Warm-Season Grasses

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Warm-season grasses consist of many genera and species. In the United States, their adaptation ranges from the humid, semitropical conditions of the Southeast, to the temperate central states, and the arid Southwest. Many originated in tropical and semitropical regions of the world and were introduced into the United States. They are used as forage, turf, and ornamentals and differ in method of pollination, mode of reproduction, chromosome number, ploidy level, time of flowering, and numerous other characteristics.

I. PARENTAL MATERIAL

All grasses are members of the Gramineae family and the warm-season grasses are in the subfamilies Panicoideae, Eragrostioideae, Bambusoideae, Arundinoideae, and Oryzoideae. Most of the grasses discussed in this chapter are members of the Panicoideae and Eragrostioideae subfamilies. They are separated into tribes and numerous genera, many of which are of economic importance. The number of species within a genus may be quite limited or almost unlimited. For example, the genus Erechthochloa has one species whereas Paspalum contains from 300 to 400 species.

The success of interspecific and intergeneric hybridization varies. Generally, it is easier to produce hybrids in cross-pollinated species than in self-pollinated species. Numerous interspecific hybrids have been produced among cross-pollinated Cynodon species (Burton, 1965; Harlan et al., 1969). Although most Paspalum species are self-pollinated and crossability in this genus is low, a number of interspecific Paspalum hybrids have been produced (Bennett and Bashaw, 1966; Burson and Bennett, 1976; Burson et al., 1973).
Crossability among *Sorghum* species is high if male sterility or perfected emasculation techniques are used. Interspecific hybrids have been produced between *S. almum* Parodi and sudangrass [*S. sudanense* (Piper) Stapf] (Pritchard, 1965a); *S. almum* and johnsongrass [*Sorghum halepense* (L.) Pers.] (Pritchard, 1965b); and johnsongrass and sudangrass (Casady and Anderson, 1952).

Since apomixis is prevalent in warm-season grasses and can affect the success of hybridization, the mode of reproduction of the species to be crossed should be known prior to making the cross (see Chap. 3). Interspecific hybrids have been produced between sexual buffelgrass, *Cenchrus ciliaris* L., and apomictic birdwoodgrass, *C. setigerus* Vahl (Read and Bashaw, 1969). An intergeneric hybrid has also been made between male-sterile pearl millet, *Pennisetum americanum* (L.) Leek, and apomictic buffelgrass (Read and Bashaw, 1974). Napiergrass, *P. purpureum* Schumach, and pearl millet cross readily (Burton, 1944).

A number of interspecific and intergeneric hybrids have been made among the Old World bluestems, Bothriochloininae (Harlan et al., 1962) with a meticulous emasculation technique developed by Richardson (1958). Hybrids have also been produced between big bluestem, *Andropogon gerardii* Vitm., and sand bluestem, *A. hallii* Hack. (Peters and Newell, 1961).

Interspecific hybrids have been made in *Digitaria* (van Heemert and Schank, 1968), *Eragrostis* (Busey, 1976), *Setaria* (Hacker, 1968), and *Zoysia* (Forbes, 1952).

Maximum diversity of a species or genus exists at its center of origin. Centers of origin of many warm-season grasses are outlined by Vinall and Hein (1937). Most germplasm has been obtained from these areas by plant collection trips. After the material is collected, it is assigned a plant introduction number and is maintained at a USDA Regional Plant Introduction Station. Most accessions of warm-season grasses are kept at the Regional Introduction Station, Experiment, GA 30212. Seed is available upon request.

Germplasm is also available through exchange with other scientists or organizations. Plant breeders working with a particular genus or species generally maintain a large collection of plants. The Soil Conservation Service, USDA, maintains collections of native species at their regional plant materials centers. This material is also available upon request.

**II. PLANT CULTURE**

Plant culture for hybridization of warm-season grasses is almost exclusively limited to the greenhouse because of the instability of field environmental conditions and the floral characteristics.

**A. Field**

Grasses hybridized under field conditions should be grown on soils to which they are adapted. No one soil type is appropriate for all warm-season grasses. Species within the same genus vary in their soil requirements. For example, dallisgrass, *Paspalum dilatatum* Poir., is adapted to heavy clay soils; whereas bahiagrass, *P. notatum* Flugge, is well-adapted to sandy soils.
Most warm-season grasses respond to fertilization more readily with nitrogen than with phosphorus or potassium. A fertilizer ratio of 4–1–2 is generally used in the South (Wilkinson and Langdale, 1974). The rate applied depends on the inherent fertility of the soil and the response of the particular grass. High rates are usually applied to grasses to be crossed.

Moisture availability is very important. Not only is it critical for plant growth and development, but it is essential for the formation of viable gametes and subsequent seed formation. Provisions should be made for additional sources of water if supplemental moisture is needed during the growing season.

Large nurseries are not required for crossing warm-season grasses. In crossing blocks utilizing cross-pollinated species, plants are normally on 1 m centers.

Other cultural conditions promoting plant growth for hybridization under field conditions are very similar to those found in the greenhouse.

B. Growth Chamber and Greenhouse

Most grasses hybridized in the greenhouse are grown in pots. A variety of planting media are used. Many media consist of differing proportions of peat moss and perlite or vermiculite. Sand or soil may be included in one of the above mixtures or may even be used alone if the plant is adapted to the particular soil. The soil is usually sterilized before potting to eliminate seed contamination. Occasionally, grasses are planted in greenhouses with soil floors.

Plants are kept moist and fertilized regularly with a low-concentration commercial fertilizer. It is generally accepted that hot, dry conditions or low soil fertility are not conducive to good pollen formation, pollination, or seed set. Soil moisture has little effect upon flowering rate; however, grasses grown under high-moisture regimes have larger inflorescences than those grown under dry conditions (Bennett, 1959b).

Most warm-season grasses flower during the summer months. They have indeterminate flowering, with new inflorescences initiated throughout the summer.

Warm-season grasses differ in their response to day length. Many are day-neutral, and others are either long-day or short-day. Within a genus, species commonly differ in their flowering response to photoperiod (Bogdan, 1977; Knight and Bennett, 1953; Evers et al., 1969). The latitude of origin of a species can be extremely important. Different strains of buffalograss, *Buchloe dactyloides* (Nutt.) Engelm., collected from Texas to North Dakota varied in their photoperiodic requirements (Olmsted, 1943).

To cross species or types that differ in their time of flowering, it is necessary to adjust the day length. Plants are usually grown in a greenhouse and day length is adjusted by covering plants or by providing supplemental lighting.

A high light level is necessary for floral initiation and the development of normal inflorescences. Plants grown under reduced light level produce elongated stems and inferior inflorescences.
Temperature can override photoperiodic effects. Night temperatures below 13 C inhibited flowering in common dallisgrass, bahiagrass, johnsongrass, carpetgrass (Axonopus affinis Chase), and bermudagrass (Cynodon dactylon (L.) Pers.) (Knight and Bennett, 1953). A night temperature of 18 to 21 C and a 14-hour photoperiod resulted in maximum flowering in common dallisgrass. However, night temperatures between 7 to 13 C inhibited seed head formation at the same photoperiod (Knight, 1955). In three Zoysia species under their correct day length, flowering was inhibited at a minimum temperature of 24 C, but was initiated at 29 C (Forbes, 1952).

Temperature also can be used to synchronize time of flowering between different species. Time of initial floret opening differs among common dallisgrass, vaseygrass (Passpalum utvillei Steud.), and P. malacophyllum Trin. (Table 1). They initiated flowering within an hour of one another when dallisgrass was subjected to a night temperature of 18 C and the other species were subjected to 13 to 16 C (Bennett, 1959c).

Warm temperatures are conducive to rapid floret opening. This principle can be used to open florets on a cool morning. Often floret opening is enhanced by carefully clasping hands around the inflorescence. Gently blowing into the clasped hands provides heat. Artificial heating of unopened florets accomplishes the same in rhodesgrass, Chloris gayana Kunth (Bogdan, 1959). Jones and Brown (1951) found that low temperatures delayed or inhibited blooming in several grasses. In Pensacola bahiagrass, maximum flowering occurred between 18 and 21 C and decreased as temperatures fluctuated in either direction (Hodgson, 1949).

Temperature also influences the time of anther dehiscence. Bennett (1959c) reported that in dallisgrass, vaseygrass, and P. malacophyllum anther extrusion and dehiscence occurred within 5 min after the florets were fully open at 21 to 24 C. At 29 C and above, the anthers dehisced before the florets open. Reducing the temperature below 21 C for dallisgrass and below 18 C for vaseygrass and P. malacophyllum increased the time the anthers remained pendent without dehiscing. Dehiscence was delayed for 4 hours at 11 C, for 2 hours at 13 C, and for 30 min at 16 C.

Generally, the optimum temperature for hybridization of warm-season grasses ranges from 26 to 32 C (day) and 17 to 21 C (night). A temperature of 21 C or lower is essential for proper emasculation when the anthers are removed from the florets before dehiscence.

Relative humidity is extremely important in hybridization because it delays anther dehiscence. Hodgson (1949) reported that high relative humidity prevented or delayed dehiscence in Pensacola bahiagrass. Heavy dew during the night delayed pollen shedding in weeping lovegrass, Eragrostis curvula (Schrad.) Nees. (Jones and Brown, 1951). Because high humidity delays anther dehiscence, this phenomenon has been used in different crossing techniques discussed in Section IV.

III. FLORAL CHARACTERISTICS

Most warm-season grasses have perfect flowers; however, there are exceptions. Buffalo grass and pampasgrass, Cortaderia selloana (Schult.) Aschers. and Graebn., are dioecious species, and Eastern gamagrass,
Table 1—Duration of pollen dispersal and time of blooming in warm-season grasses.

<table>
<thead>
<tr>
<th>Scientific name</th>
<th>Common name</th>
<th>Pollen dispersal period per inflorescence</th>
<th>Time of blooming (Daylight Standard Time)</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Andropogon gerardii</em> Vittm.</td>
<td>Big Bluestem</td>
<td>7 days</td>
<td>0400-0700</td>
<td>Jones and Newell, 1946</td>
</tr>
<tr>
<td><em>A. hallii</em> Hack.</td>
<td>Sand Bluestem</td>
<td>8 hours</td>
<td>0400-0900</td>
<td>Jones and Newell, 1946</td>
</tr>
<tr>
<td><em>Bouteloua curtipendula</em> (Michx.) Torr.</td>
<td>Sideoats Grama</td>
<td>8 days</td>
<td>0400-0900</td>
<td>Jones and Newell, 1946</td>
</tr>
<tr>
<td><em>B. gracilis</em> (H.B.K.) Lag. ex Steud.</td>
<td>Blue Grama</td>
<td>6 days</td>
<td>0300-0900</td>
<td>Jones and Newell, 1946</td>
</tr>
<tr>
<td><em>Buchloe dactyloides</em> (Nutt.) Engelm.</td>
<td>Buffalograss</td>
<td>Intermediate</td>
<td>0600-1100</td>
<td>Jones and Newell, 1946</td>
</tr>
<tr>
<td><em>Chloris gayana</em> Kunth.</td>
<td>Rhodeegrass</td>
<td>--</td>
<td>1145-1230</td>
<td>Davidson, 1941</td>
</tr>
<tr>
<td><em>Chloris gayana</em> Kunth.</td>
<td>Rhodeegrass</td>
<td>8 days</td>
<td>1245-1400</td>
<td>Bogdan, 1959</td>
</tr>
<tr>
<td><em>Eragrostis curvula</em> (Schrad.) Nees.</td>
<td>Weeping Lovegrass</td>
<td>10.5 days</td>
<td>2400-0800</td>
<td>Jones and Brown, 1951</td>
</tr>
<tr>
<td><em>E. trichoides</em> (Nutt.) Wood</td>
<td>Sand Lovegrass</td>
<td>7 days</td>
<td>0700-1100</td>
<td>Jones and Newell, 1946</td>
</tr>
<tr>
<td><em>Panicum maximum</em> Jacq.</td>
<td>Guineaagrass</td>
<td>--</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cultivars</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fine leaf</td>
<td></td>
<td>--</td>
<td>1600-2330</td>
<td>Warmke, 1951</td>
</tr>
<tr>
<td>Broad leaf</td>
<td></td>
<td>--</td>
<td>2000-2400</td>
<td>Warmke, 1951</td>
</tr>
<tr>
<td>Boringuen</td>
<td></td>
<td>--</td>
<td>2100-0100</td>
<td>Warmke, 1951</td>
</tr>
<tr>
<td>Regular guinea</td>
<td></td>
<td>--</td>
<td>2300-0400</td>
<td>Warmke, 1951</td>
</tr>
<tr>
<td>Gramalote</td>
<td></td>
<td>--</td>
<td>1330-1600</td>
<td>Warmke, 1951</td>
</tr>
<tr>
<td>Colonial</td>
<td></td>
<td>--</td>
<td>2130-2330</td>
<td>Javier, 1970</td>
</tr>
<tr>
<td>King Ranch #2</td>
<td></td>
<td>--</td>
<td>2130-2330</td>
<td>Javier, 1970</td>
</tr>
<tr>
<td>Common</td>
<td></td>
<td>--</td>
<td>2130-2330</td>
<td>Javier, 1970</td>
</tr>
<tr>
<td>Sabi</td>
<td></td>
<td>--</td>
<td>1730-1900</td>
<td>Javier, 1970</td>
</tr>
<tr>
<td>Green Panic</td>
<td></td>
<td>--</td>
<td>2130-2330</td>
<td>Javier, 1970</td>
</tr>
<tr>
<td>Sabi</td>
<td></td>
<td>--</td>
<td>1930-2130</td>
<td>Javier, 1970</td>
</tr>
<tr>
<td><em>P. purpurascens</em> Radii</td>
<td>Paragrass</td>
<td>--</td>
<td>0700-1200</td>
<td>Warmke, 1951</td>
</tr>
<tr>
<td><em>P. virgatum</em> L.</td>
<td>Switchgrass</td>
<td>12 days</td>
<td>1000-1400</td>
<td>Jones and Newell, 1946</td>
</tr>
<tr>
<td><em>Paspalum dilatatum</em> Poir.</td>
<td>Dallisgrass</td>
<td>9 days</td>
<td>0400-0830</td>
<td>Jones and Brown, 1951</td>
</tr>
<tr>
<td><em>P. malacophyllum</em> Trin.</td>
<td></td>
<td>11 days</td>
<td>0200-0830</td>
<td>Bennett, 1959b</td>
</tr>
<tr>
<td><em>P. notatum</em> Flugge</td>
<td>Bahiagrass</td>
<td>5 days</td>
<td>2000-0900</td>
<td>Hodgson, 1949</td>
</tr>
<tr>
<td><em>P. viridii</em> Steud.</td>
<td>Vaseygrass</td>
<td>13 days</td>
<td>0300-0830</td>
<td>Bennett, 1959b</td>
</tr>
<tr>
<td><em>Phragmites communis</em> Trin.</td>
<td>Common Reed</td>
<td>10 days</td>
<td>0600-1000</td>
<td>Jones and Newell, 1946</td>
</tr>
<tr>
<td><em>Sorghastrum nutans</em> (L.) Nash</td>
<td>Indiangrass</td>
<td>8 days</td>
<td>0600-1000</td>
<td>Jones and Newell, 1946</td>
</tr>
<tr>
<td><em>Sorghum halepense</em> (L.) Pers.</td>
<td>Johnsongrass</td>
<td>9.5 days</td>
<td>0900-1500</td>
<td>Jones and Brown, 1951</td>
</tr>
<tr>
<td><em>S. vulgare</em> Pers.</td>
<td>Sorghum</td>
<td>10 days</td>
<td>0400-1000</td>
<td>Jones and Newell, 1946</td>
</tr>
<tr>
<td><em>S. sudanense</em> (Piper) Stapf.</td>
<td>Sudangrass</td>
<td>10 days</td>
<td>0600-2400</td>
<td>Jones and Newell, 1946</td>
</tr>
<tr>
<td><em>Spartina pectinata</em> Link</td>
<td>Prairie Cordgrass</td>
<td>10 days</td>
<td>0400-1000</td>
<td>Jones and Newell, 1946</td>
</tr>
</tbody>
</table>

**Tripsacum dactyloides** (L.) L., is a monoeocious species.

A majority of the grasses are wind-pollinated, but insects are sometimes instrumental in pollination (Bogdan, 1977). Most warm-season grasses are naturally cross-pollinated; however, many are capable of producing some self-pollinated seed (Hanson and Carnahan, 1956). In addition to dioecious and monoeocious plants, cross-pollination results from self-incompatibility, male sterility, and protogyny.

To produce hybrids, the plant breeder must know the flowering behavior of a species (Table 1). Flowering is a broad term that includes floret opening, anther and stigma extrusion, and anther dehiscence. Because locations differ within time zones, flowering times are general indicators and are not absolute.
Although flowering of most grasses starts slowly, increases to a peak, and gradually declines, some grasses have two flowering peaks. Peak pollen shedding in weeping lovegrass occurs shortly after 0100 hours and also near dawn; in Pensacola bahiagrass, a major peak occurs between 2130 and 2330 hours and a minor peak at 0730 hours (Jones and Brown, 1951; Hodgson, 1949).

The time of flowering is not uniform for a species. The variability among guineagrass, *Panicum maximum* Jacq., cultivars readily illustrates this fact (Table 1). Environmental factors also influence the time of flowering. Researchers have observed that flowering may be delayed or may not occur on cool, cloudy, humid days.

The length of time from floret opening or anther extrusion to pollen shedding varies between species and also is influenced by the environment. The whole process may take 6 to 12 min (Bogdan, 1959), 30 min (Jones and Newell, 1946), or 1 hour (Hodgson, 1949). Grasses that flower late in the day, when the humidity is low and the temperature is high, generally shed pollen quickly (Table 1).

Stigmas of warm-season grasses usually are considered receptive when they emerge from the floret. In dallisgrass, stigmas were receptive when the anthers dehisced, but they appeared dry and dead 2 hours after anthesis. In a humid environment some stigmas of dallisgrass remained receptive for 1 day after anthesis (Bennett, 1944). Stigmas of *Bothriochloa* and * Dichanthium* species remained receptive 18 to 24 hours after emasculaton (Richardson, 1958) compared to 96 hours in bahiagrass (Burton, 1948c). In buffalograss, the stigmas remained receptive 20 days after anthesis, with maximum receptivity 5 days after stigma emergence and continued high receptivity until the 13th day (Jones and Newell, 1948).

Floral characteristics are sometimes conducive to natural hybridization. In dioecious and monoecious species, hybridization is necessary for seed production. In protogynous species, the stigmas are exerted from the florets before the anthers, which is also conducive to hybridization. Buffalograss and birdwoodgrass are protogynous and their stigmas are exerted 2 to 3 days before anthesis (Fisher et al., 1954). Kikuyugrass, *Pennisetum clandestinum* Hochst. ex Chiov., is also protogynous and the presence of withered stigmas before the anthers are exerted indicate that cross-pollination has already occurred (Edwards, 1936). Species of *Zoysia* are protogynous (Forbes, 1952).

Under natural conditions, grass pollen retains its viability for only a short period after anthesis. The thin walls of pollen grains desiccate very rapidly under conditions of high temperature and low humidity (Hodgson, 1949). Dallisgrass pollen ceased to germinate in vitro 30 min after dehiscence (Bennett, 1959a), and sudangrass pollen viability was negligible 5 hours after shedding (Hogg and Ahlgren, 1943).

Although grass pollen does not generally lend itself well to storage, longevity of *Eragrostis superba* Peyr. pollen has been extended from less than 2 hours at room temperature to 4 days when stored at 2 C (Busey, 1976). Carmichael (1970) stored spikelets of *Digitaria pentzii* Stent with half-exerted anthers in a warm, humid environment. The pollen from spikelets stored for 8 hours germinated better in vitro than pollen from spikelets stored for 2, 4, and 6 hours. The addition of 50 ppm gibberellic acid to the medium increased germination.
Buffalograss pollen stored at 45 C and 30% relative humidity or at 25 C and 40% relative humidity remained viable for 24 hours, but was non-viable after 48 hours (Jones and Newell, 1946). When stored either as free pollen or in the spikelets at 4 C and 90% relative humidity, it remained viable from 6 to 8 days (Jones and Newell, 1948).

The time from pollination to fertilization varies for most warm-season grasses. The reported range is a minimum of 30 min to a maximum of 18 hours (Dewald and Harlan, 1961; Streetman and Wright, 1960).

IV. ARTIFICIAL HYBRIDIZATION AND SELF-POLLINATION

A. Equipment

In predominately self-pollinated species, it is necessary to emasculate the florets of the female parent to produce hybrids. The basic items include forceps, scissors, pollen collectors, pollination bags or plastic bags, stapler, paper clips, pencil, tags, artist brushes, string, stakes, and magnifying glasses.

B. Preparation of the Female

A successful, but tedious method used for self-fertile species involves emasculating the florets before anthesis (Richardson, 1958). Inflorescences are selected when a few anthers on the terminal rachis have dehisced. These florets are removed with small cuticle scissors, leaving only unopened florets. The hands and scissors are then rinsed in dilute alcohol or water bath to eliminate possible pollen contamination. Spikelet arrangement in many grasses are paired, and the spikelets differ in age. With the aid of a magnifying glass, the pedicellate spikelets are removed with scissors or forceps, leaving only the sessile spikelets.

Emasculation is initiated with the basal florets on the lowermost branch and gradually proceeds to the top floret. The emasculation procedure involves grasping the awn of the floret with the thumbnail and index finger of one hand. With forceps in the other hand, the tip of the lemma is grasped, and the lemma and palea are gently pulled apart. After the stigmas and anthers are exposed, the lemma is released and the bottom of the palea is pushed up. This dislodges the anthers, which can then be removed with forceps. The anthers, usually three, should be counted as they are removed to avoid leaving any in the floret. This process is repeated until the entire inflorescence is emasculated. The tools should be rinsed after each floret is emasculated.

The emasculated inflorescence is covered with a bag and the pertinent information (i.e., genus, species, date) is recorded on the bag. The sides of the bag are folded just below the inflorescence and secured to the stem with a paper clip. The bag is fastened to an overhead support so that the excess weight of the bag does not break the culm.

Other hand emasculation techniques are less laborious, but less effective. Burton (1948a) has used a humidifier to aid in the emasculation of Paspalum species. The night before flowering, plants are placed in a green-
house where a continuous fog is maintained. All exserted anthers are removed with forceps before dehiscence, and the plants remain in the fog until flowering ceases. The plants are placed in a separate dry room to dry the stigmas for future pollination. After the stigmas are dry, the emasculated inflorescences are covered with an isolation bag until they are pollinated. This process is repeated daily until all the florets have flowered or the unemasculated florets are clipped from the inflorescence with scissors. In a modification of this method, the humidifier is placed inside a walk-in chamber in the greenhouse, and the plants are placed inside the chamber. This permits the plants to be in a heavier fog during emasculation (Read and Bashaw, 1969; 1974). Disadvantages of this technique are expense of a humidifier, discomfort of working in an environment at 100% relative humidity, and fogging of magnifying glasses used to remove the anthers. Species which shatter their seed readily lose their spikelets within 2 to 3 days after emasculation under these conditions.

Another technique that involves the principle of humidity is to spray water in the greenhouse prior to floret opening to increase the humidity at the time of anther exertion (Bennett and Bashaw, 1966). This method provides less time between anther extrusion and dehiscence than does the use of a humidifier, and the anthers must be removed quickly with forceps and the aid of a magnifier. This technique works best in locations where the relative humidity is 90% or higher early in the morning and the temperature is 21 C or lower.

Polyethylene bags also have been used to increase humidity as a means of preventing dehiscence. Hanna et al. (1973) placed the inflorescence of the female guineagrass parent into a plastic bag the evening before crosses were made. The next morning at about 0700 hours, high humidity inside the bag permitted removal of nondehiscent anthers. Stigmas were allowed to dry before pollination. The procedure was repeated for 2 to 3 days, after which unopened florets were removed.

Hot-water emasculation has been successful with forage sorghums, but has been less successful with other warm-season grasses. Induced male sterility by use of gametocides was attempted in weeping lovegrass, but the crossing frequency was not increased (Busey, 1976).

Selfed seed of self-compatible genotypes can be obtained by enclosing an inflorescence in a pollinating bag before flowering has begun. Seed set is usually higher if the bagged inflorescences are agitated daily after anthesis. Spatial isolation is another method for obtaining selfed seed. This usually results in a seed set higher than that from bagging because environmental conditions inside the bag can be adverse. However, complete spatial isolation from related species can be a problem if adequate plot land is unavailable.

C. Pollination

Pollen is collected when the anthers dehisce. This usually occurs early in the morning, near or shortly after sunrise (Table 1). However, dehiscence varies under different environmental conditions.
Cresses made in the greenhouse have the advantage of keeping the male parent in close proximity to the female and thus facilitating pollination. This can be done by growing the male parent in pots inside the greenhouse. Another method is to cut stems bearing inflorescences from plants in the field the evening before pollination and place them into a container of water in the greenhouse. Only those inflorescences which have begun to flower should be collected. The florets flower the next morning and continue to flower for several mornings. A large number of inflorescences can thus be maintained in a small area. The greenhouse should be closed with fans turned off because excessive air movement disperses the pollen at dehiscence. Pollination should be initiated immediately after collection because most grass pollen is short-lived.

Several pollen collection methods are used. With one method, a piece of slick-surfaces paper or a petri dish is held about 2 to 3 cm below the inflorescence, and the inflorescence is gently tapped. The pollen falls onto the paper or into the dish. A second method uses a small vacuum pump and a pollen collector. The pollen is pulled into the glass pollen collector and kept until needed. With a third method the inflorescence of the male parent is enclosed in a pollination bag. After dehiscence the bag is used to collect the pollen. This method is less desirable, because it can be used only on those grasses with many florets.

Pollen is applied to the stigmas of emasculated florets. If emasculation has been made the day before or within the hour, receptive stigmas will appear fresh and turgid. The manner of pollen application depends on the collection method. If the pollen is collected on slick paper, in a dish, or a pollen collector, the female inflorescence can be bent and placed in contact with the pollen or the pollen can be applied to the stigmas with an artist’s brush. When pollen is collected in a bag, the bag is carefully transferred to the emasculated inflorescence; the inflorescence is inserted and agitated. Sometimes the bag can be torn apart and the pollen can be applied with a brush or the inflorescence can be brought in contact with the pollen on the bag.

After pollination, the inflorescence is enclosed in a labeled pollinating bag and stapled. An identification tag which was attached to the culm supporting the inflorescence should be attached to the base of the bag.

The expected success varies for each species. Success is usually higher in naturally cross-pollinated species than in self-pollinated species. The relationship between species being hybridized also affects success as do environmental conditions existing at the time of the cross.

D. Factors Affecting Efficiency

Limited pollen often reduces crossing efficiency. Some grass species produce very little pollen, and a large number of plants are required as a pollen source. These are often grown in the field and cuttings are the primary source of pollen.
The day length of species with different photoperiodic responses can be altered by the use of a growth chamber or lights or dark curtains in the greenhouse. If species undergo anthesis at different times of the day, their 24-hour rhythms can be altered by placing one plant in a growth chamber or a dark room to delay flowering. Proper management can provide synchronized flowering and anthesis in two species.

Although genetic markers are extremely valuable in identifying hybrids when one parent is carrying the marker, very few are reported in the warm-season grasses. These include a recessive white stigma color in common bahiagrass (Burton, 1948c), albino seedlings in Pensacola bahiagrass (Hodgson, 1949), and two chlorophyll deficient seedling characters in *Sorghum almum* (Pritchard, 1965b). Yellow-colored panicles in rhodesgrass (Bogdan, 1963), purple stolons in *Zoysia japonica* Steud. (Forbes, 1952), and hairiness in *Setaria trinervia* Stapf. (Hacker, 1968) also have been useful markers.

V. NATURAL HYBRIDIZATION

Many warm-season grasses are highly cross-pollinated because of self-incompatibility which facilitates natural hybridization. Male sterility and protogyny also allows natural hybridization.

Male-sterile lines are available in only a few warm-season grasses, probably because plant breeders have directed little attention to this effort. Male-sterile plants have been found or developed and have been used in Pensacola bahiagrass (Burton, 1948b), *Bothriochloa intermedia* (R.Br.) A. Cams (Harlan et al., 1962), sudangrass (Craigmiles, 1961), and johnsongrass (Casady, 1961).

In naturally cross-pollinated species, artificial hybrids are often produced by enclosing inflorescences of each parent at similar stages of anthesis in the same bag. Van Heemert and Schank (1968) used Terylene pollinating bags to produce *Digitaria* hybrids. They placed detached culms of the male parent in a test tube containing water and attached the tube to a stake below the bag. The culms extended inside the bag to a point where the inflorescence of the male parent was located above the female inflorescence to facilitate pollen transfer. Placing the inflorescences of the male parent in test tubes eliminated the possibility of seed forming in the male parent because the water supply in the test tube was gone by the end of anthesis. Mutual pollination has been accomplished with other cross-pollinated species by enclosing the inflorescences of both parents in the same bag (Hacker, 1968; Burton, 1948b, 1965).

In the usual design for crossing blocks in the field, one female plant is surrounded by pollen parents at a distance of 0.5 to 1 m between plants. In grasses that shatter their seed prematurely, the inflorescence of the female parent may be enclosed in a nylon cage, which permits the pollen to pass and yet retains the seed shattered from the female plant.

Each crossing block should be isolated from potential sources of contamination. This has been accomplished by spatial isolation, barriers to pollen movement, and a knowledge of the direction of the prevailing wind.
In kleingrass, *Panicum coloratum* L., crossing blocks were located at least 90 m apart, in an orchard where the trees served as a barrier (Burson and Bashaw, 1969). In side oats grama, *Bouteloua curtipendula* (Michx.) Torr., isolation blocks were about 7.6 m² in size and 8.5 m apart in each direction, with a thick stand of sorghum planted between blocks (Harlan, 1950). Adequate isolation in Pensacola bahiagrass was 176 m for foundation seed production (Hodgson, 1949). Jones and Newell (1946) concluded that isolation distances of 302 m were necessary to avoid contamination in cross-pollinated grasses.

Culms of some cross-pollinated species can be removed shortly before anthesis and placed in water, which allows anthesis, pollination, fertilization, and seed formation. This method eliminates the necessity of planting specific field designs for particular crossing schemes. It also reduces problems related to contamination because the crosses are made inside a laboratory or greenhouse. This technique has been used to obtain seed for recurrent restricted phenotypic selection, single crosses, and polycrosses (Burton, 1948b, 1974).

**VI. SEED DEVELOPMENT, HARVEST, AND STORAGE**

Grass seed normally develop rapidly. Mature embryos developed in 14 to 18 days after pollination in dallisgrass (Bennett, 1944), 12 to 14 days in blue panicgrass, *Panicum antidotale* Retz. (Wright and Hall, 1965), and 8.5 days in weeping lovegrass (Busey, 1976). Seed development is commonly determined by removing several spikelets from the inflorescence and freeing the seed from the lemma and palea by rubbing them in the palm of the hand. This method is undesirable for hand-emasculated florets because hybrid seed may be lost. Seed development is best determined by germinating the seed after proper drying and storage.

In most grasses, the seed are harvested when the culm and inflorescence turn brown. Inflorescences from crosses are usually handled individually. Each culm is clipped from the plant by hand, and it is kept in the pollinating bag during drying. The bag should be folded and stapled at the opening where the culm enters to avoid the loss of seed inside the bag. The bags are placed in a warm dry environment to provide proper drying. In dallisgrass, a temperature from 38 to 60 °C provided adequate drying without reducing seed viability (Bennett and Marchbanks, 1969). The desired final moisture level in the seed is about 10%. After the seed is dry, the inflorescences are removed from the bags, threshed by rubbing them between corrugated rubber blocks, and cleaned with a small forced-air blower or separated by screening. Hanson and Carnahan (1956) have discussed grass seed harvesting and processing in detail.

The cleaned seed are placed into coin envelopes with the essential information written on the outside. The environmental conditions are very critical if the seed is to be stored for an extended period. High temperature and humidity result in rapid seed deterioration. As a general rule, the combined sum of the percentage of relative humidity and temperature (°F) should not exceed 100. Storage conditions that do not exceed 10 °C and 40% relative humidity or lower would be desirable.
Proper storage conditions include pest control. Naphthalene crystals often are placed in the envelopes to protect against insects. Seed stored in paper envelopes should be placed in metal containers to protect against rodents. Sometimes the seed are stored in small glass jars for this purpose. Seed storage areas should also be rodent-proof; however, poison bait should be in the room as a precaution.

REFERENCES


