Epidemiology of the Kresek Phase of Bacterial Blight of Rice

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ABSTRACT

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The incidence of kresek (wilting) increased with increased concentrations of bacterial inoculum (*Xanthomonas campestris* pv. *oryzae*) in rice cultivars Co 33, IR 8, and Taichung Native 1 (TN 1). However, even the lowest concentration of inoculum induced kresek in highly susceptible Co 33, but IR 8 and TN 1 were susceptible only at higher concentrations. Dipping the roots in the inoculum for a minimum of 5 min caused kresek, if the host and the pathogen were compatible. Isolates of *X. campestris* pv. *oryzae* from different areas in India varied in ability to induce kresek. Young seedlings were more susceptible than older plants. Ability to induce kresek increased when isolates were passed through host plants but declined with successive transfers on an artificial medium.

Xanthomonas campestris pv. *oryzae* (Ishiyama) Dye causes either leaf blight or wilting of rice (*Oryza sativa* L.) plants. Leaf blight is common in rice-growing countries of south and southeast Asia, and the wilting syndrome known as kresek occurs sporadically, causing serious damage in farm and experimental fields. Despite investigations of leaf blight (5,7,8,10), little information is available on kresek (3,6,9). We present information about the influence of certain factors on the development of kresek in rice plants.

MATERIALS AND METHODS

Plants. Seedlings of rice cultivars Co 33, IR 8, IR 20, TKM6, Chinsurah Boro II, and Taichung Native 1 (TN 1) were grown in 30-cm diameter earthen pots filled with 7 kg of alluvial field soil and watered regularly with tap water. Inoculated seedlings were transplanted in similar pots filled with soil submerged in water. They were fertilized with ammonium sulphate, 1 g/kg of soil.

Inoculation. Two isolates of bacterium from blighted leaf samples collected from Kota (Rajasthan) and Atanga (Orissa), maintained on potato-sucrose agar (PSA) were used, unless otherwise stated.

Seedlings of Co 33, IR 8, and TN 1, 15–20 days old, were uprooted and the roots washed thoroughly in tap water. The root tips were finely trimmed with a pair of sterilized scissors and dipped for 5, 30, 60, 120, or 240 min in a bacterial suspension containing 1.5×10^{10} cells/ml (O.D. 1 at 620 nm) prepared from a 48-hr-old culture grown on PSA (6).

Seedlings of Co 33, IR 8, and TN 1 were also inoculated with the bacterial suspension adjusted to optical densities of 1.0, 0.75, 0.5, 0.25, 0.125, 0.075, 0.05, 0.025, and 0.01, according to an Elico Model CL 24 Spectrophotometer (Elico Pvt Ltd., Hyderabad, India).

Observations were recorded on the percentage of plants with kresek 20 days after inoculation.

Pathogen variability. Seven isolates of the pathogen collected from different rice-growing regions of India were used to inoculate 20-day-old seedlings. The five rice cultivars used, Co 33, IR 8, IR 20, TKM 6, and Chinsurah Boro II, had differing degrees of susceptibility to kresek.

Observations on the percent incidence of kresek were recorded 20 days after inoculation.

Influence of host passage. The Kota isolate of the bacterium was inoculated on plants of Co 33 and TN 1 individually, and the bacterium was reisolated 10 days after each inoculation successively for three times. Twenty-five seedlings were inoculated at each time.

Virulence during storage. Two bacterial isolates were maintained on PSA and subcultured on the same medium for six generations. Isolates belonging to generations 1–6 were inoculated at the same time on Co 33 seedlings. The change in the virulence of the bacterium was based upon the percent incidence of kresek.

Effect of seedling age. Co 33 seedlings, 10, 20, 30, 45, and 60 days old, were grown separately by varying the dates of sowing. All the seedlings were inoculated with a bacterial isolate from the blighted leaf samples from Central Rice Research Institute. Inoculated seedlings were transplanted and the kresek incidence was recorded periodically until 20 days after inoculation.

RESULTS AND DISCUSSION

The susceptibility of three cultivars to kresek varied greatly. A 5-min contact between the roots and the bacterium caused 75–80% mortality in Co 33, 20% in IR 8, and 25% in TN 1. However, dipping roots for longer than 30 min increased the incidence of wilting in TN 1 and IR 8 (Table 1).

Younger seedlings succumbed to wilting (Table 2) faster than older plants did. For instance, 10-day-old seedlings succumbed to wilting as early as 8 days after inoculation. Symptom expression was considerably delayed in older seedlings, and the number of wilted

Table 1. Effect of duration of exposure of rice roots to *Xanthomonas campestris* pv. *oryzae* on the development of kresek

Exposure (min.) to	Percent incidence of kresek on cultivar		
	Co 33	IR 8	TN 1
Kota isolate			
5	80	20	25
30	85	30	40
60	90	50	60
120	90	50	60
240	100	75	50
Atanga isolate			
5	75	10	15
30	95	20	40
60	95	50	55
120	100	75	60
240	100	80	60

Table 2. Effect of age of rice seedlings on the development of kresek caused by *Xanthomonas campestris* pv. *oryzae*

Age of seedlings[a] (days)	Number of seedlings wilted (days after inoculation)				
	8	12	16	20	24
10	10	20	22	23	24
20	5	10	19	21	23
30	0	5	15	20	20
40	0	0	10	12	15
50	0	0	3	5	10
60	0	0	3	7	10

[a] 25 seedlings of each age were treated.

Table 3. Influence of subculturing of *Xanthomonas campestris* pv. *oryzae* on potato-sucrose agar on ability to induce kresek in rice cultivar Co 33

Bacterial isolate	Percent incidence of kresek after (no. of) subcultures					
	1	2	3	4	5	6
Atanga	95	75	30	15	0	0
Kota	90	50	20	0	0	0

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plants also decreased (4,9).

Inoculum concentration markedly influenced the wilting of rice plants, but cultivars responded differently. A bacterial suspension with a 0.5 O.D. appeared to be optimal to induce 100% mortality in the highly susceptible cultivar Co 33. Cultivars IR 8 and TN 1 showed maximum wilting only when inoculated with highest bacterial concentration of O.D. 1.0 (Fig. 1). The lowest concentration of bacterial inoculum that caused kresek varied with the isolate and rice cultivar (1,9). Co 33 appeared to be highly susceptible to both isolates; its seedlings wilted even at the lowest inoculum concentration.

Virulence of the bacterium was highly influenced by the number of subcultures made on PSA (Table 3). The percent of plants wilting decreased with an increased number of transfers on PSA. The fresh culture of the Atanga isolate caused 90–95% wilting, which decreased to 15% by the fourth transfer; the Kota isolate caused wilting only through three transfers.

Passing the bacterium through the host increased the virulence of the isolates (Table 4). Plants inoculated with isolates from kresek-infected plants showed more severe wilting than those inoculated with isolates from blighted leaves (2). Virulence of the Kota isolate increased greatly after it was passed through the host three times, resulting in 90% wilting in TN 1 and 100% in Co 33.

Rice cultivar TKM 6, which is resistant to leaf blight, had the lowest percentage of wilting after inoculations with all seven bacterial isolates, and Co 33 and IR 20 had the highest kresek incidence (Table 5). Isolates in Sri Lanka (9) and Philippines (2) differ widely in their ability to induce kresek. Of the various isolates used in our study, the isolate from Hooghly (West Bengal) was weakly virulent on all five test cultivars, causing only 24% mortality on the highly susceptible cultivar Co 33 and on IR 20 (Table 5). However, mortality was 40% in IR 8. Because IR 8 and Chinsurah Boro II responded differently to the various isolates, they could be used to differentiate the bacterial isolates.

Our study clearly indicates that factors such as virulence of the bacterium and the growth stages of the host cultivar greatly influence kresek incidence in rice. The bacterial isolates from different localities varied considerably in ability to induce kresek. These findings suggest that the kresek-inducing strains of the bacterium might occur in the pathogen population in nature, which results in the sporadic occurrence of wilting in fields when the compatibility between the host, pathogen, and the environment is favorable.

Table 4. Influence of host passage on ability of *Xanthomonas campestris* pv. *oryzae* to induce kresek on rice

No. of isolations from infected host	Percent incidence of kresek on	
	TN 1	Co 33
1	25	40
2	75	85
3	90	100

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Table 5. Variation among isolates of *Xanthomonas campestris* pv. *oryzae* in ability to induce kresek in rice

Origin of isolates (region)	Percent incidence of kresek on cultivars				
	Co 33	IR 8	IR 20	TKM 6	Chinsurah Boro II
Atanga	90	75	70	40	32
Cuttack	70	56	84	20	68
Hyderabad	48	20	36	10	16
Patna	40	32	48	28	48
Pusa	56	28	40	12	32
Hooghly	24	40	24	12	24
Kota	85	55	60	25	20

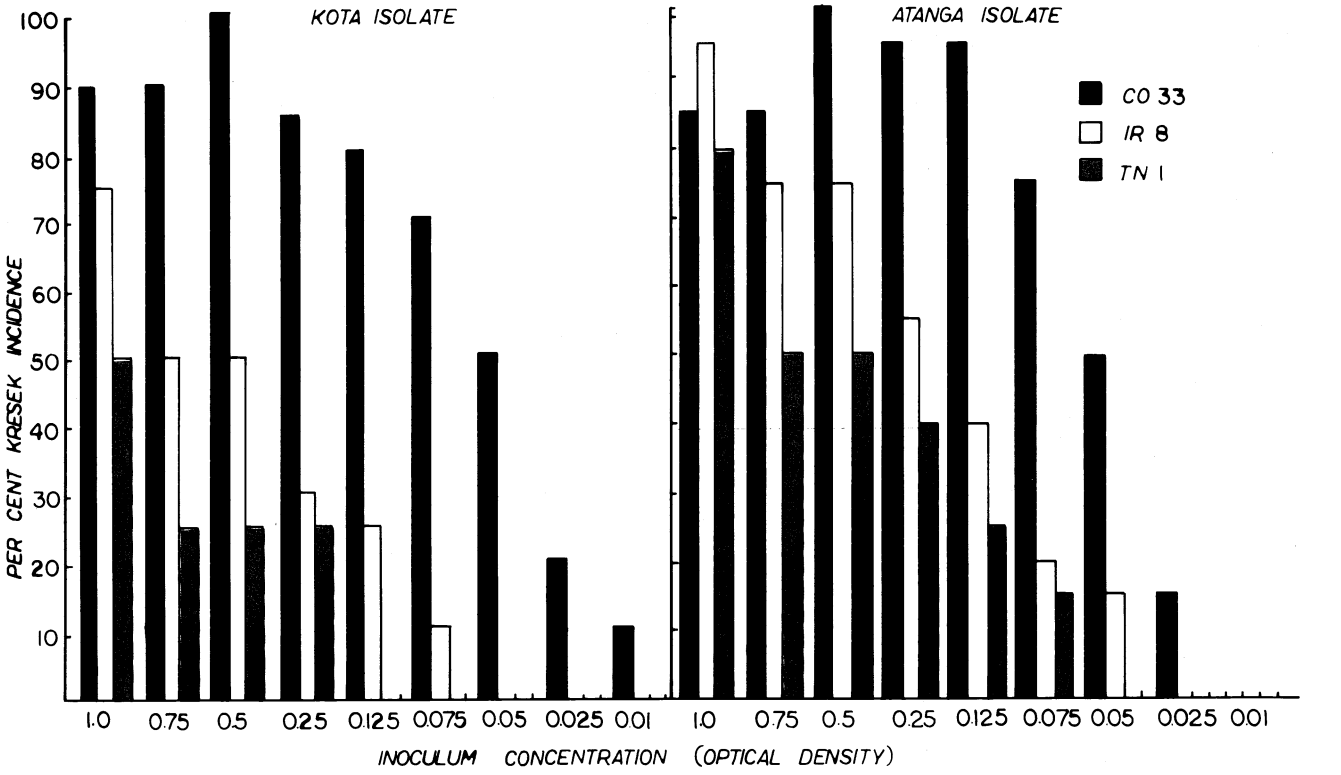

Fig. 1. Effect of inoculum concentrations of *Xanthomonas campestris* pv. *oryzae* on the incidence of kresek in rice seedlings.

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