. Rhizome and tiller development could also be observed on the same plants and these differed considerably between varieties. In one such test, Merion developed an average of 5.2 tillers per plant, compared to Newport which developed only 1.3 per plant. On the other hand, Newport had twice as many rhizomes per plant as Merion.

Chemical Use

In the third approach we have found several chemicals useful for distinguishing varieties. An illustration is use of ethephon on Festuca rubra. This resulted in development of two distinctly different types of plants. The first responded relatively little to the chemical and leaf blades were long and narrow. Stems of the second type elongated and the leaf blades were short and wide. Ruby and Ranier red fescue had nearly 100% short-wide leaf plants. Pennlawn and Illahee each had about 60% long-narrow leaf plants. Highlight and Jamestown chewings fescue had 100% long-narrow leaf plants.

Maleic hydrazide, applied to soil in which perennial ryegrass seedlings were growing, was useful in distinguishing Manhattan from other varieties. This chemical caused plants to die, but most Manhattan plants stayed green longer than those of other varieties. Captan applied to soil after timothy seeds were planted resulted in stunting of some plants and normal growth in others. Varieties differed in percentage of stunted plants.

Defoliation

We found that removal of the primary stem of birdsfoot trefoil seedlings had a profound effect on growth habit of secondary stems. Empire plants were decumbent, but Viking plants were upright. Clipping also brings out growth habit differences among

alfalfa varieties. Growth habit of perennial ryegrass can also be modified by clipping.

Nutrient Manipulation

We started growing seedlings with nutrient solutions about six years ago. This has resulted in many interesting discoveries. It is possible to withhold any particular nutrient and observe for deficiency symptoms. In addition, a nutrient may be withheld either before or after a complete nutrient solution is used. Use of different forms and levels of nutrients offers additional possibilities. We published a paper recently on use of a solution lacking calcium with Kentucky bluegrass varieties. Seedlings of some varieties turn yellow and die when they are grown with this solution. Others remain green and relatively healthy. Delta and Nugget are varieties that turn yellow. Arista, Fylking and Pennstar remain green.

If Merion and Windsor are grown with a complete solution, seedlings of both varieties have green leaf sheaths and it is difficult to see much difference between them. However, if they are grown with a complete solution the first two weeks and then with a solution lacking nitrogen the next three weeks, lower leaf sheaths of Merion become red and those of Windsor remain green. A solution lacking calcium, but with a high level of nitrogen supplied as ammonium nitrate, caused red color to extend two inches or more up Nugget plants. In the same experiment, red color extended only 1/2 inch up Baron plants.

Acceleration of Heading or Flowering

By use of continuous light and suitable temperatures, we can get alfalfa, birdsfoot trefoil, red clover and sweet clover plants to bloom in 4 to 5 weeks from the time the seed is planted. Spring barley and oats will head out in 4 to 5 weeks under similar conditions. In lawn grasses, we have used this technique to distinguish annual from perennial ryegrass. Because the fluorescence test that has been used in the past is not always reliable, there is need for a new method of distinguishing these species. We grew seedlings with continuous light, a complete nutrient solution, and a

temperature of 27°C for 20 hours and 16°C for 4 hours. Within three weeks of the time seeds were planted, 10% of annual plants had headed out and most of the rest had elongated seed stalks. In contrast, no perennial ryegrass plants had developed any stems at all.

Summary

In species such as Kentucky bluegrass, barley and oats, varieties tend to be very uniform and in many cases it is possible to distinguish off-type plants with a high degree of precision. In most cross-pollinated species, plants differ considerably within a variety. Thus, it is difficult to develop reliable tests for distinguishing varieties and it may be necessary to accept a lower degree of precision. In addition it may not be possible to distinguish varieties with similar characteristics.

Discussion Period

Dr. Daniel: How long do you let your seedlings go in your growth habit tests?

Dr. Nittler: In bluegrass, we hope to terminate an experiment in five weeks from the time we start. We can get good results on some of our oat tests in ten days by using a minus-phosphorus solution. This results in development of red color in leaf sheaths of some varieties and not in others.

Dr. Payne: We certainly would like for you to keep pursuing this matter of forcing greenhouse winter heading in the fescues. We tried 16, 20, 24-hour day regular greenhouse conditions after a cold induction, but didn't obtain heading.

Dr. Nittler: Timothy and some bentgrasses will head out in growth chambers, but most other perennial grasses such as red fescue, orchardgrass, and perennial ryegrass apparently need to be vernalized. These will not even produce stems. Ethephon will induce red fescue seedlings to produce stems. The first time I saw this I thought they would also head out, but they didn't. It may be that use of ethephon and cold dark periods would induce heading. This is something we plan to try.

Dr. Duich: Have you ever tried a heavy metal approach?

Dr. Nittler: No.

Dr. Duich: Sometimes with fungicide applications, I know there's quite a differential cadmium response of varieties. The PMA effect on Merion might be a good systematic approach to try.

Dr. Nittler: There are so many agricultural chemicals that there is no limit to how much work you can do with them. The growth regulators, in general, have been a useful group for bringing out varietal differences.

Dr. Long: In Texas a few years ago we noted that certain insecticides would react differently on some of the bermudas. One interaction was chlordane interaction on Texturf 10 bermuda. It would turn it very yellow.

Dr. Nittler: This is something I haven't looked at. Some of the fungicides, such as captan, would be useful on certain grasses.

General Discussion

Dr. Long: Dr. Fisher, you mentioned that if you have allowed more photosynthetic area on bluegrasses that you would probably encourage rhizoming. Dr. Daniel stated that he had space plants that weren't clipped down, and got pretty good rhizoming. Do you think that if these were under a little mowing pressure, they wouldn't rhizome as much?

Dr. Fisher: That's our experience and also the experience of a number of other people who have worked with rhizomes, even of different species. If you cut the photosynthetic area of your plant down, rhizome production and rhizome growth drops off rapidly. The rhizome is a parasite on the parent plant and depends on an excess of photosynthate. In fact, MacIntyre has evidence that high carbohydrate levels with low nitrogen levels is the best combination for inducing rhizome production in couchgrass.

Dr. Payne: Dr. Duich, how are you handling the breeder's seed of Penn State varieties as they are sent West for increase? For example, Penncross is composed of three clones. How are you maintaining these clones?

Dr. Duich: We're maintaining space plant nurseries--alternate rows.

Dr. Payne: How large a nursery do you maintain of those clones?

Dr. Duich: About two acres, depending on projected foundation seed needs.

Dr. Nittler: Is there going to be a shift from one generation to the next in Pennfine, or will it be pretty stable?

Dr. Duich: We haven't seen a shift in two generations of space plant and turf testing. We can't distinguish anything. That's as far as we've gone.

Dr. Nittler: I might comment, too, that we're looking at this in the growth chambers trying to see if we can determine some difference in the relatively young stage. We think we can see some, especially in plant size. We've measured the second leaf blade length. Pennfine seems to be shorter than the other varieties that you have showed me in your tables. Also, it tends to have less red color on the lower leaf sheath than some of the larger varieties.

Dr. Daniel: Dr. Long, how much dwarfness do you think people want in turfgrasses?

Dr. Long: To start with, about 70% reduction in lawn mowing and clipping.

Mr. Mayer: How are we going to measure dwarfness in a mowed lawn? We're doing it in a nursery, but does this correlate with the lawn? The homeowner mows his lawn at two inches. Even if a mature plant grows six inches, he is still going to mow it at two inches. He is still going to be taking yields off.

Dr. Duich: If one has to mow it only three times in the spring, though, I think you've achieved success.

Dr. Daniel: We started measuring yield after leaf spot had hit the other grasses. Our healthier grasses produced the most clippings. Those that had been weakened by the disease produced less clippings. This was a completely erroneous period of observation.

Let's assume you get something down to where you're mowing the grass once every two weeks at three quarters of an inch, and your clipping length doesn't exceed your mowing height. Are you committed then to keep out crabgrass or grasses such as annual bluegrass, ryegrass and red fescue when they are considered as weeds? Is the landscaper ready to give the homeowner that clean of a seed bed? Are the seedsmen ready to clean up his seed field well enough, or is the seedhouse going to keep it clean so there is no other crop in it so that it will stand to compete?

Dr. Duich: I'd like to make one associated comment from a practical point of view. I think we've been a little lax in not getting some of this data. We've noticed, particularly on some of these so-called decumbent, low-growing types, that there seems to be quite a varietal differential after you mow on leaves uncovered from previous mowings. Some of the remaining leaves turn yellow and brown off. A particular variety may look very poor until you get enough regrowth to cover it up. As soon as you remove it, even from the previous mowing, you find it again. I think Merion has been rather notorious for this unclosed mowing, high-maintenance levels. However, with some of these other varieties, Nugget, for example, I don't know what happens to the clippings. They just seem to disappear. So from the aesthetic point of view, they look very promising. If we would achieve true dwarfness, this could be a very important consideration, too.

Dr. Kuc: When you talk about a dwarf, do you refer to the ultimate height of the plant, or do you refer to the rate of growth, or both? Do all dwarf plants have a reduced rate of growth?

Dr. Daniel: My idea of a dwarf is one that has 50% fewer clippings compared to what has been normal in the past. If Merion, a standard, is giving you a certain yield of clippings, then a dwarf variety should be about 50% less.

Dr. Long: Dr. Dudeck, what is your experience with Tifdwarf on attempts to use it in home lawn situations in the Southeast? What are the problems with this dwarf variety?

Dr. Dudeck: Tifdwarf has not been used in the home lawn situation, to my knowledge, because of the high maintenance involved. Florida No Mow, FB 137 or Floraturf has been promoted and used incorrectly, as the name implies, as a no mow variety. Unfortunately, too many problems were found with this grass in the field prior to its release. Actually, it has never been officially released from Florida. Tifdwarf, which does have a slower growth rate compared to the other bermudas, is not used in the home lawn situation because of the maintenance required.

Dr. Kneebone: Tifdwarf has been used a little in the Phoenix area as a substitute for Tifgreen with the same idea as the no mowing situation. In Arizona, Tifdwarf will seldom get above three inches, whereas Tifgreen left unmowed might get up to eight inches. Tifdwarf rolls and swirls. To be handled properly it would be mowed at one-half inch, which means mowing just as frequently as any other bermudagrass. There should be enough growth to self heal and keep the green down so the green is below the cut of any dwarf.

Bermudagrass dwarfs are different than the bluegrass ones. It's more the length of the internode and length of the individual leaves rather than the over-all dwarfness of the plant. We get some that are much more dwarf than Tifdwarf--so dwarfy that you can't grow a lawn.

Dr. Long: I don't think we gave Dr. Kuc a very good answer a while ago on whether we're dealing with growth rate, or growth height response. Some years ago when I had a program going and was looking at some St. Augustine dwarfs, we did pick up this aspect of growth rate. Comparing growth rate with one of the experiments with Floratine or Bitter Blue over a period of four months, we measured no stolen growth on the dwarf, as compared to 10 to 12 inches of stolen growth on Floratine or Bitter Blue. Apparently we have an effect on one aspect of the dwarfness. Maybe a slowing up of the growth rate. One of Dr. Daniel's students found this in some of his work with bluegrass. He had very little growth on some of the material he was studying.

Dr. Daniel: On the other end of the scale, one of the first plants we worked with, the grandmother of 90% of our research work, we've never found anything more heterogenous in its offspring, nor more vigorous in its own ability to spread, than the old original--this 16B we picked up off Chicago Golf Club. In seven generations, we haven't found anything more vigorous than the original plant, it seems, on spread. We've found a lot of dwarfness over this time.

Dr. Payne: In regard to your question of how much dwarfness we want, we can use dwarfness if we can get two or three varieties

so that we can get a blend. But as you can imagine, it probably won't blend too well with other types, especially when mowing patterns aren't consistent. If we have one, it would appear that we should have three.

Dr. D. Taylor: Just a comment on the performance of Nugget, one of the dwarf Kentucky bluegrasses. I've noticed that in fall planting, I've had trouble getting a good stand. With varieties of this type, if the practice in the area is to fall plant, we'll have to move the planting date ahead in order to get a good establishment before winter.

Dr. Higgins: It would seem to me that if the people would cut the grass longer you wouldn't have to worry as much about the dwarf plants. I wonder if there is some way to educate the public to want and cut it taller, it would simplify the breeding problem if you didn't have to worry about dwarfness. The first question that comes to my mind is why do people want to cut it short? Is this because they hate their lawn and they're trying to hold back its growth by damaging the grass? Of course it's physiologically done. I wonder if it might be easier to solve the problem of more Americans having better lawns if they could be educated to raise the blade a little bit, rather than try to come up with varieties that are more dwarf. There's enough problems of breeding without having to worry about that.

Dr. Daniel: As we've worked with our plots, if we have two heights of cut, we normally spend 95% of our time noticing what happens to the low cut. If you want to use the turf for croquet, for a party, for walking or for viewing, generally the more you can manicure it, the better it may perform, providing you have survival. So if we can dwarf the plant and cut it at three quarters of an inch, and cut off three quarters of an inch, that may be as equally satisfactory to a lawn as having it two inches high, and when mowed, cutting off two inches of clippings.

Mr. Bangs: Institutional turf managers frequently make the statement that, "I cut my turf as short as I can so I can reduce the number of times that I have to mow it." I think it might be helpful if we could have a comment on that statement because it is quite common.

Dr. Fisher: The following suggestion doesn't necessarily refer to a turfgrass clone of bluegrass, but to the vigorous wild one that we're working with. We found that when cutting frequently and lower, we had much reduced growth. After a period, the dry weight accumulation dropped, and we put it down to reducing the photosynthetic tissue quite drastically. I think the person that does cut his lawn very short doesn't need to cut it as often. He doesn't have to cut as much material. On the other hand, maybe he does cut it as often for appearance sake. There's a big distinction here because if he has it that short, he may want to continue maintaining it short and would have to manicure more often. But he's not cutting as much material as the man who lets his lawn grow three inches high. After all, a half-inch growth doesn't show as much on three inches as it does on a half inch.

Dr. Daniel: Of course, with a lawn mower we're just simulating what they've done with bonzi trees in Japan. They can take just about any species and by trimming both top and root, subject it to growth control. With a variety of grass, short cut is more drought susceptible than a long cut on a given day. So I'm sure we bonzi both top and root as we trim it down to a considerable extent.

Dr. Whalley: I have a comment from a range situation that exactly agrees with what's been said. If you overgraze or over-clip range grasses, then the rate of both top and root growth is considerably lessened. With a reduced root system, drought resistance is then decreased. In fact, I've heard it said with alfalfa that if you pull a leaf off the top, a root falls off the bottom. I'm sure this is an exaggeration, but the principle certainly holds.

Dr. Madison: I'll say it from the opposite point of view. When we mowed a healthy turf at 2 1/2 inches at intervals of two to three weeks, grass vigor built up so extensively that within two or three days after mowing, an inch or two of growth recovered and the grass appeared ready for another mowing.

BASICS IN GRASS IMPROVEMENT

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Introduction

Often lacking in turf breeding programs is the bad example against which to compare the many fine selection and breeding studies so that their excellence can be appreciated. I should like to give an example of a poor program which arose unintentionally.

In 1959, we abandoned plots of creeping bentgrass (Agrostis stolonifera) because of cross contamination with bluegrass. We sprayed the plots with weed oil and withheld irrigation water for the summer. With fall rains, many grasses recovered and grew. The next summer we not only withheld water but used weed oil twice, burning the field after the second application. With October rains there was again growth of many individuals. Impressed with such persistence, we selected 350 clones of good color, texture, etc., and grew them in field rows. Late in the summer of 1961, while evaluating these, we noted that each original plant formed a small tuft from which spreading by stolons had taken place. Further, we noted that plants had spread into their own shade so that there was good spreading to the north and east, but almost no spreading to the hot soil of the south and west.

Occasional exceptions were noted and we found about 23 clones that grew well to the south. We retested these in replicated trials and ended with six that had ability to spread into hot soil. As putting green turf, three were discarded for disease susceptibility and one for an open, loose habit of growth. Of the remaining two, we put one out to test with golf superintendents. Within two years, every test was lost due to changes in personnel, new construction

or similar reasons. When we examined the history we realized there is a great reluctance to use grasses that require vegetative planting.

This is an example of the commonest type of grass selection in turf improvement: single plant selection of an individual superior in one or more of its characteristics.

Faults in our program included developing a clone when there was resistance to vegetative propagation; lack of a defined program with stated goals; and selection that was strong on qualities, weak on conditions. Two hundred nursery lots of Congressional, Arlington and Cohansey bentgrass distributed to golf course superintendents in 1956 evoked enthusiasm but resulted only in vegetative planting of part of one green. Qualities vs conditions is shown in Table 1. An immature program may tend to neglect the conditions of test, though Burton has clearly stated them in his guidelines (Burton, 1969). Informal discussion at the 1st International Turfgrass Research Conference was concerned with the possible disparity between conditions of test and conditions of use, a problem discussed by Juska and Hanson (1962). Qualities are emphasized in row tests, conditions in plot tests and there is a tension between the two. Ahlgren et al. (1945) noted that of 74 superior clones of 11,800 in field rows, only one remained superior in a mowed plot.

Table 1

<u>Condition of Testing</u>	<u>Qualities Under Test</u>
Climate	Color (early, midseason, late)
Irrigation	Vigor (establishment, spread, recovery)
Mowing	Resistance (to insects, diseases, nematodes, weeds)
Fertilization	Tolerance (to heat, cold, drought, clipping, wear)
Shade	Form (compact of spreading)
Competition	Competitiveness
	Texture
	Persistence

Regionalized Grass Research Programs

I can find only praise for the many turf improvement programs across the country, but I wonder if there is not also room for small programs based on mass selection to meet local conditions. I say this because regional adaptation of colonizing species is usual (Bradshaw, 1959). I have been impressed by failure of several Eastern varieties to perform as well in the West as varieties of Western origin and vice versa; also, mass selection does not require the expense of large row trials, results of which are often negated in plots. As an example of regional difference, note the superiority of Pennlawn red fescue in the humid East, the indifferent performance of Rainier; while in the hot dry West, Rainier is the superior grass of the two.

We have been sufficiently impressed by the small regional test that we have written out a program, though to date we have not moved to implement it. Such a program based on population genetics, allows us to emphasize the conditions of testing, particularly the many conditions of climate and competition. The manner in which we propose to get a handle on the program is through disease. Firstly, it is disease that destroys the uniformity of our green carpet and makes present bluegrasses unsuited. Secondly, is the nature of the turfgrass diseases.

There are three basic parasitic diseases of turf: rust, smut and mildew. The remaining diseases are facultative parasites that appear only when the grass is stressed to the point of injury. We propose then to use as the conditions of the test, those conditions which predispose to disease, and to use disease as the selective agent to cause genetic shift in our population (where diseases are those caused by the facultative parasites).

The stress factors in our situation which lead to disease are climate (high temperatures), soil compaction and water stress, and those factors which increase competition or reduce carbohydrate supply, such as fertilization and short and frequent mowing.

Advantages of Regional Testing Program

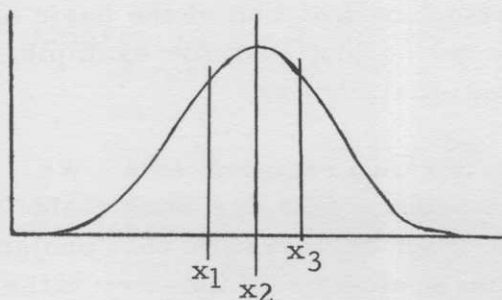
The advantages of such a program have been noted by Hanson (1972). Beginning with a seed source of high genetic diversity, we quickly make large gains towards our goal with minimum effort and cost. In ordinary population genetics one assumes that if a property Z, having a normal distribution, is selected for, that the mean of the population will be changed as in Figure 1, with the maximum shift in the first selected generation. If one is working with a population of apomicts, essentially all of the shift occurs in the first selected generation. Benefits in subsequent generations would result from increasing the selective pressure. Variability in subsequent generations would result as genetic noise accompanying selection pressure on other characteristics. If the initial population is partially apomictic, subsequent generations may lose in sexuality and gain in apomixis.

An added advantage of the program is that we would be selecting for resistance to the local strains of disease organisms.

Disadvantages of Regional Testing Program

Disadvantages of the program we have outlined are several. Foremost is that it fails to select for such turf qualities as color, texture and density. The literature indicates that bluegrass may be bred for disease and insect resistance, climatic adaptability, turf habit, shade tolerance, texture, density, color, sexuality, rhizome extension and number emerging. Our program selects only for disease resistance, turf habit, climatic adaptability and against sexuality (Hanson and Juska, 1962; Long, 1972).

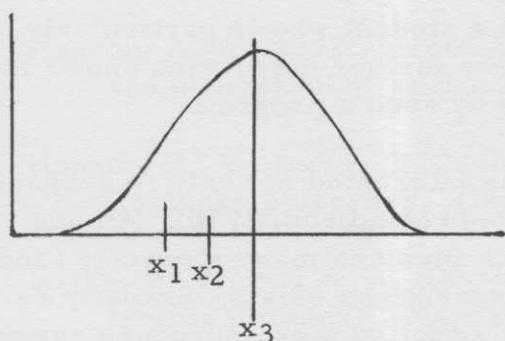
A second disadvantage is one to be considered in all population work--reproductive potential. There is always a danger we will lose our genetic base by overproductivity from a few individuals, hence special techniques must be used to suppress expression of high productivity (we might harvest equal seed weights from equal areas).



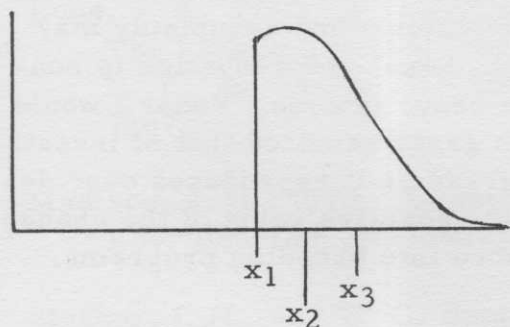
x_1 - eliminated by selection

x_2 - mean of the normal distribution

x_3 - mean of population after selection



In the first selected generation, the distribution is assumed normal about the new mean.



With apomixis, the first selected generation has a non normal distribution about the new mean.

Figure 1. In using the method of population genetics to improve Kentucky bluegrass, we assume we are shifting the population mean of some characteristics as illustrated above.

Another disadvantage is a lack of knowledge of the interactions between the factors we are selecting for, and of the basic assumptions we are making that may not be justified; for example, that characters do occur in a normal distribution.

A final disadvantage refers to our own requirements. We have limited time and funds and we require that our work yield results that can be generalized. Without such results that project is one of simple application and should be done by industry rather than the university. To obtain generalizable results requires extensive data collection. Our budget would justify this only if we thought the results would be of value to our constituency. We don't know whether limited production of a variety for a local region is feasible. While work has been planned, we probably shall not execute it unless we find a student who is particularly eager for such a program, or unless further discussion shows how we can advance population studies by such a program.

In recent years I have become interested not only in grasses, but in the whole subclass of monocots (Madison, 1970). Among perennial colonizing species, which includes many monocots (and dicots), a form of alternation of generations exists. Sexually reproducing individuals adapted to a situation may reproduce asexually over an indefinite period of time. In the asexual phase, chromosome numbers may change, and polyploidy and aneuploidy may appear (Stucky and Banfield, 1946). Sexual reproduction is conservative and tends to restore the basic genome. Today I would find the most exciting challenge in grass genetics that of investigating the genetic plasticity of a grass as it reproduces over decades or centuries by rhizomes, the adaptive value of the changes, and whether they could be introduced into breeding programs.

In an apomict such as Poa pratensis, we may already be taking advantage of genome changes. In Agrostis spp. the evidence suggests that the mutants are lost by sexual reproduction (Stucky and Banfield, 1946).

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BREEDING BEHAVIOR NOTES ON SOME IMPORTANT TURFGRASS SPECIES

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Introduction

Progress in improving grasses for turf applications is influenced to a large degree by the breeding behavior of the subject grass species. Methods employed for combining desired traits in the grasses selected are effected by a number of the breeding behavior characteristics. Some of the important breeding behavior aspects include: (1) chromosome number and ploidy levels; (2) megasporogenesis; (3) microsporogenesis; (4) embryogenesis; (5) self & cross sterility/fertility; (6) amphimictic or apomictic.

Plant Breeding Characteristics

Chromosome Number and Ploidy Levels

The chromosome numbers and ploidy levels of the important turfgrass species vary widely. Within the species where sexual reproduction occurs (amphimictic), chromosome number will have a marked effect on the methods used in breeding and the outcome. In general, hybridization of parental types within a species with the same chromosome number is more likely to succeed than hybridization of parental types with differing chromosome number. Long and Bashaw (1961) reported no success in crosses made on selected St. Augustine parental types. A later study of chromosome numbers showed that one of the parental types used was a triploid, thus, accounting for lack of success in hybridization. Chromosomal imbalance in the triploid type resulted in abnormal microsporogenesis and megasporogenesis.

The effect of chromosomal imbalance has been found to be most pronounced in interspecific hybridization. A lack of proper pairing where numbers vary and low or no homology with nearly balanced numbers lead to high sterility.

In grass species where apomixis is prevalent, varying chromosome numbers have not lead to high sterility levels.

Megasporogenesis

This represents the sequence of events that lead up to the development of the egg cell in the female portion of the grass flower. In grass species such as Lolium perenne, Stenotaphrum secundatum and Festuca rubra where normal sexual reproduction occurs, a linear tetrad of spores are formed with one spore becoming functional and the remaining three disintegrating. The functional megaspore then gives rise to an eight nucleate embryo sac as a result of two successive mitotic divisions. One nucleus of the eight functions as the egg cell and fuses with a nucleus from the pollen tube after it enters the embryo sac. Another nucleus from the pollen tube fuses with polar nuclei in the embryo sac which then develops into endosperm. This then represents the sequence of events of megasporogenesis for the normal sexual species.

In apomictic species, at least two mechanisms of apomixis are common. In Poa alpina, Poa nemoralis, and Poa palustris diplospory occurs. Instead of a linear tetrad of spores forming from an archegonial initial as the case of sexual species, the archegonial initial itself goes through mitotic divisions and forms an unreduced embryo sac. In contrast, the embryo sac of the sexual species has a reduced chromosome complement. Apospory represents the apomictic mechanism common to Poa pratensis. In this instance a nucellar cell develops into an unreduced embryo sac. In facultative apomicts such as Poa pratensis, a normal reduced egg may occur. Thus, varied chromosome numbers of sexual progeny may result from fertilization of a reduced egg yielding diploids and those produced from unreduced eggs yielding triploids. Progeny may vary widely in chromosome number.

As one plans a breeding program, it is quite essential to take into account the events in megasporogenesis, particularly where apomictic species such as Poa pratensis are considered.

Microsporogenesis

Where megasporogenesis relates to the functions leading to the development of the egg cell in what one would perhaps consider the female phase, microsporogenesis includes the events leading to the development of pollen which is classified as the male phase. The sequence starts in the anthers of the grass flower. Special cells (microspore mother cells) undergo reduction and division to form four functional microspores which then mature to form pollen within the anthers. In the steps of microsporogenesis, reduced spores are formed.

Cytological studies of microsporogenesis reveal substantial information to alert the plant breeder as to what may be encountered in an improvement program for the species in question. Long and Bashaw (1961) were able to determine why hybridization was not successful in selected Stenotaphrum secundatum types after completing a survey of microsporogenesis of the parental types. As reported above, one parental type was triploid. A study of microsporogenesis revealed a high degree of sterility as would be expected. Without developing this information, considerable time may have been lost in further hybridization attempts.

Embryogenesis

Following the two parallel functions of megasporogenesis and microsporogenesis, the next sequence of events leads to the development of the embryo which represents the new generation. This is a critical phase in terms of introducing desired genetic variation into a planned hybrid combination. The most sensitive phase is found in pollination and fertilization. Lack of homology between two parents may be reflected in rejection of the desired pollen by the stigmatic surface of the embryo sac unit. If acceptance occurs, the pollen germinates on the stigmatic surface and forms a pollen tube which penetrates into the embryo

sac. If chromosomal imbalance is a factor, the fertilization step may be blocked. During pollen tube growth into the embryo sac two mitotic divisions occur; whereas, during sperm formation in the animal system no mitotic divisions take place. This represents one of the key differences in reproduction in plants as contrasted to reproduction in animals. Pollen is much more sensitive to chromosomal imbalances and deficiencies than sperm in animals and as a result the pollen aborts. The egg cells in plants are not as sensitive as the pollen to the chromosomal imbalances and as a consequence these imbalances are more likely to be transmitted through the female gametes.

After the pollen tube penetrates the embryo sac, one nucleus fuses with the egg cell and another fuses with two polar nuclei. The fertilized egg cell then develops into the embryo while the other fused nuclei give rise to endosperm which forms a part of the new seed. Chromosomal imbalance, lack of chromosomal homology and genetic homology are revealed when studies are made of these events.

In apomicts such as Poa pratensis, the unreduced embryo sac and resulting unreduced egg develops into an embryo without fusing with the nucleus arising from the pollen tube. The nucleus from the pollen tube that fuses with the polar nuclei to form the endosperm has been reported to stimulate further development of the unreduced pro-embryo into an embryo. In species with sexual reproduction, such as Stenotaphrum secundatum (diploids), normal fusion of the egg and the nucleus from the pollen tube and the other tube nucleus with the polar nuclei to form endosperm occurs. This represents a significant functional difference between apomicts and amphimicts.

Self & Cross Sterility/Fertility

The planning of a breeding improvement program must take into account these phenomena. In the species Cynodon dactylon, self sterility is quite common. When certain breeding procedures are used to work towards plant uniformity, a high degree of sterility occurs leading to low seed set. In a very practical sense, to obtain acceptable seed yields in Arizona Common bermudagrass,

it is necessary to have an unrelated (to a degree) pollen source to provide for higher cross fertilization. This leads then to lack of uniformity in resulting progeny which is reflected in coarse types noted in lawn plantings.

Self sterility is also common in Lolium perenne and requires that appropriate breeding methods be employed to circumvent the self sterility phenomena.

It is particularly important to take an inventory of the aspect of cross compatibility or incompatibility on new grass species planned for an improvement program. For example, if one were to plan an improvement program on Eremochloa ophuroides, a prerequisite would be to study the cross and self sterility or fertility phase before progress may be realized.

Amphimictic or Apomictic

Plants that exhibit sexual reproduction are classified as amphimicts and those that reproduce by asexual means are classified as apomicts (Gustafsson, 1946).

In the amphimictic process, the gametophyte has a reduced chromosome number. In the gametophyte an egg-cell, two synergids, two polar nuclei and three antipodal cells develop. If the egg-cell with the reduced chromosome number in this process forms an embryo following fertilization, this then represents sexual reproduction. Plants in which reproduction is not associated with any fertilization are termed apomictic.

Two subgroups of apomixis include agamospermy and vegetative reproduction. In the first subgroup (agamospermy), reproduction takes place by means of seed. In the second subgroup, reproduction is by vegetative formations, such as bulbils, runners, etc. (Gustafsson, 1946). The two mechanisms of apomixis under agamospermy are termed diplospory and apospory. These were discussed above under megasporogenesis.

Important turf species in which apomixis (agamospermy) occurs include Poa pratensis and Paspalum notatum.

The above discussion on the breeding behavior aspect was not made in depth, but hopefully covered in sufficient detail to emphasize the importance of having background information to provide leads on appropriate breeding systems to employ for turf-grass improvement.

In the following review, several important species used for turf applications will be considered relative to our knowledge of breeding behavior and progress that has been made in application of appropriate breeding systems.

Turf Species Breeding Behavior

Kentucky Bluegrass (Poa pratensis)

Reproduction in Kentucky bluegrass is by partial (facultative) apomixis: The apomictic process allows retention of particular genotypes which usually are in a heterozygous state. In an improvement program, the desired genotype is one exhibiting a high degree of apomixis from the standpoint of varietal purity maintenance in seed multiplication.

The infrequently occurring sexual reproductive process releases potential variability, allowing the species to adapt to different environmental conditions. The partial sexual process allows the plant breeder to hybridize plants with complementary characteristics. Where sexuality is low and it is desired to be increased for selected parents for breeding purposes, mutagenic agents such as X-ray have been found effective (Julen, 1954).

Possible methods for species improvement include selection, hybridization and mutation breeding.

The selection and hybridization methods have yielded improved varieties. In selection, superior plants are taken from old turf areas or in space plantings. Superior plants are evaluated for turf characteristics, followed by seed multiplication and release.

In hybridization, highly apomictic males are crossed to superior females with a degree of sexuality. Selection may begin in the F_1 generation because material is heterozygous, therefore, F_1 plants likely are different. Segregation in the F_2 generation is limited because of apomixis.

In mutation breeding, selection may begin in M_1 (the generation that was treated) because of the heterozygous condition of the genotype. Segregation is again limited in the M_2 generation because of apomixis.

Cytogenetic studies reveal chromosome numbers ranging from $2n = 18$ to 150 (Grazi, 1961). Chromosome pairing may be disturbed when plants are subject to varied conditions (Grun, 1952).

In seed formation, competition between apomictic and sexual seeds during embryo formation may be influenced by environment. In commercial seed production, location of fields in environments favorable for apomictic seed formation should be considered.

In aiding breeding programs, more information is needed on conditions necessary for seed formation such as environments required for inflorescence initiation and flowering. Such information would aid handling plants with different maturities relative to crossing which would allow more flexibility in time of year when hybridization is carried out.

St. Augustinegrass (*Stenotaphrum secundatum*)

This species is presently used extensively for lawns in the Gulf Coast States and to a limited extent in Southern and Central California. The distribution of this species for lawns is limited by temperatures. Normally, it does not persist in regions where temperatures range down to 10°F for several weeks.

Research was carried out at Texas A & M during the late 1950's and early 1960's to study breeding behavior of St. Augustinegrass. From this research (Long and Bashaw, 1961) it was determined that chromosome numbers varied from $2n = 18$ to $2n = 36$.

The method of reproduction was found to be amphimictic. In diploid accessions studied, normal microsporogenesis, megasporogenesis and embryogenesis were observed. Intraspecific hybridization studies revealed that diploid accessions were cross and self fertile.

Plant collections obtained from the Southeastern United States were found to be largely triploid. Collections from the Western Gulf Coast were found to be largely diploid. Collections obtained from Africa were diploid while one accession obtained from Australia was determined to be triploid.

Intraspecific F_1 hybrids were found to be superior in some characteristics over parental types (Long, 1972).

Tall Fescue (*Festuca arundinacea*)

Tall fescue, although classified as a cool-season perennial grass, is considered one of the most widely adapted grasses relative to temperatures. It is used for turf ranging from the Southern transition region to Canada.

It is a naturally cross-pollinated grass ranging from highly self-sterile to highly self-fertile (Cowan, 1956). Agronomic studies in the species have emphasized forage applications with little attention given to turf applications. The adaptable breeding methods for improving the species are considered as follows:

1. Phenotypic individual selection: Burton and Devane (1953) showed that high heritability (76-90%) as well as wide genetic variability (18-90%) exists for certain characteristics in tall fescue. This indicates that individual selection to improve the species for turf application is probable. Sources of material should be included from a wide area of the world where it is grown. Individual plants are selected on the basis of their own performance for one or more characters in space-planted field nurseries. The selected plants are used either in clonal evaluation or in crossing with other desirable types.

2. Controlled hybridization: Although the floral organs of tall fescue are small, certain controlled hybridization steps can be carried out. By intercrossing selected plants and by practicing recurrent selection within maternal lines, an increase in the frequency of desirable genes may be achieved. The F_2 population will provide new combinations of characteristics not seen in the parent and F_1 plants. With diallel crosses, both specific and general combining abilities of the selected types, can be estimated.

3. Synthetic variety procedure: Synthetic varieties are more desirable in the breeding of polyploid species, such as tall fescue. The selected non-inbred clones can be 2 to 6. The parents selected are dependent mainly on their high general combining abilities and low inbreeding depression. Clonal performance and specific combining ability represent further factors for consideration. Polycrosses and selfs can be used to evaluate general combining ability and inbreeding depression.

Cytogenetic investigations show the chromosome number for tall fescue to be $2n = 42$. Microsporogenesis, megasporogenesis and embryogenesis are the same as for other sexually reproduced plant species.

Fine-Leaved Fescues (Festuca rubra, Festuca ovina)

The fine-leaved fescues (creeping red fescue and chewings fescue) are used widely in the same general region as Kentucky bluegrass. Varieties of these species are used largely in blends with Kentucky bluegrass. Most seed products used for shaded areas include the fine-leaved fescues. The botanical variety of Festuca ovina var. duriuscula has recently been improved by breeding and is finding use in turf seed products.

Cytogenetic investigations reveal chromosome numbers of the fine-leaved fescues from $2n = 28$ to $2n = 56$. The basic chromosome number of the fine-leaved fescues is considered as seven. These species are classified as amphimicts and exhibit normal sexual reproduction.

Improved fine-leaved fescues have been developed by two breeding methods which are:

1. Selection: Selection either mass or single line tested for turf quality and seed yield and then released for commercial application.

2. Synthetic variety procedure: Recently developed varieties are the result of this breeding procedure. Lines or clones tested by polycross techniques to measure combining ability are selected for use in synthetic varieties.

Testing of individual component clones for turf performance and flowering are carried out in synthetic variety development procedures.

Perennial Ryegrass (*Lolium perenne*)

Renewed interest in the use of perennial ryegrass for fine turf applications no doubt has resulted from the introduction of improved fine-leaved turf type varieties within the past five years (Funk & Engel, 1968).

Improvement of perennial ryegrass has resulted through synthetic variety breeding procedures. Outstanding clones are tested by polycross techniques to assess combining ability. Those combining well are selected for including in the makeup of improved synthetic varieties.

Cytogenetic investigations show perennial ryegrass to reproduce sexually. The chromosome number is $2n = 14$ with a basic chromosome number of seven. Some tetraploids have been produced.

Bahiagrass (*Paspalum notatum*)

This species is used in the Southeastern United States in lawn applications where a utility type turf is desired. It withstands traffic quite well, is free of major turf pest problems, and is highly drought tolerant.

Improved varieties have been developed by selecting outstanding clones (Burton, 1951). Isolated clones have been tested largely for forage applications. Superior clones, based on forage performance, have been increased and released. One variety, Argentine, appears to be preferred for turf application.

Both apomictic and sexual types occur within the species. The apomictic types are classified as obligate.

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Discussion Period

Dr. Whalley: Has there been any work done on the induction of apomixis, in triploid Stenotaphrum spp. so that the production of viable seed is possible?

Dr. Long: No.

Dr. Duich: Has anyone ever used colchicine on St. Augustine, particularly in those triploids?

Dr. Long: I worked some time with colchicine while at Texas A & M and for some reason we couldn't get it to work. I did take another species that was somewhat more difficult, the dallas-grass (Paspalum dilatatum), and was able to take it from 50 chromosomes up to 100, or double that one. I thought it would be even more difficult to work with. So far we haven't been successful with St. Augustine.

Dr. Madison: Would you characterize the African Stenotaphrum spp.?

Dr. Long: On the African biotypes, there were two; one was brought in from Kenya and another from South Africa. The one from Kenya was diploid. The one from South Africa was an extremely coarse plant and we were never able to get flowering. I didn't go to the root tip or somatic tissue to check this out, but from the general appearance, it would have all the characteristics of a tetraploid. The one tetraploid that we picked up from the Florida collection was as you would expect relative to larger plant parts. The pollen was substantially larger than the diploids.

Dr. Dudeck: You characterized Paraguayan and Wilmington as tetraploids, 40 chromosome obligate apomictic. Did you mention Argentine and Pensacola?

Dr. Long: Pensacola is the diploid. Argentine, as I understand, is the obligate apomict with 40 chromosomes.

Dr. Payne: Is it necessary to have pollen from an outside source in order to stimulate seed development in an apomict?

Dr. Funk: No, but a foreign pollen will work.

Dr. Daniel: What's the chromosome count of zoysia?

Dr. Long: Zoysia has a chromosome number of $2n - 40$.

PLANT VARIETY PROTECTION ACT AND TURFGRASS BREEDING

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Introduction

I would like to cover two items in this presentation. One, discuss some of the highlights of the Plant Variety Protection Act and, two, explain how we are processing plant variety applications. The Plant Variety Protection Act was signed into law on December 24, 1970. It extends voluntary, legal protection for a period of 17 years to developers of new varieties of plants which reproduce by seeds. Protection has been available for vegetatively propagated material through the Patent Office, U. S. Department of Commerce. Carrots, celery, cucumbers, tomatoes, okra and peppers are not included in the Plant Variety Protection Act.

Requirements for Protection

Three requirements qualify new varieties of plants for protection as "novel varieties." The novel variety must be distinct, uniform and stable. It must differ from all known varieties by at least one identifiable character. This could be morphological, physiological or some other characteristic, such as the baking quality of a new variety of wheat. Any variation that may exist in a variety must be describable, predictable and commercially acceptable.

Finally, the variety, when sexually reproduced and reconstituted, must remain unchanged in its essential and distinctive characteristics to a degree expected of similarly developed varieties.

Protection cannot be given to a variety if it has been a public variety; i.e., if it has been for sale more than a year in this country or if it was described in a technical journal more than one year before filing an application. Our present law states that an application will not be accepted after one year from the filing date of an application in a foreign country. However, we are proposing to change the time period to not more than four years. The time period would be based on testing requirements of each country. The law is designed to protect a novel variety from being sold without the consent of the originator of the variety.

Procedures Necessary

The breeder who wants his variety protected applies to the Secretary of Agriculture. The breeder must fully describe the variety botanically, including the distinctive features indicating novelty, and give the genealogy and breeding procedures in developing it. An objective description form to be filled out also will facilitate establishment of varietal novelty. A breeder will also be expected to deposit and maintain viable seed of his variety in a public repository. The applicant must also state the basis of his ownership.

The Plant Variety Protection Office will maintain a library of scientific works to aid the office staff in administering their duties. After certificates are issued, specifications of protected plant varieties will be published in the Official Journal of the Plant Variety Protection Office. From time to time, as through an information service, the Office will disseminate to the public those portions of the technological and other public information available to or within the Plant Variety Protection Office to encourage innovation and promote the progress of the useful arts.

We hope to gradually provide a voluntary listing under our data processing system for those varieties developed, but for which no rights are desired. We will try to have as complete a record of all plant varieties in our office filing system as is feasible. This will be done only as we complete background information on each species for search purposes, and will apply only to those species on which applications have been filed.

Once the variety information is centralized, it will be uniform, meaningful and useful to the office and to all plant breeders for search and research purposes.

There are foreign breeders who would like to receive protection under our Plant Variety Protection Act and market their novel varieties in this country. We propose to restrict such granting of certificates to the extent that rights for the same species are granted our nationals by the country of the applicants.

In order to organize the mass of data describing many crop varieties we are developing a series of objective description forms. These forms, when completed, will allow us to file the information in a computerized data processing system for easy retrieval. In developing these forms, we have sought to identify characters by which varieties may be distinguished. We have endeavored to devise means that will best describe or define each character.

The breeders are usually not concerned with all of these characteristics and are often not even aware of some of them. A number of breeders who have viewed some of the forms we have developed in the Plant Variety Protection Office have said that breeders would now recognize the value in recording many of these characteristics that they had not recorded routinely. And thus, these forms would have the effect of encouraging breeders to make more complete descriptions of their new varieties.

When an applicant for plant variety protection fills out the objective variety description form, he is not required to fill in all the blanks. However, we feel it is in the interest of the breeder to fill in as many blanks as possible.

The form developed for bluegrass (Fig. 1) illustrates the approach we have taken to distinguish between varieties. Forms for other turfgrass genera will be quite similar to this one in many respects. No attempt has been made to make a complete description for each variety in the objective form. However, the form should allow the Examiner to limit his search to a relatively few varieties that must be compared for other expressions of difference. Kentucky bluegrass characteristics were relied on heavily in developing the form, since it is the most important bluegrass grown for turf.

The principal characteristics in the form are grouped under 18 headings. For example under No. 6, Leaf Blade -- color, surface and size measurements are included. Standard varieties are listed that represent each color.

Under No. 9, Panicle -- six standard varieties are given in order to indicate the number of centimeters that the new variety is shorter and/or longer than one or two of the standard varieties. Plant height is indicated similarly with comparative varieties.

Twelve items are listed under No. 17, Environmental Adaptation; for example, heat, drought and air pollution. An extra blank is provided to record other environmental factors. Each factor listed is scored: 0 - Not tested, 1 - Susceptible, and 2 - Resistant.

Under No. 18, fourteen of the most important diseases of bluegrass are listed that are scored as given under No. 17.

The characteristics included in the form do not represent all characteristics that may eventually be included. With additional information and experience in using the objective description forms of different crops, modifications will be made in them. Additional characteristics will be included and others more accurately or appropriately defined. Additional and/or other cultivar standards will be desirable. Colors might be standardized with an appropriate color system such as the Nickerson's Color Fan.

We solicit comments and constructive suggestions for modifying the form to improve on its practical usage for varietal description.

OBJECTIVE DESCRIPTION OF VARIETY
Bluegrass (Poa spp.)

Figure 1

1. KIND:

- ☐ 1 = Poa compressa
3 = P. trivialis

- 2 = P. pratensis
4 = Other (specify) _____

- ☐ Region of best adaptation: 1 = Northeast 2 = Transitional Zone
3 = North Central 4 = Pacific N.W. 5 = Other (specify) _____

2. MATURITY (At first anthesis):

- ☐ Date: 1 = Early (Delta) 2 = Medium early (Fylking)
3 = Medium (Newport) 4 = Late (Merion)

- ☐ Number of days earlier than ☐ 1 = Nugget
2 = Fylking
3 = Delta
4 = Merion
☐ Number of days later than ☐ 5 = Newport
6 = Baron

3. PLANT HEIGHT (Longest shoot from soil surface to top or head):

- ☐ cm. Height 1 = Nugget 2 = Fylking
☐ cm. Taller than . . . ☐ 3 = Delta 4 = Merion
☐ cm. Shorter than . . ☐ 5 = Newport 6 = Baron

4. HABIT:

- 1 = Prostrate (Fylking) 2 = Semi-prostrate (Merion)
☐ 3 = Erect (Delta)

5. VEGETATIVE REPRODUCTION (1 = Absent, 2 = Present):

- ☐ Rhizomes ☐ Stolons

6. LEAF BLADE:

- ☐ Color:
1 = Light Green (Rough Bluegrass)
2 = Moderately Dark Green (Merion)
3 = Dark Green (Adelphi)
4 = Blue Green (Canada Bluegrass)
5 = Other (specify) _____

- ☐ Upper surface 1 = Shiny 2 = Dull
☐ Lower surface

- ☐ mm. Width ☐ mm. Length

7. LEAF SHEATH (Base):

- ☐ Color (Seedling): 1 = Green 2 = Red
 Surface: ☐ 1 = Glabrous 2 = Pubescent 1 = Smooth 2 = Rough
☐ 1 = Non -glaucous 2 = Glaucous
 Size: ☐ mm. Length
☐ Keel: 1 = Not Keeled 2 = Keeled
-

8. LEAFINESS (At first anthesis):

- ☐ Number of leaves per tiller or shoot:
 1 = Few (1-3) 2 = Intermediate (4-6) 3 = Many (More than 6)
-

9. PANICLE:

- ☐ mm. Length
☐ mm. Longer than ☐ 1 = Nugget 2 = Fylking
☐ mm. Shorter than ☐ 3 = Delta 4 = Merion
☐ mm. Shorter than ☐ 5 = Newport 6 = Baron
☐ Number of panicles per Plant ☐ Milligrams seed per panicle
 Branches LOWEST WHORL:
☐ 1 = Drooping (Prato) 2 = Horizontal (Merion) 3 = Other specify
 PANICLE HABIT:
☐ 1 = Nodding () 2 = Upright (Nugget)
☐ mm. Spikelet Length
-

10. LEMMA:

- ☐ Basal Webbing: 1 = None 2 = Scant 3 = Copious
 Pubescence 1 = Glabrous 2 = Slightly Pubescent
 3 = Pubescent 4 = Other (specify) _____
☐ Keel ☐ Lateral Nerves ☐ Intermediate Nerves
☐ Intermediate Nerves: 1 = Distinct 2 = Obscure
-

11. SEED:

- ☐ Apomictic %: 1 = more than 95 2 = 85 to 95 3 = less than 85
☐ Phenol Reaction: 1 = None - Lemma removed (Merion) 2 = Beige (Cougar)
 3 = Brown (Windsor) 4 = Black (Delta - 2 hours)
 5 = Black (Anheuser - 24 hours)
 Size: . ☐ mm. Width ☐ mm. Length
 Weight: ☐ Grams per 10,000 seed
☐ Chromosome Number (2n)
-

12. TURF DENSITY MAINTENANCE AT ONE INCH CUT):

- ☐ 1 = Poor 2 = Moderate (Merion)
 3 = Superior (Nugget) 4 = Excellent

13. VERTICAL GROWTH RATE:

- ☐ 1 = Slow (Nugget) 2 = Medium (Merion)
☐ 3 = Fast (Delta) 4 = Other (specify relation to a standard) _____

14. SPRING GREEN UP:

- ☐ 1 = Early 2 = Medium 3 = Late

15. FALL DORMANCY: (1 = Dormant 2 = Intermediate 3 = Not Dormant)

- ☐ Northern (42°30' + 30' Lat.) ☐ Intermediate (40° + 30' Lat.)
☐ Southern (37°30' + 30' Lat.)

16. SEEDLING VIGOR: (growth rate)

- 1 = Slow 2 = Medium 3 = Fast
☐ Seedling

17. ENVIRONMENTAL RESISTANCE TO (0 = Not Tested 1 = Susceptible
2 = Resistant):

- | | | |
|----------------------------------------------------------|------------------------------------------|------------------------------------------------|
| <input type="checkbox"/> Cool Temperature (Winter color) | | |
| <input type="checkbox"/> Heat | <input type="checkbox"/> Drought | <input type="checkbox"/> Shade |
| <input type="checkbox"/> Poor Fertility | <input type="checkbox"/> Acid Soil | <input type="checkbox"/> Alkalinity |
| <input type="checkbox"/> Salinity | <input type="checkbox"/> Soil Compaction | <input type="checkbox"/> Poor Drainage |
| <input type="checkbox"/> Air Pollution | <input type="checkbox"/> Cold (Injury) | <input type="checkbox"/> Other (specify) _____ |

18. DISEASE, INSECTS, AND NEMATODE RESISTANCE TO: (0 = Not Tested
1 = Susceptible
2 = Resistant):

- | | |
|---------------------------------------------------------|---------------------------------------------------------|
| <input type="checkbox"/> <u>Helminthosporium vagans</u> | <input type="checkbox"/> <u>H. sorokinianum</u> |
| <input type="checkbox"/> <u>H. dictyoides</u> | <input type="checkbox"/> <u>Rhizoctonia solani</u> |
| <input type="checkbox"/> <u>Erysiphe graminis</u> | <input type="checkbox"/> <u>Ustilago striiformis</u> |
| <input type="checkbox"/> <u>Fusarium nivale</u> | <input type="checkbox"/> <u>F. roseum</u> |
| <input type="checkbox"/> <u>Typula iotana</u> | <input type="checkbox"/> <u>Scelerotinia homeocarpa</u> |
| <input type="checkbox"/> <u>Puccinia graminia</u> | <input type="checkbox"/> <u>P. striiformis</u> |
| <input type="checkbox"/> <u>Pythium ultimum</u> | <input type="checkbox"/> <u>Crambus bonifatellus</u> |
| <input type="checkbox"/> Other (specify) _____ | |

INSTRUCTIONS

COLOR: Nickerson's or any recognized color fan may be used to determine plant colors of the described variety.

Discussion Period

Dr. Duich: I think one of the best ways to get some of your information, such as varietal turf performance, would be through accepted regional tests. As far as the objective description of a variety, would it be appropriate for your office to maintain a location for certain varieties to run referee growth-type tests?

Dr. Higgins: This is not necessary. The breeder must provide the data that will convince the Examiner that his variety is distinct and meets all the requirements.

A lot of thought has been given to making cultivar distinctions without grow out tests. We are going to try this route first. I think we can tell better how our system will work after we have used it a few years.

Dr. Daniel: Troy is not in many tests or in much production. I would suggest substituting Newport for Troy as one of the standard varieties because of it's wide use.

Dr. Higgins: Well, I am sure you have many comments like this and all you need to do is put them on the form distributed here. Please do not wait until 1973 to make your suggestions. Within the next few weeks will be fine. If you get another idea later, after you have sent these forms in, please let me hear from you.

Dr. Daniel: I believe that "where the data was taken" could be added to the section on seed head height. There is a difference on whether the seed was grown in Indiana or the Northwest, for example. Also where you often have a choice between three numbers -- one, two, three -- it seems it would be good if you could expand this to at least five throughout. You don't give the breeder much leeway if he is only choosing the extremes or the middle.

Dr. Long: Dr. Higgins, have you seen the check list that Dyvendok uses?

Dr. Higgins: Yes. I wondered if he has finally published this work?

Dr. Long: It's Dyvendok's checklist on morphological features used by the IVRO in Holland.

Dr. Higgins: It is just limited to certain ones, and I have mentioned some of them.

Dr. Long: I think there are quite a few of the characteristics that match up with what Dr. Nittler has observed in sheath color. Now he goes into the hairs and the color on the sheath. Dr. Dade, on our staff in Oregon, finds he does not agree with the hair characteristics that Dyvendok has and he's come up with some refinements that we could probably provide.

Dr. Higgins: This is the very thing that I would like to obtain from you.

Dr. Duich: Would you outline, by name, the table of organization of your office at the present time?

Dr. Higgins: I am sure you know the two at the top: Stan Rollin, the Commissioner, and Bernard Leese, the Chief Examiner. My title is Examiner. We have four other Examiners so far; Herbert Fisher, Ken Evans, Bob Snyder and Eldon Taylor. Taylor is also our Librarian. Snyder and Fisher are concerned with ornamental plants and vegetables. I have worked on peas but I will work on agronomic crops, as will Ken Evans. The Librarian-Examiner is obtaining books and periodicals for our library and encouraging people to send in information from all over the country. We also have two secretaries.

Mr. Mayer: What are the costs involved when applying for a plant patent?

Dr. Higgins: The application fee is \$50. However, as soon as the regulations are approved, it will be \$250. There will be an additional \$250 charge to make the search and \$250 when a certificate is awarded.

Dr. Nittler: First, on this maturity, I think blooming might be much more precise than a heading date. Secondly, this requirement for uniformity, that will have to be interpreted pretty broadly with this cross pollinated crop, won't it?

Dr. Higgins: Yes. However, this will depend on the crop.

Dr. Payne: Would you briefly describe this "search" you speak of after you receive this application. What is your procedure from the time you receive this?

Dr. Higgins: I cannot tell you in complete detail because we are in the process of developing a search procedure, and I think it will vary for different crops -- the information available, and so on. We obtain descriptions of the varieties that are in commerce. We have located a number of references that are good. They describe some of the older varieties. For many, it will be hard to get much information.

Dr. Payne: Will this ultimately be computerized in the manner of the fingerprint system where a button is pushed and a selected few are delivered for study?

Dr. Higgins: The information stored in the computer will come from the objective description forms and, as I have indicated, this information will not describe a variety completely. It is going to be a method to help us zero in on a few related varieties.

Mr. Simmons: Could you review your potential plans for handling material from foreign countries?

Dr. Higgins: We will not accept an application from a foreign country unless that foreign country reciprocates with similar protection to individuals from the United States for that specific crop. If Kentucky bluegrass is protected in Germany, then we will give applicants from Germany protection for Kentucky bluegrass in the United States.

Dr. Duich: At the present time with agronomic crops we have registration with The American Society of Agronomy, as published in Crop Science. Have you people given any consideration to any type of coordination of this registration? For example, we get involved in a lot of double paper work because now, for example, they want performance data, and they have a committee in multiple locations.

Dr. Higgins: We are working closely with Crop Science, and this whole program, to coordinate our work with this group.

Dr. Duich: When I submitted Pennfine for registration in Crop Science I contacted Stan Rollin and asked if we could put a statement on the last line, that plant variety protection had been applied for. I did this based on some of the experiences Dr. Funk had with Manhattan perennial ryegrass? Maybe scare off some of the scavengers. Is this going to be allowed? I guess this was more or less the precedent for he went along with it in this case.

Dr. Higgins: After you apply for a protection, you can start selling the seed. The seed label must indicate that protection has been applied for, and that sexual reproduction is not allowed without arrangement with the owner.

Dr. Duich: Just like "patent applied for?"

Dr. Higgins: Yes.

Dr. Daniel: At what point will you take over from the Plant Patent Office. We have previously worked with bluegrasses through the Plant Patent Office?

Dr. Higgins: We are accepting applications to protect varieties of Kentucky bluegrass now. Based on the wording and interpretation of the Act, Kentucky bluegrass is included.

Dr. Daniel: This has not been specified by law, or even by an agreement.

Dr. Duich: I can reply to that a little bit. When they were working on developing this Act, there was nothing specifically said about bluegrass and of course even the bluegrass patent being awarded under the Plant Patent Protection Act is a little fuzzy or in the grey area, and I very specifically asked ASTA to get bluegrass under this rather than the 1937 Plant Patent Act. They said the reason they did not want to put bluegrass in there specifically, since there was this matter of vegetative reproduction, was that there were other crops, particularly in the ornamental area. If they covered anything pertaining to asexual reproduction, they were going to get a lot of static on the Act itself. In order to eliminate all this opposition, they dropped bluegrass per se, in as far as naming it in the Plant Variety Protection Act of 1970.

Dr. Daniel: And yet, Dr. Higgins, you have some applications for bluegrasses already?

Dr. Higgins: Yes, seven. However, I don't know if these are vegetatively or sexually reproduced as to whether they will get protection or not.

Dr. Long: In discussions back in January with Stan Rollin, I think any bluegrass that is propagated by seed will fit the Plant Variety Protection Act. A case like Warren's vegetatively propagated bluegrass would have to go under the Plant Patent Act. We would plan to handle the St. Augustine and bermuda-grasses that are propagated strictly vegetatively under the Plant Patent Act, and not the Plant Variety Protection Act. So the language in the 1970 act is sufficiently broad to take the blue-grasses, provided you propagate them by seed.

Dr. Payne: You mentioned patenting by a foreign country. What about O. M. Scott & Sons, for example, applying for the patent of a line from a foreign country?

Mr. Doellinger: We just attended a forum sponsored by the American Seed Trade Association last week in Portland and it was stated that a U. S. National may obtain protection of a novel variety even though it results from breeding and research

conducted in a foreign country. We understand further, that a National of the foreign country may assign the ownership of a novel variety to a National of the United States who may go ahead and apply for protection.

Dr. Yu: Dr. Higgins, we know that bluegrass is apomictic and variation for chromosome numbers exist among varieties. In case a patented variety had produced some progenies with a different chromosome number, but was not detected by the original patentee, can another person who detected this change in chromosome number apply for a new patent on this plant, although the other plant characteristics remain the same?

Dr. Higgins: According to the Act, the plant cannot change. That is one of the characteristics of uniformity. If this were the only distinguishing character, the owner would not have protection. The owner would have to submit a new application.

Dr. Long: One suggestion that I think we should consider on the form for bluegrasses would be to use approximate for chromosome number.

Dr. Higgins: Possibly so, but the variability would have to be in that range. It could not vary any more than that range.

Dr. Daniel: To illustrate the approximate, the Swedes just count to the nearest number of seven. They say chromosomes in bluegrass tend to run in sevens so they just group it to the nearest multiple of seven as a means of expediting their observational work.

Dr. Higgins: This has application to other crops. Maybe the way to handle it is, instead of asking for the $2n$ number of chromosomes in the form, they should give at least the n number. That would cover the same thing, would it not? I think that would only be one factor.

Dr. Daniel: How soon do you hope to finalize this form? You have started; you have a certain beginning here.

Dr. Higgins: I will work on it as soon as I receive your suggestions for improving the form. I would say within two months.

Mr. Mayer: I assume we submit data as an addendum to this form. Will there be a format to use on the type of data that we're syppose to submit?

Dr. Higgins: There are no special details in that regard.

BREEDING SEEDED VARIETIES OF BERMUDAGRASS FOR TURFGRASS USE

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Introduction

The bulk of the bermudagrass seed moving in world trade for the past half century has been produced in Arizona. As alfalfa seed production in the Yuma area declined under a combination of fewer pollinators and increased soil salinity, bermudagrass, present as a weed, increased under salinity levels too high for the alfalfa, and bermudagrass seed changed from a by-product to a cash crop. The drawbacks and some of the advantages of Arizona bermudagrass seed for turf relate to cyclic seed reproduction under irrigation; first as a weed and later as a crop in the hot, dry climate at Yuma. The first bermudagrass in the area probably arrived a century or so ago from Western Mexico and from Southern California (Kneebone, 1966). Characteristics related to origins and history include: capacity to endure drouth, salty conditions and mismanagement, susceptibility to diseases under humid conditions, rapid spread and lack of winterhardiness.

A frequent contaminant in bermudagrass seed is giant bermudagrass, common in the Yuma area and probably stemming from introductions from India, tested there by the Bureau of Reclamation before World War I. Natural crossing frequently occurs between the diploid giant bermudagrass and the tetraploid bermudagrass. Giant bermudagrass, as its name implies, is coarse, undesirable as a turf and it is much less tolerant of Southeastern pathogens than bermudagrass.

Bermudagrass Research Program

The present bermudagrass research program in Arizona began in 1959 in response to concern by seed growers with potential market losses to vegetatively propagated varieties. A certification program was developed to provide Southern consumers assurance that they would have no contamination from giant bermudagrass and collections of breeding material were assembled to assess the potentials and begin an improvement program. Arizona certified seed today is not only free of giant bermudagrass, but is considerably more uniform in type, a result of stringent roguing of off-types in two cycles of reproduction of typical bermudagrass preceding present foundation seed production. This seed must be used to establish new certified seed fields. As yet, however, a distinctly different seeded turf type variety has not been released.

Harlan et al. (1970) describe eight species and ten botanical varieties of *Cynodon* from which three of their six varieties of *C. dactylon* (dactylon, elegans, polevansii), both varieties of *C. incompletus* (incompletus and hirsutus) and *C. transvaalensis* can be considered to have turfgrass potential. The varieties of *C. dactylon* are tetraploid ($2n = 36$), the others diploid ($2n = 18$) with occasional tetraploids in *C. incompletus* var. hirsutus (Harlan et al., 1970). Giant bermudagrass, *C. dactylon* var. Aridus, is diploid and a probable progenitor of *C. dactylon* var. dactylon. Crosses have been made among and between all of these, with the best known being the triploid cultivars from *C. transvaalensis* x *C. dactylon* var. dactylon; Tiffine, Tifgreen, Tifway and Tifdwarf. Ugandagrass is a cultivar of *C. transvaalensis*, while U3 and Tiflawn are selections from *C. dactylon* var. dactylon. Dr. Huffine at Oklahoma has been testing some of his collections of *C. incompletus* for roadside turf and several have been grown at Tucson. But there has been little progress with this taxon, the chief objection being the very hairy leaves of most types.

Gene flow between distinct types of bermudagrass is restricted by sterility and infertility. Inhibited germination occurs with seeds of some crosses (Harlan et al., 1969, 1970) and in

many cases F_1 plants tend to be vegetative and head poorly. The causes of sterility appear to be chromosomal, not in the traditional sense of meiotic irregularity, but more cryptic factors, possibly short inversion segments or deletions. Translocation races are known in *C. dactylon* var. *dactylon*. Within *C. dactylon* var. *dactylon* and in other varieties, relatively infertile plants are very common, even in commercial seed fields. The typical vigorous, promising selection for turfgrass is almost always infertile, and strongly vegetative, some types never heading. Whatever the causes of infertility, crosses of fertile x infertile normally give infertile and often poorly heading progenies, a good feature for turf, but unfortunate for selection toward seeded types incorporating the good points of their respective parents.

Although seed analysts report little difficulty with seed dormancy in commercial lots, much of the seed obtained from breeding lines (whether open-pollination, selfed, or crossed) tends to be dormant and progenies are often limited for this reason. Perhaps 50% or more of the seed obtained fails to produce seedlings. Many selfs are quite vigorous and the extremely weak selfed seedlings familiar in other cross-pollinated species are seldom seen. Perhaps the poor recombinations tend to be in the dormant seeds. This would be in accord with the observations of Harlan et al., (1969, 1970) of increased dormancy with seeds from certain crosses.

There is enormous genetic variability available for exploration in bermudagrass. To use pest problems as an example, plants varying from susceptibility to near immunity to bermuda mite, Rhodesgrass scale, Ruth's scale and spider mites have been observed. Resistance has not yet been found in turf-type plants for a new pest (so far confined to seed fields), the pink mealy bug which has been partially accountable for the present high price of bermudagrass seed. Only in giant bermudagrass have plants free of this pest been isolated.

Another Arizona problem is iron chlorosis. Some genotypes become chlorotic very readily and others maintain an acceptable green color. Obviously, there are equally wide ranges in color,

leafiness, cold tolerance, winter hardiness, etc., which are of potential value. One of the more promising developments in this program is the discovery that turf quality in some infertile types tends to be dominant in F_1 's between these plants and good seed setting plants which possess few other virtues. Using plants with high seed setting potential as female parents provides an obvious opportunity for potential commercial seed production.

Bermudagrass Breeding Program

Objectives of the breeding program have been to select or develop high combining clones which will produce profitable yields of "hybrid" seed that will, in turn, provide consumers with superior turfgrass. Two important features of bermudagrass make this approach feasible: (1) Easy vegetative propagation, and (2) a high level of self-sterility. Selfed seed set is normally less than .1% on a floret basis. Self-sterile clones propagated vegetatively in alternate rows would provide seed for planting in the consuming area. Using only two clones should result in a much more uniform product than broader based combinations or than advanced synthetic generations. Nursery studies from mutual bag crossing have confirmed this.

Because infertility and other seed production problems are the foremost bottlenecks to a seeded variety, all new materials are first checked for seed potential, hoping to retain only those with at least some fertility for the breeding program. Since Arizona seed is produced primarily for the Southeastern United States, selections with demonstrated value there have been of greater interest than germ plasm from old stands in Arizona. Composites of local selections have given turf no different from certified seed. Ultimately, a body of source materials will be accumulated which will combine quality potentials with at least reasonable seed production. The genetic variability for most traits of value from a turf standpoint is available. Some crosses produce high quality progeny. Seed set from such crosses must be raised from 5% to 85%. Some plants in commercial seed fields have seed sets this high or higher. Vigorous selections from the southeast generally have seed sets of 5% or lower. Research is being done on seed production practices, which may help to ease this problem. Growers

for example, often irrigate too heavily for best results. Pollen germination studies at Tucson some years ago suggested that heavy concentrations of pollen were necessary for good seed set and pollen longevity was limited, making extreme isolation distances unnecessary. Outcrossing studies with isolated plants of giant bermudagrass in bermudagrass seed fields suggest that when distances from other giant bermudagrass plants are as little as 30 feet, the diploid giant bermudagrass plants will produce up to 75% triploid outcrosses to the tetraploid bermudagrass. It is assumed that there is similar dependence upon nearby pollen sources in bermudagrass. Lacking good genetic markers, this has not been tested as yet.

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IMPROVEMENT OF KENTUCKY BLUEGRASS FOR THE TRANSITION ZONE

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Introduction

In the Eastern United States it is difficult to grow satisfactory turfgrass in an area of transition between cool-humid and warm-humid conditions. In this zone, cool-season grasses do poorly because of relatively high temperatures, high humidity and drought, while warm-season grasses have a long dormant season and may be subject to winterkill (Holt, 1969). No turfgrass species is well adapted in this transition zone. The predominant turfgrasses used are Kentucky bluegrass varieties and mixtures of Kentucky bluegrass and fine-leaved fescues. Bermudagrass and zoysia perform well as summer turf but are disliked because of their long period of winter dormancy. Tall fescue provides an acceptable coarse-leaved lawn in areas where Kentucky bluegrass is difficult to maintain and is better adapted than Kentucky bluegrass to areas of hard use, steep slopes, and sunny exposures in the transition zone (Juska et al., 1969).

In Virginia, Kentucky bluegrass and other cool-season species are adapted to the Northern Piedmont and areas close to or west of the Blue Ridge Mountains. Mixtures of Merion Kentucky bluegrass with Kenblue or South Dakota Certified (with some red fescue added for shaded areas) are the predominant turfgrasses recommended and used (Shoulders et al., 1969). Eastern Virginia and the Southern Piedmont (with about 1/2 of the area of the state and perhaps 2/3 of the lawns and other turfgrass) is in the transition zone. Here Kentucky bluegrass with red fescue or Kentucky 31 fescue is recommended for shaded lawns with bermudagrass, zoysia or Kentucky 31 tall fescue recommended for open sunlight and partial shade. All of these species have problems with establishment, management and adequate performance in the transition zone.

Temperature Factor

Temperature is a critical factor in adaptation of turfgrass species. Estimates of effective day and night temperatures to which several grasses are well adapted have been made by Madison (1971). (These are calculated according to a suggestion made by Went who takes the difference between the mean minimum and mean maximum temperature and divides it by four. This figure is added to the mean temperature to get an effective day temperature and subtracted from the mean to get an effective night temperature.) The approximate summer temperature ranges for several turfgrasses are:

Species	Effective night temperature, °F	Effective day temperature, °F
Red fescue	55 - 65	61 - 71
Kentucky bluegrass	58 - 68	69 - 79
Bermudagrass	70 - 82	80 - 92
Zoysia	75 - 85	84 - 94

from Figure 4.6 of Madison

The area between Kentucky bluegrass and bermudagrass (which do not overlap) is considered a transition zone with no turfgrass species well adapted. Effective day and night temperatures for several locations in Virginia were determined from long-time summer temperature data. Blacksburg and areas west of the Blue Ridge were found to be well within the area of Kentucky bluegrass adaptation, Roanoke and Lynchburg on the fringe of this area, Richmond and the area near Washington, D.C. in the transition zone, and Norfolk on the edge of the area of bermudagrass adaptation.

Climatic Factor

In addition to air temperature, Ward (1969) in his consideration of climatic factors that affect turfgrass performance, mentions soil temperature, moisture, humidity, radiant energy, wind and microclimate. Moisture from precipitation during the growing

season is important to the performance of turfgrass in Virginia where each year there is a 10% probability of 12 or more consecutive drought days. Droughts are even more common in the Southern Piedmont and Eastern Virginia than the state as a whole (VanBavel and Lillard, 1957). Microclimate, defined as climate near the soil surface, is influenced by slope, shade, mulches, and other factors. In West Virginia, Bennett and Elliott (1972) reported that over a three year period in midsummer the average amount of a Kentucky bluegrass stand affected by Helminthosporum vagans on a south-facing slope was 55% while two nearby north-facing slopes averaged 6% and 8% affected.

Improvement of Kentucky bluegrass for the transition zone is furthered by the great diversity present within the species Poa pratensis L. and the apomictic character which makes it possible to utilize superior chance or planned hybrid genotypes. Extensive clonal collections, especially from sites not considered favorable for bluegrass, will be evaluated for turf quality. A close correlation was found by Funk et al. (1967) for turf quality of clonal and mass-seeded plots. Some selections from old turf areas may be useful directly. Other selections with characteristics that appear to be of value for performance in the transition zone will be used as parents in an intraspecific hybridization program similar to that proposed by Funk and Han (1967) and reported by Pepin and Funk (1971) to be successful in producing promising apomictic hybrids.

Adaptability of Kentucky Bluegrass

There are only limited data available on characteristics of Kentucky bluegrass strains which are important to their performance under summer temperature stresses in the transition zone. With three bluegrass strains of distinctly different origins grown at four temperature regimes, Youngner and Nudge (1968) found that Merion, a strain that originated in an area of relatively high July temperatures, tended to have the highest carbohydrate reserves and to produce more top growth than Fylking or Newport, which originated in cooler areas. Five Kentucky bluegrasses - Pennstar, Nugget, Kenblue and two selections made near Norfolk, Virginia - were grown at three temperature regimes and at high and low nitrogen levels by Watschke et al. (1970). They concluded that bluegrasses

originating in warm regions were better adapted to high temperature stress because of inherent lower nitrate absorption and higher carbohydrate levels. In a further study, Watschke et al. (1972) grew ten bluegrass strains under high temperature stress for four weeks and recorded foliar yield and carbohydrates and rates of photosynthesis and respiration. Bluegrasses with best growth at high temperatures usually were highest in carbohydrates, had higher photosynthesis and lower respiration. They suggested that bluegrasses with genetically high photosynthetic efficiency be selected for use in areas of high temperature stresses.

No laboratory technique presently available would appear of great practical value for screening large quantities of Kentucky bluegrass breeding materials for adaptation to high temperature stresses. Some evaluation for carbohydrate level, photosynthetic efficiency, etc. might be desirable for strains being evaluated as potential parents in hybridization program. But if we need strains that will perform under high temperature stresses we will need to evaluate under high temperature stresses. A major part of this evaluation might make use of the variable and various stresses that nature provides so generously in the transition zone.

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TURFGRASS BREEDING IN FLORIDA

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Introduction

Breeding work on warm season turfgrasses is being conducted at the Agricultural Research Center at Fort Lauderdale. Emphasis is being placed on St. Augustinegrass, bahiagrass and bermudagrass, although a selection program with zoysiagrass and centipedegrass is also being carried out. Cooperating in this program at Fort Lauderdale are E. O. Burt, turf technologist; R. E. McCoy, pathologist; J. A. Reinert, entomologist; S. R. Johnson, nematologist at Belle Glade; and G. C. Horn, turf technologist at Gainesville.

St. Augustinegrass

The primary objective in St. Augustinegrass improvement is to develop types which are resistant to St. Augustinegrass decline virus (SAD), chinch bugs, brown patch and root knot nematodes. Dwarf, low-growing types with dark-green color and minimal thatching tendencies are also desired.

In cooperation with Texas A&M University, Florida is releasing a new St. Augustinegrass to be named "Floritam." This vigorous variety is resistant to SAD and downy mildew, and does not show zinc and iron deficiencies under Texas conditions.

Laboratory and field techniques have been developed for quick screening of St. Augustinegrasses for Rhizoctonia solani and chinch bug tolerance. Photo-period studies are underway to manipulate flowering for controlled crossing in the greenhouse.

Bahiagrass

The primary objective in bahiagrass improvement is to develop types which are determinate in their flowering. In South Florida, bahiagrass flowers continuously from May to November and requires excessive mowing to produce an aesthetically desirable turf.

Bermudagrass

The primary objective in bermudagrass improvement is to develop types which are resistant to sting and lance nematodes, bermudagrass mite and leaf spot (Helminthosporium spp.). Selections for vigorous types under minimum fertilization were made. Temperature types which will grow year-round in South Florida are desired. A rapid screening technique involving starch accumulation under cool nights is being used to identify promising types.

Primary activities to date involved categorizing approximately 1,300 clones. Upon completion, single or polycross hybridization will be attempted.

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