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Post harvest Diseases of Banana (*Musa paradisiaca* L.)- A Survey and Pathological Investigations

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ABSTRACT

A Survey for assessment of postharvest disease prevalence on banana was carried out during 2015-16 in Dharwad and Hubballi markets. pathogenic studies of postharvest diseases of banana were carried out in the Department of Plant Pathology, College of Agriculture, University of Agricultural Sciences, Dharwad, Karnataka. Predominant diseases identified on banana fruits in Dharwad and Hubballi markets were anthracnose (Colletotrichum musae) and crown rot (Fusarium semitectum), Finger rot (Lasiodiplodia theobromae) and cigar end rot (Verticillium theobromae) were observed in traces. Seasonal variation in disease incidence and disease severity were observed, the mean disease incidence of banana anthracnose was maximum during rainy season (17.53 % in Dharwad & 10.30 % in Hubballi) in all the varieties when compared to winter (7.16 % in Dharwad & 6.56 % in Hubballi) and very low incidence was observed during summer (2.25 % in Dharwad & 2.22 % in Hubballi). Disease severity (per cent disease index) of anthracnose was recorded maximum in rainy season which was more in Dharwad (20.56 %). It is evident that disease incidence was more in rainy season in both the varieties when compared to winter season.

Key words: *Banana, Fruit, Disease, Fungi, Potassium*

INTRODUCTION

Banana*"Musa paradisiaca* L." is one of the most popular fruits in India where it has been the food of sages since ancient times and important dessert fruit in India which provides a more balanced diet and is a good source of potassium. During storage, banana fruits deteriorate through the activity of microorganisms and their activity is favoured by the changing physiological state of the fruit. Post harvest diseases caused by various microbes have had considerable influence nutritive value, harvesting, transhipment and storage of fruits. Diseases like crown-rot, anthracnose, pitting disease, squirter disease, fruit rot, finger-stalk rot, brown specks on fruit, cigar-end rot of fruit and brown spot disease are reported from different parts of the world[19].

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The pathogenic fungi responsible for the market diseses were isolated and identified in various studies. In a survey carried out in Allahabad fruit markets, six fungal pathogens *viz*., *Fusarium moniliforme, Alternaria alternata, Aspergillus flavus, Curvularia lunata, Penicillium* sp. and *Cladosporium* sp. were found associated with postharvest diseases of banana. Maximum disease incidence was recorded with *F. moniliforme*[24]. Incidence and severity are influenced by weather factors too, Hence, variation in different seasons is reported by some researcher. Fruit rot of banana was observed with maximum disease incidence during the months September to June (37 %) in Navasari market of Gujarat. Isolations from the infected samples revealed that *L. theobromae* was the major fungal pathogen associated with finger rots followed by *Fusarium* sp. in few samples[13]. The pathogens associated with fruit rot of banana *viz., C. musae* and *Lasiodiplodia theobromae* were isolated by many workers[2,8,21] by following tissue isolation method on potato dextrose agar. The publication of several papers on Indian banana diseases by Indian and foreign pathologists have resulted in a steady progress in the research works of banana diseases. However, much attention need to be paid to the studies on the economic aspect of fungal diseases encountered during storage condition and it is necessary to stress upon finding the ways and means of saving the fruits from infection by various pathogenic microorganisms. In this connection, present investigation was undertaken to study the disease prevalence in Dharwad and Hubballi markets.

MATERIAL AND METHODS

Survey

Survey for assessment of postharvest diseases incidence on fruits was carried out during 2015-16 in Dharwad and Hubballi markets. Laboratory studies on isolation and identification of pathogens, morphological, studies of postharvest disease of banana were carried out in the Department of Plant Pathology, College of Agriculture, University of Agricultural Sciences, Dharwad, Karnataka. During survey, characterstic symptoms of each disease were recorded and also samples were collected separately in polythene bags and brought to the laboratory for isolation of pathogens associated with the diseases. Seasonal variation in disease incidence and disease severity were also assessed in present study by fortnightly visits to the markets during different seasons like winter, summer and rainy seasons of 2015-16 in 0-5 disease rating scale[16] *viz;* No symptoms on fruit surface(0), 0.1 – 5 % area covered by lesions(1), 5.1 – 10 % area covered by lesions(2), 10.1 – 25 % area covered by lesions(3), 25.1 – 50 % area covered by lesions(4) and >50 % of area covered by lesions(5).

$$\text{Per cent disease index (PDI)} = \frac{\text{Sum of individual rating}}{\text{No. of fruits assessed} \times \text{Maximum disease grade}} \times 100$$

Pathological studies

Pathogens were isolated by standard tissue isolation method. Single spore isolation was employed for purification of the isolates; pure cultures thus obtained were stored on potato dextrose agar slants for further studies. The cultures were identified based on colony characters, fruiting structurs and spore morphology. The characters such as size, shape of conidia, asexual fruiting bodies, cultural characters such as colony colour, growth rate and colony texture were considered for the identification. Spores were mixed with lactophenol thoroughly in order to obtain a uniform spread, on which cover slip was placed carefully for microscopic examination. *Fusarium* cultures were identified based on colony characters, pigmentation, shape and septation of macro and microconidia as per the standard description[11]. *Lasiodiplodia theobromae* cultures were identified based on colony

characters, fruiting structurs and spore morphology[20]. The average size of the spore like breadth and length were thus obtained under microscope. Microphotographs were taken to show the typical spore morphology of the pathogen.

Pathogenisity was proven by pinprick method**.** Banana fruits were thoroughly cleaned with sterile distilled water and then surface sterilised with 1.0 per cent NaClO for one min. Thereafter, they were wounded superficially with sterilized pins (pinprick method) and then were inoculated by smearing with spore suspension ($1x10^5$ spores/ml) or mycelial plug of *C. musae*. Other set of fruits were inoculated without wounding. The fruits thus inoculated were kept in moist chamber, to maintain high humidity. Control was maintained by treating with sterile distilled water instead of pathogen. Observations were made regularly for the appearance and development of symptoms. After symptom development, reisolation was done from the artificially infected fruits. The isolate obtained was compared with the original culture for confirmation. Similar procedure was followed for all the pathogens.

RESULTS AND DISCUSSION

It is known that the infection on banana like all other fruits that occur in the market stage is the result of contamination and infection that takes place during growing season or due to injury caused during harvesting, processing, packing and transporting. In consideration of the above, some markets of Hubballi and Dharwad were surveyed and infected bananas were collected to ascertain the pathogens involved and extent of damage they cause. Predominant diseases identified were anthracnose caused by *Colletotrichum musae* and crown rot caused by *Fusarium semitectum.* In addition, finger rot caused by *Lasiodiplodia theobromae* and cigar end rot caused by *Verticillium theobromae* were observed in traces in Hubballi market. However, more than one pathogen was also found to be associated with banana fruit rot in traces during survey in Dharwad. Other fungal pathogens isolated from completely rotten fruits include *Rhizopus* sp., *Fusarium* sp., *Alternaria* sp. and *Aspergillus* sp. Further it was observed during survey that, fully ripen fruits were showing rot symptoms frequently whereas green fruits did not show symptoms. The presence of inhibitors such as phenols in the unripe fruits might be responsible for retarding the growth of pathogens as reported earlier.

Regular survey was conducted for three seasons of the year to record the extent of deterioration in Cavendish, Ney poovan and Sugandhi varieties of banana in Hubballi and Dharwad markets. The mean disease incidence of banana anthracnose was maximum during rainy season (17.53 % in Dharwad & 10.30 % in Hubballi) in all the varieties when compared to winter (7.16 % in Dharwad & 6.56 % in Hubballi) and very low incidence was observed during summer (2.25 % in Dharwad & 2.22 % in Hubballi). Disease severity (per cent disease index) of anthracnose was recorded maximum in rainy season which was more in Dharwad (20.56 %) compared to Hubblli (15.80 %). It is evident that disease incidence was more in rainy season in both the varieties when compared to winter season. This may be attributed to the favourable environmental conditions to pathogen prevailing such as temperature and relative humidity in rainy season. These findings are in harmony with survey reports of Sarkar *et al*[22]., who concluded that maximum disease incidence of banana fruit rot was observed during August to February.

The disease incidence of anthracnose among the locally available banana cultivars has been observed to vary. Among the three varieties studied, Cavendish was recorded with higher incidence and severity. This is probably due to the biochemical composition of the peel of the different varieties and the enzymatic ability of the fungi to act on the peel, nutritional compounds (e.g. sugars and amino acids) originating from the host plant can have positive or negative influence on the germination, growth and other stages of the infection process of plant pathogens as opined by Carlile and Watkinson[4].

Crown rot disease was observed only during rainy season survey in all the varieties studied but not noticed in winter or summer in any of the variety. Among the varieties, Cavedish recorded more crown rot incidence (8.97 %) in Dharwad. While in Sugandhi 8.17 per cent incidence was recorded. Among the varieties, Cavendish recorded maximum disease severity of crown rot (31.42 %) which was observed during rainy season in Hubballi market. This may be due to favourable environmental condition which might have resulted in the build-up of inoculums and thus more the severity in rainy season. Seasonal variation in disease incidence was also demonstrated by Nath *et al*[13]., who concluded that fruit rot of banana was recorded maximum disease incidence during the months September to June (37 %) in Navasari market of Gujarat. Reason behind the proneness of Cavendish to crown rot might be the mode of storage as it is stored or transported to short distances after the hands were separated from bunch at crown region. The injuries made during separation of hands paves the way for entry of crown rot pathogen. Variation in the disease incidence in different varieties of banana was also discussed by De Costa and Chandima[5] and Rani and Thammaiah[18] in their investigations.

Symptomatology

Anthracnose, crown rot and finger rot were frequently observed during the present study. Different kinds of symptoms were observed in anthracnose in different varieties of banana. The first evidence of anthracnose symptoms on ripe fruits was appearance of circular to brown spots anywhere on the fruit surface but more frequently towards the flower end. Spots enlarged and coalesced on fruit to result irregular or elongated, deeply sunken lesions. At later stage, the whole fruits turned to brown. Black to brown acervuli were formed abundantly on the lesions with orange conidial mass. Similar observations were reported by Latiffah *et al*[10]., and Priyadarshanie and Vengadaramana[17]. Lesions had progressed quickly on injured fruits. Infected fruits showed black colouration of the skin and shrunken on severe infection. Another symptom of anthracnose; circular, light brown lesions with white to light pink fungal growth were seen in some samples of Cavendish and Ney poovan varieties. Black and dry rot starting from the flower end of fruits was the other symptom of anthracnose noticed only in fully ripe bananas of Ney poovan variety, which may be referred as blossom end rot symptom of anthracnose. These observations are in agreement with De Costa and Gunawardhana[6] who defined the anthracnose symptoms developed at the ripe stage on the bottom tip region of the fruit as blossom end rot. These differences may be attributed to different sources of infection, entry of pathogen as well as the composition and complex physiology of ripening in different varieties. These observations are supported by investigations of Nazriya *et al*[14]., who reported that *C. musae* has also been associated with blossom end rot, crown rot and tip rot symptoms in banana.

Symptoms of crown rot appeared as brown to black colour lesions on the crown region at the cut end. Frequently, whitish fungal growth later developed on the cut crown surface. The fungus penetrated deeply into the crown and the necks of the fingers, causing a dry, fibrous black rot. Rot was also noticed on fingers in some cases. From severely infected crowns, fingers detached prematurely. Disease was found with mature unripe as well as ripe fruits and progressed rapidly after fruit ripening.

Finger rot of banana initially visualised at the lower end of the fruit at the blossom end, the decay spread uniformly causing brownish black discoloration of the peel and softening of the pulp. The affected area of the peel became wrinkled and the pulp reduced to a soft, rotten mass and a dark grey mold grew on the peel surface under high humidity. Similar observations were made by Nath *et al*[13].

Pathological studies

The fungal systematics is still based mainly on morphological criteria as observable characteristics. Hence, fungi are recognized

and identified basically by their phenotypes. Simple morphological characteristics, such as conidial dimensions, appressorial dimensions, presence or absence of setae in acervulus and growth rate have been widely used as taxonomic criteria within the genus *Colletotrichum* in many studies[2,12]. During the present investigations, the fungal pathogens associated with the postharvest diseases of banana were isolated and identified based on their colony and spore morphology and the characters observed were compared with descriptions of standard literature available. Fungal pathogen cultures from anthracnose infected banana samples obtained by tissue isolation method were identified as *Colletotrichum musae* according to standard taxonomic key and description. Banana fruits exhibiting symptoms of crown rot were subjected to tissue isolations, the fungal colonies developed were identified as *Fusarium semitectum* in most of the samples collected from market. In other similar studies Zakaria *et al*[26]., and Jiménez *et al*[9]., reported that, crown rot which was caused by *F.semitectum* which is was isolated more frequently from crown rot infected bananas. The morphological and cultural characteristics observed were conformed to the standered descriptions[11].

Cultural studies of the isolated pathogens revealed that *C. musae* on potato dextrose agar produced salmon pink coloured colonies with white mycelial growth towards margins. Dark orange pigmentation on reverse side of the colony was also observed. Mycelium sparsely produced, hyphae septate and hyaline. Conidia were hyaline, aseptate, broadly elliptical or cylindrical with rounded ends. Setae were not observed in acervuli. The average size of conidia was 11.43- 16.27 x 3.86-5.47 µm. Conidia germinated and produced appressoria which were dark brown coloured and irregularly lobed. Findings of present study are in agreement with the results of Lim *et al*[12].,; Photita *et al*[15]., and Zakaria *et al*[27]. *F. semitectum* showed the growth of dense aerial mycelia on PDA and Richards's agar media, initially with white to cream and later light brown in colour. Dark brown pigmentation was observed on reverse of the culture plates. Microscopic examination revealed the presence of septate and hyaline hyphae. Macroconidia abundant, mostly straight or slightly curved, generally 3 - 5 septate and measured about 28.72- 37.23 x 4.24- 6.38 µm in size. microconidia these were single or two celled, measuring 7.56- 11.32 x 3.23- 4.85 µm. Chlamydospores single celled, occurred either singly or in chains, terminal or intercalary. These findings are found to in conformity with the reports Ingle and Rai[7] and Abd-Elsalam and Schnieder[1]. Fungus associated with banana finger rot was isolated and identified as *Lasiodiplodia theobromae* based on colony characters and spore morphology. Cultures of *B. theobromae* on PDA were quick growing, reached the brim of 90 mm Petriplate within 3-4 days after inoculation. Colour of the culture varied from grey, greyish white to greyish black in early growth stages but turned black after 20 to 30 days. Conidia were oval in shape, hyaline and single celled in the beginning and later turned dark brown and two celled, broadly rounded at the apex and truncate at the base. Similar observations were made by Shirshikar[23], Sangeetha *et al*[20]., and Suhanna *et al*[25].

Koch's postulates were proved for banana anthracnose by pinprick method. Lesions appeared as black necrotic, circular and sunken lesions, later elongated and sunken. Lesions showed fungal fructification of orange coloured conidial mass. Pulp beneath the lesions was also affected showing brown discolouration. Penetration by appressoria or the penetration peg might depend upon availability of natural or other openings. Some fungi even invade without any opening. Their capacity to penetrate depends upon their ability to dissolve the cell wall. In a study carried out by Ashwini *et al*[3]., demonstrated the pathogenicity test for banana anthracnose, inoculated fruits showed typical anthracnose symptoms after five to seven days. Similarly, Priyadarshanie and Vengadaramana[17] reported that charecterstic

symptoms of anthracnose started appearing on fruits after two days of inoculation.

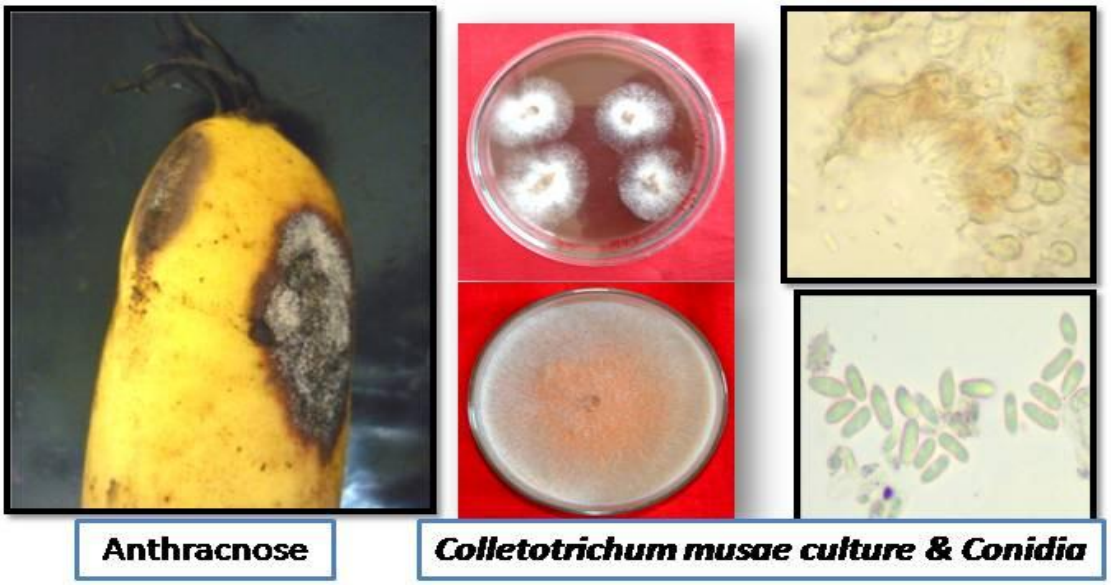
Pathogenicity test for crown rot and finger rot were also proved by pin prick inoculation method. Inoculated fingers showed symptoms after 4 to 5 days of incubation at room temperature. Symptoms of crown rot appeared as brown to black colour lesions on the crown region showing black dry rot of stalks which turned fibrous later. Symptoms of finger rot appeared as brownish black discoloration of the peel at flower end of the finger and a softening of the pulp was observed. Damaging postharvest diseases arise from infections initiated by mechanical and physiological injuries on the surface of the fruits; the injury created by severing the product from the plant is a frequent point of initiation of postharvest diseases by wound pathogens. Pathogenicity of crown rot pathogens in banana were proved by Subhash *et al*[24]., by wound inoculation (pin prick method) and obtained similar results.

Table 1: Prevalence of postharvest diseases of banana in Dharwad and Hubballi markets

	Variety	Percent Disease Incidence/ (Disease Severity)						Other diseases
		Rainy season		Winter		Summer		
		Anthracnose	Crown rot	Anthracnose	Crown rot	Anthracnose	Crown rot	
Dharwad	Cavendish	16.48 (23.60)*	8.97 (14.28)	8.23 (7.60)	0	3.31 (6.40)	0	**Mixed infections
	Ney poovan	18.57 (8.80)	3.12 (12.14)	6.09 (7.20)	0	1.19 (4.80)	0	
	Mean	17.53 (16.20)	6.05 (13.21)	7.16 (7.40)	0	2.25 (5.60)	0	
Hubballi	Cavendish	15.56 (26.80)	4.69 (31.42)	12.86 (7.20)	0	3.42 (5.60)	0	**Finger rot and cigar end rot
	Ney poovan	8.80 (11.80)	4.68 (16.42)	6.82 (4.40)	0	1.02 (3.20)	0	
	Sughandi	6.55 (8.80)	8.17 (19.82)	0	0	--	--	
	Mean	10.30 (15.85)	5.85 (22.55)	6.56 (3.87)	0.00	2.22 (4.40)	0.00	

*****Disease severity; 0 No disease--variety not available; ****** In traces

Postharvest diseases of banana observed during survey in Dharwad and Hubballi markets



CONCLUSION

An intensive survey was conducted in the markets of Dharwad and Hubballi revealed that anthracnose disease was noticed in all the varieties and both the markets surveyed. The mean disease incidence of banana anthracnose was maximum during rainy season in all the varieties when compared to winter and very low incidence was observed during summer. Disease severity of anthracnose was recorded maximum in rainy season.

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