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Mango Anthracnose Disease: Present Status and Future Research Priorities

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OCCURRENCE AND IMPORTANCE

Anthracnose is presently recognized as the most important field and post-harvest disease of mango worldwide (Ploetz and Prakash, 1997). It is the major disease limiting fruit production in all countries where mangoes are grown, especially where high humidity prevails during the cropping season. The post-harvest phase is the most damaging and economically significant phase of the disease worldwide. It directly affects the marketable fruit rendering it worthless. This phase is directly linked to the field phase where initial infection usually starts on young twigs and leaves and spreads to the flowers, causing blossom blight and destroying the inflorescences and even preventing fruit set.

CAUSAL AGENTS AND SYMPTOMS

Mango anthracnose is caused by *Glomerella cingulata* (Stoneman) Spauld. and H. Schrenk (anamorph: *Colletotrichum gloeosporoides* (Penz.) Penz. var. *minor* J.H. Simmonds (Fitzel and Peak, 1984) and *C. acutatum* J.H. Simmonds (Freeman *et al*., 1998). The pathogen also causes blossom blight, leaf blight and in some severe cases, tree dieback (Ploetz, 1994; Ploetz *et al*., 1996). In Australia and India, *C. acutatum* (teleomorph: *Glomerella acutata*) has been reported to also play a minor role in causing the disease (Fitzell, 1979; Prakash, 1990).

The anthracnose pathogen invades inflorescences, fruit, leaves and stems of mango. Leaf anthracnose appears as irregular-shaped black necrotic spots on both surfaces of the mango leaf. Lesions often coalesce to form large necrotic areas, frequently along the leaf margins. Severely affected leaves usually curl. Lesions develop primarily on young tissue and conidia are formed and can be observed in lesions of all ages. In older leaves, lesions do not develop, but latent infections are formed and the fungus remains dormant until the tissue senesces. Growth then resumes and fruiting structures are produced in the necrotic tissue. Under favorable conditions, conidia are dispersed and invade young twigs causing twig dieback in some cases (Ploetz *et al*., 1998).

Panicle anthracnose or blossom blight can affect both the inflorescence stalk and the individual flowers. Infection reduces fruit set and production considerably, since affected flowers are killed. In the stalk, elongated dark gray to black lesions appear. Blighted flowers are dry and their color varies from brown to black. Small emerging fruits can be infected and aborted. Larger fruits aborted because of other physiological causes are usually mummified and the mummies are invaded saprophytically by *C. gloeosporoides* on which they sporulate profusely.

Postharvest anthracnose appears as rounded brown to black lesions with an indefinite border on the fruit surface. Infection in larger fruit does not normally develop into lesions. After initial establishment in the fruit, the fungus remains latent or dormant until the fruit begins to ripen. Dark depressed circular lesions than develop on the ripening fruit and increase rapidly in size. They may even cover the entire fruit surface in extreme severe cases. Lesions larger than 2 cm are fairly common on severely infected fruit. Lesions of different sizes can coalesce and cover extensive areas of the fruit, typically in a tear-stain pattern, developing from the basal toward the distal end of the fruit (Arauz, 2000). Lesions are usually restricted to the peel, but in severe cases the fungus can penetrate even the fruit pulp. In advanced stages of infection, the fungus produces acervuli and abundant orange to salmon pink masses of conidia appear on the lesions.

Visual scales have traditionally been used to evaluate the severity of the disease on infected fruit (Koomen and Jeffries, 1993). Because this kind of evaluation is qualitative and tends to be subjective, Corkidi *et al*. (2006) recently developed a more accurate image-analysis method that could be used to quantitatively assess mango fruit with lesions from anthracnose infection. They were able to demonstrate that this method is more accurate than the traditional and widely used technique developed by Brodrick (1978) which is based on the percentages of the area affected on the fruit by the fungal pathogen.

DISEASE EPIDEMIOLOGY

Disease cycle: Fitzell and Peak (1984) established that conidia were the most important type of inoculum in mango orchards in North New South Wales (NSW),

Australia. They were produced on lesions on leaves, twigs, panicles and mummified fruit. Infected, new leaf flushes were viewed as the most significant source of inoculum. Even though ascospore production in dry leaves on the ground has been reported (Ann, 1995), the role of the teleomorph stage in the disease cycle is still unclear. Since conidia are formed abundantly in the mango canopy, this is considered to be the primary source of inoculum. Conidia can be rain-splashed to other leaves or flowers to cause secondary infections; thus making the disease polycyclic in these organs. Developing fruit can be infected and some aggressive isolates can cause pre-harvest fruit losses (Gantotti and Davis, 1993).

In the case of postharvest anthracnose, developing fruit are infected in the field, but infections remain quiescent until the onset of repining, which occurs after harvest. Once the climacteric period of the fruit starts, lesions begin to develop. There is usually no fruit-to-fruit infection, hence postharvest anthracnose is considered a moncyclic disease (Aruz, 2000).

A study of the genetic diversity in the population of the mango anthracnose pathogen in Florida showed that there might be exceptions to this general pattern of the disease cycle (Gentotti and Davis, 1993). Molecular analysis on isolates of *C. gloeosporoides* from different mango tissues revealed variation in patterns of pectic-degrading enzymes. They concluded from the study that the fungus on mango was genetically diverse, suggesting variation in ability to cause disease in different tissue by different isolates. Other related work also indicated that the mango population of *C. gloeosporoides* may comprise a pathogenically and genetically distinct population of *C. gloeosporoides* (Hayden *et al*., 1994). Mango fruit can also be infected with conidia from isolates of *Colletotritichum* sp. from other host plants such as avocado, papaya and citrus (Freeman and Shabi, 1996). The epidemiological significance of these potential inoculum sources, on the disease cycle, still need to be assessed. Generally, genetic and geographical data seem to suggest that the mango population of *C. gloeoesporoides* was disseminated throughout the world from a single source as an endophyte. An increased understanding of the origins and diversity of *C. gloeosporoides* on mango would have relevance to future research on host and chemical control strategies across regions and locations.

Termination of fungal quiescence on climacteric fruits appears to be related to the reduction of antifungal compounds or the production of ethylene by the repining fruit (Prusky, 1996; Flaishman and Kolattukudy, 1994). As mango fruit ripens, there is a reduction in the concentration of phenolic compounds, which are active against *C. gloeoesporoides in vitro*. Similar systems have been found with avocado anthracnose (Prusky and Keen, 1993). The involvement of ethylene in the termination of quiescence strongly suggests that *Colletotrichum* sp. must have coevolved to develop a mechanism to use the host's ripening hormones as a signal to reactivate the infection process. This mechanism may prevent contact of the pathogen with host tissues that have high levels of antifungal compounds. Resistance to the pathogen in mango fruit tissue is advantageous to the host during seed development, but not afterwards because the ripe pulp needs to be destroyed by invading saprophytes or weak pathogens to help liberate the fruit to germinate in rich organic substrate (Flaishman and Kolattukudy, 1994). Therefore, there is evolutionary value in allocating chemical defenses to the immature fruit but not to the ripe fruit, as is apparently the case in mango (Prusky and Keen, 1993).

Role of temperature and moisture in infection: Most of the research on the effects of weather on the anthracnose pathogen on mangoes has been done in NSW, Australia and the Philippines (Fitzell *et al*., 1984; Dodd *et al*., 1991). Empirical models have been developed to predict the occurrence of infection in mango orchards. From the research it has been established that *C. gloeosporoides* requires free water or relative humidity above 95% for conidial germination and appressorium formation. Conidia can, however, survive for up to 2 weeks at humidities as low as 62% and then germinate if exposed to a high humidity. Optimum production of conidia occurs between 25 and 30°C when free moisture is available. In general, infection is favored at temperatures ranging from 20 to 30°C. Within this range, there is considerable variation in the optimal temperature requirements for germination and appressorium formation among isolates of *C. gloeosporoides* from different locations.

Temperature and moisture requirements for infection have also been used to build forecasting systems for mango anthracnose, a vital component for the disease management.

DISEASE MANAGEMENT

Control of postharvest anthracnose can be achieved from field management, after harvest treatments, or preferably, a combination of both. Management strategies must be efficient and cost-effective, as well as safe to consumers, agricultural workers and the environment.

FIELD CONTROL

A number of options are available for the management of mango anthracnose under field conditions. This mainly involves cultivar selection, cultural or agronomic practices and the use of fungicide sprays.

Resistant varieties: Resistance has not been used as a consistent means of control of mango anthracnose. This is partly because of the variable reactions in cultivars to the disease from one location to another. Although most or all commercial mango cultivars are susceptible to mango anthracnose, some are less susceptible than others. The cultivar Keitt, for example, is less susceptible than Kent, while Kessington Pride, the widely grown cultivar in Australia, is listed as moderately resistant to the disease. At present, none of the cultivars under production are significantly resistant to be produced without using some fungicide spray protection in humid environments (Dodd *et al*., 1997).

Cultural control: Since the development of the disease is dependent on wetness or high relative humidity, orchards should ideally be established in areas with a well-defined dry season to allow for fruit development in conditions unfavorable for disease development. In most of the tropics, mango flowering usually occurs during dry seasons. Within a given area, however, mango trees can flower at any moment during the dry season, depending on factors such as tree maturity, temperature and nutritional status of the trees. If flowering occurs early, before the dry season is well established, flowers and young fruit can be infected. Anthracnose development on fruits is worst when trees flower in the dry season and the last part of fruit development takes place in the rainy season. High incidences of anthracnose are thus not uncommon in fruit that develop during the rainy season.

In seasonal tropical areas, a possible disease avoidance strategy is to manipulate flowering such that fruit develop during the least rainy time of the year. The incidence and severity of mango anthracnose can be very low in fruit developed completely in the dry season, even without the application of any other control measure. This strategy unfortunately is not usually applicable to subtropical mango-growing areas, where low temperatures provide the stimulus for flowering, rather than water deficit and flowering is not necessarily followed by dry periods. Considering the above, considerable effort has been made to understand and manage mango flowering and more still needs to be done in this area as an additional strategy for anthracnose disease control. Mango flowering can be advanced by several weeks in tropical environments, with potassium nitrate sprays on mature foliage (Nunez-Elisea, 1985).

Field sanitation involving the collection and incineration of fallen fruit and tree trash has been recommended for anthracnose control (Lim and Khoo, 1985). Sanitation of the whole tree itself can be difficult to practice. Elimination of dry panicles and mummified fruit is time-consuming and the possible benefits have not yet been fully evaluated. Fruit wrapping has been investigated as a disease control option but this has shown other side physiological effects, which can only be ignored depending on the seriousness of the disease situation (Hoffman *et al*., 1997).

Chemical control: Much of the attention and efforts on anthracnose control has concentrated mostly on the use of fungicides. Fungicide application focuses on reducing damage to inflorescences and fruit. This practice started a long time ago but unfortunately, only few fungicides are presently approved for use on mango in importing countries. The choice of fungicides used therefore, depends on the intended destination of the exported fruit.

Looking back at history, Ruehl and Ledin (1960) first showed that zineb, maneb or captan applied at weekly intervals during flowering followed by monthly applications during fruit development gave adequate disease control. McMillian (1972) showed that it may be phytotoxic to flowers. During fruit development, mancozeb was equal to copper at Bowen in Queensland (Grattidge, 1978). Benomyl has been shown to be superior to other protectant fungicides during both flowering and fruit development (McMillan, 1973). The surfactant Nu Film 17 was shown to enhance the level of control with copper and Benomyl (McMillan, 1972), but others could not improve control with the surfactants Nu Film 17, Agral 60 or summer oil. It is interesting to note that some of these fungicides are still very much alive and playing an active role in anthracnose disease control in many mango-growing regions of the world even today. When some of these fungicides are used, up to 25 seasonal sprays of protectant and systemic fungicides can be used to keep the disease under control in some extreme situations, where fruit develops under disease-favorable conditions.

In general, dithiocarbamate fungicides are highly effective for anthracnose control. Mancozeb, however, cannot be used for USA-bound fruit because of ethylene produced as a by-product of its degradation, even though it is still an effective fungicide for anthracnose control. Ferbam is presently the alternative for use in the USA market. Copper fungicides are also recommended, but

their efficacy is lower than that of the dithiocarbamates under high disease pressure and phytotoxicity on mango flowers still remains an issue of concern. Fungicides with after-infection activity for mango anthracnose include benzimidazoles and the imidazole, prochloraz. Benomyl has been used in calendar-base spray schedules, usually in a mix with protectant fungicides to delay the build up of resistance in the pathogen population. It has also been applied as an eradicant spray following infection periods. Prochloraz has been used as a protectant or as an eradicant spray (Estrada *et al*., 1996).

Resistance of *C. gloeosporoides* to benzimidazole fungicides has been reported in many cases. Resistance to prochloraz has not yet been reported. Extensive screening of a large population of Taiwan mango isolates of the pathogen exposed to various field application frequencies and concentrations could not establish resistance to the fungicide even thirteen years after its registration and extensive use in Taiwan (Kuo, 2001). It is still widely expected that it is a matter of time for resistance to occur since it has already occurred with other fungal pathogens and there is considerable variation to it among isolates of *C. gloeosporoides* from mango.

Disease forecasting systems: Two predictive models based on temperature and moisture requirements for infection of mango by *C. gloeosporoides* have been developed. These models have been the basis for two forecasting systems for mango anthracnose, which have been used in the field to time fungicide applications.

In NSW in Australia, Fitzell *et al*. (1984) studied the requirements for temperature and wetness duration for production of dark appressoria from conidia applied to detached young mango leaves under laboratory conditions. They developed an in-field microprocessor-based data recording forecasting system called a Mango Anthracnose Estimator (MAE) and used it to time applications of prochloraz during the mango flowering period in NSW (Peak *et al*., 1988). Use of the model resulted in a reduction of four to eight fungicide sprays per season to control flower anthracnose as compared with weekly spraying, which was the commercial recommendation. In later trials, low incidences of blossom blight on mango trees did not allow for an ideal comparison and confirmation of the use of the MAE model for the timing of sprays as compared with the protective spray schedule (Peterson *et al*., 1991).

A similar system was developed in the Philippines based on the studies by Dodd *et al*. (1991). It differs from the Australian system, in that it was developed using combined data from fruit and leaf inoculations and it includes relative humidity in addition to wetness and temperature. It was also a laboratory study but was tested under field conditions for the control of postharvest anthracnose in the Philippines. Benomyl, prochloraz or triforine was applied following a predicted infection period with a threshold of 40% of conidia forming dark appressoria. One timed application of benomyl or prochloraz was as effective as six calendar-based sprays of either fungicide.

When a comparison of the Australian and Philippine models is made, there are important differences between the models in level of infection predicted from a given combination of temperature and wetness duration. At 25°C, for example, 10% of conidia would form dark appressoria in about 1 h according to the Australian model and in about 16 h according to the Philippine model. Such a discrepancy indicates that weather-based forecasting systems for mango anthracnose should not be extrapolated from one region to another and the infection criteria should be elucidated locally. The difference between the two models may also reflect differences in experimental methods, especially in the plant tissues used to develop them. Tissue specificity can occur in different isolates of *C. gloeosporoides* and this factor should be considered in the development and application of mango anthracnose forecasting system.

The forecasting systems can be simplified in certain tropical areas, based on long term observations of the weather pattern. It could be assumed that if a wetness period occurs, it is likely that an infection will take place as has been shown for Costa Rican conditions (Arauz, 2000). Therefore it should be possible to time fungicide sprays based on the mere occurrence of a wetness or high humidity period. Working under such an assumption, a considerable reduction in the number of fungicide sprays can be achieved, as was the case in Costa Rica, compared with calendar-based program for postharvest anthracnose control.

Forecasting systems are useful for diseases that are important but sporadic. An anthracnose forecast in the seasonal tropics would be most useful in dry seasons, when sporadic rain is possible, or during transitional periods between dry and wet seasons. Once the rainy season is established, calendar-based fungicide applications are the best strategy for chemical control, since conditions are usually favorable for disease development. The most advisable strategy would depend on the time of flowering of a given orchard in a given region of production.

The choice of fungicides to be used for after-infection sprays in the field should be determined by the fungicides that will be used in postharvest treatments.

Field and postharvest applications of the same fungicide, or fungicides with a risk of cross-resistance should be avoided as much as possible. Benomyl and prochloraz give good after-infection control of mango anthracnose, but prochloraz is the only one presently registered for postharvest use in several places. Therefore, its use in the field should be restricted to situations where no other options are available.

POSTHARVEST CONTROL

Traditionally, postharvest control of mango anthracnose has been aimed at reducing the level of quiescent infections on the fruit. Even though fruit-to-fruit spread of anthracnose after harvest is unlikely, postharvest control of latent infections is often needed and used, especially if fruits are to be stored or shipped to other places (Dodd *et al*., 1997). Attempts have been on to control anthracnose in ripe fruit by postharvest treatments for many years. The effectiveness of hot water dip in anthracnose control was demonstrated a long time ago. More recent research (Prusky and Keen, 1993) is suggesting the possibility of prolonging the period of fruit resistance and delaying the onset of anthracnose development until fruit ripens. The elimination of quiescent infection is achieved commercially by thermal and chemical treatments, or a combination of both. Dips of fruit in hot water alone are not quite efficient. Temperatures between 50 and 55°C for 3 to 15 min have been recommended, with the higher temperature corresponding to the lower exposure times. Cultivars also vary in their tolerance to hot water and temperature treatments should never exceed 55°C for 5 min Hot water treatments by themselves leave no chemical residue on the fruit and could be a good anthracnose control option for organic mango or for mango targeted for places where no fungicides are currently labeled for postharvest use. Temperature and time control are critical, because fruit can be easily damaged by over-exposure to heat.

Several different fungicides have been applied after harvest to control anthracnose. Benomyl was used in the past in combination with hot water treatments but is no longer permitted in most places. Thiabendazole is also effective. Prochloraz can be used but efficacy varies depending on disease pressure. Imazalil, has also shown variable efficacy. One advantage of benzimidazole fungicides such as benomyl or thiabendazole is that they are also effective for the control of stem-end rot caused by *Lasiodiplodia theobromae* (pat.) on mango, which is widely considered the second most important postharvest disease of mango in tropical areas. Prochloraz is not affective against *L. theobromae* on mango.

The combination of hot water and fungicides is the most effective commercial postharvest treatment for the control of mango anthracnose. Both rate of fungicide and duration of exposure to hot water are lower and efficacy is higher than with either treatment considered separately. Even at high levels of infection, high efficacies can be achieved. Combined with fungicide dips, an acceptable range of 52 to 53°C is quite efficient. Hot water and fungicides can be applied sequentially or together. Irradiation of fruit to control anthracnose has been attempted with mixed results. A short-wave infrared radiation treatment developed in South Africa for anthracnose control is as effective as hot water treatment and is much faster and less expensive (Saaiman, 1996).

Postharvest practices such as cold storage and controlled atmosphere maintain resistance to decay by delaying the ripening process. There are some limitations to the potential benefit of this approach. Mangoes are sensitive to chilling and are injured at temperatures lower than 10 to 13°C. Once the fruit are allowed to ripen under ambient conditions, disease develops normally. In the last decade, some progress has been made to check this. A better understanding of the physiological basis of quiescent infections in tropical and subtropical climacteric fruit has been achieved (Prusky and Keen, 1993). Research has shown that antifungal compounds are present in immature avocado and mango fruit and that the concentration of these compounds decreases as the fruit ripens. The decline in antifungal compounds can be delayed so that it occurs closer to full ripeness. In Costa Rica, postharvest treatments with butylated hydroxy anisole (BHA) resulted in reduced severity of mango anthracnose (Arauz, 2000).

Postharvest biological control of mango anthracnose has been attempted with limited and varying results.

Judging from the limitations and sometimes unreliability of the post harvest treatments, an alternative field program coupled with a postharvest chemical treatment may be the way to go to achieve an effective and reliable anthracnose disease control in mango. One is clearly not good enough without the other.

CONCLUSIONS

An integrated approach at managing anthracnose disease on mangoes is the way to go. The integrated management of post-harvest anthracnose under tropical and sub-tropical production conditions requires a sound knowledge of the biology of the pathosystem, the technologies available for field and post-harvest control in any given area or region, their economical feasibility and ecological acceptability (Arauz, 2000).

A successful integrated management program of mango anthracnose must take into account the following key factors: i.) The system is rain-moisture driven. Infection criteria have been identified and should be used to time fungicide applications rather than continue to rely on calendar-based spray schedules; ii.) The severity of post-harvest anthracnose on mango is the result of cumulative quiescent infections that develop after harvest, as fruit ripens. These are subject to eradication by post harvest treatments; iii.) Flowering can be managed so that susceptible tissue is produced during the drier months of the year. This will decrease the probability of fruit infections; iv.) Several pre-harvest and post-harvest treatments are commercially available and should always be used in combination wherever and whenever possible and v.) The efficacy of the various treatments is dependent on infection severity and this could vary from season to season and from one locality to another (Arauz, 2000).

SUMMARY OF FUTURE RESEARCH PRIORITIES

The Australian and Philippines disease forecasting systems have shown that the number of fungicide applications per season applied to control anthracnose, could be drastically reduced when such a system is used to determine time of spray applications. It has also been stated that such a system developed for one production region cannot be extrapolated and used in other regions because of dependence on local environmental conditions. There is thus a need to test and validate the MAE system from NSW Australia, under conditions in other mango growing regions, to make necessary modifications to suit the local or regional conditions of these areas of production.

Fungicide resistance has been reported in cases where systemics have been used in an unregulated way as the main means of field disease control. There are reported cases of resistance to some systemics, including benomyl, against populations of *C. gloeosporoides* in some regions. With the recent registration of the systemic fungicide Amistar, for use as a component of integrated field disease management in mangoes in Australia and elsewhere, it would be necessary to develop a fungicide resistance management strategy for Amistar and Prochloraz, the two current registered systemics in the system, if long term and lasting benefits are expected from their use in field anthracnose disease management.

Limited research has clearly demonstrated or suggested the importance of initial field inoculum reduction in epidemics of anthracnose and other related diseases in the field and on harvested fruit. It would be useful to evaluate the benefits of orchard sanitation on the incidence and severity of anthracnose disease in field trees and on harvested fruit. This could be integrated with minimal and optimal sprays of appropriate rotating fungicides, so as to clearly establish the role of this on anthracnose management.

Isolates of *C. gloeosporoides* from other crops have been demonstrated to cross-infect mangoes under controlled conditions. It would be useful to determine the epidemiological significance of such cross-infections of mangoes by *C. gloeosporoides* isolates from other tropical hosts such as avocado, papaya, banana etc, which are often grown in close proximity with mango orchards in some tropical environments.

Ascospores from the sexual phase of *C. gloeosporoides* have been reported but their role in field epidemics of mango anthracnose have not been demonstrated, even though their involvement can explain quite a lot, on the epidemiology of the disease and variability of the pathogen population that has also been reported. It would be useful to continue with investigations on the production and possible involvement of ascospores of the pathogen in the disease cycle of mango anthracnose. This will help to explain the variation among genotypes attacking mangoes and also in the development of strategies for control that target strains with different degrees of pathogenicity.

Pathogenic variability has been reported among *C. gloeosporoides* isolates from different mango orchards in different regions. This can help explain among other things, variation in response to similar control strategies in different areas and development of resistance to systemics in some places. It is also useful to develop control strategies targeting the more virulent isolates of the pathogen. It will be useful to assess pathogenic variability among *C. gloeosporoides* isolates from mangoes in all the different production regions as has been done in the Florida production region of the USA (Davis, 1999).

There are new fungicide products available in the market. There are also compounds reported to have systemic acquired resistance mechanisms. It would be useful to screen and identify more effective fungicides and to determine the effects of the fungicides and Systemic Acquired Resistance (SAR) compounds on anthracnose development under field spray conditions, as much of the past research efforts have focused on the determination of these effects under post-harvest conditions.

Genetic resistance as a component of mango anthracnose disease management has not been fully exploited. There are mango germplasm collection pools located and maintained in different mango research institutions and centers in Australia, the USA and elsewhere. It would be useful to undertake a systematic screening of these germplasm collections to identify entries with more resistance to mango anthracnose disease so that these could be used in developing new mango cultivars for tropical regions where the disease continuous to be a major pre and postharvest problem on mangoes.

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