ANNUAL PROGRESS REPORT

concerning

BREEDING AND DEVELOPMENT OF ZOYSIAGRASS

Submitted by:

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EXECUTIVE SUMMARY
FIRST ANNUAL PROGRESS REPORT
BREEDING AND DEVELOPMENT OF ZOYSIAGRASS

Principle Investigator: Dr. M. C. Engelke

Postdoctoral Research Associate: Dr. Michael P. Kenna

RESEARCH PERIOD OF THIS REPORT: May 1, 1984 to September 1, 1984

Research Accomplished:

A. Germ Plasm Acquisition:

Over 900 introductions and domestic zoysiagrass accessions were assembled, and established in both greenhouse pots and space planted field nurseries.

B. Germ plasm Assessment:

1. The Oriental collection was held in quarantine under isolated greenhouse conditions for 2 years pending indentification of a yet unknown abnormality. Through cooperative efforts we were unable to isolate, innoculate, identify or transmit the causal agent of the abnormality. Symptoms have not been observed on any of the plant material over the past 13 months.

2. All Oriental accessions were planted to field plots for evaluation of agronomic performance with attention directed toward growth habit, growth rate, reproductive characteristics, seasonal color expression, hardiness under stress situations of moisture and temperature extremes, and disease and insect resistance. Special attention is being directed to identifying morphological and anatomical characteristics which will aid in taxonomic identification of the different species.

3. Initial screening for genetic tolerance of zoysiagrasses to high soil temperature and low moisture conditions was initiated in July, 1984 under greenhouse conditions.

4. Replicated turf trails of 39 domestic accessions and commercial varieties was evaluated over the past 3 years. Field notes during 1982 and 1983 indicate that significant variation exists among genotypes.

C. Research Planned 1985:

1. Multiple character analysis and chromosome studies in Zoysia accessions.

2. Response of Zoysia accessions to high soil temperature and low moisture stress.

3. Evaluate variability in agronomic and morphological traits, reproductive characteristics, (i.e. sod, sprig, and/or seed production), and turf quality and adaptability.

4. Cytological assessment of the meiotic stability of interspecific hybrids within the Zoysia genus.

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I. INTRODUCTION

This annual report, as required in the contract, is for the period of May 1, 1984, until September 1, 1984. Ms. Jo Ann Treat, Executive Vice President, Texas Research Foundation, and Mr. Charles W. Smith, Director, Administration and Services for United States Golf Association, signed the contract agreement effective May 1, 1984. A check for the first quarter funding arrived May 29, 1984, and the appropriate accounting arrangements were established through the Texas Research Foundation.

The following, although considered as an annual report for 1984, summarizes the accomplishments of the past 4 months:

II. IMPLEMENTATION

The establishment and expansion of the Germ Plasm Introduction Nursery (GPIN) was well under way prior to the initiation of this project as a result of the Oriental collection trip conducted in 1982 which was jointly funded by the United States Golf Association, the United States Department of Agriculture and the Texas Agricultural Experiment Station. Although this germ plasm collection was held in quarantine for 24 months, considerable information was gained on the anatomical and morphological variability which existed. Other germ plasm resources were obtained through cooperative exchange with scientists throughout the country including Mr. Jack Murray (Beltsville Agricultural Research Center), Dr. Robert Carrow (Kansas State University), Dr. Phil Busey (University of Florida Agricultural Experiment Station - Ft. Lauderdale), Dr. Herb Portz (University of Southern Illinois - Carbondale), and Dr. Vic Gibeault and the late Dr. Vic Youngner (University of California - Riverside).

Field planting of all plant materials in the GPIN was accomplished in June, 1985. All Oriental germ plasm resources are simultaneously maintained in separate pots under controlled conditions of the greenhouse. This duplicity of handling germ plasm is essential to maintain the genetic integrity of the individual plant as well as to insure against possible losses due to environmental extremes, pestilence, or other catastrophic events which could result in the loss of the plant material before proper agronomic assessment has been completed. At the present time, nearly 1,500 individual plants of zoysiagrass and other species are maintained individually in the greenhouse.

Budgeting of this position includes a full time research support position. The project was successful in attracting Dr. Michael P. Kenna (Vita attached) as a Postdoctoral Research Associate to work on Zoysiagrass breeding. Projected research accomplishments are outlined in the 1985 Annual Plan of Work included in this report.

The outline of the presentation will follow the research objectives as originally detailed in the proposal (Figure 1).

Several phases of the breeding program were implemented during this reporting period while others are still planned for the future (Table 1).

A. GERM PLASM ACQUISITION and development of the Germ Plasm Introduction Nursery (GPIN) is a continuing process by which the breadth of diversity is assembled, evaluated, and concentrated throughout the life of the program. Prior to the USDA/USGA/TAES Oriental collection trip in the spring of 1982, the zoysiagrass germ plasm in the United States was concentrated at the USDA Beltsville Agricultural Research Center (BARC), University of California - Riverside (UCR), Kansas State University (KSU), and University of Florida - Ft. Lauderdale. The primary sources for the initial cycle GPIN includes the existing domestic germ plasm resources as listed (Table 2), and a major portion of the Oriental germ plasm collection (Table 3). The current and future work will center on the evaluation and utilization of this GPIN for the development of new zoysia turfgrasses for generations to come.

B. GERM PLASM ASSESSMENT was first initiated at TAES-Dallas in 1981, while more detailed and systematic evaluations were initiated in the summer of 1982 with the introduction of the Oriental collection.

1. PLANT INTRODUCTIONS - Plant Quarantine

Plants for the USDA/USGA/TAES Oriental collection trip were returned by mail directly to the USDA BARC and placed in quarantine to assess the potential introduction of alien diseases or other undesirable organisms. In the fall of 1982, inspectors observed an unknown malady on several of the introductions. These "abnormal" plants were placed in deep isolation, and all "clean" plants were released in February, 1983 to TAES-Dallas. A number of plants released to TAES-Dallas subsequently expressed the same malady and were also isolated (Table 4). Symptoms occurring on the plants include: leaf stripe and leaf curl which may be closely related (Figure 2), leaf mosaic, and a distinct stunting of growth. Identification of the causal agent was cooperatively investigated by pathologists and virologists from Texas A&M University and the USDA BARC. The strongest initial suspect was a viral agent, however, all state-of-the-art isolation, identification, and transmittal procedures have failed to conclusively verify this. The symptoms eventually subsided on greenhouse grown plant materials with no occurrences noted over the past 13 months. Current research by the BARC group would suggest it may be caused by a rather common bacterial complex. However, this remains to be verified.

All Oriental plant materials (Table 3) were planted to space planted nurseries at TAES-Dallas in June, 1984. Material not previously expressing "symptoms" were planted in two replicated (n=325), whereas, any material showing "symptoms" were represented only once (n=418). The entire nursery is monitored frequently specifically looking for any occurrences of disease symptoms. Field observations on the agronomic performance, flowering habit, rate of growth, etc. are made monthly or more frequently if substantial differences are noted.

2. TAXONOMIC CLASSIFICATION

The Oriental and domestic plant material received at TAES-Dallas were tenatively classified by genus and species, F1 hybrid, or as unknown (Table 5). Detailed notes on morphological and anatomical characters were completed on a subsample of 61 plants which expressed a wide range of phenotypic variability (Table 6). This taxonomic data set will be used to develop methods for more efficient classification of the remaining accessions and those acquired in the future.

3. FIELD ASSESSMENT

As previously mentioned, the Oriental zoysiagrasses which did not express symptoms of abnormality (325) were planted in replicated field nurseries June, 1984. Approximately 400 of the plants which had expressed "symptoms" were planted in non-replicated field nurseries. Field notes on general growth habit, rate of spread, summer and winter color, density, texture, heat and drought tolerances, winter survival, spring greenup, summer and fall dormancy, disease incidence, insect infestation, general competitive ability, and production potential will be evaluated.

4. SOIL TEMPERATURE AND MOISTURE STRESS TOLERANCE

Screening germ plasm for high soil temperature and low soil moisture stress was initiated July, 1984. A specialized greenhouse bench containing graded sand was completed and seeded to a Korean zoysiagrass population. The initial temperature gradient of 43 to 49°C is maintained across the 2.4m span of the 1.2m wide bench. Identification of the optimum and maximum soil temperature range should be possible, along with the identification of individual plants which are able to survive at higher soil temperatures.

C. SPECIES HYBRIDIZATION will begin as soon as unique parents are identified. Single crosses between selected plants are planned to ascertain the genetics of simply inherited characters (i.e., one or two genes controlling

the trait), and in examining the genetic linkage between traits. The progeny from polycrosses among several plants with no control of cross-pollination will be used to study quantitatively inherited traits (i.e., several genes controlling the trait). In addition to using the resulting progenies in genetic studies, desirable progenies are: a) included in subsequent GPIN's to increase gene frequency of favorable traits in the base population, b) recycled through all phases of GERM PLASM ASSESSMENT, and c) may be included in replicated turf trials.

D. REPLICATED TURF TRIALS (RTT) for the Oriental zoysias and progeny originating from them will be established once promising experimental varieties (EXPERVARS) are identified. An RTT of 39 domestic accessions was planted at TAES-Dallas July, 1981, to ascertain the performance of existing cultivars and accessions of zoysiagrass in the north Texas region (Table 7). Field notes were taken during 1982/83 and results indicate that there exists a wide range of plant response for important turf characteristics.

III. RESEARCH PLANNED

Projected research accomplishments outlined for 1985 follows:

A. Multiple Character Analysis and Chromosome Studies in Zoysia Accessions

Objective: Identify morphological and cytological similarities and differences among selected zoysiagrass accessions. This information will be useful in classifying the remaining germ plasm collection.

Procedures: During the summer and fall of 1984 information on several morphological and cytological traits will be completed for 60 zoysiagrass accessions which are representative of the entire germ plasm collection. Selected plants will be grown under uniform environmental conditions and some of the characters will include:

External morphology

- -number of florets/inflorescence
- -flowering habit
- -leaf vernation
- -length of leaf sheath
- -presence/absence of rhizomes
- -ligule characteristics
- -leaf angle
- -primary axis length of the inflorescence
- -size of florets
- -number of primary nerves
- -length of internodes
- -length, width, and hairiness of flag leaf
- -foliage color
- -stolon diameter

Internal Anatomy

-number and type of vascular bundles present
-presence of parechyma in the midrib
-abaxial epidermis characteristics (basal hairs, papillae, and thickness
-pollen stainability - fertility

Chromosomes

-root tip squashed - mitosis
-pollen mother cell - meiosis and microsporogenesis

Work was completed on 61 zoysiagrass accessions for 26 morphological and anatomical characters (Table 6). The completion of data collection for external and internal characters will be completed by October, 1984. Determination of chromosome numbers may require a slightly longer period of time. Information on stage of flowering and meiosis will be obtained using methods similar to those described by Forbes (4).

Multivariate analysis of variance (MANOVA) will be applied to the morphological and cytological data to determine individual and group differences on the basis of all characters studies. Discriminant function analysis (DF) will be used to determine which characters contributed the most towards individual or group separation (8, 10, 11).

B. Response of Zoysia Accessions to High Soil Temperatures

Objective: Screen several zoysiagrass accessions for tolerance to high soil temperatures and determine what morphological and agronomic characters are correlated with tolerance to high soil temperatures.

Procedures: During the fall and winter of 1984, zoysiagrass accessions which are being characterized morphologically, will be screened in a specialized greenhouse bench which permits artificial manipulation of soil temperatures. Vegetative propagules will be established October, 1984, for at least 2 replications, and planted into the soil bench in late November, 1984. Transplanted material will be allowed to establish for 3 to 4 weeks. Soil temperatures will then be gradually increased until a maximum temperature of 140°F is attained. Plant response to temperature stress will be recorded weekly, noting differences in leaf morphology (and anatomy), growth habit, and plant vigor. At the termination of the experiment, shoot and root weights will be determined for each entry.

Field and greenhouse information on the zoysiagrass accessions screened in this study will help determine if any morphological or agronomic characters are correlated with tolerance to high soil temperatures.

C. Evaluate variability of Morphological and Agronomic Traits in *Zoysia* Accessions

Objective: Characterize the phenotypic variability among zoysiagrass accessions under field conditions for characters of agronomic importance to the turfgrass industry.

Procedures: Field notes concerning the agronomic value of plant accessions will be taken for the 1984-85 and 1985-86 growing seasons. Two replications will be required to test for significant differences among the agronomic characters which will include:

- -growth habit
- -rate of spread
- -height
- -color
- -density
- -texture
- -heat and drought tolerance
- -winter survival
- -spring vigor and greenup
- -summer and fall dormancy
- -disease incidence
- -competitive ability
- -production potential
- -anthesis date
- -open and self-pollinated seed set
- -pollen stainability

All agronomic information will be coded to facilitate the use of a field data recorder. Field notes for more than one date will be required for most of the characters. Those characters which are subjective should be recorded by more than one researcher.

Several researchers working with bermudagrass (3, 5, 7, 12) have cut culms at ground level and placed these immediately into vials containing water. This method has been used successfully to produce self and cross-pollinated seed. However, if zoysiagrass inflorescences cut prior to anthesis do not mature, then 3 to 5 inflorescences will be bagged on the field plots. Open and self-pollinated seed set will be determined on at least 3 inflorescences for each accession.

The mean and range for each character in the study will be determined. A data base of all character information will be built to facilitate examination of the accessions from a computer terminal. MANOVA and DF analyses will be conducted to determine plant groupings and those characters which are important in determining groups (8, 10, 11).

D. Fertility and Cytological Characteristics of some Interspecific *Zoysia* hybrids

Objective: Determine the biological compatibility of *Zoysia tenufolia* or (*Z. matrella* or *Z. japonica*) with *Z. macrostaycha* and *Z. sinica*.

Procedures: During the spring of 1985, information collected in the first three studies will be used to select 8 to 10 superior genotypes for hybridization. Interspecific hybrids among *Z. macrostaycha* and *Z. sinica* and with the other *Zoysia* species will be included since they have not been studied (Figure 3). Determination of cytological and fertility information on the parents and their F_1 hybrid offspring will follow those methods described by Forbes (4), Burson (2), and Hanna and Burton (5). This information is necessary to ascertain because the biological compatibility between zoysiagrass species will determine if agronomic characters can be transferred from *Z. macrostaycha* and *Z. sinica* to the turf-type zoysiagrass species.

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VITA

Michael Patrick Kenna

Post-doctorial Research Associate

Major Field: Crop Science, Plant Breeding and Genetics, Turfgrass Science and Culture, and Statistics.

Biographical:

Personal Data: Born in El Cajon, California, April 11, 1956, son of Mr. and Mrs. John M. Kenna, Jr. Married to Susan Winsor Kenna on July 14, 1979.

Education: Graduated from Granite Hills High School, El Cajon, California, in June, 1974; attended the University of California at San Diego from September, 1974, to June, 1976; received Bachelor of Science Degree in Ornamental Horticulture and Minor Degree in Botany from California State Polytechnic University, Pomona, in June, 1979; received Master of Science Degree in Agronomy from Oklahoma State University, Stillwater, in May, 1981; and completed requirements for Doctor of Philosophy Degree in Crop Science and Minor Degree in Statistics, from Oklahoma State University, Stillwater, in July, 1984.

Professional Experience: Golf course crew, Singing Hills Golf and Country Club, El Cajon, California, 1972 to 1976; Student Teaching Assistant, California State Polytechnic University, 1976 to 1979; Graduate Teaching Assistant, Oklahoma State University, 1979 to 1980; Graduate Research Assistant, Oklahoma State University, 1980 to 1984; Post-doctorial Research Associate, Texas A&M Research and Extension Center, Dallas, 1984 to present.

Professional Organizations: Gamma Sigma Delta (Agricultural Honor Society); Sigma Xi (Scientific Research Society); Crop Science Society of America; and American Society of Agronomy.

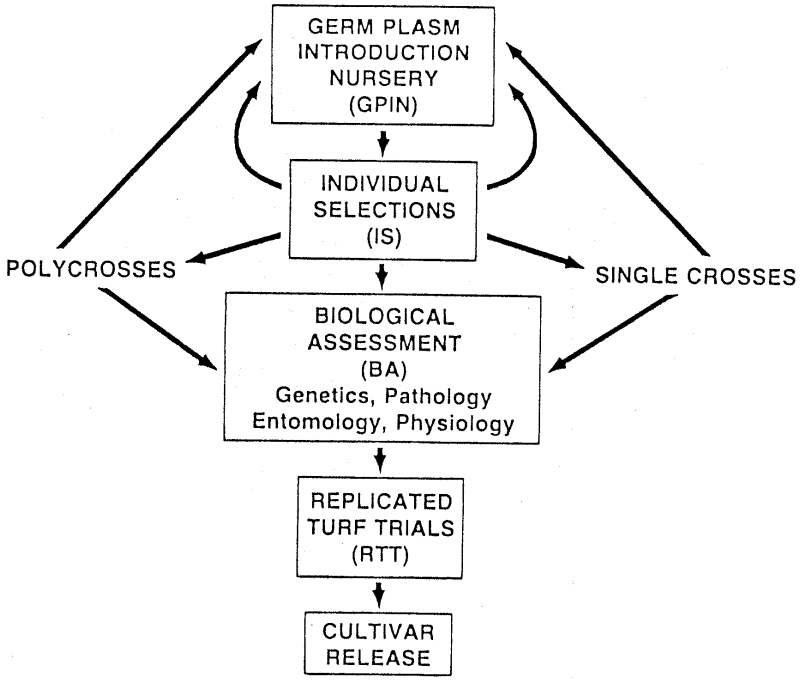

FIGURE 1. FLOW DIAGRAM DEPICTING MAJOR ELEMENTS AND EVENTS IN DEVELOPING ZOYSIAGRASSES INTO ACCEPTABLE TURFGRASS CULTIVARS

Figure 2. Leaf stripe (LS) and leaf curl (LC) symptoms observed on Oriental zoysiagrass accessions during Spring,1983, at TAES-Dallas.

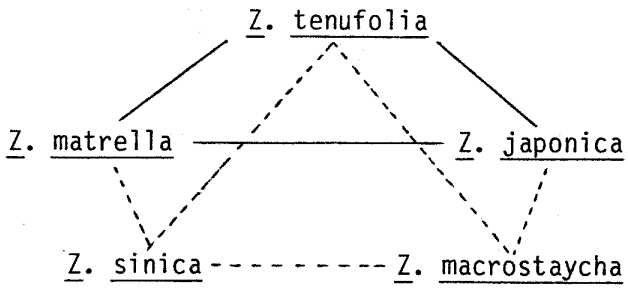

Figure 3. Interspecific crosses and biological compatibility among Zoysia species. Solid line indicates the cross-compatibility of Zoysia species worked out by Forbes (4). The broken line indicates those interspecific hybrids not studied.

TABLE 1. Time Table for developing improved Zoysiagrasses.

PRIMARY OBJECTIVE		YEAR INITIATED
Germ Plasm Acquisition	1+	(1981)
Plant Introduction		(1982)
Cooperators		(1984)
Germ Plasm Assessment	1 - 10+	(1984)
Heat and drought tolerance		(1984)
Growth habit, texture, quality		(1984)
Flowering habit		(1983 - 1985)
Disease resistance		(1984 - 1986)
Salinity tolerance		(1985)
Field Assessment		(1984)
Species Hybridization	2 - 10+	(1985 - 1986)
Replicated Field Trials		(1981)
Local	3+	(1981)
State & Regional	3+	(1986)
Cultivar Release (Initial)	6 - 10+	(1990)

TABLE 2. Domestic zoysiagrass inventory summary, TAES-Dallas, August, 1984.

Origin	Date	Number of Plants		
		Received at TEAS	Survived at TEAS+	Planted in Field
Beltsville	3/81	18	18	18
	6/81	6	6	6
Florida	6/83	129	129	129
California	1/83	11	11	11
Kansas	12/80	7	7	7
Plant Introduction Center	1/81	6	6	6
Total		177	177	177

+ Under greenhouse conditions.

TABLE 3. Oriental zoysiagrass inventory summary, TEAS-Dallas, August, 1984.

Origin	Date Received	Number of Plants					
		Received at TAES	Survived at TAES*	"Syndrome" Expressed**	Recorded Missing	Planted in Field	Taxonomy Data
Phillipines (P)	2/83	61	60	0	0	60	11
Tiwan (T)	2/83	21	20	0	0	20	7
Japan-Fukuako (JS)	6/82	97	95	93	2	93	8
Japan (J)	6/82	397	391	177	0	391	20
Korea (K)	6/82	181	179	148	0	179	15
Total		757	745	418	2	743	61

* Under greenhouse conditions
** At BARC and TAES-Dallas