Techniques to screen sorghums for resistance to insect pests

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Introduction

Sorghum is an important cereal crop in Asia, Africa, Americas and Australia. Grain yields on farmers' fields in Asia and Africa are generally low (500–800 kg ha^{-1}) mainly due to insect pest damage. Nearly 150 insect species have been reported as pests on sorghum (Sharma 1993), of which sorghum shoot fly (*Atherigona soccata*), stem borers (*Chilo partellus, Busseola fusca, Eldana saccharina* and *Diatraea* spp.), armyworms (*Mythimna separata, Spodoptera frugiperda* and *S. exempta*), shoot bug (*Peregrinus maidis*), aphids (*Schizaphis graminum* and *Melanaphis sacchari*), spider mites (*Oligonychus* spp.), grasshoppers and locusts (*Hieroglyphus, Oedaleus*, *Aliopus*, *Schistocerca*, and *Locusta*), sorghum midge (*Stenodiplosis sorghicola*), mirid head bugs (*Calocoris angustatus* and *Eurystylus oldi*), and head caterpillars (*Helicoverpa, Eublemma*, *Cryptoblabes*, *Pyroderces*, and *Nola)* are the major pests worldwide. Other insects reported to be damaging sorghum are of regional/local importance. Annual losses due to insect pests differ in magnitude on a regional basis. They have been estimated to be $1,089 million in the semi-arid tropics (SAT), $250 million in United States, and $80 million in Australia (ICRISAT 1992). In India, nearly 32% of sorghum crop is lost due to insect pests (Borad and Mittal 1983).

Host-plant resistance is one of the most effective means of pest management in sorghum. It is compatible with other methods of pest control, there is no cost involvement for the farmers, and is environment-friendly. There are over 36,700 sorghum germplasm accessions in the genebank at ICRISAT, which serves as a global repository of the sorghum germplasm. We have undertaken an extensive exercise to screen the sorghum germplasm collection for resistance to the key sorghum pests such as sorghum shoot fly, spotted stem borer, sorghum midge and head bugs. A brief description of the biology, nature of damage by the target insect pests, and the techniques followed to evaluate for resistance to these insect species are described below.

Sorghum shoot fly, *Atherigona soccata*

Biology and nature of damage

Fig. 17. Formation of deadhearts in sorghum due to shootfly damage.

Sorghum shoot fly, *A. soccata* is a key pest of sorghum in Asia, Africa and the Mediterranean Europe. Shoot fly females lay cigar shaped eggs singly on the lower surface of the leaves at 1 to 7-leaf stage, ie, 5 to 25 days after seedling emergence. Eggs hatch in 1 to 2 days, and the larva moves along the shoot to the growing point. The larva cuts the growing point, resulting in wilting and drying of the central leaf, known as a deadheart (Fig. 17). The deadheart can be pulled out easily, and produces a bad smell. Normally, the damage occurs one week to four weeks after seedling emergence. The damaged plants produce side tillers, which may also be attacked. Larval development is completed in 8 to 10 days and pupation takes place mostly in the soil. The pupal period lasts for 8 days. The entire life cycle is completed in 17 to 21 days. The shoot fly population begins to increase in July, peaks in August-September, and declines thereafter. Infestations are high when sorghum plantings are staggered due to erratic rainfall. Shoot fly infestations are normally high in the postrainy season crop planted in September to October. Temperatures above 35°C and below 18°C, and continuous rainfall reduces shoot fly survival. During the off-season, the insect survives on alternate hosts such as *Echinochloa colonum*., *E. procera*, *Cymbopogon* sp., *Paspalum scrobiculatum*, *Pennisetum glaucum,* and on volunteer/fodder sorghum.

Resistance-screening techniques

Interlard-fishmeal technique (multi-choice field-screening). Adequate shoot fly density for resistance screening can be achieved by manipulating the sowing date, using infester rows, and spreading fishmeal (which attracts the shoot flies) in the field (Plate 1). Shoot fly abundance can be monitored through fishmeal-baited traps to determine the periods of peak abundance of the shoot fly. This information can be used for planting the test material so that the susceptible stage of the crop coincides with the optimum shoot fly pressure. Late-sown crops are subjected to high shoot fly infestation. At ICRISAT-Patancheru, sowing test material in

mid-July in the rainy season, and during October in the postrainy season is effective in screening for resistance to shoot fly. The interlard-fishmeal technique, which is useful for increasing shoot fly abundance under field conditions, involves planting four rows of a susceptible cultivar (such as CSH 1 or CSH 5), sown 20 days before the sowing of test material. Fishmeal is spread uniformly 1 week after seedling emergence or kept in plastic bags in the interlards to attract shoot flies from the surrounding areas. One generation of the shoot fly is completed on interlards, and the emerging flies infest the test material. The same procedure can also be adopted for the test material as well (Taneja and Leuschner 1985a, Sharma et al. 1992).

No-choice-cage-screening technique. To confirm resistance to the observed under field conditions, and to study the resistance mechanisms, the cage-screening technique developed by Soto (1972) has been modified to simulate field conditions. The shoot flies are collected from fishmeal-baited traps in the field (Sharma et al. 1992). Shoot flies are collected in the morning, and are separated from other dipteran flies. The cage-screening technique can be used for multiple- or no-choice tests. For a multiple-choice test, the test genotypes are sown in the field in 3.4×2 m beds, with a row spacing of 15 cm. Ten days after seedling emergence, the plants are covered with a 3.4×2×1 m screened cage, and the shoot flies are introduced into the cage. Eggs and deadhearts are recorded after 1 week. For a no-choice test, only one genotype is sown in 1×1 m beds. Six beds can be covered with a 2×3×0.5 m cage having six compartments. Twenty shoot flies are released into each compartment, and observations are recorded as described above. Rapid screening can also be carried out using a top-cage technique. This system consists of two plastic trays (40×30×14 cm), one for sowing the test material and the other (a top-cage fitted with fine wire-mesh) is clamped over the lower tray, thus forming a cage. Ten days after seedling emergence, the top-cage is assembled and 20 flies are released into each cage through an opening. Observations are recorded as described below.

Damage evaluation for resistance screening. Record the number of eggs and the plants with eggs, plants with deadhearts, and the total number of plants at 14 and 21 days after seedling emergence. Record the number of tillers, and tillers with panicles at maturity as a measure of genotype's recovery resistance. Grain yield under protected and unprotected conditions can also be used as a measure of resistance to sorghum shoot fly.

Spotted stem borer, *Chilo partellus*

Spotted stem borer, *Chilo partellus* is common in Asia and East and Southern Africa. The first indication of stem borer infestation is the appearance of small-elongated windows in whorl leaves where the young larvae have eaten the upper surface of

the leaves. Later, the plant presents a ragged appearance as the severity of damage increases. The third-instar larvae migrate to the base of the plant, bore into the shoot, and damage the growing point resulting in the production of a deadheart. Normally, two leaves dry up as a result of stem borer damage. Larvae continue to feed inside the stem throughout the crop growth. Extensive tunneling of the stem and peduncle leads to drying up of the panicle, production of a partially chaffy panicle or peduncle breakage (Fig. 18a & b). Stem borer infestation starts about 20 days after seedling emergence, and deadhearts appear on 30 to 40 day old-crop. A female lays up to 500 eggs in batches of 10 to 80 near the midrib on the under surface of the leaves. Eggs hatch in 4 to 5 days. The larval development is completed in 19 to 27 days. Pupation takes place inside the stem and the adults emerge in 7 to 10 days. During the off-season, the larvae diapause in plant stalks and stubbles. With the onset of rainy season, the larvae pupate and the adults emerge in 7 days. In northern India, moth catch in light traps begins to increase during the last week of July and peaks during August to September, while in southern India, the peak in moth catches has been recorded during January to February.

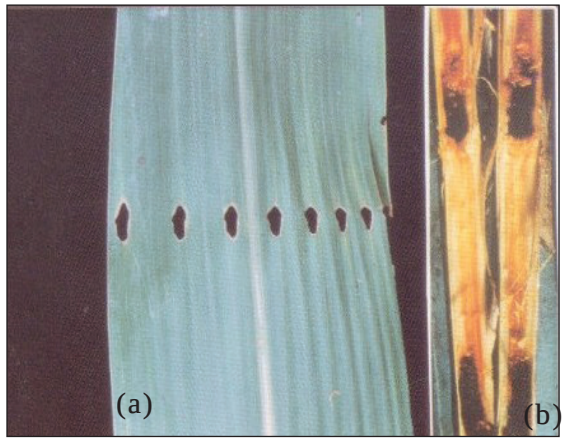

Fig. 18. Leaf damage (a) and stem tunneling (b) by spotted stem borer in sorghum.

Resistance-screening techniques

Techniques to screen for resistance to spotted stem borer have been described by several workers (Jotwani 1978, Taneja and Leuschner 1985b, Sharma et al. 1992). The following techniques may be followed to screen for resistance to stem borer under natural and artificial infestation.

Screening under natural infestation

Hot-spots. Hot-spot locations, where the pest populations are known to occur naturally and regularly at levels that often result in severe damage, are ideal to test large numbers of germplasm accessions. Hot-spot locations for *C. partellus* are Hisar in Haryana and Warangal in Andhra Pradesh, India; Agfoi and Baidoa in Somalia; Panmure and Mezarbani in Zimbabwe; Kiboko in Kenya; and Golden Valley in Zambia.

Sowing date. To screen for resistance under natural infestation, especially at the hot-spot locations, adjust the sowing date of the crop such that the crop is at a susceptible stage when the stem borer abundance is at its peak. Determine the periods of maximum borer abundance through pheromone traps, light traps, or by monitoring borer infestation in the crop planted at regular intervals. In northern India, *C. partellus* is most abundant in August to September, and the crop sown between the 1st and 3rd week of July suffers maximum stem borer damage. At ICRISAT-Patancheru, a maximum number of moths in the light traps have been recorded during September, followed by smaller peaks during November and February.

Mass rearing and artificial infestation

Mass rearing. Artificial infestation with laboratory-reared insects has been successfully used for several pest species, including *C. partellus*. Several diets have been developed for mass rearing of *C. partellus*. An artificial diet to rear *C. partellus* has been standardized at ICRISAT (Taneja and Leuschner 1985b). Most of the ingredients of this diet [Fraction A: water 2000 mL, *kabuli* chickpea flour 438.4 g, brewer's yeast 32.0 g, sorbic acid 4.0 g, vitamin E (Viteolin capsules) 4.6 g, methyl parahydroxy benzoate 6.4 g, ascorbic acid 10.4 g, and sorghum leaf powder 160.0 g. Fraction B: Agar-agar 40.8 g, water 1600 mL, and formaldehyde (40%) 3.2 mL)] This amount is enough to prepare 15 jars with 300 g diet each. For preparing sorghum leaf powder, collect leaves from a susceptible cultivar (such as CSH 1) from 35 to 40-day-old plants. Wash the leaves, dry in sunshine or in an oven at 65°C, grind them into a fine powder, and autoclave for 15 min at 120°C at 5 kg cm^{-2} pressure. Store the leaf powder in a sealed container in a cool dry place. Blend the ingredients of fraction A (except the sorghum leaf powder) for 1 min. Soak the sorghum leaf powder in warm water (70°C) and blend with fraction A for 2 min. Boil agar-agar (fraction B) in 1.6 L of water, cool it to 40°C, combine with formaldehyde and fraction A, and blend for 3 min. Pour 300 g diet in a one-liter plastic jar. Allow the diet in the jar to cool to room temperature. Place about 100 eggs at the black-head stage in each jar, and keep the jars in a dark room for 2 days. This discourages the photopositive behavior of 1st-instar larvae, and they settle on the diet for feeding. After 2 days, transfer the jars to the rearing room, maintained at 28±1°C, 60 to 70% relative humidity (RH), and 12 h photoperiod. On artificial diet, the larval period lasts for 22 to 28 days and the pupal period for 5 to 6 days. Moth emergence begins 30 days after larval inoculation, and continues up to the 40th day. Females emerge 2 to 3 days later than the males. The sex ratio is close to 1:1. Average moth emergence from this diet is 70 to 75%, with a maximum of up to 90%. Most of the moths emerge in 30 to 40 days after larval inoculation.

The moths are collected with the help of aspirators attached to a vacuum cleaner or with the help of hand-held aspirators. Collect the male and female moths separately (males are smaller in size with dark forewings and pointed abdomen), and transfer them to the egg-laying cages. The oviposition cage is a wire-framed (36 mm holes) cylindrical cage (25 cm high and 25 cm in diameter). A fine georgette cloth with 6×6 mm holes at regular intervals is fitted around the outer side of the cage. A sheet white glycine paper (25×80 cm) is wrapped around the cage to serve as an oviposition site. Two plastic saucers covered with a mosquito net are placed at the bottom and the top of the cage. Release 50 pairs of moths in each oviposition cage. A female lays 10 to 12 egg masses (500 to 600 eggs) over a period of 4 days. Most eggs are laid on the 2nd and 3rd day after emergence. Replace the glycine paper daily. Feed the moths with water using a cotton swab. Egg hatching is drastically reduced when relative humidity falls below 50%. To obtain high humidity, hang the glycine papers containing egg masses on a rod in a plastic bucket containing water. Cover the plastic bucket with a lid. Store the eggs at 26±1°C. Under these conditions, the embryo matures to the black-head stage within 4 days. For long-term storage, keep black-head stage eggs at 10°C. This delays egg hatching up to 15 days. This is quite helpful in timely infestation of the test material.

Field infestation. For field infestation, the Bazooka applicator, developed at the International Maize and Wheat Improvement Center (CIMMYT) (CIMMYT 1977), has been modified to suit the requirements for infesting sorghum. Take 500 black-head stage egg masses along with 85 g of poppy seeds (*Papaver* sp.) or corn cob grits, and keep them overnight in a plastic jar with a tightly fitted lid. In the morning, mix the 1st-instar larvae with the carrier and transfer them into the plastic bottle of the Bazooka. Infest 15 to 20-day-old plants in the field individually by placing the nozzle of the Bazooka close to the leaf whorl. With a single stroke, 5 to 7 larvae are released into each plant whorl, which are sufficient to cause appreciable leaf feeding and >90% deadhearts in the susceptible genotypes. Deadheart formation decreases progressively as the infestation is delayed. For stem and peduncle tunneling, plants may be infested between 25 to 35 days after seedling emergence. Infest the crop in the morning between 0800 and 1100 to avoid larval mortality due to high temperature. Rotate the Bazooka applicator after every 10 plants to ensure uniformity in larval distribution. The number of larvae per plant can be regulated by varying the number of egg masses mixed with the carrier in each Bazooka. A second infestation may be required if it rains immediately after first infestation. Shoot fly infestation interferes with screening for resistance to stem borer. Spray fenvalerate or endosulfan to suppress shoot fly infestation 1 week before artificial infestation with stem borer. Also, it is helpful to sow the test material early in the season when shoot fly infestation is negligible.

Damage evaluation for resistance screening

Stem borer attack in sorghum causes leaf damage, deadheart formation, stem and peduncle tunneling, and production of chaffy panicles. Leaf injury is the first larval feeding symptom, and is related to yield loss only under severe infestation. Stem tunneling adversely affects the quantity and quality of fodder, but is not correlated with reduction in grain yield under many situations. Peduncle damage could be critical if there are winds of high enough velocity to break the peduncle. Deadheart formation is the most important criterion for differentiating degrees of resistance, and is directly related to loss in grain yield. The following observations may be recorded to evaluate for resistance to stem borer.

Leaf feeding. Record the rate of leaf feeding 2 weeks after artificial infestation, and 4 to 5 weeks after crop emergence under natural infestation. Record the total number of plants, the number of plants showing the leaf-feeding symptoms, and the leaf-feeding score on a 1 to 9 scale (1=<10% leaf area damaged, 2=11–20, 3=21–30, 4=31–40, 5=41–50, 6=51–60, 7=61–70, 8=71–80, and 9= >81% leaf area damaged).

Deadhearts. Record plants with deadhearts 3 weeks after artificial infestation, and 4 to 6 weeks after crop emergence under natural infestation. Record the total number of plants, plants showing borer deadhearts, and the visual score (1 to 9 scale) as described for leaf feeding score (1 = <10% plants with deadhearts, and 9 = >80% plants with deadhearts).

Chaffy panicles. At crop harvest, record observations on the number of partial and completely chaffy panicles, the number of broken panicles, and the visual score (1 to 9 scale) for chaffy/broken panicles, and grain mass per 100 grains.

Recovery resistance. Record the number of plants with tillers and the number of tillers with productive panicles. Evaluate for recovery resistance on a 1 to 5 scale (1=>80% plants with 2 to 3 uniform and productive tillers, and 5=<20% plants with one or nil non-uniform productive tillers).

Stem tunneling. At maturity, record plant height and the peduncle length of five plants at random in each plot. Measure the stem and peduncle tunneling separately and express it as a percentage of stem/peduncle length.

Shoot bug, *Peregrinus maidis*

Shoot bug, *Peregrinus maidis* is a common pest of sorghum in India, parts of Africa, West Indies and the Philippines. It sucks sap from the leaf whorls. The damaged plants become stunted. In case of severe infestation, the top leaves start

drying up first, extending gradually to the lower leaves, and the plant may die. The leaves curl and present a tanned appearance. They also secrete honeydew on which the sooty molds grow. Infestation at the later stages of plant growth may twist the top leaves, and inhibit panicle emergence. Its infestations are more severe under drought conditions. It is a serious pest of sorghum in the postrainy season in India. The female is yellowish brown, and the male dark brown. Wings may be longer or shorter than the abdomen. Long winged forms have transparent wings. Females are larger than the males. The nymphs and adults live in groups in plant whorls, and on the inner side of leaf sheaths. The females make a slit in the upper surface of the mid-rib and insert the eggs in groups of 1 to 4, and cover them with a white waxy substance. A female lays up to 100 eggs in 7 days. The eggs are white, elongate, cylindrical and taper at the ends. Egg incubation period is 1 to 7 days. There are 7 nymphal instars, and the development is completed in 16 days.

Evaluation for resistance

Screening for resistance to shoot bug can be carried out under natural infestation in field or in the greenhouse. For this purpose, the material should be planted during late rainy season in July or early post-rainy season in October. Appropriate resistant and susceptible checks should be included in the trials. The material should be protected against shoot fly and stem borer through whorl application of carbofuran granules during the seedling stage. Shoot bug infestation/damage should be evaluated on the main plants. Shoot bug infestation can also be created under greenhouse conditions, by using leaf cages or by confining the shoot bug females to the whorl leaves. Data may be recorded on the numbers of shoot bugs produced in 15 days. For this purpose, the leaf cages can be fixed on 3rd or 4th leaf from the bottom, or infestation carried out in the whorl leaves, and the plants placed away from each other.

Damage evaluation

Shoot bug density. Just before emergence of the flag leaf, ie, 45 days after seedling emergence, evaluate the test genotypes for shoot bug resistance by recording insect density. For this purpose, count the number of shoot bugs in the whorls of five plants tagged at random in each plot.

Plant damage ratings. Shoot bug damage to the plants can be evaluated at the panicle emergence stage on a 1 to 9 scale as follows:

1. A few shoot bugs present in the leaf whorls and no apparent damage to the leaves.
2. One to two central leaves showing damage symptoms, and 10−20% of the infested leaf area damaged.

3. Two to three leaves showing damage symptoms, and 20−30% of the leaves damaged.
4. Three to four leaves showing damage symptoms, and 30−40% of the leaves damaged.
5. Four to five leaves showing damage symptoms, and 40−50% of the leaves damaged.
6. Five to six leaves showing damage symptoms, and 50−60% of the leaf area damaged, and no panicle emergence in some plants.
7. Six to seven leaves showing damage symptoms, 60−70% of the leaf area damaged, and 50% plants with panicle emergence.
8. Seven to eight leaves showing damage symptoms, 70−80% of the leaf area damaged, and a few plants with panicle emergence.
9. Most of the leaves showing shoot bug infestation > 80% and plants with a twisted appearance and no panicle emergence.

Grain yield. Record grain yield of the genotypes being tested. The test material can be maintained under infested and noninfested conditions. Harvest all panicles from the middle row(s) at maturity, and record panicle and grain mass. Express the loss in grain yield in the infested plots or panicles as a percentage of the grain yield in noninfested plots or panicles.

Sugarcane aphid, *Melanaphis sacchari*

The sugarcane aphid, *Melanaphis sacchari* occurs in Asia, Africa and America. It prefers to feed on the under surface of older leaves (Fig. 19). The damage proceeds from lower to the upper leaves. The adults and nymphs are yellow. They suck sap from the lower surface of leaves, and this leads to stunted plant growth. The damage is more severe in crop under drought stress, resulting in drying of leaves and plant mortality. The aphids secrete honeydew, which falls on the ground, on which sooty molds grow. Their numbers increase rapidly at the end of the rainy season during dry spells. This aphid also reproduces by parthenogenesis. Each female gives birth to 60 to 100 nymphs in 13 to 20 days. The life cycle is completed in 6 to 7 days during the dry season. Its populations are high during the postrainy season in India.

Fig. 19. Aphid infestation in sorghum.

Screening for resistance

Screening for resistance to sugarcane aphid can be carried out under natural infestation in the field. For this purpose, the material should be planted during late rainy season in July or early postrainy season in October. Appropriate resistant and susceptible checks should be included in the trials. The material should be protected against shoot fly and stem borer through whorl application of carbofuran granules, and aphid infestation/damage should be evaluated on the main plants only. Aphid infestation can also be created under greenhouse conditions. Aphid multiplication and growth rates can be studied using leaf cages by confining aphid females with the leaves, and counting the numbers of aphids produced in 15 days. For this purpose, the leaf cages can be fixed on 3rd or 4th leaf from the bottom.

Damage evaluation

Aphid density. At the flag leaf stage or 50 days after seedling emergence, evaluate the test genotypes for aphid resistance by recording aphid density. For this purpose, count the number of aphids in a unit area (3×3 cm) on the mid-portion of three leaves per plant (3rd, 5th and 7th leaf), and record observations on five plants tagged at random in each plot.

Leaf damage rating. Leaf damage due to aphid feeding can be assessed at the milk-dough stage of grain development using a 1 to 9 scale.

1. A few aphids present on the lower 1−2 leaves, with no apparent damage to the leaves.
2. Lower 1 to 2 leaves showing aphid infestation, and 10−20% of the infested leaves/ area showing damage symptoms.
3. Lower 1 to 3 leaves showing aphid infestation, and 20−30% of the infested leaves/area showing damage symptoms, with moderate leaves of honeydew/ black molds on the leaves/soil.
4. Lower 1 to 4 leaves showing aphid infestation, and 30−40% of the infested leaves/area showing damage symptoms, with moderate leaves of honeydew/ black molds on the leaves/soil.
5. Lower 1 to 5 leaves showing aphid infestation, and 40−50% of the infested leaves/area showing damage symptoms, with moderate levels of honeydew/ black molds on the leaves/soil.
6. Aphid infestation up to 6th leaf, and 50−60% of the infested leaves/area showing damage symptoms, and heavy honeydew/black molds on the leaves, and on soil below.
7. Aphid infestation up to 7th leaf, and 60−70% of the infested leaves/area

showing damage symptoms, and heavy honeydew/black molds on the leaves, and on soil below.

8. Aphid infestation up to 8th leaf, and 70−80% of the infested leaves/area showing damage symptoms, and heavy honeydew/black molds on the leaves, and on soil below.
9. Heavy aphid infestation up to the flag leaf, and >80% of the leaves showing aphid damage (drying up symptoms), heavy honeydew/black molds on the leaves, and on soil below.

Grain yield. Record grain yield of the genotypes being tested. The test material can be maintained under infested and noninfested conditions. Harvest all panicles from the middle row(s) at maturity, and record panicle and grain mass. Express the loss in grain yield in the infested plots or panicles as a percentage of the grain yield in noninfested plots or panicles.

Sorghum midge, *Stenodiplosis sorghicola*

Sorghum midge, *Stenodiplosis sorghicola* larvae feed on the developing ovary resulting in production of chaffy spikelets. Females lay eggs in panicles at flowering during the morning hours. The damaged panicles present a blasted appearance. Midge damaged spikelets have a pupal case attached to the glumes or have a small exit hole of the midge parasite on the upper glume. Adults emerge between 0600 to 1100. Mating takes place within one hour after emergence. Generally, the males emerge one hour earlier than the females, and hover around the spikelets where the females are about to emerge. Males die after mating while the females proceed in search of sorghum panicles at flowering for oviposition. Females lay 30 to 100 eggs singly in the spikelets at anthesis during the morning hours, and die by the afternoon. Eggs hatch in 1 to 4 days. The larvae suck the sap from the developing ovaries and complete development in 7 to 12 days. Larvae pupate inside the glumes, and the pupal period lasts for 3 to 8 days. Adults live for 4 to 48 h. The population builds up 2 to 3 months after the onset of monsoon rains, and maximum density occurs during September to October. A small proportion of the larvae enter diapause in the spikelets in each generation, which may last as long as 3 to 4 years. The larval diapause is terminated by warm and humid weather (25 to 30°C, and > 60% relative humidity).

The major difficulties in identifying source material with stable resistance against sorghum midge includes: a) variation in the flowering of sorghum cultivars in relation to midge incidence, b) day-to-day variation in midge populations, c) competition with other insects such as head bugs, d) parasitization and predation by natural enemies; and e) sensitivity of midge flies to temperature and relative humidity. A large proportion of lines selected as less susceptible under natural conditions

comprise early- and late-flowering escapes. Because of these problems, genotypes rated as resistant under natural infestation often turn out to be susceptible in the following seasons or at other locations. Techniques to screen for midge resistance have been described by Jotwani (1978), Page (1979) and Sharma et al. (1988a,b, 1992).

Resistance-screening techniques

Field screening techniques (multi-choice conditions)

Hot-spots. Hot-spot locations are useful to screen for resistance to sorghum midge. Hot-spot locations for sorghum midge are Dharwad, Bhavanisagar and Pantnagar in India, Sotuba in Mali, Farako Bâ in Burkina Faso, Alupe in Kenya, and Kano in Nigeria. Midge infestations are also high at several locations in Australia, the USA and Latin America.

Sowing date. To screen test the material for resistance to sorghum midge under natural conditions, it is necessary to determine the appropriate time for sowing at different locations. Determine the periods of maximum midge density through fortnightly sowings of a susceptible cultivar. Adjust sowing dates so that the flowering of the test material coincides with greatest insect density. At ICRISAT-Patancheru, maximum midge damage has been observed in the crop planted during the 3rd week of July. The peak in midge density occurs during October, and a second but smaller peak has been observed during March in the postrainy season, for which planting is carried out during mid-December.

Infester row technique. Midge abundance can be increased through infester rows and spreading sorghum panicles containing diapausing midge larvae in the infester rows (Sharma et al. 1988a). Sow infester rows of susceptible cultivars such as CSH 1 and CSH 5 (1:1 mixture) 20 days before the test material. Alternatively, early-flowering (40 to 45 days) lines (IS 802, IS 13249 and IS 24439) can be sown along with the test material. Plant four infester rows of the susceptible cultivar after every 16 rows of the test material. Collect midge-infested chaffy panicles containing diapausing midge larvae at the end of the cropping season, store in gunny bags or in bins under dry conditions until the next season. Moisten the panicles for 10 to 15 days to stimulate the termination of larval diapause. Spread midge-infested sorghum panicles containing diapausing midge larvae at the flag leaf stage of the infester rows. Adults emerging from the diapausing larvae serve as a starter infestation in the infester rows to supplement the natural population. Midge population multiplies for 1 to 2 generations on the infester rows before infesting the test material. This technique increases the midge damage by 3 to 5 times. Infester rows alone are also effective in increasing midge infestation.

Sprinkler irrigation. High relative humidity is important for adult emergence, oviposition and subsequent damage. Use overhead sprinkler irrigation to increase relative humidity in midge-screening trials during the postrainy season or periods of low relative humidity. Operate sprinkler irrigation daily between 1500 to 1600 from panicle emergence to the grain-filling stage of the crop. Sprinkler irrigation between 1500 to 1600 does not affect oviposition by the midge females because peak midge activity and oviposition occur between 0730 and 1100. Midge damage increases significantly with the use of sprinkler irrigation.

Selective use of insecticides to control other insects. Head bug, *Calocoris angustatus* and the midge parasitoid, *Tetrastichus diplosidis* are the two major insects limiting midge abundance in resistance screening trials. Head bugs damage the sorghum panicles from emergence to hard-dough stage and compete for food with the sorghum midge. They also prey on the ovipositing midge females at flowering, while *T. diplosidis* is an efficient parasite of sorghum midge at some locations. Spray less persistent and contact insecticides such as carbaryl and malathion to control head bugs at the complete-anthesis to milk stage (Sharma and Leuschner 1987). The sorghum midge larvae feeding inside the glumes are not affected by the contact insecticides sprayed after flowering.

Split sowings and grouping the material according to maturity and height. Group the test material according to maturity (early, medium and late) and height (dwarf, medium and tall) for proper comparisons, and avoid the shading effect from tall genotypes. Sow the test material twice at a 15-day interval to minimize the chances of escape from midge damage. Split sowing of the test material increases the efficiency of selection for midge resistance. Plant population also affects the insect density per unit area, and in some cases influences the incidence and survival rate of insects. The level of midge damage has been observed to be higher at lower planting densities.

No-choice headcage technique

Caging midge flies with sorghum panicles permits screening for midge resistance under uniform insect pressure. A headcage technique has been developed at ICRISAT-Patancheru. It consists of a cylindrical wire frame made of 1.5 mm diameter galvanized iron wire. The loop attached to the top ring rests around the tip of the panicle, and the extensions of the vertical bars at the lower ring are tied around the peduncle with a piece of G.I. wire or electric wiring clips. Select sorghum panicles at 25 to 50% anthesis stage, and remove spikelets with dried-up anthers at the top and immature ones at the bottom of the panicle so that only the spikelets at anthesis are exposed to the midge flies for oviposition. Place the wire-framed cage around the sorghum panicle and cover it with a blue cloth bag (20 cm wide

and 40 cm long). The cloth bag at the top has an extension (5 cm in diameter, 10 cm long) to release the midges inside the cage. Collect 20 adult female midges in a plastic bottle (a 200 ml aspirator) between 0800 and 1100 from flowering sorghum panicles (only female midges visit the flowering sorghum panicles and these are collected for use in infestation). Release 40 midges into each cage, and repeat the operation the next day. Infest 5 to 10 panicles in each genotype, depending upon the stage of material and the resources available. Midge damage decreases as the time of collection and release advances from 0830 to 1230. Examine the cages 5 to 7 days after infestation and remove any other insects such as head bugs, panicle-feeding caterpillars and predatory spiders from inside the cage. Remove the cages 15 days after infestation and evaluate the midge damage. The headcage technique is quite simple, easy to operate, and can be used on a fairly large scale to confirm the field resistance of selected genotypes. Changing weather conditions influence midge activity and can affect midge damage under the headcage. In general, it is a thorough test for use in resistance screening, and is particularly applicable in identifying stable and durable resistance.

Damage evaluation for resistance screening

Feeding by the midge larva inside the glumes leads to sterile or chaffy spikelets. However, the symptoms (chaffiness) of natural sterility and extensive grain damage by sucking insects are superficially similar to the damage caused by sorghum midge. The midge-infested panicles have either small white pupal cases attached to the tip of damaged spikelets or have small parasite exit holes in the glumes. Genotypes flowering on different dates should be tagged with different-colored labels or tapes or marked with paint along with panicles of resistant and susceptible checks for proper comparison. Selection for resistance should be based in relation to reaction of resistant and susceptible checks flowering on the same day.

Chaffy spikelets. This is the most appropriate criterion by which to evaluate sorghum lines for midge resistance. Tag five panicles in each genotype at half-anthesis stage. Record midge damage in 250 spikelets at 15 days after flowering or at maturity. Collect five primary branches each from the top, middle and bottom portions of each panicle, bulk the samples from all the five tagged panicles in a genotype, remove secondary branches from the primary branches, and mix the sample thoroughly. Pick up the secondary branches at random and count the number of chaffy spikelets in a sample of 250 spikelets. In samples collected at the milk stage, squeeze the chaffy spikelets between the thumb and first finger or with forceps, and record the number of spikelets producing a red ooze (this indicates midge damage). Chaffy spikelets with early-instar larvae at times may not produce a red ooze. Express the data as a percentage of chaffy or midge-damaged spikelets.

Visual damage rating. At crop maturity, evaluate midge damage on a 1 to 9 scale where 1=<10%, 2=11–20%, 3=21–30%, 4=31–40%, 5=41–50%, 6=51–60%, 7=61–70%, 8=71–80% and 9=>81% midge-damaged spikelets.

Grain yield. Record grain yield from the genotypes being tested. The test material can be maintained under infested and noninfested conditions by using cloth bags or sprayed with insecticides at flowering to control the sorghum midge. Harvest all panicles from the middle row(s) at the time of maturity and record the panicle and grain weight. Express the loss in grain yield in the infested plots or panicles as a percentage of the grain yield in non-infested plots or panicles.

Head bug, *Calocoris angustatus*

Head bug, *Calocoris angustatus* is a serious pest of grain sorghum in India. The nymphs and adults suck the sap from the developing grain. The damage starts as soon as the panicle emerges from the boot leaf. High levels of head bug damage lead to tanning and shriveling of the grain. Head bug damage leads to both qualitative and quantitative losses in grain yield. Head bug damage spoils the grain quality, and renders the food unfit for human consumption. Such grain also shows poor seed germination. Head bug damage also increases the severity of grain molds. Head bug females lay eggs inside the spikelets from panicle emergence to post-anthesis. A female lays 150 to 200 eggs, and the eggs hatch in 5 to 7 days. Nymphal development is completed in 15 to 17 days. Nymphs feed on milky and soft-dough grains, and this results in tanning and shriveling of the grain. Its infestations are high during August-September in the rainy season. During the off-season, the bugs feed on fodder sorghum. There is no evidence of diapause. Techniques to screen for resistance to head bugs have been discussed by Sharma and Lopez (1992) and Sharma et al. (1992).

Resistance-screening techniques

Field screening

Screening for head bug resistance can be carried out under field conditions during periods of maximum bug density. Screening for head bug resistance under field conditions is influenced by: a) variation in flowering, b) fluctuations in bug density, and c) the effect of weather conditions on the bug population build-up and damage. Early- and late-flowering cultivars normally escape head bug damage, while those flowering in mid-season are exposed to very high bug infestation. The following techniques can be used to increase the screening efficiency for head bug resistance under field conditions.

Hot-spots. In India, ICRISAT-Patancheru, Bhavanisagar, Kovilpatti, Coimbatore, Palem and Dharwad are the hot-spot locations to screen for resistance to head bugs. At ICRISAT- Patancheru, head bug density is very high during September to October.

Sowing date. Adjust sowing dates such that flowering of the test material coincides with maximum head bug density. Determine the periods of maximum head bug abundance through fortnightly sowings. Maximum bug numbers at ICRISAT-Patancheru have been recorded during September and a second but smaller peak has been recorded during March. Crops sown during the second week of July suffer the maximum head bug damage. At Bhavanisagar, the peak in head bug density occurs during May to June, and the optimum time to sow the test material for resistance screening is during the second fortnight of February.

Infester-row technique. Sow infester rows of mixed-maturity cultivars 20 days earlier than the test material. Alternatively, sow early-flowering (40 to 45 days) sorghums (IS 802, IS 13249 and IS 24439) as infester rows along with the test material. Sow four rows of a susceptible cultivar after every 16 rows of the test material. Collect head bugs from other fields and spread them in the infester rows at the panicle emergence to augment bug abundance. Sow the test material in two sets, at an interval of 10 to 15 days to reduce the chances of escape in the early- and late-flowering lines. For better results, group the test material according to maturity and height. The sowing date of each maturity group can also be suitably adjusted so that flowering occurs during peak activity period of the head bugs.

No-choice headcage technique

To overcome the problem of variation in flowering among the test cultivars, and fluctuations in insect abundance, the headcage technique developed for midge resistance screening has been found to be useful to screen for resistance to head bugs as well. This technique also permits to monitor the increase in head bug population in the infested panicles under no-choice conditions in relation to different infestation levels and stages of panicle development. Select 5 to 10 panicles at the top-anthesis stage in each plot/genotype, and tie the headcage around the sorghum panicle and cover it with a white muslin cloth bag. Collect bugs in muslin cloth bags from sorghum panicles at the milk stage, and separate the adult males and females (males are smaller and darker in color than the females). Collect 10 head bug pairs in a 200-ml plastic bottle aspirator and release them inside the cage. Examine the infested panicles after 1 week and remove panicle-feeding caterpillars or predatory spiders if any. Remove the muslin cloth bag along with the bugs 20 days after infestation, kill the bugs with ethyl acetate or benzene (2 ml bag^{-1}), or keep the bags in deep-freeze for 30 min. Count the total number of bugs

in each cage. Evaluate the panicles for head bug damage at maturity as described under damage evaluation.

Damage evaluation for resistance screening

Sorghum head bugs suck the sap from developing ovary and result in shriveling and tanning of the grains. Some grains may also remain undeveloped. Damage symptoms are normally evident on some or all of the grains. Head bug damage is generally higher inside the panicle. In some cases, a portion of the panicle may be more damaged than the rest, and some grains may be normal, while others show damage symptoms. Head bug damage can be evaluated by the following criteria.

Head bug counts. Tag five panicles at random in each genotype at the half-anthesis stage. Sample the panicles for head bugs at 20 days after flowering or infestation in a polyethylene or muslin cloth bag containing a cotton swab soaked in 2 ml of ethyl acetate or benzene. Count the total number of adults and nymphs.

Grain damage rating. Evaluate head bug damage at maturity on a 1 to 9 scale (1 = all grains fully developed with a few feeding punctures, 2 = grain fully developed and with feeding punctures, 3 = grains showing slight tanning or browning, 4 = most grains with feeding punctures and a few showing slight shriveling, 5 = grains showing slight shriveling and browning, 6 = grains showing more than 50% shriveling and tanned, 7 = most of the grain highly shriveled and having a dark brown coloration, 8 = grain highly shriveled and slightly visible outside the glumes, and 9 = most of the grains highly shriveled and almost invisible outside the glumes.

Grain yield. Harvest all panicles from the middle row(s) of each plot or genotype at maturity and record panicle and grain weight in each plot or panicle. Plots or panicles of lines being tested can also be maintained under infested and uninfested conditions by using cloth bags to exclude the head bugs. Measuring grain yield and grain quality parameters under insecticide-protected and infested conditions can also be used as a measure of genotypic resistance to head bugs. Express the loss in grain yield of infested plots or panicles as a percentage of the grain yield in non-infested plots or panicles.

Grain weight and floaters. Take a sample of 1,000 grains at random from each replication or panicle. Equilibrate the moisture content (24 h at 37°C), and record the grain weight. Prepare a sodium nitrate solution of a specific density of 1.31 in a beaker. Place the 1,000-grain sample in the beaker containing sodium nitrate solution, and count the number of grains floating on the surface, and express them as a percentage of the total number of grains.

Germination test. Take 100 grains at random from each replication or panicle and place them between the folds of a water-soaked filter paper in a Petri dish. Keep

the Petri dishes in an incubator at 27±1°C or at room temperature in the laboratory. Record the percentage of grains with radical and plumule emergence after 72 h. Data on grain hardness, 1,000-grain weight, percentage floaters, and percentage germination should be recorded only when the researcher intends to collect more data for in-depth studies on head bug resistance.

References

Borad PK and **Mittal VP. 1983.** Assessment of losses caused by pest complex to sorghum hybrid CSH 5: Pages 271−278 *in* Crop losses due to insect pests (Krishnamurthy Rao BH and Murthy KSRK, eds.). Special issue of the Indian Journal of Entomology. Entomological Society of India, Rajendranagar, Hyderabad, Andhra Pradesh, India.

CIMMYT. 1977. CIMMYT review 1977. El Batan, Mexico: Centro Internacional de Mejoramiento de Maiz y Trigo. 99 pp.

ICRISAT. 1992. Medium Term Plan. Patancheru, Andhra Pradesh, India: International Crops Research Institute for the Semi-Arid Tropics.

Jotwani MG. 1978. Investigations on insect pests of sorghum and millets with special reference to host plant resistance. Final Technical Report (1972–77). Research Bulletin of the Division of Entomology. New Delhi, India: Indian Agricultural Research Institute. 114 pp.

Page FD. 1979. Resistance to sorghum midge (*Contarinia sorghicola* Coquillett) in grain sorghum. Australian Journal of Experimental Agriculture and Animal Husbandry 19:97–101.

Sharma HC. 1993. Host plant resistance to insects in sorghum and its role in integrated pest management. Crop Protection 12:11–34.

Sharma HC and **Leuschner K.** 1987. Chemical control of sorghum head bugs (Hemiptera: Miridae). Crop Protection 6:334–340.

Sharma HC and **Lopez VF.** 1992. Screening for plant resistance to sorghum head bug, *Calocoris angustatus* Leth. Insect Science and its Application 13:315–325.

Sharma HC, Doumbia YO and **Diorisso NY.** 1992. A headcage technique to screen sorghum for resistance to the mirid head bug, *Eurystylus immaculatus* Odh. in West Africa. Insect Science and its Application 13:417–427.

Sharma HC, Taneja SL, Leuschner K and **Nwanze KF.** 1992. Techniques to screen sorghum for resistance to insects. Information Bulletin no. 32. Patancheru 502 324, Andhra Pradesh, India: International Crops Research Institute for the Semi-Arid Tropics. 48 pp.

Sharma HC, Vidyasagar P and **Leuschner K.** 1988a. Field screening sorghum for resistance to sorghum midge (Cecidomyiidae: Diptera). Journal of Economic Entomology 81:327–334.

Sharma HC, Vidyasagar P and **Leuschner K.** 1988b. No-choice cage technique to screen for resistance to sorghum midge (Cecidomyiidae: Diptera). Journal of Economic Entomology 81:415–422.

Soto PE. 1972. Mass rearing of sorghum shoot fly and screening for host plant resistance under greenhouse conditions. Pages 137–138 *in* Proceedings of the International Symposium on Control of Sorghum Shoot Fly (Jotwani MG and Young WR, eds.). New Delhi, India: Oxford and IBH.

Taneja SL and **Leuschner K.** 1985a. Resistance screening and mechanisms of resistance in sorghum to shoot fly. Pages 115–129 *in* Proceedings of the International Sorghum Entomology Workshop, 15–21 July 1984, Texas A&M University, College Station, Texas, USA. Patancheru 502 324, Andhra Pradesh, India: International Crops Research Institute for the Semi-Arid Tropics.

Taneja SL and **Leuschner K.** 1985b. Methods of rearing, infestation, and evaluation for *Chilo partellus* resistance in sorghum. Pages 178–185 *in* Proceedings, International Sorghum Entomology Workshop, 15–21 July 1984, Texas A&M University, College Station, Texas, USA. Patancheru 502 324, Andhra Pradesh, India: International Crops Research Institute for the Semi-Arid Tropics.