A Review of Sorghum Grain Mold

G.A. Forbes[1], R. Bandyopadhyay[2], and G. Garcia[3]

Abstract

Terminology and definitions, symptoms, causal agents, importance, and control of fungal-related grain deterioration of sorghum are reviewed. The term grain mold (GM) has gained general acceptance as the most satisfactory descriptor of this condition.

Two concepts of fungal-related grain deterioration may be found in literature. In one, GM is a condition caused by parasitic and/or saprophytic interactions of numerous fungal spp and the plant at anytime between anthesis and harvest. In the other, only a few fungi infecting and colonizing spikelet tissues prior to grain maturity are involved. Fungi involved in postharvest deterioration (weathering) are not considered part of the GM complex. Numerous forms of GM damage have been described, but little work is reported on quantification of losses. A potential mycotoxin contamination in molded grain samples has been demonstrated.

New techniques, including serial dilutions and ergosterol concentration, have been useful in evaluating GM severity. Screening of more than 7000 accessions has identified more than 150 GM-resistant lines.

Introduction

The purpose of this review is to summarize research done on fungal-related deterioration of sorghum grain, frequently referred to as grain mold (GM). Grain mold, in its broadest sense, is certainly one of the major biotic constraints of sorghum for feed and food production. The historical development of GM and its perceived importance were reviewed by Williams and Rao (1981).

GM is usually the result of a complex of fungus-host interactions. Because of this complexity, it is difficult to synthesize a coherent review of the related literature. This review discusses GM from these viewpoints: (1) description, (2) importance, and (3) control.

This information is intended to complement the review of Williams and Rao (1981). Readers are advised to refer to that review for further discussion and references on various aspects of sorghum GM.

What is Grain Mold?

Terminology and definition

Williams and Rao (1981) reported that numerous and diverse terms have been used to describe fungal infection and colonization of sorghum spikelet tissues. Since publication of their review, consensus has developed among several major institutions for the exclusive use of the term "grain mold" (GM) to describe the condition resulting from fungal deterioration of sorghum grain (Canez and King 1981; Castor 1981; Frederiksen et al. 1982; Forbes 1986; ICRISAT 1986). However, other terms still appear in the recent literature: seedborne fungi (Bhale and

1. Plant Pathologist Centro International de la Papa, International Potato Center, PO Box 5969, Lima, Peru.
2. Plant Pathologist, Cereals Program, ICRISAT Center, Patancheru, Andhra Pradesh 502 324, India.
3. Plant Pathologist, CIBA-GEIGY, Arias 1851 (1429) Buenos Aires, Argentina,

Forbes, G.A., Bandyopadhyay, R., and Garcia, G. 1992. A review of sorghum grain mold. Pages 265-272 *in* Sorghum and millets diseases: a second world review, (de Milliano, W.A.J., Frederiksen, R.A., and Bengston, G.D., eds). Patancheru, A.P. 502 324, India: International Crops Research Institute for the Semi-Arid Tropics.(CP 738).

Khare 1982; El Shafie and Webster 1981; Hepperly et al. 1982; Kissim 1985); seed mycoflora (Kabore and Couture 1983; Khairnar and Gambhir 1985; Shree 1984), fungus associated with sorghum seed (Munghate and Faut 1982), head mold (Dayan 1980; Mathur and Naik 1981; Naik et al. 1981), seed-rotting fungi (Anahosur et al. 1984), and weathering (Ibrahim et al. 1985).

It would be difficult to demonstrate the intrinsic value of any one term over the others, but the advantage of researchers agreeing on the use of a single term seems obvious. The relatively significant level of acceptance for the term "GM" at present should be predictor of greater degree of uniformity in future publications as well. Some divergence in terminology may continue, however, as a reflection of an even-more fundamental level of dispute among researchers, that of definition.

Definitions of GM are only rarely given in explicit terms (Castor 1981; Williams and Rao 1981). Therefore the following discussion is based on implicit definitions inferred from GM-related literature and is subject to interpretive bias. Nonetheless, most definitions of GM found in recent literature appear to fit into one of two general concepts of fungal-related grain deterioration.

The first concept (A, Fig. 1) describes a condition resulting from fungal infection and colonization of grains occurring any time between anthesis and harvest. Here GM can be broadly defined as a fungal component of preharvest grain deterioration, involving numerous fungal species interacting in different ways with the plant (i.e., parasitically and/or saprophytically).

The second concept (B, Fig. 1) restricts the definition of GM to a condition caused by infection and colonization of spikelet tissues prior to grain maturity. In this limited definition, only a few fungi are thought to be involved. The multitude of field fungi that colonize grain after physiological maturity are not part of GM per se, but rather constitute a component of weathering, or general postharvest grain deterioration.

On a practical level, the two concepts are similar. For example, early and late infections in concept A can be seen as analogous to the GM and weathering of concept B. Fungal-related grain deterioration, whether occurring before or after grain maturity, can cause important losses. The objective of plant-improvement programs, therefore, is sorghum cultivars resistant to all aspects of fungal-related grain deteriorations.

These concepts differ mainly in the way that infections occurring before grain maturity are related to fungal colonization of the mature grain. In concept A, the difference is quantitative. The earlier the infection occurs, the greater the potential for damage and the fewer the fungal species involved.

In concept B, infections occurring prior to grain maturity could be considered qualitatively different from postmaturity colonization. The early infections involve relatively few fungi acting as true parasites on living tissue. Postmaturity grain colonization involves many genera of field fungi that colonize primarily nonliving tissue.

For practical purposes, the more generalized concept A sufficiently explains what one sees in the field. The qualitative distinction between GM and weathering (concept B), however, helps explain many aspects of fungal deterioration of sorghum grain, including resistance, symptom expression, infection process, and etiology.

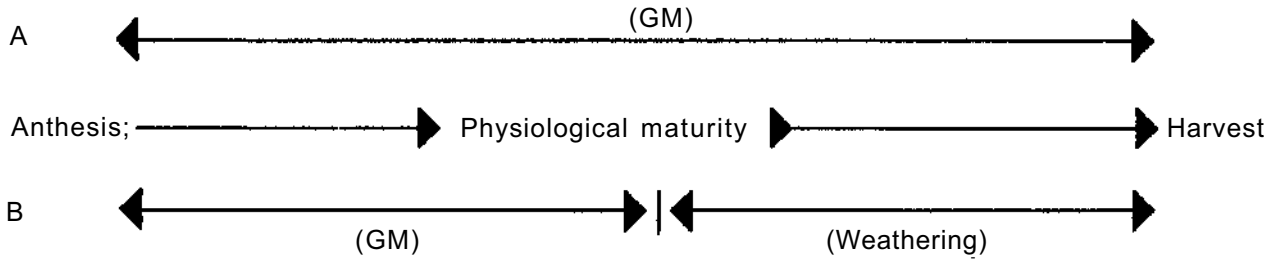

Figure 1. Two concepts A, and B, of fungal-related grain deterioration in sorghum.

For purposes of this review, GM refers to a condition resulting from all fungal associations with sorghum spikelet tissues occurring from anthesis to harvest. However, the qualitative distinction between early infection and postmaturity colonization will be employed when needed to facilitate discussion of certain aspects of the disease.

Symptoms

In discussing symptoms, one cannot help returning to the qualitative difference between early infections and postmaturity colonization. Symptoms of the two conditions can be very different.

Early infection by a GM pathogen probably occurs on the apical portions of spikelet tissues: glumes, lemma, palea, etc. Colonization then proceeds toward the base of the spikelet, either in the spikelet tissues or in voids between these tissues. A more-detailed discussion of this infection pattern will follow later.

Infection of the grain itself occurs at the base, near the pedicel, and can interfere with grain filling (Frederiksen et al. 1982) and/or cause a premature formation of the black layer (Castor 1981). Either condition causes a reduction in grain size, a symptom often associated with GM.

Visible superficial growth (the first signs of the fungus) occurs at the hilar end of the grain, and subsequently extends acropetally on the pericarp surface. Climatic conditions determine whether this growth will eventually spread to that part of the grain not covered by the glumes.

Infection induced by inoculation in greenhouse plants growing under low humidity produces very small grains without visible signs of the fungus on the exposed stylar end of the grain (Forbes 1986). That part of the grain hidden by the glumes is covered by a dense fungal mat. In contrast, the result of severe infection in the field usually is grains that are pink, white, or black (depending on the pathogen). This is because of coverage of the grain by fungal mycelium (Williams and Rao 1981).

Early infections also involve spikelet tissues other than the grain. One of the first visible symptoms following inoculation is pigmentation of the lemma, palea, glumes, and lodicules. This factor is highly cultivar dependent, and may be linked with mechanisms of resistance (discussed later).

Fungal colonization of sorghum grain produces a different set of symptoms. Colonization occurs primarily on the exposed part of the grain and may be limited to that area. Removal of the glumes will often show a sharp line of demarcation between protected and exposed areas (authors' observations). Postmaturity colonization is generally what produces the "moldy appearance" of grain maturing in humid environments. The color of the moldiness depends on the fungi involved.

Differences between early infections and postmaturity colonization can be difficult to substantiate in the field. Both conditions occur together, and late-season colonization can mask symptoms of infection occurring during grain development.

Causal fungi

It is thought that only a few fungi infect sorghum spikelet tissues during early stages of grain development. These are (in approximate order of importance) *Fusarium moniliforme* Sheld., *Curvularia lunata* (Wakker) Boedijn, F. *semitectum* Berk., & Rav., and *Phoma sorghum* (Sacc). F. *moniliforme* and C. *lunata* are of significance worldwide (Castor 1981; Frederiksen et al. 1982; Williams and Rao 1981; Bandyopadhyay 1986). The pathogenicity of these fungi has been established by inoculation of plants in the field and in the greenhouse.

If sorghum grains of harvest maturity are incubated on nonselective agar, the above fungi may be isolated in low frequencies relative to many other fungi. This is because the pericarp of sorghum routinely supports a rich and varied mycoflora that is not eradicated with conventional techniques of surface sterilization.

Williams and Rao (1981) list the species most frequently isolated in studies of mycoflora associated with sorghum grain. Subsequent studies list much the same spectra of fungal species. Recent papers in this area of research include El Shafie and Webster 1981, Granja and Zambolim 1984, Kabore and Couture 1983, Kissim 1985, Khairnar and Gambhir 1985, Novo and Menezes 1985, Pachkhede et al. 1985, and Shree 1984.

The importance of this mycoflora is not well known. These fungi are generally thought to be

restricted to the pericarp, but penetration into the endosperm can occur if the mature grain is exposed to high relative humidity or moisture for an extended period. Under severe climatic conditions, the endosperm can be completely colonized and partially degraded by field fungi (Glueck and Rooney 1980).

Fungal colonization of pericarp tissues of many cereal grains is common. Depending upon the timing and degree of penetration, these fungi are considered to be saprophytes or apathogenic weak parasites (Neergaard 1977).

Researchers in Australia have recently described F. *nygami* in association with sorghum grain (Burgess and Trimboli 1986). This new species resembles F. *moniliforme,* but produces chlamydospores. Its role in the etiology of grain mold is unknown.

Head Blight

Williams and Rao (1981) described head blight as "an invasion of tissues of the inflorescence by F. *moniliforme* Sheld. which results in the florets being killed to various degrees, up to complete destruction of the head/' Symptoms include discoloration and necrosis of the panicle, extending into inflorescence branches, and reddening of the pith in affected areas. Severe head blight results in open panicles with drooping rachis branches (Frederiksen et al. 1982).

Many researchers feel that head blight is distinct from GM (Williams and Rao 1981), but there appears to be no differentiation at the pathogen level (Frederiksen et al. 1982). Grain mold symptoms are routinely induced by inoculation with F. *moniliforme* Sheld., but head blight does not always occur. This would seem to indicate that certain causal or predisposing factors for head blight and for GM may differ.

Researchers in Argentina report that resistance to F. *moniliforme* may be tissue dependent. A. resistance reaction for head blight is not always indicative that a cuitivar will be resistant to F. *moniliforme in* spikelet tissues (Forbes et al., unpublished data).

The actual losses to head blight are not known, but its potential for economic loss has been demonstrated. In 1979, losses of between U.S. $3.2 million and U.S. $7.2 million were attributed to head blight in Texas (Castor and Frederiksen 1981). In general, head blight appears to be more important in Mexico and the humid southeastern USA than in Texas (Frederiksen et al. 1982) In southern France, panicle discoloration and necrosis is common in some genotypes (author's observations), but the etiology of this condition has not been studied.

Importance of Grain Mold

There is little doubt that GM in its broadest sense constitutes one of the most important biotic constraints to sorghum improvement and production. Sorghum workers worldwide, queried in 1977, indicated GM as one of the most important diseases of sorghum (Williams and Rao 1981). More recently, the real and potential importance of GM has been emphasized for Africa (Louvel and Arnoud 1984), the Americas (Frederiksen et al. 1982), and India (ICRISAT 1987).

Damages caused by grain mold

Williams and McDonald (1983) pointed out that in spite of general agreement that GM is important, there have been few attempts to quantify losses resulting from the disease. This problem does not arise from a lack of evidence that GM causes damage. Certain GM pathogens have repeatedly been associated with losses in seed mass (Castor and Frederiksen 1980; Hepperly et al. 1982; Singh and Makne 1985); grain density (Castor 1981; Ibrahim et al. 1984), and percentage germination (Castor 1981, Maiti et al. 1985). Other types of damage relating to storage quality, food and feed processing quality, and market value that may result from GM have been discussed by Williams and Rao (1981).

Mycotoxin Research

One consequence of GM that has received much attention in the last decade is contamination. There is growing concern for the deleterious nature of subacute doses on animals, Mycotoxins in feed slow the growth rate, predispose animals to other infections and are teratogenic and carcinogenic (Lacey 1985). Mycotoxin content of grains contaminated during preharvest stages usually increases when the grains are stored.

Since the 1980s, several instances of sorghum contamination by mycotoxins have been reported from USA, Australia, Africa, and India,

McMillian et al. (1981,1983a, 1983b, 1985) collected preharvest grain samples from several sorghum fields in Georgia and Mississippi from 1980 to 1982 and in 1984, and reported variable mycotoxin contamination with respect to the nature of mycotoxin, region, and species (e.g., maize shows more aflatoxin than sorghum). In 64 fields sampled on Georgia's coastal plain, 56% showed 1-90 ppb aflatoxin and 31% had 2-1468 ppb zearalenone. Grain harvested in Mississippi had neither of the mycotoxins.

Mold damage was severe in 1982, and mycotoxicosis was suspected in grain-fed swine. Of the 25 Georgian fields sampled, 84% showed aflatoxin [7-148 ppb (median 16 ppb)], and 8% contained zearalenone- [1515-10 420 ppb (median 6120 ppb)]. None of the 1984 samples showed aflatoxin. but one sample contained 80 ppb zearalenone. Shotwell et al. (1980) reported more than 1000 ppb zearalenone in 18% of the samples; 1000 ppb is the threshold value of physiological significance (Mirocha and Christensen 1974).

Australian reports of mycotoxin contamination of pre- and postharvest sorghum have been reviewed by Blaney (1985). He cites cases of suspected mycotoxicosis in four commercial swine operations — two due to aflatoxin, another due to aflatoxin and ochratoxin A, and the fourth due to zearalenone. Very high μg g^{-1} concentrations of these mycotoxins (aflatoxin <9.6, ochractoxin <0.1, and zearalenone <8) were detected in grain harvested and improperly stored.

In Nigeria, Salifu (1981) studied mold invasion and mycotoxin contamination in developing grains of short- and long-duration genotypes. The short-duration cultivars filled grains in unusually wet weather; no rains occurred from milk stage onward until harvest of the long-duration cultivars. All mature samples of the four short-duration cultivars had aflatoxin (10-80 μ*g* g^{-1}). Aflatoxin and zearalenone were first detected at the hard-dough stage. None of the long-duration genotypes in this study produced mycotoxin, but the author cites another instance of aflatoxin contamination (100 μg g^{-1}) in a long-duration cultivar grown in a wetter region in northern Nigeria.

Bhradraiah and Ramarao (1982) reported the occurrence of aflatoxin B_1 B_2, and G_1 from preharvest and mature grain samples of some widely grown cultivars in India. They reported more aflatoxin in the early-maturing hybrids CSH 5 than on medium- and long-duration cultivars; additional studies are needed. Aflatoxin B_1 content in their study was 25-180 ppb.

Grains are also contaminated with toxic metabolites produced by species of *Alternaria,* particularly *A. alternata.* Although alternariol and its monomethyl ether, altenuene and altertoxin I were found in moldy grain, no sign of toxicity was noticed in rats or chicks fed with these mycotoxins (Seitz 1984).

Tenuazonic acid is a potent mycotoxin, antineoplastic and protein-inhibiting, primarily produced by some species of *Alternaria,* but it has not been detected in sorghum grain. However, *Phoma sorghina,* a widely distributed GM fungus, is known to produce tenuazonic acid (Steyn and Rabie 1976) and may be responsible for onyalai, a human disorder prevalent in Africa. Onyalai is diagnosed by haemorrhagic vesicles in the mouth that appear when *Phoma*-infected grain is ingested.

Most of the mycotoxin research has been carried out in countries that use sorghum grain as feed. It is important to analyze the situation in countries where sorghum is consumed by human beings. Many questions remain concerning mycotoxin contamination. How prevalent is mycotoxin in food prepared from contaminated grain? What is the epidemiology of mycotoxin production in the field? Is it possible to breed for reduced mycotoxins, as has been done in groundnut (Mehan et al. 1986). Several toxigenic fusaria are known to occur on sorghum, so how widespread is the occurrence of trichothecins in field-grown sorghum grains? Intensification of research on mycotoxins as it relates to GM was advocated in the last review on the subject (Williams and Rao 1981).

Measuring grain mold

The above discussion illustrates the potential damage resulting from GM. To accurately assess the importance of GM, however, it becomes necessary to correlate the level of damage with the corresponding level of disease. Assessment of GM importance, therefore, is effective only to the degree of accuracy in measuring GM. Measurement of GM severity is also important for

other areas of research, including epidemiology and host resistance.

Visual appraisal has been the most common means of quantifying GM to date. Visual appraisal involves a complex of factors and can estimate severity (degree of colonization per grain indicated by signs or discoloration), incidence (proportion of grain affected), or damage (reduction is grain size), depending upon the method of assessment.

Visual appraisal, obviously the quickest and easiest method of disease assessment, is used for screening large numbers of samples (Bandyopadhyay and Mughogho 1988a). Advances in the search for resistance to grain mold achieved to date can be attributed to screening techniques based primarily on visual appraisal.

This form of estimation often has a surprisingly close association with other measures of severity. In several independent studies, a significant correlation has been established between visual appraisal and ergosterol concentration (discussed below) (Bandyopadhyay and Mughogho 1988b; Forbes 1986; ICRISAT 1986; Seitz et al. 1983).

Several factors can bias visual appraisal. For example, light-colored grains show more grain mold than dark-colored grains with equal severity. To avoid this problem, and be more accurate in general, workers at ICRISAT compare grain samples with light-grained and dark-grained standards of known severity levels (Bandyopadhyay and Mughogho 1988a). Comparing threshed grain is the most accurate method of visual assessment of GM (Frederiksen et al. 1982).

If visual assessments of GM severity are to be useful elsewhere, a common scale is required. Scales using well-defined units, such as percentage of grain surface affected (Forbes 1986; Bandyopadhyay and Mughogho 1988a) would seem to standardize comparison methods.

Because visual appraisal is a global evaluation of the condition of sorghum grain, it can provide only limited information about severity of GM per se. Extraneous factors, perhaps cultivar dependent, may mask the effects of GM. To get more accurate measurement of GM, researchers have used several techniques that have the commonality of estimating the quantity or incidence of the pathogen (fungal tissue or propagules) in a given amount of host tissue.

Most attempts to quantify GM pathogens in grain tissue have involved measures of incidence, and are based on the proportion of grains infected with certain pathogens (Hepperly et al. 1982; Gopinath and Shetty 1985; Granja and Zambolim 1984). Infection frequencies are measured by plating and incubating the entire kernel on blotting paper, or more often, agar.

Whole-grain plating can be biased by the competitive nature of the fungi making up the mycoflora (Neergaard 1977). Some scientists have attempted to compensate for this bias by using selective agar (Castor 1981) or chemical treatment of grain (Gopinath and Shetty 1985). The importance of competitive nature in a fungal sp is demonstrated by the fact that the incidence of F. *moniliforme* often increases when a Fusarium-specific agar is used (Castor 1981).

The relationship between GM severity and incidence is poorly understood. One can assume, however, that incidence would not reflect the important effects of infection timing on severity, since a grain infected late would count the same as one with early infection. Incidence-severity relationship studies for other diseases have proved to be complex, and have been impossible to determine for certain diseases (Seem 1984). It is doubtful that incidence studies will give much information about the severity of GM.

Some researchers have tried to quantify the degree of fungal colonization of sorghum grain. Forbes (1986) spread suspensions of ground seed tissues on a Fusarium-specific agar to quantify colonization by F. *moniliforme.* This technique, proposed as an indicator of disease severity, estimates the amount of viable fungal tissue (propagules g^{-1} of seed tissue).

Fungal biomass in a sample of sorghum grain is also estimated by measuring the concentration of ergosterol, a sterol produced by fungi but not by plants (Seitz et al. 1977). Ergosterol measurements are routine at ICRISAT (ICRISAT 1986). The procedure is sensitive and has the attractive attribute of estimating total (viable and nonviable) fungal biomass. Differences in ergosterol concentrations are often found among grain samples with similar degrees of superficial mold growth (Seitz et al. 1983).

Relationship of disease severity and damage

The potential for GM to damage grain is often demonstrated, but the relationship between severity and damage has seldom been quantified. New methods of assessing severity, such as measuring ergosterol, may make this an active area of future research. Even now, certain patterns are emerging from the few studies reported.

Severity appears to be more closely associated with viability than with yield. In a recent study, two measures of severity (ergosterol concentration, and propagules of *E moniliforme* g^{-1} seed tissue) were more highly correlated with percentage germination than with seed mass or grain density (Forbes 1986).

The sensitivity of percentage germination as an indicator of GM was also demonstrated by Castor and Frederiksen (1980). They suggested its use as a means of evaluating resistance. Automatic measuring of seed leachates and correlation with germination could become an efficient technique for studying the effects of GM severity on viability (Forbes 1986).

Control of Grain Mold

Avoidance

Avoidance of GM has often been described as one of the most important traditional control strategies (Castor 1981; Williams and Rao 1981). In areas where photosensitive cultivars have grown, GM is avoided because flowering and grain fill occur in the dry season. Avoidance is one of the most important control strategies still in commercial seed production. Most seed is produced with irrigation in arid regions to avoid GM and other problems.

Chemical control

Chemical control appears to provide some protection against GM. In another experiment, fungicide sprays at milk stage and 10 days later were shown to reduce GM infection (Naik et al. 1981).

Most studies involving fungicides and GM-related fungi, however, deal with the efficacy of seed dressings for improving seedling emergence and vigor (Patil et al. 1986; Munghate and Faut 1982; Vidhyasekaran 1983). Certain fungi have also been eradicated from sorghum grains with hot water treatment (Bhale and Khare 1982).

Resistance

In most cases, avoidance or chemical control in farmers' sowings is impractical. For this reason, major research efforts have focused on development of resistant cultivars. Improvement of screening techniques is a major effort in this research.

Screening at one of the major research institutes is currently done with natural inocula (Bandyopadhyay and Mughogho 1988a). High moisture levels are assured by sprinkling on rain-free days. Sprinklers are used as necessary throughout the period of grain development, as well as after physiological maturity. Materials to be screened are compared with grain samples with known levels of GM severity. More than 7000 accessions have been screened, and 156 lines selected as resistant (Bandyopadhyay and Mughogho 1988b).

Worldwide, many screening techniques are used. In Argentina a seed company has developed a method of separately screening for resistance to GM fungi occurring before grain maturity, those colonizing seed tissues after grain maturity, and those causing head blight. The different types of resistance are identified by screening at different stages of plant development and in the case of head blight, on the basis of symptom development in the peduncle (G. Garcia, unpublished data).

Screening with this technique revealed that resistance to early-season GM pathogens is not always associated with resistance to fungi causing damage late in the season. The independence of these two types of resistance was demonstrated earlier (Castor 1981).

Another approach to the identification of resistance is used by some researchers in northern Africa. Multivariate statistical techniques are used to determine cultivar reaction based on incidence of important GM pathogens, grain quality, germination and seedling viability, and visual assessment of moldiness (Louvel and Arnoud 1984).

Resistance mechanisms

In the last 10 years there has been a great deal of research directed toward the elucidation of resistance mechanisms. Much of this research has involved biochemical analyses of infected and noninfected tissues. Waniska reviews this work elsewhere in this volume.

Histological study has produced some insight of the process of early infection by GM pathogen on susceptible and resistant cultivars.

Several independent histological studies indicate similar patterns of initial infection and subsequent colonization of sorghum spikelet tissues (Castor 1981; Forbes 1986; Bandyopadhyay 1986). These studies were designed to determine the infection pattern following inoculation with F. *moniliforme* and with C. *lunata.* Resistant and susceptible cultivars inoculated with F. *moniliforme* were also compared (Castor 1981; Forbes 1986).

On a susceptible cultivar, initial infection by F. *moniliforme* occurs on the apical ends on the spikelet tissues: lemma, palea, glumes, filaments, and senescing styles. Fungal mycelium advances basipitally, either by colonizing spikelet tissues or by growing in voids between these tissues. Early colonization of glumes (3-4 days following inoculation) was found to be very heavy and caused little cellular disruption or pigmentation in the host (Forbes 1986).

Within 5 days of inoculation, mycelium can be seen in all parts of the spikelet, with the denser growth around the ovary base. Lodicules appear to serve as an important energy source, and are always surrounded by dense fungal growth, but extensive colonization of lodicule tissue per se has been questioned (Forbes 1986). It is apparently from this energy source, near the point of attachment to the pedicel, that infection of the ovary wall occurs.

In the next stages of invasion, a dense mycelial mat progresses acropetally, between the aleurone layer and the pericarp. Subsequent invasion of the endosperm, embryonic tissues, and pericarp originates from this peripheral mat Halloin (1983) has pointed out that peripheral growth on the inner layers of the true seed coat precedes embryonic colonization in many seed species.

When environmental conditions are appropriate, mycelial growth pushes through the pericarp, producing a white or pink fungal mass which can completely cover the grain.

Early invasion of a resistant spikelet appears to be as follows. As in the susceptible cultivar, mycelial growth can be seen in all parts of the spikelet at 5 days after inoculation. However, much of this growth is found in the voids between spikelet structures (Forbes 1986).

Pigmentation occurs rapidly in localized areas where host and fungal tissue were in close association. Fungal growth can involve cell disruption and cell wall depositions, inducing localized necrosis (Forbes 1986). Using another resistant cultivar, Castor (1981) likewise noticed heavy pigmentation associated with restricted fungal growth in and near the lodicules.

Castor proposed that localized pigmentation associated with resistance could be caused by luteolinidin that reddens sorghum stalks in response to pathogenic and nonpathogenic fungi. Pigmentation can also occur as a result of inoculation, suggesting that the mere presence of pigments does not confer resistance. However, pigmentation in susceptible cultivars appears to differ, from that in resistant cultivars, in coloration, intensity, location, and timing (Castor 1981; Forbes 1986).

After these early events, fungal invasion of the resistant spikelet is either arrested (Forbes 1986), or proceeds at a much slower pace than in a susceptible cultivar (Castor 1981), delaying infection of the ovary, and protecting it somewhat from damage.

Infection by C. *lunata* differs from that of F. *moniliforme* in the following way. During the initial period of spikelet invasion, C. *lunata* can infect the apical part of the ovary wall from the colonized lemma, palea, lodicules, filaments, pollen grain, and decaying style (Bandyopadhyay 1986). Within 5 to 10 days mycelium penetrates the pericarp and ramifies throughout the cross and tube cells. Colonization does not usually continue directly into the endosperm, but rather through the placental sac, which can also lead to invasion of the embryo.

Differences between the infection patterns of E *moniliforme* and C. *lunata* may partially explain the fact that resistance to the two occasionally differ (Castor 1981; Louvel and Arnoud 1984).

For both fungi, infection pattern and the degree of damage caused undoubtedly is affected by the maturity of the spikelet at the time of

infection. Either fungus can interfere with grain filling and cause premature formation of the black layer, reducing kernel size. If infection occurs early enough, invasion of the ovary base will cause the caryopsis to be aborted (Castor 1981).

In summary, colonization of a susceptible cultivar proceeds rapidly in all spikelet tissues without observable immediate host reaction. Colonization patterns are different for F *moniliforme* and C. *lunata.* In resistant cultivars examined in these studies, the presence of F *moniliforme* induced pigmentation and localized necrosis, involving cellular disruption and cell wall depositions. Infection of the embryo did not occur, or was retarded. Thus resistance mechanisms may involve spikelet tissues other than the ovary.

Epidemiology

Epidemiological studies may provide information that can be used to improve control strategies. Unfortunately, little is known about the epidemiology of GM. At the time of the review by Williams and Rao (1981), knowledge at the time was probably well-stated by their comment, "generally it seems that wet weather following flowering is necessary for GM development and the longer wet period, the greater the mold development." Even now, there have been but few studies on GM epidemiology, and little added to our knowledge of the subject.

In what appears to be one of the few epidemiological studies of GM, ICRISAT workers successfully monitored diurnal and seasonal trends of aerial spore densities of *Curvularia lunata* throughout the growing season (ICRISAT 1986). This type of study has been done for F. *moniliforme* spores in and above the canopy of maize (Ooka and Kommedahl 1977), indicating that techniques exist which could easily be applied to sorghum.

Some epidemiological insight may be gained indirectly from a study done in Texas (Forbes 1986). A conidial suspension of F. *moniliforme* was applied to panicles at anthesis, either by spraying or submerging. Plants were then incubated for 24 h and later moved, with nonincubated controls, to a greenhouse where conditions were not favorable for further infections. Severe GM developed on all incubated plants, but not on the noninoculated plants, indicating that moisture is needed for initial infection but not for disease progression at the grain level. The severity of GM within a field is probably greatly influenced by the effect of moisture on repeated infections through time. Little is known, however, about the apparently critical relationship between moisture, inoculum availability, and host maturity.

Future Needs

Etiology and the role of host maturity

There are few published accounts of controlled inoculation studies with suspected GM pathogens. Institutions with appropriate facilities (growth chambers, inoculation chambers, and greenhouses with controlled environments) could do closely monitored and replicated studies using known and suspected grain mold pathogens at different stages of plant development. Such research might clear up a major area of confusion in GM-related literature—which organisms are capable of infecting which tissues at which stages of host development. Immunofluorescence techniques would add sensitivity and selectivity to histological methods.

Resistance mechanisms

There is a need to continue studies on the resistance mechanisms of sorghum without a testa or with low tannin content. Preliminary studies on two cultivars have indicated potential mechanisms of resistance, including localized necrotic reaction and inhibition of fungal growth associated with pigmentation. Confirmation of these characteristics would be most useful in other cultivars (Bechtel et al. 1985) Determination of the nature of the physiological changes associated with resistance likewise would be a valuable contribution.

Epidemiology

As mentioned, few quantitative studies on epidemiological aspects of grain mold are in the record. Research designed to determine the importance of environmental variables and inoc-

ulum dynamics in disease development should be of first priority. Knowledge of disease spread in time and space may facilitate many other areas of GM research.

References

Anahosur, K.H., Kulkarni, K.A,, and Patil, S.H. 1984. Role of fungicide-cum-insecticide seed treatment in the control of seed rotting fungi and shootfly of sorghum. Mysore Journal of Agricultural Sciences 16:418-421.

Bandyopadhyay, R. 1986. Grain mold. Pages 36-38 *in* Compendium of sorghum diseases. (Frederiksen, R.A., ed.) St. Paul, MN, USA: The American Phytopathological Society.

Bandyopadhyay, R., and Mughogho, L.K. 1988a. Evaluation of field screening techniques for resistance to sorghum grain molds. Plant Disease 72:500-503.

Bandyopadhyay, R., and Mughogho, L.K. 1988b. Sources of resistance to sorghum grain molds. Plant Disease 72:504-508.

Bechtel, D.B. Kaleikau, L.A. Gains, R.L., and Seitz, L.M. 1985. The effects of *Fusarium graminicola* infection on wheat kernels. Cereal Chemistry 62:191-192.

Bhadraiah, B., and Ramarao, P. 1982. Isolates of *Aspergillus flavus* from sorghum seeds and aflatoxin production. Current Science 51: 1116-1117.

Bhale, M.S., and Khare, M.N. 1982. Seed-borne fungi of sorghum in Madhya Pradesh and their significance. Indian Phytopathology 35:676-678.

Blaney, B.J. 1985. Mycotoxins in crops grown in different climatic regions of Queensland. Pages 97-108 *in* Trichotechenes and other mycotoxins: proceedings of the International Mycotoxin Symposium, Aug 1984, Sydney, Australia (Lacey, J., ed.), Chichester, UK: John Willey & Sons.

Burgess, L.W., and Trimboli, D. 1986. Characterization and distribution of *Fusarium nygamai* sp nov. Mycologia 78:223-229.

Canez, V.M. Jr., and King, S.B. 1981. A comparison of methods for assessing grain mold in sorghum. Phytopathology 71:865. (Abstract).

Castor, L.L. 1981. Grain mold histopathology, damage assessment and resistance screening within *Sorghum bicolor* (L.) Moench lines. Ph.D. thesis, Texas A&M University, College Station, TX, USA. 192 pp.

Castor, L.L., and Frederiksen, R.A. 1980. *Fusarium* and *Curvularia* grain mold in Texas. Pages 93-192 *in* Sorghum diseases, a world review: proceedings of the international workshop on sorghum diseases, 11-15 Dec 1978, ICRISAT, Hyderabad, India. Patancheru, Andhra Pradesh 502 324 India: International Crops Research Institute for the Semi-Arid Tropics.

Castor, L.L., and Frederiksen, R.A. 1981. *Fusarium* head blight occurrence and effects on sorghum yield and grain characteristics in Texas. Plant Disease 64:1017-1019.

Dayan, M.P. 1980. Studies on sorghum head mold organisms in the Philippines. D.Sc. thesis, University of the Philippines, Los Banos, Luzon, Philippine Island.

El Shafie, A.E., and Webster, J. 1981. A survey of seed-borne fungi of *Sorghum bicolor* from the Sudan. Transactions of the British Mycological Society 77:339-342.

Forbes, G.A. 1986. Characterization of grain mold resistance in sorghum *(Sorghum bicolor* L. Moench). Ph.D. thesis, Texas A&M University, College Station, TX, USA. 75 pp.

Frederiksen, R.A., Castor, L.L., and Rosenow, D.T. 1982. Grain mold, small seed and head blight: the *Fusarium* connection in sorghum. Proceedings of the Thirty-seventh Annual Corn and Sorghum Industry Research Conference 37:26-36.

Glueck, J.A., and Rooney, L.W. 1980. Chemistry and structure of grain in relation to mold resistance. Pages 119-140 *in* Sorghum diseases, a world review: proceedings of the international workshop on sorghum diseases, 11-15 Dec 1978, ICRISAT, Hyderabad, India. Patancheru, Andhra

Pradesh 502 324, India: International Crops Research Institute for the Semi-Arid Tropics.

Gopinath, A., and Shetty, H.S. 1985. Occurrence and location of *Fusarium* species in Indian sorghum seed. Seed Science and Technology 13: 521-528.

Granja, E.M., and Zambolim, L. 1984. The incidence of phytopathogenic fungi on sorghum seeds. Revista Ceres 31:115-119.

Halloin, J.M. 1983. Deterioration resistance mechanisms in seeds. Phytopathology 73:335-339.

Hepperly, P.R., Feliciano, G, and Sotomayor, A. 1982. Chemical control of seedborne fungi of sorghum and their association with seed quality and germination in Puerto Rico. Plant Disease 66:902-904.

ICRISAT (International Crops Research Institute for the Semi-Arid Tropics). 1986. Annual Report 1985. Patancheru, Andhra Pradesh 502 324, India: ICRISAT.

ICRISAT (International Crops Research Institute for the Semi-Arid Tropics). 1987. Annual Report 1986. Patancheru, Andhra Pradesh 502 324, India: ICRISAT.

Ibrahim, O.E., Nyquist, W.E., and Axtell, J.D. 1985. Quantitative inheritance and correlations of agronomic and grain quality traits of sorghum. Crop Science 25:649-654.

Kabore, K.B., and Couture, L. 1983. Mycoflore des semences du sorgo cultive en Haute-Volta. Naturaliste Canadien 110:453-457.

Khairnar, D.N., and Gambhir, S.P. 1985. Studies on seed mycoflora of some jowar varieties from Nalik area (Maharashtra). Indian Botanical Reporter 4:68-70.

Kissim, M.Y. 1985. Seed-borne fungi of sorghum in Saudi Arabia. Pakistan Journal of Botany 17:263-266.

Lacey, J. (ed.) 1985. Trichothecenes and other mycotoxins: proceedings of the International **Mycotoxin Symposium, Aug 1984,** Sydney, Australia. Chichester, UK: John Wiley & Sons.

Louvel, D., and Amoud, M. 1984. Moisissures des grains chez le sorgho: definition d'une resistance. Etat des recherches sur le sorgho au Senegal, Actes de la reunion de travail du 22 Avril 1983.

Maiti, R.K., Raju, P.S., and Bidinger, F.R. 1985. Studies on germinability and some aspects of preharvest physiology of sorghum grain. Seed Science and Technology 13:27-35.

Mathur, K., and Naik, S.M. 1981. Fungi associated with moldy sorghum grain and reaction of sorghum cultivars to head molds. Bulletin of Grain Technology 19:143-146.

McMillian, WAV., Widstrom, N.W. and Wilson, D.M. 1981. Aflatoxin contamination in 1984 preharvest sorghum. Sorghum Newsletter 24:123.

McMillian, W.W., Wilson, D.M., and Widstrom, N.W. 1985. Aflatoxin contamination in 1984 preharvest sorghum. Sorghum Newsletter 28:25.

McMillian, W.W., Wilson, D.M., Mirocha, GJ., and Widstrom, N.W. 1983a. Mycotoxin contamination in grain sorghum from fields in Georgia and Mississippi. Cereal Chemistry 60:226-227.

McMillian, W.W., Wilson, D.M., and Widstrom, N.W. 1983b. Aflatoxin and zearalenone contamination in grain sorghum samples collected in Georgia fields in 1982. Sorghum Newsletter 26:122.

Mehan, V.K., McDonald, D., and Ramakrishna, N. 1986. Varietal resistance in peanut to aflatoxin production. Peanut Science 13:7-10.

Mirocha, C.J., and Christensen, C.M. 1974. Oesterogenic mycotoxins synthesized by *Fusarium.* Pages 129-148 *in* Mycotoxins. Amsterdam, The Netherlands: Elsevier Science Publishers.

Munghate, A.G., and Faut, J.G. 1982. Efficacy of nine different fungicides against fungi frequently associated with sorghum seed. Pesticides 16:16-18.

Naik, S.M., Singh, S.D., and Mathur, K. 1981. Efficacy of fungicides in controlling head molds of sorghum. Pesticides 15:25-39.

Neergaard, P. 1977. Seed Pathology. London, UK: McMillian Press Ltd. 1187 pp.

Novo, R.J., and Menezes, M. 1984. Effectiveness of fungicides in seed treatment of grain sorghum, *Sorghum bicolor.* Fitopatologia Brasileira 9:543-549.

Novo, R.J., and Menezes, M. 1985. Fungi isolated from sorghum *(Sorghum bicolor)* seeds from the municipalities of Caruaru and Serra Talhada, Pe. Fitopatologia Brasileira 10:57-64.

Ooka, J. J., and Kommedahl, T. 1977. Wind and rain dispersal of *Fusarium moniliforme* in corn fields. Phytopathology 67:1023-1026.

Pachkhede, A.U., Vyawajare, S.V., and Shreemali, J.L. 1985. Two new species of *Phoma* from India. Current Science (India) 54:485-486.

Patil, M.R., Sakpal, P.N., and Patil, V.N. 1986. Efficacy of different fungicides against *Fusarium moniliforme* and *Curvularia lunata* (important fungi) associated with sorghum seed. Pesticides 20:27-30.

Salifu, A. 1981. Mycotoxins in short season sorghums in northern Nigeria. Samaru Journal of Agricultural Research 1:83-88.

Seem, R.C 1984. Disease incidence and severity relationships. Annual Review of Phytopathology 22:133-150.

Seitz, L.M. 1984. Alternaria metabolites. Pages 443-455 *in* Mycotoxins—production, isolation, separation and purification (Betina, V., ed.). Amsterdam, The Netherlands: Elsevier Science Publishers.

Seitz, L.M., Mohr, H.E., Burroughs, R., and Sauer, D.B. 1977. Ergosterol as an indicator of fungal invasion in grains. Cereal Chemistry 54:1207-1217.

Seitz, L.M., Mohr, H.E., Burroughs, R., and Glueck, J.A. 1983. Preharvest fungal invasion of sorghum grain. Cereal Chemistry. 60:127-130.

Shree, M.P. 1984. Seed mycoflora of sorghum varieties with particular references to *Exserohilum turcicum* (Pass) Leo at Sug. and related genera, causing seed rot, seedling and leaf blight diseases. Seed Research 12:24-39.

Shotwell, O.L., Bennett, G.A., Goulden, M.L., Plattner, R.D., and Hesseitine, C.W. 1980. Survey for zearlenone, aflatoxin and ochratoxin in U.S. grain sorghum from 1975 and 1976 crops. Journal of the Association of Official Analytical Chemists 63:922-926.

Singh, A.R., and Makne, V.G. 1985. Correlation studies on seed viability and seedling vigor in relation to seed size in sorghum *(Sorghum bicolor)* Seed Science and Technology 13:139-142.

Steyn, P.S., and Rabie, C.J. 1976. Characterization of magnesium and calcium tenuazonate from *Phoma sorghina.* Phytochemistry 15:1977-1979.

Vidhyasekaran, P. 1983. Control of *Fusarium moniliforme* infection in sorghum seed. Seed Science and Technology 11:435-439.

Williams, R.J., and McDonald, D. 1983. Grain molds in the tropics: problems and importance. Annual Review of Phytopathology 21:153-178.

Williams, R.J., and Rao, K.N. 1981. A review of sorghum grain molds. Tropical Pest Management 27:200-211.