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ROLE OF *FUSARIUM MANGIFERAE* IN CAUSATION OF MANGO MALFORMATION DISEASE

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Abstract: Determination of fungi associated with malformed tissues of mango was done on different local and exotic cultivars during the year 2000-2001. Five mango growing districts of the Punjab were selected for the study with three locations in each district. A total of 75 malformed samples and 750 tissues (10 tissues / sample) of 5 standard local cultivars and 1500 tissues of exotic cultivars were assayed. *Fusarium mangiferae* proved to be the dominant fungus with 100% sample's infection in all the five districts. Maximum within tissue infection of 98% was observed in Okara and Sahiwal districts. Local variety Anwar rataul and exotic Tomy atkin appeared to be the most infected varieties giving 92.0 and 96.0% tissue infection, respectively.

Keywords: Determination, *Fusarium mangiferae*, malformation, *Mangifera indica,* Punjab.

INTRODUCTION

Mango is an important fruit of Indo Pak Subcontinent known to be cultivated in this region from ages. It is extensively grown in the Punjab over an area of 49500 ha with annual production of 634900 tonnes. In Pakistan, area and production are 97000 ha and 989800 tonnes, respectively [Anonymous 2001-2002]. Like other fruit crops, Mango is attacked by several animate and inanimate diseases. Malformation is the most threatening disease causing colossal losses every year. Despite hectic efforts, complete control has not been achieved yet. Two types have been characterized viz. vegetative and floral. Both the symptoms are the expression of the same disease [Varma *et al*. 1969, Schlosser 1971]. Viral [Kausar 1959, Latif *et al*. 1961, Singh and Jawanda 1961], acarological [Singh 1997] and physiological [Sharma 1953] etiologies have been claimed. Recent literature reveals the association of a fungus *Fusarium mangiferae* to be the cause of the disease [Britz *et al*. 2002].
Disease manifestation by artificial inoculations has been proved [Summanwar *et al*. 1966, Freeman *et al*. 1999]. The fungus *F. subglutinans* [*mangiferae*] was found associated with malformation and identical morphology and etiology of vegetative and floral isolates was confirmed [Nariani and Seth 1962, Chadha *et al*. 1979, Ploetz and Gregory 1993, Freeman *et al*. 1999, 2004]. The main objectives of the present study were to determine the frequency of different fungi in malformed tissues and establish the cause of mango malformation in Pakistan.

MATERIALS AND METHODS

The studies to determine the fungal association were conducted during the flowering cycle of the year 2002-2003 (March-April). Sample collection for local varieties was done from 5 mango growing districts of the Punjab. Maximum disease severity was observed in the selected orchards. Five local varieties viz. Dusehri, Chaunsa, Langra, Anwar rataul and Malda and five exotic viz. Tomy atkin, Swarnika, Maya, Zill and Kensington were kept under the study. Three locations were selected in each district to collect the samples of local varieties. Each location contributed five panicles along with 6-8 cm shoot portion representing one of each variety. From each of the 5 districts, 15 samples of every local cultivar were collected.

Collection of exotic cultivars was done from Mango Research Station, Shujabad, Multan. Thirty samples of each exotic cultivar were taken. The samples were placed in an ice box immediately after clipping to avoid heating during transit. Ten tissue pieces 5 mm long, excised from peduncles and panicle-shoot juncture were surface disinfested in 1% NaOCl solution for 2 minutes, rinsed twice in sterilized deionized water, dried on sterilized blotting papers and placed onto 9 cm diameter Pyrex glass Petri plates containing Potato Dextrose Agar (PDA) medium [Nelson *et al*. 1983]. The plates were kept in a cooled incubator at 25^{o}C under fluorescent illumination to give a 12 hour photoperiod to ensure maximum macro conidial production. The plates were examined after 6-7 days of incubation. The fungi isolated were identified following standard keys [Ellis 1980, Nelson *et al*. 1983]. The colonies of *F. mangiferae* were purified on Carnation leaf agar (CLA) medium to ensure abundant macro conidia with least phenotypic variation. The identification was verified on the basis of typical micro and macro conidia (Figs. 1 and 2).

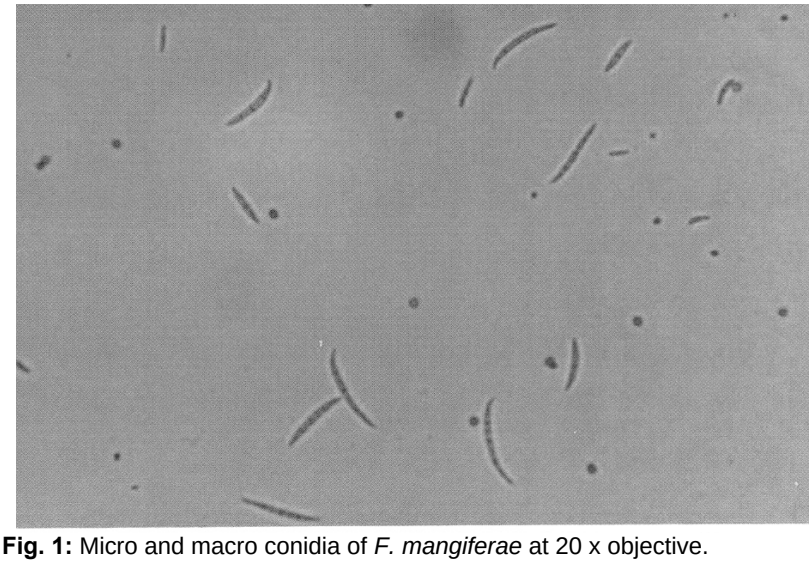

Fig. 1: Micro and macro conidia of *F. mangiferae* at 20 x objective.

Fig. 2: Magnified image of four celled macroconidium.

RESULTS

Four fungi viz. *F. mangiferae, F. pallidoroseum, F. oxysporum and Alternaria alternata* were found to be associated with malformed parts. Maximum recovery was exhibited by *F. mangiferae.* In Okara, Sahiwal, Pakpattan, Jhang and Lodhran districts, 100% samples appeared to be infected with this fungus. Maximum infection frequency (within tissue infection) of 98.0% was recorded in Okara and Sahiwal followed by 96.0, 93.33 and 62.0 % in Pakpattan, Lodhran and Jhang districts (Table 1).

Table1: Fungi associated with malformed parts of mango.

Sr. No.	District	Samples	Sub-samples	Fungi isolated	(%) Infection frequency	*S.E	(%) Samples infected	S.E.
1.	Okara	15	150	*F. mangiferae*	98.0(90-100)	1.31	100.00	00.00
				A. alternata	2.0(0-30)	0.37	6.66	41.44
2.	Sahiwal	15	150	*F. mangiferae*	98.00(90-100)	1.31	100.00	00.00
				A. alternata	6.66(0-30)	1.04	46.66	165.92
				F. pallidoroseum	1.33(0-30)	0.25	13.33	77.02
3.	Pakpattan	15	150	*F. mangiferae*	96.0 (90-100)	2.56	100.00	00.00
				A. alternata	1.33(0-10)	0.08	13.33	77.02
				F. pallidoroseum	4.66(0-30)	0.79	26.66	130.35
4.	Jhang	15	150	*F. mangiferae*	62.0(40-100)	15.71	100.00	00.00
				F. oxysporum	4.66(0-40)	1.10	20.00	106.67
5.	Lodhran	15	150	*F. mangiferae*	93.33(80-100)	4.15	100.00	00.00

*S.E. ± Standard error

Other fungi like *F. pallidoroseum*, *F. oxysporum* and *A. alternata* showed least infection level. ‘Chaunsa’, ‘Langra’, ‘A. rataul’ and ‘Malda’ showed 100.0% infection in Okara while ‘Langra’, ‘A. rataul’ and ‘Malda’ in Sahiwal district. ‘Malda’ retained 100.0% infection in Pakpattan. The cultivar ‘A. rataul’ gave maximum tissue infection of 92.0% while ‘Langra’ and ‘Malda’ both showed 91.33% infection. Comparatively less infection

of 86.0% was observed in tissues of 'Chaunsa' which is even much higher to support extensive colonization of the fungus (Table 2). Maximum infection frequency of 89.46% was exhibited by fungus *F. mangiferae* colonizing 671 out of 750 tissues. *F. pallidoroseum*, *A. alternata* and *F. oxysporum* showed only 1.2, 2.0 and 0.93% infection frequency (Table 3). Tissue assay of exotic cultivars revealed maximum infection of 96.0% caused by *F. mangiferae* in 'Tomy atkin' followed by 93.0 and 91.33% in 'Kensington' and 'Maya', respectively (Table 4).

Table 2: Percent recovery of *F. mangiferae* from varieties of different districts.

Sr. No.	District	Variety									
		Dusehri	S.E.	Chaunsa	S.E	Langra	S.E	A. rataul	S.E	Malda	*S.E
1.	Okara	90.00	30.00	100.00	00.00	100.00	00.00	100.00	00.00	100.0	00.00
2.	Sahiwal	93.33	20.75	96.66	10.76	100.00	00.00	100.00	00.00	100.0	00.00
3.	Pakpattan	100.00	00.00	86.66	38.53	96.66	10.76	96.66	10.76	100.0	00.00
4.	Jhang	66.66	74.08	53.33	82.96	63.33	77.41	63.33	77.41	63.33	77.41
5.	Lodhran	83.33	46.30	93.33	20.75	96.66	10.76	100.00	00.00	93.33	20.75
	Total	86.66		86.00		91.33		92.00		91.33	

Table 3: Cumulative infection of different fungi in malformed tissues of five mango cultivars from five districts of the Punjab

Sr. No.	Fungus	Tissues colonized (out of 750)	(%) Infection frequency	*S. E.
1.	*F. mangiferae*	671	89.46	1.26
2.	*F. pallidoroseum*	9	1.20	0.16
3.	*A. alternata*	15	2.00	0.26
4.	*F. oxysporum*	7	0.93	0.12

Table 4: Percent recovery of *F. mangiferae* from five exotic cultivars.

Sr. No.	District	Cultivar	No. of tissues		% infection	*S. E
			Examined	Infected		
1.	Multan	Tomy atkin	300	288	96.00	1.28
2.	Multan	Swarnika	300	222	74.00	6.41
3.	Multan	Maya	300	274	91.33	2.64
4.	Multan	Zill	300	263	87.66	3.60
5.	Multan	Kensington	300	279	93.0	2.17

*S.E. ± Standard error.

DISCUSSION

The present studies were aimed to determine the fungi associated with malformed tissues of local and exotic cultivars cultivated in mango growing areas of the Punjab. *F. mangiferae* proved to be the dominant fungus infecting majority of the tissues. The infection frequency of other fungi remained much low (Table 3). The infection frequency (within tissue infection) of *F. mangiferae* confirms its role in causation of malformation symptoms. The pathogenic interaction with floral buds resulted in high incidences of malformation which started early in the floral season, extended up to April and re-established in November. *F. mangiferae* was often associated with the floral and vegetative apices. With in panicle infection of 84.5% in small pedicel and peduncle tissue pieces caused by *F. subglutinans* [*mangiferae*] was confirmed by Ploetz [1994] in USA.

The infected tissues always yielded typical and abundant macro conidia on Carnation leaf agar (CLA). Fungus was isolated from healthy shoots only when they were either in close contact with diseased ones or exhibited initial stage of infection with least or scanty macro conidial production in quite a few tissues. Symptoms of vegetative and floral malformation appeare where mycelia of *Fusarium* sp. are present in the tissue at high concentrations. Malformin conc. in malformed tissues correlate with the intensity of the disorder.

Frequent recovery of *F. mangiferae* from malformed trees grown in different ecological zones of the world has already been proved. As malformed shoots show elevated levels of infection but non malformed ones show least pathogenic association, it is suggested that symptom manifestation occurs only after massive colonization by the fungus *F. mangiferae* [Ploetz and Gregory 1993].

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