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IMPACT OF ENVIRONMENTAL VARIABLES ON SPORE DISPERSAL TREND OF FUSARIUM MANGIFERAE CAUSING MANGO MALFORMATION DISEASE IN PAKISTAN

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ABSTRACT

Malformation is one of the most destructive mango diseases. Although trees are not killed, the vegetative phase of the disease impedes canopy development during vegetative phase of the host plant and floral phase dramatically reduces fruit yield with overwintering inoculums during dormant phase of the host plant. Environmental conditions and trend of spore liberation of its pathogenic fungus "Fusarium mangiferae" were recorded during flowering phase (Feb-April, 2014), fruit development phase (May-July, 2014), vegetative phase (Aug-Oct, 2014), and dormant phase (Nov-Jan, 2014-15), of the mango plants. Through installation of spore traps of various distance levels containing Nash-Synder media in petri plates. During these phases, different environmental variables including temperature (T), relative humidity (R.H) and wind speed (W.S) were observed. Maximum number of colonies were observed through the spores trapped from the centre of the experimental block (0m) while minimum numbers of colonies were developed from the spores trapped at 150 m distance. Amongst different phenological phases of mango, fruit development remained very much contaminated with spores of the fungus while minimum spore liberation was noted during flowering phase of mango indicating very rare infection during this phase. This study also reflected that maximum number of airborne macro conidia of *F. mangiferae* were recorded when R.H was below 55%. Low R.H appeared to be a major factor associated with the diurnal conidial dispersal of airborne pathogen. Hence primary infection of F. mangiferae starts from vegetative and floral buds differentiation and control strategy including spray of systemic fungicides should start at this stage of development.

Keywords: Mango, Malformation, Fusarium, Spore liberation, Environment.

INTRODUCTION

Nature has endowed Pakistan with wide range of agro climatic conditions which permit quality production of both tropical and temperate fruits. The climate of Pakistan is approximately favorable to all types of fruit production. Among these fruits Mango (*Mangiferae indica*) occupies the 2nd largest position after citrus in Pakistan having an area of 171 thousand hectares with production of 168 thousand tons (GOP, 2015). Its

Submitted: May 13, 2018 Revised: May 28, 2018 Accepted for Publication: June 13, 2018 * Corresponding Author: Email: mtmalik64@hotmail.com © 2017 Pak. J. Phytopathol. All rights reserved. popularity is due to its excellent aroma, flavor, delicious taste and high nutritive value being rich in vitamin A and C. Its origin is belonged to South Asia where it has been under cultivation for the last four hundred years (Salunkhe *et al.*, 1994).

In recent time, mango production is greatly affected by various abiotic and biotic factors in Pakistan (Masood *et al.*, 2011). Abiotic factors include thermal regimes, flood, drought, rainfall, windstorm, nutritional deficiency and mal cultural practices etc (Ploetz, 2003; Nafees *at al.*, 2010). Abrupt change in temperature and heavy rains not only disturb the bearing behaviors of mango trees but predispose different diseases, out of which mango malformation has paramount's importance (Ploetz, 2003). It is the most threatening disease affecting the

mango orchards severely which has been the significant impact on the promulgation of trees and commercial fruit yield. The losses due to this disease vary with the cultivars and disease severity. In general, production losses may range from 80-90% as the diseased inflorescence may not succeed to produce fruits (Ploetz, 1999).

Mango malformation disease is of two types i.e. vegetative malformation and floral malformation. These types may express the same disease (Kumar and Beniwal, 1987) causing serious losses in the yield. Most recent literature confirms that malformation is induced by a fungus named as *Fusarium mangiferae* (Ploetz 1999; Ploetz *et al.*, 2002; Freeman *et al.*, 2004). However, gradual increase in its appearance on new seedling and trees is dominating with the passage of time which is also unpredictable. The latently infected plants and the infected propagation material are responsible for the blowout of the pathogen which is the main source of the long distance spread of disease (Kumar *et al.*, 1993) & (Ploetz, 1999).

Efforts to manage malformation may not met with significant success. Pruning of infected buds, indicative shoots and many subtending nodes and use of effective fungicides showed some effectiveness against this disease (Pernezy and Ploetz, 2000). Although a lot of research and experiments have been conducted since decades on different aspects of mango malformation disease but less importance has been given to its spore dispersal trend in the light of environmental variables in Pakistan. So, understanding with its epidemiology through observing spore dispersal trend in the present degradation environmental has paramount's importance. In this study, the management strategy could be formulated after the confirmation of the exact cause of this disease, which is a fungus named Fusarium mangiferae.

MATERIALS AND METHODS

This study was conducted in experimental orchard of Mango Research Institute Multan, comprising of different mango varieties Viz S.B Chaunsa and Sindhri. Trees had the average height of 05m with spacing of 10m between rows. For the estimation of conidial abundance in the air in experimental block, passive space trapping system was used. The height of those traps was maintained at 02 m with the help of bamboo sticks. In this frame, a petri plates containing *Fusarium* specific medium Nash-Snyder (NS) was exposed directly to the air (Gamliel-Atinsky *et al.*, 2009).

The spore traps were installed on north side of both blocks at 6 different places in and outside the experimental block viz. Om (Centre of block) at 30m, 60m, 90m, 120m and 150 m far from the block with 4 replications. The traps were used to install once in a week between 9.00 am to 4.00 pm, during four phenological phases of mango viz vegetative phase (Aug-Oct), dormant phase (Nov-Jan), flowering phase (Feb-April) and fruit development phase (May-July). In statistical analysis, the mean of all the data of spore liberation during each month was considered as representative data of that particular month.

The exposed Petri plates containing NS media were selected with laboratory film and placed in incubator at 25 ± 1 °C for colony growth of the fungus. After four to five days, the colonies were counted with the division of Petri plates into four parts to estimate spore liberation inoculum availability at a site (Noriega-Cantu *et al.*, 1999; Gadoury *et al.*, 1983). The metrological data regarding temperature, relative humidity and wind speed were also noted from the adjacent Central Cotton Research Institute Multan. Number of spores liberated were assessed by observing the number of colonies developed in passive spore trapping method at different spatial levels. Spores and colony growth were observed in microscope.

The data of above treatments were collected and analyzed statistically by using statistic 8.1 version analytical software with the help of factorial design. The treatment means were compared by using LSD at 1% alpha confidence level.

RESULTS & DISCUSSION

Inoculum load of mango malformation causing fungus *F. mangiferae* was monitored through trapping its spores with harboring on NS medium where the spores developed fungus colonies. The number of fungus colonies developed reflected the number of trapped spores or inoculum load. Basically, this study was aimed at offering a lucid and complete view of this different aspects of mango malformation spread.

The spores of *F. mangiferae* were tried to detect at different spatial levels so that spore free zone could be assessed specially for the establishment of mango nursery. The data given in table 1 revealed the significant decrease in inoculum load with the increase of distance from orchard. Maximum number of spores was examined in the centre of the orchard while minimum at the distance of 150 m away from the

orchard. This trend of spore spread very clearly indicated that spore free zone was quite possible beyond 150 m to establish a clear mango nursery. The maximum spore libation in the centre of the block might be due to the fact that source of inoculum was present nearby and when the distance from source increased the spore trapping was decreased.

Table 1. Mean inoculum load of *F. mangiferae* monitored at 06 different spatial levels.

0m	30m	60m	90m	120m	150m
207.50 a	188.72 b	169.28 c	149.92 d	136.21 e	124.50 f



Figure 1. F. mangiferae spores trapped at six spatial levels.



Figure 2. F. mangiferae colonies developed from spores trapped at six spatial levels.

These results were in line with the results of Gamliel *et al.*,2009 who concluded as the distance from source increases the number of spores will decrease. Spread and distribution of the disease inoculum was also examined during four (04) different phonological stages of mango viz. vegetative phase indicating the months of August, September and October, dormant phase comprising of November, December and January, flowering phase consisting of February,

March and April and fruit development phase having May, June and July to note mode and trend of spore dispersal. It was observed in table 2 that maximum spore liberation was during fruit development phase which was by 259.99 in number on an average followed by 168.19 noted during vegetative phase and in dormant stage 143.37 the least spore liberation (79.19) was observed during flowering stage indicating rare primary infection during this stage.







Figure 2. F. mangiferae spores trapped in different phenological stages.



Figure 2.1. *F. mangiferae* colonies developed from spores trapped in vegetative phase at six spatial levels.



Figure 2.2. F. mangiferae colonies developed from spores trapped in dormant phase at six spatial levels.



Figure 2.3. F. mangiferae colonies developed from spores trapped in flowering phase at six spatial levels.



Figure 2.4. *F. mangiferae* colonies developed from spores trapped in fruit development phase at six spatials levels.



Figure 2.5. *F. mangiferae* colonies and microscopic observation of fungal spores.

It is worth mentioning here, that during fruit development phase of mango, minimum relative humidity was observed which is the most important factor for spore liberation. On the other hand, tendency of emergence of new vegetative flushes also remained in progress due to frequent irrigation and conducive temperature. These liberated spores may easily land on new flushes. After crop harvest, vegetative growth was achieved and resultantly, more inoculum load of pathogenic fungus was observed during this phase as compared to dormancy at flowering phase because of more spore production due to less quantity of *F. mangiferae* during this phase. It was clear that spore liberation / spread was always more on new vegetative flushes irrespective of their time of emergence. The maximum spore liberation in the fruit development stage might be due to the favorable environmental conditions which cause the maximum spore liberation in this stage as compared to other phenological stage.

The mean inoculum load of *F. mangiferae* ranging 104.10-254.40 during vegetative phase of development is quite different when compared with its load ranging from 60.65-97.23 in flowering phase (Table 3).

Table 3. Mean inoculum load of <i>F. mangiferae</i> monitored in 04 phenological stages at different spatial levels.							
	Vegetative	Dormancy	Flowering	Fr. Dev.			
0m	254.40 d	179.56 g	97.23 n	298.81 a			
30m	217.15 f	164.52 h	89.65 o	283.56 b			
60m	175.37 g	150.58 i	84.27 o	266.90 c			
90m	138.60 j	132.90 j	75.31 p	252.85 d			
120m	119.54 k	121.50 k	68.02 q	235.77 e			
150m	104.10 m	111.19 l	60.65 r	222.06 f			



Figure 3. Comparison of *F. mangiferae* spores trapped in interaction of phonological stages with spatial levels.

]	Γemperature(°C) Relative Humidity (%]				Wind		
Phenological Stage	Phenology Spar	Max.	Min.	Morn.	Even.	Speed (Km/h)	Mean of spore liberatior	
	August	35.2	27.9	77.1	67.3	5.5		
Vagatativa nhaqa	September	35.2	25.4	79.5	63.8	4.9	1(0.10	
vegetative phase	October	33.8	22.7	79.3	63.4	3.3	168.19	
	MEAN	34.7	25.3	78.6	64.8	13.7	-	
	November	26.9	13.1	84.1	74.6	2.6	_	
Dormant Phase	December	20.4	9.3	89.7	75	3.1	1/2 27	
Dormant Phase	January	19.7	6.2	97.1	61.4	2.8	145.57	
	MEAN	22.3	28.6	90.3	70.3	2.8		
	February	21.2	8.6	94.3	69.2	4.1		
Flowering Phase	March	25.4	14.2	86.3	62.1	4.9	70.10	
Flowering Fliase	April	33.6	20.2	67	43.5	5.6	- / 7.17	
	MEAN	26.7	14.3	82.5	58.2	4.8		
	May	36.7	24.6	64	43.7	6.7		
Fruit Development Phase	June	39.9	30.5	62.1	42.4	7.8	- 250.00	
	July	36.8	29.4	70.6	51.5	7.4		
	MEAN	37.8	28.1	65.5	45.8	7.3		
	AVERAGE	32.95 55.65		7.3				

	Table 4. Correlation of s	pore dispersal	trend with	different en	vironmental	factors.
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The mean of temperature, relative humidity and wind speed during fruit development phase were 32.95°C, 55.65% and 7.3km/h respectively (Table 4).

The results are in alliance with the finding of (Gamliel *et al.*, 2009) and (Noriega-Cantu *et al.*,1999) who described that maximum air born spores of *F. mangiferae* were trapped when the relative humidity was low 55%. The production of *mangiferae* (antifungal compound produced by the host plant in defense) was also low in this stage which resulted an

increase in spore dispersal (Chakrabarti *et al.*, 1997) and (Chakrabarti and Kumar 1998). Maximum spore liberation during fruit development i.e. summer season and summer growth may escape the infection because fungus could not move in fresh growth because during this time, rate of growth of fungus is much slower than that of host tissues.

The disease prediction model was also developed (table 5) at the completion of the study and it will help us in the control of this disease in future.

	Phase Name	Mean Temperature (°C)		Mean Relative Humidity (%)		Mean Wind Speed (Km/h)	Mean of total	
	-	Max.	Min.	Morn.	Max.		spores trapped	
Maximum spore	Fruit Development Phase	37.8	28.1	65.5	45.8	7.3	259.99	
production and dispersal	Vegetative Phase	34.7	25.3	78.6	64.8	13.7	168.19	
Maximum disease appearance	Flowering Phase	26.7	14.3	82.5	58.2	4.8	79.19	

Table 5. Disease Prediction Model.

According to this prediction model the maximum spore production and liberation occurs in fruit development phase followed by vegetative phase. It was observed that spore liberation of *F. mangiferae* was not very much affected by the temperature and wind speed but contrary to these it was greatly influenced by the relative humidity.

CONCLUSION

This study reflected that maximum number of air borne macro conidia of *F. mangiferae* were recorded in infected mango orchards when relative humidity was low (55%). During fruit development phase as, low relative humidity appears to be a major factor associated the diurnal conidial dispersal of air born pathogen. Finally, it is concluded that the primary infection *F. mangiferae* starts from vegetative and floral buds at the control strategy including spray of systemic fungicides should start at this stage.

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