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Impact of inoculation methods and fruit maturity stages on the development of Fusarium fruit rot of citrus

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Injury to fruit surface on epicarp (Pin-pricking) was found essential for the infection and development of Fusarium rot (45.1 %). The severity of Fusarium rot was found more in mature fruits (42.48 %) rather than semi-mature (20.46 %) and immature (12.73 %) fruits.

Key words: Citrus, maturity stages, *Fusarium pallidoroseum*, fruit rot, post-harvest.

INTRODUCTION

Citrus, one of the most important fruits of the world, is cultivated widely in the tropical and sub-tropical regions. It ranks third among the sub-tropical fruits of the world with different varieties. Acid lime (*Citrus aurantifolia* Swingle) belongs to the family Rutaceae. It is belived to be a native of Malaya, Assam and China. Acid lime is one of the commercially important citrus fruit grown in India besides sweet oranges, mandarin and grape fruit. There are four commercially important species of lime namely, *Citrus aurantifolia* Swingle(Acid lime), *C. latifolia* Tanaka(Tahiti lime), *C. limonia* Osbeck (Rangpur lime) and *C. limettioides* Tanaka(Sweet lime) (Khan, 2007). The lemon is rich in many food ingredients, particularly citric acid. Different varieties contain the citric acid in various proportions ranging from 3.71 to 8.40 per cent. It is mainly due to its citric acid and Vitamin C contents that the lemon is widely used in medicine. It is valued for its juice which is mostly used as an accessory food.

It increases the flavor and improves the taste of various dishes. It is often used in the preparation of salads and prevents discoloration of sliced bananas and apples. It is widely used in the preparation of lemonades, squashes, jams, jellies, pickels and marmalades. Eighty three per cent of the citrus fruit samples found associated with Fusarium spp. exhibiting 25 to 100 per cent infection in citrus fruits (Tournas and Katsoudas, 2008). As very meagre research work has been carried out on fruit rots of citrus and their management in India, with a view to extend the shelf life of citrus fruits and to reduce the losses caused by post-harvest diseases; it is felt worthwhile to carry out the investigations on Fusarium fruit rot of citrus and its management under middle Gujarat conditions.

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MATERIALS AND METHODS

Inoculation methods on the development of Fusarium fruit rot of citrus

Healthy, uniform semi-mature fruits of citrus cv- Kagzi lime were collected from the Sardar Patel vegetable market, Anand and brought to the laboratory in paper bags. The fruits were washed with tap water, then surface sterilized by dipping in 0.1 per cent NaOCl solution for one minute followed by three washings with distilled sterile water.

The experiment was conducted in completely randomized design (CRD) with six replicates. The fruits were injured by the following methods:

Stylar-end pricking**:** The fruits were injured at stylar-end with sterilized pins fixed on cork and dipped in spore suspension (106 spores/ml) of the test pathogen.

***Rubbing*:** The fruits were rubbed with each other by shaking manually for five minutes in crate and then the fruits were dipped in spore suspension (106 spores/ ml) of the test pathogen.

***Pin pricking on epicarp*:** The fruits were injured by pin pricking with sterilized pins fixed on cork on epicarp of fruit upto 2 mm depth. The fruits were dipped in spore suspension (106 spores/ ml) of the test pathogen.

***Without injury (Control)*:** The fruits without injury were dipped in spore suspension (106 spores/ml) of the test pathogen.

The inoculated and uninoculated fruits were placed in sterilized polythene bags. One fruit was accommodated in one bag. A piece of sterilized moist absorbent cotton swab was placed inside each bag and the mouth of the bags were loosely tied with rubber bands. The bagged fruits were kept at 27 ± 10C temperature. The observations on disease incidence and severity were recorded on 4th and 8th day with the help of assessment key (0%, 1-10%, 11-20%, 21-40%, 41-50% and >50%).

$$\text{Severity (\%)} = \frac{\text{Area of infected fruits}}{\text{Total area of fruit tissue}} \times 100$$

Methods for fruit maturity on the development of Fusarium fruit rot of citrus

The fruits of three different stages were selected for the study.

***Immature fruits*:** Freshly harvested healthy fruits with green colour.

***Semi-mature fruits*:** Healthy fruits with yellowish green colour.

***Mature fruits*:** Healthy fruits with yellow colour.
The healthy fresh fruits of cv. Kagzi lime (immature, semi-mature and mature) were collected and inoculated. Six repetations were maintained for each stage. The severity of the fruit rot was recorded on 4th and 8th day as mentioned earlier.

RESULTS AND DISCUSSION

Inoculation methods on the development of Fusarium fruit rot of citrus

The data presented in Table 1 revealed that significantly highest Fusarium fruit rot severity, 23.91 and 45.1 per cent was recorded after 4th and 8th day of inoculation by pin- prick method, respectively over all other methods followed by Stylar- end pricking method (13.21 & 32.18 %). Rubbing method (3.56 & 16.80 %) found least effective in causing fruit rot infection. Fruit rot symptoms did not observed on fruits when inoculated without any injury. The results clearly indicated that the injury on fruit epicarp was essential for infection and further development of rot as Fusarium pallidoroseum is a weak parasite. Pin- pricking on epicarp and stylar-end pricking exhibited 100 per cent disease incidence, while rubbing method revealed only 50 per cent disease incidence.

Smilanick et al. (2008) reported a rapid method of inoculation of *Geotrichum citri-aurantii* by dipping a sterile rod tip into the inoculum suspension by making a single puncture on citrus fruit. Rubbing, pin-pricking, cork-wounding and fall injury methods showed 100 per cent disease incidence, while stem-end method and stylar-end method exhibited 50 per cent Phomopsis rot incidence in aonla fruits (Choubey, 2007).

Table 1: Effect of different inoculation methods on severity and incidence of Fusarium rot of citrus

Inoculation Methods	Fusarium Rot Severity (%)		Disease Incidence (%)
	4th day	8th day	8th day
Stylar- end pricking	13.21	32.18	100.0
Rubbing	3.56	16.80	50.0
Pin pricking on epicarp	23.91	45.10	100.0
Control	0.00	0.00	0.0
S.Em. ±	0.26	0.41	--
C.D. at 5%	0.78	1.20	--
C.V. %	13.00	10.54	--

Singh (2011) tested different inoculation methods of Fusarium moniliforme on banana fruit by stem-end pricking, stylar-end pricking, pricking on epicarp, rubbing and without injury. Among all, stem-end pricking method was found most effective for development of fusarium rot. Sheth (2008) tested different inoculation methods of *Aspergillus niger* on citrus fruit by stem-end, stylar-end, pin-pricking, cork-wounding and fall injury. Among all, cork-wounding method was found most effective

for initiation and further development of Aspergillus rot.

Impact of fruit maturity on the development of Fusarium fruit rot of citrus

In order to find out the most vulnerable stage of fruit for initiation of infection and development of Fusarium rot, three stages of fruits were selected i.e. immature, semi-mature and mature. The results presented in Table 2 reveals that maximum fruit rot severity was recorded in mature fruits as compared to semi-mature and immature fruits. The highest Fusarium fruit rot severity was recorded in mature fruits after 4th (22.53 %) and 8th day (42.48 %) of inoculation, respectively. While in semi mature fruits it was 15.50 and 20.46 per cent after 4th and 8th day of inoculation, respectively. Lowest rot severity was recorded in immature fruits (7.48 & 12.73 %). It is clear from the result that as the citrus fruits reaches towards maturity the phenol content of fruit get reduced due to which they become more vulnerable for infection and further development of fruit rot incited by *F. pallidoroseum*.

Table 2: Impact of fruit maturity on the development of Fusarium fruit rot of citrus

Fruit Stage	Fusarium Rot Severity (%)	
	4^{th} day	8^{th} day
Immature	7.48	12.73
Semi-mature	15.50	20.46
Mature	22.53	42.48
S.Em. ±	0.43	0.58
C.D. at 5 %	1.30	1.75
C.V. %	7.92	6.93

It has been reported that ripe mango fruits are more vulnerable for infection by *Rhizopus* and *Botryodiplodia* rot than semi-ripe and unripe fruits when inoculated at epicarp. Chukwuka *et al.* (2010) inoculated the ripe and unripe papaya fruits with *Aspergillus flavus, A. niger, Fusarium sp., Mucor* sp. and *Rhizopus nigricans*. Among these, ripe fruits exhibited more rotting than unripe fruits. Singh (2011) reported that significantly highest Fusarium fruit rot severity was noticed in semi-ripe fruits (9.44 & 28.88 %) as compared to ripe (8.88 & 27.77%) and unripe fruits (3.33 & 4.99 %) on 4th and 7th day after inoculation, respectively.

Damaram (2012) recorded significantly highest Fusarium fruit rot severity in semi-ripe fruits (9.83 & 31.44 %) as compared to ripe (8.77 & 29.61%) and unripe fruits (3.66 & 5.71 %) of tomato after 4th and 8th day of inoculation, respectively. Naresh (2014) also reported significantly highest Fusarium fruit rot severity in ripe fruits (22.50 & 40.41 %) as compared to semi-ripe fruits (14.83 & 20.33 %) and unripe (7.30 & 12.70 %) of papaya after 4th and 8th day of inoculation, respectively.

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REFERENCES

Choubey, S. (2007). Phomopsis fruit rot of aonla and its management. M. Sc. (Agri.) thesis submitted to Anand Agril. Uni. Anand.

Chukwuka, K. S., Okonko, K. I. and Adekunle, A. A. (2010). Microbial ecology of organisms causing Pawpaw (Carica papaya L.) fruit decay in Oyo State, Nigeria. *Am-Euras. J. Toxicol. Sci*., 2 (1): 43-50.

Damaram (2012). Investigation on Fusarium Fruit Rot [*Fusarium pallidoroseum* (Cooke) Sacc.] of Tomato (*Lycopersicon esculentum* Mill.) and its Management. M. Sc. (Agri.) Thesis submitted to Anand Agril. Uni. Anand.

Khan, I. (2007). Citrus genetics, breeding and biotechnology. 26-27.

Naresh (2014). Investigation on Fusarium Fruit Rot [*Fusarium solani* (Mart.) Sacc.] of Papaya *(Carica papaya* L.) and its Management. M.Sc. (Agri.) thesis submitted to Anand Agril. Uni. Anand.

Sheth, D. (2008). Studies on Aspergillus fruit rot (*Aspergillus niger* Tieghem) of citrus (*Citrus aurantifolia* Swingle) and its management. M.ScA. (gri.) thesis submitted to Anand Agril. Uni. Anand.

Singh, K. B. (2011). Studies on Fusarium fruit rot (*Fusarium moniliforme* Sheldon) of banana (Musa paradisiaca L.) and its management. M.Sc. (Agri.) thesis submitted to Anand Agril. Uni. Anand.

Smilanick, J. L.; Mansour, M. F.; Gabler, F. M. and Sorenson D. (2008). Control of citrus post-harvest green mold and sour rot by potassium sorbate combined with heat and fungicides. *Post-harvest Biol. Tech.* 47: 226-238.

Tournas, V. H. and Katsoudas, E. J. (2008). Microbiological quality of various medicinal herbal teas and coffee Substitutes. *Microbiol. Insights*, 47-55.