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Variability in the Pathogen of Pineapple Disease of Sugarcane

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ABSTRACT

The organism *(Ceratocystis paradoxa)* of pineapple disease was isolated from 70.0, 75.3 and 64.5% samples of ungerminated setts, dead seedlings and root pieces respectively which were collected from Sreepur, Setabganj and SRTI farm. Two distinct strains, light and dark type of *C. paradoxa* were obtained, having micro and macro conidia in both the types measuring 12-14μm × 4-5μm and 15-18μm × 9-12μm respectively. When two isolates were grown together fertile perithecia were formed in 5 to 7 days, which (Perithecia) did not grow when cultured separately. The characteristic disease symptom developed in the inoculated setts using both the isolates and the pathogen was confirmed following Koch's postulates. Except *C. paradoxa*, other eight different fungi belonging to seven genera were also associated with the ungerminated setts, dead seedlings, roots and setts taken from apparently healthy standing cane.

INTRODUCTION

The causal fungus of pineapple disease, *Ceratocystis paradoxa* (de seynes) Moreau, generally invades sett pieces after planting in the soil, and causes the death of buds and young shoots. The disease was first reported from Java by Went (1893). Since then the disease has been reported from 39 out of 102 sugarcane growing countries in the world and cause considerable damage (Agnihotri, 1983). It has been reported on standing cane but such occurrence are not common (Edgerton, 1955). Generally standing stalks damaged by rats, borers, mechanical injuries or weakened by drought are infected by pineapple disease (Munjo, 1975). In Bangladesh pineapple disease was identified and fungus was isolated in 1976 from a newly introduced clones (Ahmed *et al.*, 1976). However, till 1987 pineapple disease could not be identified in any local varieties in Bangladesh, although gappy stands of cane in the field due to low germination and death of young plants in seed bed and in the field were common. Therefore, the present investigation was made to study the status of pineaple disease in Bangladesh.

MATERIALS AND METHODS

(a) In February, 1988 a total of 300 ungerminated setts and dead seedlings were collected from Sreepur, Setabganj and SRTI and throughly washed with tap water. About half a centimetre piece from each cut end of the setts was cut by sharp razor, and the pieces were taken out from the cut ends. Red lessioned root pieces were also taken. All these sett and root pieces were surface sterilized and plated on potato dextrose agar (PDA) medium.

(b) In January, 1988, the canes of the variety L. Jaba-C were collected from standing crop, and 200 two budded setts were prepared randomely and then observed the prevalence of fungi by the blotter method (Anonymous, 1976) with some modifications.

(c) Pathogenicity test was done with pure culture of both the isolates. In the middle of the two budded setts, 5 mm diameter were made and 0.5 cm mycelial block was inserted into each hole. The mycelial block were collected from 7 days old culture grown on agar medium. After inoculation through inserting the mycelial blocks, the holes were closed by cane pieces covered with insulating tape. In the case of control, similar treatments were done except inoculum. Both the cut ends of 150 setts of the variety L. Jaba-C were sealed by wax, and thus placed in metal tray filled with sands and periodic watering was done according to necessity.

RESULTS AND DISCUSSION

(a) The organism *(Ceratocystis paradoxa)* of pineapple disease was isolated from 70.0, 75.3 and 64.6% samples of ungerminated setts, dead seedling and root pieces respectively (Table 1, Fig. 1A). The fungus conitains micro condia (thin walled, cylindrical, size 12-14μm· x 4-5μm) and macro conidia (spherical in shape, size 15-18μm x 9-12μm) (Fig. 1.B). Two strains light and dark type were isolated from the setts of both varieties of L. Jaba-C and Misrimala collected from SRTI and Sreepur. Mono conidial culture of two isolates were prepared on potato dextrose agar (PDA) media (Fig. 2). When two isolates of *C. paradoxa* were grown together on PDA medium, fertile perithecia formed in 5 to 7 days at 28°C in the incubator. The base of the perithecia was pale brown, globose and immersed in the medium (Fig. 1. D), neck of the perthecia were black, measuring 1200 to 1300μm x 45 to 60μm in diameter and ostiole is covered with numerous pale brown erect tapering hyphae (Fig. 1. C).

Table 1. Fungi observed on the ungerminated setts, roots and seedlings collected from Sreepur, Setabganj and SRTI.

Fungi	Percent incidence on		
	Ungerminated setts	Dead seedlings	Roots
Aspergillus flavus	18.6	12.6	8.0
Aspergillus spp.	10.0	5.3	8.6
Curvularia lunata	11.3	7.3	2.0
Cladosporium sp.	4.6	-	1.3
Cephalosporium sacchari	36.6	54.0	76.6
Ceratocystis paradoxa	70.0	75.3	64.6
Fusarium moniliforme	10.0	21.3	6.0
Trichoderma viridae	46.0	28.6	17.3

150 samples were taken for each treatment.

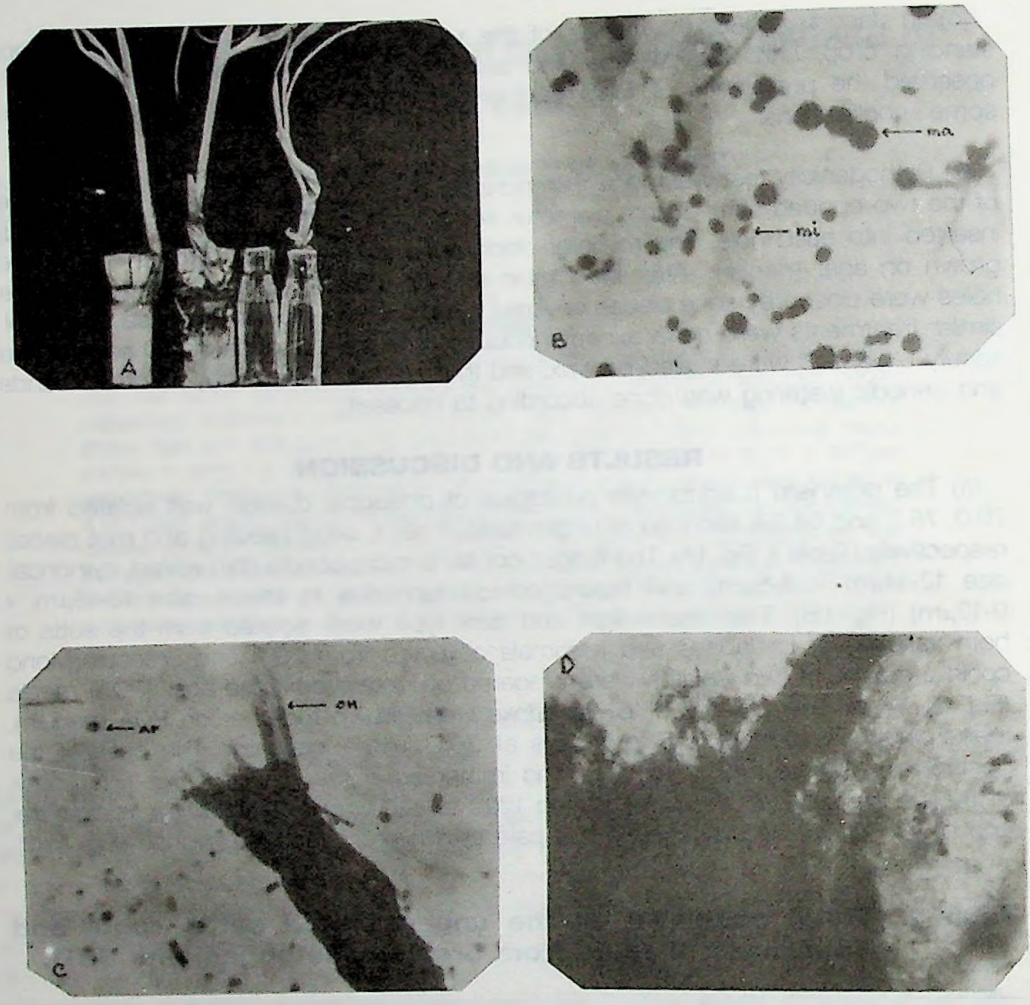

Figure 1. A. Dead seedlings affected by pineapple disease.
B. Micro and macro spores of *Ceratocystis paradoxa* : ma, chain of macrospores; mi, microspores.
C. Extrusion of ascospores from the ostiole of the perithecium, (OH, ostiolar hyphae, AP, ascospores).
D. Knobbed appendages at the base of the perithecium.

Ascospores were hyaline, ellipsoidal 8-10μm x 4-5μm in size. However, it has been reported that perithecia were not obtained when the dark or light type of isolates were grown separately (Liu *et al* 1971). Except *C. paradoxa*, other 7 different fungi belonging to 6 genera were also detected of which *Cephalosporium sacchari* was recorded in 36.6, 54.0 and 76.6% samples of ungerminated setts, dead seedlings and roots respectively.

(b) Thirty five percent of the setts collected from apparently healthy standing cane without any borer or rat damage were found infected with *C. paradoxa* when tested in the laboratory (Table 2, Fig. 3). So the disease is even present in the standing

Table 2. Incidence of different fungi colonizing the 200 setts collected from healthy standing crop.

Fungi	No. of fungal infection	% of total infection[1]	% of setts yielding[2]
Aspergillus flavus L	15	4.6	7.5
Aspergillus spp.	06	1.8	3.0
Alternaria tenuis C.G. Nees	12	3.7	6.0
Curvularia lunata (Wakker) Boedijin	18	5.6	9.0
Colletotrichum falcatum (Went)	05	1.5	2.5
Ceratocystis paradoxa (de syebes) Moreau	70	21.2	35.0
Cephalosporium spp.	123	38.3	61.5
Fusarium moniliforme sheldons	33	10.2	16.5
Trichoderma sp.	39	12.1	19.5

[1]Total number of fungal infections were 321.
[2]Percentage seed (setts) yielding different fungal organism was calculated on the basis of 200 setts.

Table 3. Germination percent and pineapple disease development after 60 days of inoculation.

Variety	Race	Setts inoculated (nos.)	Germination (%)	Disease development	
				Mortality of seedlings	Proved pathogenic (%)
L. Jaba-C	Dark type	50	60.6	24.6	100
	Light type	50	72.0	17.3	100
	Control	50	91.3	0.0	0.0

Figure 2. Dark and light strains of *Ceratocystis paradoxa* on potato dextrose agar medium. Letf to right.

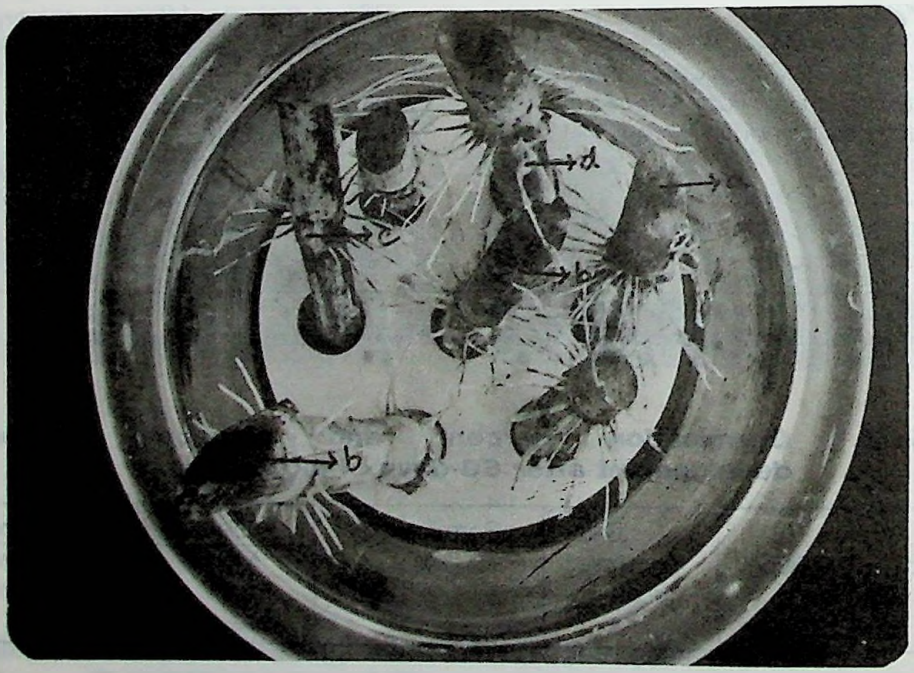

Figure 3. Disease development due to *Ceratocystis paradoxa*, *Cephalosporium sacchari* and *F. moniliforme* (setts taken from standing cane).
a : Light type *C. paradoxa* b : Dark type *C. paradoxa*
c : *Cephalosporium sacchari*, d : *Fusarium moniliforme*.

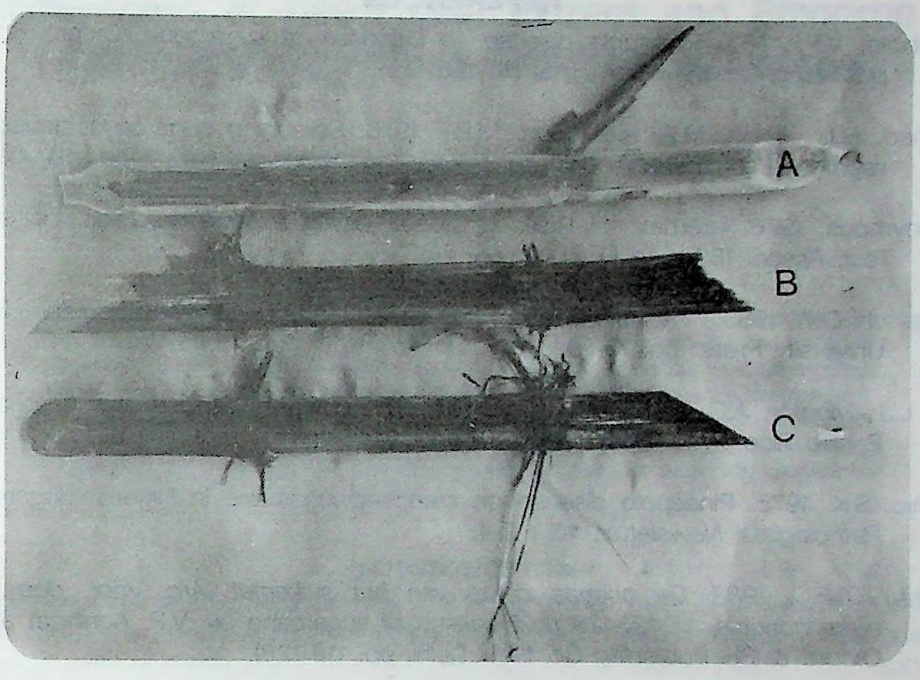

Figure 4. Internal symptom of pineapple disease after 60 days of inoculations. a : Control, b : Light type isolate was inoculated, c : Dark type isolate was inoculated.

crop. This ovservation has the confirmity with the experimental results obtained by Munjo (1975). Except. C. paradoxa, other 8 different fungi belonging to 7 genera were also associated with the setts. The most common fungi, in order of prevalence were *Cephalosporium sacchari, Ceratocystis paradoxa, Trichoderma* sp., *Fusarium moniliforme, Curvularia lunata, Aspergillus flavus, Alternaria tenuis, Aspergillus* spp. and *Colletotrichum falcatum* (Table 2). Among them *Cephalosporium sacchari* and *F. moniliforme* also causes sett rot, germination failure and seedling infection (Fig. 2).

(c) The characteristic disease symptom (black central cylinder) appeared on the inoculated setts using both the isolates, where no disease symptoms appeared on the control setts (Fig. 4). In the inoculated setts, germination was less (60.6-72.0%) compared to control (91.3%), and seedlings death were 24.6 and 17.3% against dark and light type of isolates respectively (Table 3). Re-isolation from the inoculated setts, which produced typical symptoms of pineapple disease yielded the same fungus. Two distinct isolates, dark and light type causing pineapple disease in the field, seed bed and standing crop is the first report of its kind in Bangladesh. However, Ahmed *et al.*, (1976) reported the disease in newly introduced clones.

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