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Assessment of mycoflora associated with postharvest losses of papaya fruits

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Market survey of Gorakhpur city for post-harvest fungal pathogens of papaya fruits revealed presence of *Alternaria alternata, Aspergillus flavus, A. niger, Botryodiplodia theobromae, Colletotrichum gloeosporioides, Curvularia lunata, Fusarium moniliforme, F. oxysporum, Penicillium expansum* and *Rhizopus stolonifer*. Results exhibited highest total incidence and abundance of *Aspergillus flavus* in rainy season (Jul-Oct) and *Fusarium moniliforme* during winter (Nov-Feb). Highest fungal incidence and rotting occurrence was during month of July-October being 44.39% and 17.02% respectively.

Key words: Disease abundance, incidence, papaya fruit, post-harvest fungal pathogens.

Introduction

Papaya (*Carica papaya* L.) of Caricaceae family is amongst various fruits cultivated commercially all over the country. It has high nutritional value (vitamin A & C, calcium, carbohydrates etc.) and great commercial potential. In India about 60.5 x 10^3 ha area is under papaya cultivation with estimated annual production of about 1,37,3001 mt/year and productivity of 22.51 mt /ha, (Ghosh 2000). Despite large acreage of land devoted to papaya the fruit loss is reported to be between 40-100 per cent of total annual produce (Sharma and Alam 1998). Most of the losses are due to fungal pathogens. These microorganisms invade the fruits and can cause considerable damage during transit, handling, storage and in the market thus rendering it unmarketable and reducing its nutritional and commercial value. Most of the investigations on post-harvest diseases of papaya have remained confined to mere reporting of the disease and its pathogens. Few included elaborate market study regarding annual disease incidence in terms of per cent fruit spoiled by rotting pathogens. Realizing the need for analysis of annual losses of papaya fruit in the region of

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eastern Uttar Pradesh; investigations were conducted in year 2003 in order to know the post-harvest fungal pathogens of papaya collected from the market of Gorakhpur city.

Materials and methods

Randomly selected eight retailers of Gorakhpur city were visited fortnightly which subjected to availability of fruit from January to May and July to December during 2003, to collect papaya fruits for examination. Total number of fruits sampled, types and number of disease symptoms shown on fruit surface, number of diseased fruits, severity of infection (approx area of fruit infested) and rotten fruits (commercially unviable and rejected ones) were taken into consideration. Degree of severity of particular fungus was qualitatively characterized as area of fruit covered by mycelia/symptom into; + = <10 %, ++ = 10 – 25 %, +++ = 25 – 50 % and ++++ = >50 % of area infested.

Some of the diseased fruits were brought in sterile polyethylene bags for further symptomatological studies, while fruits showing diseased patches in the market were selected and diseased tissue was scraped and kept in sterile filter paper bags. Diseased spots collected were numbered and brought in the lab for isolation and identification of pathogens. Fungal pathogens (on diseased tissue) were surface sterilized and transferred to CzapexDox Agar (CDA) medium slants. After 3 days new mycelia coming out of infected tissue were transferred to new CDA slants. Fungi were identified as per description in standard monograph. Pathogenicity tests were conducted on three fruits each against each isolated fungus by Granger and Horne (1924) method. Fruits were incubated at room temperature (28 ± 2^{o}C) for 5 days. The fungus was designated as pathogen only after satisfying Koch's postulate. Survey of local grown varieties along with transit ones was done. Monthly temperature (minimum and maximum) and relative humidity (RH) for the year 2003 were also recorded for epidemiological study following Lukose and Singh (1997). The data was pooled on monthly basis, analyzed and calculated in terms of per cent fungal incidence and abundance by the following formula.

Per cent fungal incidence = (Number of diseased fruits (by particular fungus)/ Total number of fruit sampled) x 100 (1)
Per cent fungal abundance = (Number of diseased fruits (by particular fungus)/ Total Number of diseased fruit) x 100 (2)

Results and discussions

Survey of fruits markets and recognition of major post-harvest pathogens of papaya fruits in Gorakhpur city (of Eastern U.P. region) is being reported for the first time in the present investigation. However, some studies have been carried out in other parts of India viz., Udaipur city (Gupta and Pathak, 1986), Jammu (Badyal, 1991), Coimbatore city (Pramod *et al.*, 2007) and Baroda city (Arya and Arya, 2007) regarding dominant post-harvest disease of stored papaya. During survey maximum disease per cent (50.99) in transit variety was recorded during rainy season, i.e. July to October (Table 1). Ten fungal species caused seven types of disease symptom in papaya fruits (Table 2). *Botryodiplodia theobromae, Fusarium moniliforme* and *Rhizopus stolonifer* wereobserved on fruits throughout the year, while *Penicillium expansum* and *Fusarium oxysporum* were observed for three months only (Table 3). Highest seasonal incidence (22.77%) was shown by *Aspergillus flavus* in the rainy season (Jul-Oct) in transit variety (Table-3). *Alternaria alternata, A. flavus* and *F. moniliforme* showed hundred per cent abundance in the month of July, April and December respectively in local variety (Table-4). Highest total seasonal per cent abundance was shown by *A. flavus* in rainy season (Jul-Oct) and by *F. moniliforme* in winter season (Nov-Feb) with maximum severity (Table-4). Maximum disease incidence (44.39%) and rotting (17.02%) was observed during rainy season (Jul-Oct) for all the papaya fruits sampled (Figure-1).

Table 1. Seasonal per cent deterioration of papaya fruit in the market

Season of survey	Variety of fruit	Number of retailers visited	Total fruit surveyed	Number of deteriorated fruit	Percentage of diseased fruits
SUMMER (Mar-Jun)	Local	4	56	12	21.42
	Transit	7	132	51	38.63
	Total	11	188	63	33.51
RAINY (Jul-Oct)	Local	5	118	47	39.83
	Transit	7	202	103	50.99
	Total	12	320	150	46.87
WINTER (Nov-Feb)	Local	8	158	39	24.68
	Transit	13	260	108	41.53
	Total	21	418	147	35.16

Table 2. Symptoms and diseases caused by different fungal pathogens in post-harvest papaya

Diseases	Pathogen	Symptom
Anthracnose	*Colletotrichum gloeosporioides*Penz.	Rot is soft; discolouration of skin develops in to circular light brown spots. At later stage lesion turns brown, sunken at centre with dot like black acervuli appearing on surface in concentric ring producing salmon pink spore.
Anthracnose/ Stem end rot	*Botryodiplodia theobromae* Pat.	Water soaked spots, which turn black with greenish irregular margin. Later in the centre dark green mycelial growth appear on surface.
Aspergillus rot/ Green mold rot	*Aspergillus flavus* Link	Initial small-scattered spots of infection, increases rapidly on general surface in a greenish yellow conidial mass covering whole surface of the fruit.
	Aspergillus niger Van Tiegh.	Small localized patches with black conidial heads. Do not spread rapidly but degrades the pulp rapidly and becomes sunken.
Fusarial rot/ soft rot	*Fusarium moniliforme*Sheld.	Soft rot, water soaked lesion enlarges rapidly and turns light brown, later centre of lesion gets depressed, white mycelial growth profuse, appear on surface.
	*Fusarium oxysporum*Schlecht.	Soft rot, white mycelial growth but less severe localized lesion/patches, leaves purple colour/patches below mycelium.
Penicillium rot/ blue mold rot	*Penicillium expansum* Link.	Initial small white suppressed mycelial outgrowth with beautiful greenish blue conidial compact mass, degrades the pulp quickly, disease localized at infection region, seldom covers whole fruit.
Soft white rot	*Rhizopus stolonifer* (Ehrenb. ex Fr.) Lind.	Soft water soaked lesions with spreading irregular margin. Later lesion increases and white mycelial growth and dark brown sporangiophores appear on surface. Fruit collapses, sometime mummifies releasing foul odour.
Sooty black rot	*Alternaria alternata* (Fr.) Keissler.	Brown spots change to grayish brown, dry in texture. Surface covered with dark brown conidiophores. Invade the pulp slightly.
	Curvularia lunata (Wakker) Boedijn.	Circular black spots, later olive brown mycelia appear on the surface. Restricted spots and invade the pulp slightly.

Table 3. Monthly per cent incidence of post-harvest fungal pathogens on *Carica papaya*

Fungal Species	Variety	Per Cent Incidence*										
		JAN	FEB	MAR	APR	MAY	JUL	AUG	SEP	OCT	NOV	DEC
Alternaria	Local	13.51	-	5.00	-	-	12.50	-	-	4.55	5.41	-
alternata	Transit	10.00	12.68	3.85	-	-	-	-	-	-	3.77	7.41
Aspergillus	Local	-	-	-	16.67	-	-	13.79	22.50	13.64	2.70	-
flavus	Transit	-	-	-	10.00	10.34	8.33	29.51	31.48	22.22	7.55	1.85
Aspergillus	Local	-	-	-	8.33	-	-	-	7.50	-	-	-
niger	Transit	-	-	-	2.50	6.90	8.33	6.56	1.85	4.44	1.89	-
Botryodiplodia	Local	10.81	-	-	-	-	-	3.45	-	-	-	-
theobromae	Transit	6.67	9.86	11.54	7.50	3.45	8.33	9.84	12.96	8.89	11.32	11.11
Colletotrichum	Local	13.51	2.56	10.00	-	-	-	10.34	22.50	-	8.11	7.50
gloeosporioides	Transit	10.00	7.04	-	-	-	-	13.11	18.52	6.67	5.66	7.41
Curvularia	Local	-	-	-	-	-	-	-	10.00	9.09	-	7.50
lunata	Transit	-	5.63	-	-	-	-	11.48	7.41	4.44	-	1.85
Fusarium	Local	13.51	17.95	5.00	8.33	9.09	12.50	6.90	15.00	13.64	21.62	15.00
moniliforme	Transit	21.67	15.49	9.62	5.00	-	25.00	6.56	24.07	15.56	20.75	24.07
Fusarium	Local	-	-	-	-	-	-	-	-	-	-	-
oxysporum	Transit	5.00	7.04	3.85	-	-	-	-	-	-	-	-
Penicillium	Local	-	2.56	-	-	-	-	-	-	-	-	5.00
expansum	Transit	1.67	1.41	-	-	-	-	-	-	-	-	1.85
Rhizopus	Local	-	2.56	-	-	9.09	-	3.45	5.00	9.09	10.81	2.50
stolonifer	Transit	1.67	1.41	9.62	17.50	10.34	16.67	1.64	7.41	33.33	1.89	-

- Fungus not reported

*Values are given as per cent of mean of all replicates

Table 4. Monthly per cent abundance of post-harvest fungal pathogens on *Carica papaya*

Fungal Species	Variety	Per Cent Abundance										
		JAN	FEB	MAR	APR	MAY	JUL	AUG	SEP	OCT	NOV	DEC
Alternaria alternata	Local	71.43 +++	- -	33.33 +	- -	- -	100.00 +	- -	- -	20.00 +++	16.67 +	- -
	Transit	28.57 +++	50.00 ++	11.76 ++	- -	- -	- -	- -	- -	- -	9.52 +	25.00 +
Aspergillus flavus	Local	- -	- -	- -	100.00 +	- -	- -	44.44 +++	69.23 ++++	60.00 +++	8.33 ++	- -
	Transit	- -	- -	- -	30.77 ++	30.00 +	25.00 ++++	78.26 ++++	70.83 ++++	55.56 +++	19.05 ++	6.25 +++
Aspergillus niger	Local	- -	- -	- -	50.00 +	- -	- -	- -	23.08 ++	- -	- -	- -
	Transit	- -	- -	- -	7.69 +++	20.00 +	25.00 ++	17.39 ++	4.71 +++	11.11 +++	4.76 +	- -
Botryodiplodia theobromae	Local	57.14 +++	- -	- -	- -	- -	- -	11.11 ++	- -	- -	- -	- -
	Transit	19.05 ++	38.89 ++	35.29 +++	23.08 +	10.00 ++	25.00 +	26.09 +++	29.17 +	22.22 +	28.57 ++	37.50 ++
Colletotrichum gloeosporioides	Local	71.43 ++	11.11 ++	66.67 +	- -	- -	- -	33.33 +	69.23 +	- -	25.00 +	50.00 ++
	Transit	28.57 +++	27.78 ++	- -	- -	- -	- -	34.78 +	41.67 ++	16.66 +	14.29 +	25.00 ++
Curvularia lunata	Local	- -	- -	- -	- -	- -	- -	- -	30.77 ++	40.00 +	- -	50.00 +
	Transit	- -	22.22 ++	- -	- -	- -	- -	30.43 ++	16.67 ++	11.11 +	- -	6.25 +
Fusariummonili forme	Local	71.43 +++	77.78 ++	33.33 +	50.00 +	40.00 +	100.00 +++	22.22 +	46.15 ++	60.00 ++	66.67 +++	100.00 +++
	Transit	61.90 +++	61.11 +++	29.41 ++	15.38 +++	- -	75.00 +	17.39 +	54.17 ++	38.89 ++	52.38 +++	81.25 ++++
Fusarium oxysporum	Local	- -	- -	- -	- -	- -	- -	- -	- -	- -	- -	- -
	Transit	14.29 +++	27.78 ++	11.76 +	- -	- -	- -	- -	- -	- -	- -	- -
Penicillium expansum	Local	- -	11.11 +	- -	- -	- -	- -	- -	- -	- -	- -	33.33 +++
	Transit	4.76 +++	5.56 +	- -	- -	- -	- -	- -	- -	- -	- -	6.25 +++

Rhizopus stolonifer	Local	-	11.11 ++	-	-	40.00 +	-	11.11 +	15.38 ++	40.00 +	33.33 ++	16.67 ++
	Transit	4.76 ++	5.56 +++	29.41 ++	53.85 ++	30.00 ++	50.00 +	4.35 ++	16.67 ++	83.33 ++	4.76 ++	- -

- Fungus not reported, Degree of severity caused by particular fungus; + = <10 %, ++ = 10 – 25 %, +++ = 25 – 50 %, ++++ = >50 %

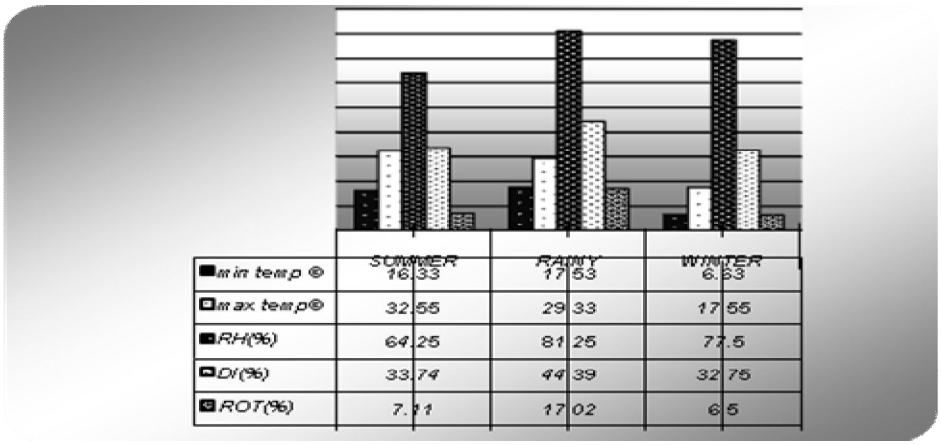

Fig. 1. Effect of temp and RH on disease incidence and rotting of post-harvest papaya

Study confirms the role of fungal pathogens in deterioration of papaya fruit during storage following observation of other workers, viz. *A. niger, F. moniliforme, Curvularia lunata* by Baiyewu and Amusa (2005);*Colletotrichumgloeosporioides* by Shivakumar *et al* (2002); *A. flavus*, *Botryodiplodia* sp, *Alternaria* sp and *Rhizopus* sp by Eckert and Ogawa (1985). Frequent prevalence of *F. moniliforme* on fruits throughout the year on papaya was reported by Baiyewu and Amusa (2005), in coherence with the present study.

Percentage of diseased fruits was greater in transit variety due to surface injury during fruit transport, handling and packaging as suggested by Paull *et al.* (1997). Increased fungal incidence and abundance in transit variety may be due to physiological state of fruits. Local varieties suffer less mishandling and are greener than transit ones which show fast ripening during transport. Thus local semi-ripe fruits show low pathogenic activity for most fungi due to low sugar content and less enzymatic activity in fruits (Adisa and Fajola 1983).

Temperature and relative humidity are important components of environment which affect sporulation and respiration of microbes. Results obtained from this study suggest that optimum temperature for rot ranged between 17-30^{o}C along with RH of 80%. This explains the increased rotting during rainy season with optimum environment. Less rotting in winter is seen,

as low temperature reduces respiration, sporulation and enzymatic degradation capacity of microbes. Present findings confirm the earlier reports of Lukose and Singh (1997) on pomegranate rot, Baiyewu and Amusa (2005) on papaya rot, Singh and Sumbali (2007) on apple rot; suggested maximum fungal rot between the temperature of 30-35^{o}C and 80% RH.

Thus above work signifies and confirms the role of fungal pathogens and climatic factors in commercial deterioration of papaya fruits. Prevalence of mycotoxigenic species viz., *Aspergillus flavus, Alternaria alternate ,Fusarium moniliforme*, and *Penicillium expansum* pose an additional health hazard for human consumption besides causing heavy commercial loss to retailers.

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