Resistance of Mango Pathogens to Fungicides Used to Control Postharvest Diseases

DONALD H. SPALDING, Research Plant Pathologist, Agricultural Research Service, U.S. Department of Agriculture, Subtropical Horticulture Research Unit, 13601 Old Cutler Road, Miami, FL 33158

ABSTRACT

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Isolates of *Colletotrichum gloeosporioides, Diplodia natalensis*, and *Phomopsis citri* showed resistance to benomyl. Cross-resistance of the three fungi was shown between benomyl, thiabendazole, and thiophanate-methyl fungicides but not to imazalil and CGA-64251 in in vitro and in vivo tests. CGA-64251 was the most effective fungicide tested for control of postharvest decay of Tommy Atkins and Keitt mangos.

The most serious postharvest diseases of mango (*Mangifera indica* L.) are anthracnose, caused by *Colletotrichum gloeosporioides* Penz., and stem-end rot, caused by either *Diplodia natalensis* P. Evans or *Phomopsis citri* Fawc. Both of the major cultivars of mangos, Tommy Atkins and Keitt, grown commercially in Florida are susceptible to these pathogens. Because mangos are subject to chilling injury when held at temperatures below 13 C (4), low temperatures cannot be used to retard decay development during marketing. Moreover, no fungicides have been approved for postharvest decay control on mangos, although a packinghouse treatment could help to reduce losses due to decay. Of several fungicides used in mango production, the locally systemic fungicide benomyl has been one of the most effective, especially for control of anthracnose (5). Benomyl has also been found to improve the effectiveness of the postharvest hot water treatment (2-min dip at 52 C) used for decay control (6).

This paper presents evidence of the development of resistance to benomyl by the fungi causing anthracnose and stem-end rot of mangos and also reports the results obtained with a promising new fungicide, CGA-64251.

MATERIALS AND METHODS

Fungal resistance to fungicides. Cultures of *C. gloeosporioides, D. natalensis*, and *P. citri* were isolated from decayed tissue of mangos collected from local packinghouses during the 1976, 1977, and 1980 seasons. Small disks (7 mm diameter) taken from pure cultures of these fungi grown on lima bean agar (LBA) plates were used to inoculate the center of other plates of LBA with or without benomyl at 100 μg/ml. The plates were then held for 1 wk at 25 C. Isolates that grew heavily on LBA without benomyl and showed definite growth on LBA with benomyl were considered to be resistant to the fungicide. The degree of resistance was calculated in terms of the percentage of linear growth on LBA with benomyl relative to linear growth on LBA without benomyl.

Cultures showing high or low resistance to benomyl were then used to evaluate cross-resistance to thiabendazole (TBZ), thiophanate-methyl (TPM), imazalil, and CGA-64251 (1.125% EC of Vangard, Ciba-Geigy Corp., Greensboro, NC 27409; 1-[2-(2,4-dichlorophenyl)-4-ethyl-1,3-dioxolan-2-ylmethyl]-1*H*-1,2,4-triazole). The percentage of resistance was then calculated from the mean linear growth on three duplicate plates incubated for 1 wk at 25 C. A culture growing as much on LBA with benomyl as without would show 100% resistance. For experimental purposes, a fungal isolate was considered to have low benomyl resistance when its calculated percentage of resistance was less than half that of another isolate considered to possess relatively high benomyl resistance.

Comparison of fungicides. Mature green Tommy Atkins and Keitt mangos were obtained from a local packinghouse and sorted into similar lots according to size, appearance, and freedom from decay and injury. Each lot was then dipped for 2 min in tap water at 27 or 52 C, with or without fungicide at 0.1% a.i. (w/v). Fifteen mangos of each cultivar were used per treatment. The following fungicides were compared: benomyl, TBZ, TPM, imazalil, and CGA-64251. The Tommy Atkins mangos were harvested in June 1980 and the Keitt mangos in July 1980. The Tommy Atkins and Keitt mangos were treated the day after picking and stored at 13 C for 17 and 21 days, respectively, after which they were ripened at 21 C and examined for decay. All decay was counted except that which was barely noticeable and covered less than 2% of the surface area of a fruit at the soft-ripe or edible stage. Trace amounts of decay are generally present on all mangos after storage and ripening for 2–3 wk. Three weeks is about the maximum market life of mangos (4).

This article reports the results of research only. Mention of a pesticide does not constitute a recommendation for use by the USDA nor does it imply registration under FIFRA as amended.

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RESULTS AND DISCUSSION

Resistance to benomyl. The incidence of *C. gloeosporioides* and *D. natalensis* isolates resistant to benomyl increased markedly from 1976 to 1980 (Table 1). Sixty-nine percent of *C. gloeosporioides*, 38% of *D. natalensis*, and 31% of *P. citri* isolates tested during 1980 showed some degree of resistance to benomyl. The development of resistance to benomyl has not been reported previously in the above fungi but has been reported in a large number of other fungi, including *C. musae* (Berk. & Curt.) Arx (3). This acquired resistance to systemic fungicides has been discussed by Dekker (2). The

Table 1. Incidence of *Colletotrichum gloeosporioides, Diplodia natalensis*, and *Phomopsis citri* isolates resistant to benomyl[a]

	No. of isolates tested/resistant		
Year	*C. gloeosporioides*	*D. natalensis*	*P. citri*
1976	15/0	35/2	...[b]
1977	38/8	40/4	...
1980	62/43	40/15	16/5

[a] Isolates were considered resistant to benomyl when able to grow on lima bean agar containing benomyl at 100 μg/ml.

[b] No isolates tested.

Table 2. Comparative resistance of isolates of *Colletotrichum gloeosporioides* to three closely related fungicides[a]

Fungicide	No. of isolates tested/resistant	Resistance (%)
Benomyl	34/27	79
Thiabendazole	34/19	56
Thiophanate-methyl	34/30	80

[a] Isolations, made during 1980, were considered resistant if visually observable growth developed on lima bean agar containing fungicide at 100 μg/ml.

Table 3. Comparative resistance to several fungicides of isolates of *Colletotrichum gloeosporioides* (*C.g.*), *Diploida natalensis* (*D.n.*), and *Phomopsis citri* (*P.c.*) of low and high resistance to benomyl

Fungicide	Benomyl resistance	Resistance[a] (%)		
		C.g.	*D.n.*	*P.c.*
Benomyl	Low	20	42	4
	High	44	100	70
Thiabendazole	Low	3	43	10
	High	7	3	5
Thiophanatemethyl	Low	13	39	14
	High	29	100	54
Imazalil	Low	0	0	0
	High	0	0	0
CGA-64251	Low	0	0	0
	High	0	0	0

[a]Percentage of fungal growth on lima bean agar with fungicide (100 μg/ml) relative to growth without fungicide. Each value is calculated from the mean of growth on three duplicate plates after 1 wk at 25 C. Two isolates of each of the three fungal species isolated in 1980 were used in these tests. The percentage of resistance of isolates of low benomyl resistance was less than half that of isolates of high benomyl resistance.

Table 4. Comparison of various fungicides for control of decay of Tommy Atkins and Keitt mangos during storage and ripening[a]

Treatment		Decayed fruits %[b]	
Fungicide	Temp. (C)	Tommy Atkins	Keitt
Control (water alone)	27	73	80
	52	57	60
Benomyl	27	87	60
	52	80	53
Thiabendazole	27	80	60
	52	40*[c]	60
Thiophanate-methyl	27	80	53
	52	33*	73
Imazalil	27	60	53
	52	20*	40*
CGA-64251	27	13*	31*
	52	7*	12*

[a]Mature green mangos were dipped for 2 min in tap water at 27 or 52 C, with or without fungicide at 0.1% a.i. concentration. Each figure represents the mean for 15 mangos. The Tommy Atkins test was run in June–July 1980 and the Keitt in July–August 1980.
[b]Decay includes both anthracnose and *Diplodia* or *Phomopsis* stem-end rot. Trace amounts of decay were barely noticeable and were not included in the above data. Tommy Atkins and Keitt mangos were stored for 17 and 21 days, respectively, at 13 C, and then ripened at 21 C.
[c]Data followed by an asterisk are significantly different from the 27 C control at $P = 0.05$ by analysis of variance.

development of resistance to benomyl by mango pathogens in Florida is probably the result of repeated use of benomyl on mango trees to control anthracnose during production.

Cross-resistance to fungicides. Seventy-nine, 56, and 88% of *C. gloeosporioides* isolates showed some degree of resistance to benomyl, TBZ, and TPM, respectively (Table 2). Research with other fungi has also shown that resistance to benomyl is generally accompanied by resistance to related benzimidazole compounds, such as TBZ and TPM (1). This relation held true in our research, although *C. gloeosporioides* isolates showed greater resistance to benomyl and TPM than to TBZ. The *D. natalensis* and *P. citri* isolates of low resistance to benomyl showed a similar degree of resistance to all three fungicides, whereas the isolates of high resistance to benomyl showed high resistance to TPM but only slight resistance to TBZ (Table 3). Because isolates were not consistently less resistant to TBZ than to benomyl or TPM, resistance to TBZ could be a problem if it were substituted for benomyl in a combination postharvest hot water treatment to control decay of mangos. The isolates resistant to benomyl, however, were not resistant to imazalil and CGA-64251 in the in vitro tests.

Chemical control of mango decay. The adverse effect of the acquired resistance to benomyl by fungi causing postharvest decay of mangos was apparent in treated Tommy Atkins and Keitt mangos (Table 4). Benomyl, whether applied in water at 27 or 52 C, failed to reduce decay of Tommy Atkins mangos and only slightly reduced decay of Keitt mangos. Results with TBZ and TPM were similar to those with benomyl, except that TPM and TBZ in water at 52 C significantly reduced decay of Tommy Atkins, but not Keitt, mangos. Imazalil in water at 52 C, but not at 27 C, significantly reduced decay of both mango cultivars. CGA-64251 was the most effective fungicide tested and in water at 27 or 52 C significantly reduced decay of all mangos tested. Further tests are planned to determine the effectiveness of CGA-64251 at various concentrations.

There is no evidence at present to indicate whether CGA-64251 or imazalil will have a resistance problem with the mango pathogens if approved for use on mangos. However, the chances of resistance development would be reduced if these fungicides were only used as a spray, drench, or dip after harvest. In postharvest use, the fungicide solution should be changed frequently to avoid inoculation of mangos with resistant organisms during treatment. The possible approval of CGA-64251 as a preharvest spray in mango groves would increase greatly the probability of selecting resistant strains of mango pathogens. The development of mango pathogens resistant to CGA-64251 would put us into the same situation we now have with the development of pathogens resistant to benomyl. The answer is probably to develop a sufficient number of effective fungicides such that their use can be alternated to avoid selection of resistant mango pathogens for as long as possible.

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