

1. EPIDEMIOLOGY AND CONTROL OF BACTERIAL BLIGHT OF RICE IN INDIA

D. N. Srivastava*

Introduction

The bacterial blight disease of rice having been recognised in India recently, the information available on various aspects of the disease under Indian conditions is meagre. Between 1959–64, the disease was investigated in Maharashtra in a restricted manner under a Scheme of the Indian Council of Agricultural Research. Following the Bihar epidemic in 1963, researches were started at the Indian Agricultural Research Institute, New Delhi. The Central Rice Research Institute at Cuttack has also been working on its chemical control. No other State or institution in the country has yet started work on the disease. A point which deserves mention here is that the final report on investigations in Maharashtra (Anonymous, 1964) does not contain any description of symptoms of the disease. The bacterium studied was found to grow in a synthetic medium containing inorganic nitrogen source. The results of critical *in vitro* studies on the nitrogen utilization by *Xanthomonas oryzae* in Japan (Watanabe, 1963), in the Philippines (Anonymous, 1965) and in India (Mohan, 1966) have shown that the bacterium strictly requires organic nitrogen for growth. On the other hand, the streak pathogen, *X. oryzaicola* is reported to utilize inorganic nitrogen (Goto, 1964). It is not improbable that the investigators in Maharashtra were dealing with the streak pathogen and included its symptoms as part of the blight syndrome. The information on the distribution, losses, perpetuation and chemical control of the disease included in this paper from the Maharashtra report, therefore, needs to be viewed with reservations.

This paper presents the history, distribution and economic importance of the disease in India. Its epidemiology has been discussed with a view to highlight the difficulties in chemical control under Indian Conditions. Finally, the development and employment of resistant varieties to control the disease have been discussed with suggestions.

History

The first Indian record of the disease, based on isolation and pathogenicity of the bacterium, was by Sreenivasan et al (1959) from Maharashtra where it was reported to be widespread and destructive since 1951 (Bhaskar et al, 1960). However, the symptoms described by Sreenivasan et al conform to the bacterial leaf streak disease which is distinct from blight both in respect to the causal bacterium as well as symptoms on the host, but the symptoms published by Bhaskar et al considerably resemble those of bacterial blight. The disease was considered to be localised in Maharashtra until it broke out in an epidemic form in Shahabad district of Bihar in 1963. The specialists who were called upon to diagnose the Bihar epidemic speculated sulphide injury, an endoparasitic nematode, *Sclerotium oryzae* or a complex of these factors as possible cause of the disease. At this time Dr. T. Mizukami from Japan visited India for a short period on an invitation from the Indian Council of Agricultural Research. After a rapid survey of Shahabad, he declared that the disease was none else but

* Professor of Plant Pathology, Division of Mycology and Plant Pathology, Indian Agricultural Research Institute, New Delhi, India

bacterial blight incited by *X. oryzae*. He noticed the disease to be prevalent in certain other areas, including South India visited by him. However, the final acceptance of the bacterial nature of the Shahabad epidemic was delayed until Srivastava and Rao (1963) isolated the bacterium from the leaves collected by Dr. Mizukami from Shahabad and reproduced the disease by pure culture inoculation on healthy rice plants. It is important to note that, while the etiology of the disease was fully established in Japan 4 decades earlier and the disease was already under investigation in Maharashtra, there should have been confusion on its cause in Bihar. It shows a lopsided development of rice pathology in India with emphasis on mycological diseases only.

Impressed with the high yields of the nonlodging, dwarf, high nitrogen responsive, Formosan variety, Taichung Native-1, in small experiments during the winter of 1964-65, the Government of India extended its trial in small areas in farmers' fields at several locations in North and South during the Monsoon season of 1965. Despite subnormal rainfall during that year, the disease appeared in a severe form in most of such plots growing this variety. A similar picture was presented in 1966 with expanded cultivation of this variety over larger areas in several States. Thus, with the introduction of one susceptible variety, a disease which was unknown in all but two States earlier, became pan-epidemic within two years. At present the bacterial blight is a major hurdle in stepping up rice yields through intensive cultivation of high yielding varieties.

Distribution

Except for Maharashtra, where the endemic area was estimated to be about 0.5 per cent of a total of 1,885,099 acres (Anonymous, 1964), no survey has been made to estimate the area in other States. However, based on casual visits to some places or examination of diseased material received from different locations by the author and his colleagues, the prevalence of the disease has been fully established in most of the major rice growing states, including the Andaman Islands. The localities where the disease appears in severe form year after year are listed below Statewise:

Jammu & Kashmir	Ranbirsingh Pura
Punjab	Amritsar and Gurdaspur
Uttar Pradesh	Basti, Gorakhpur and Varanasi
Bihar	Shahabad
West Bengal	Chinsura
Tripura	Arundhatinagar
Assam	Silchar
Orissa	Cuttack and Sambalpur
Andhra Pradesh	East and West Godawari districts, Krishna and Hyderabad.
Madras	Tanjore
Maharashtra	Karjat, Thana, Ratnagiri and Nasik

The rice bowl of Chhatisgarh in Madhya Pradesh and the limited rice areas in Gujarat, Rajasthan, Himachal Pradesh and Haryana States have not yet been explored for bacterial blight. It will be seen that the endemic areas listed above include both single as well as double cropped ones. The disease is of importance only in the rainy season, but moderate to severe infections have been observed in the winter crop in the double cropped areas of Andhra Pradesh, Maharashtra and Madras.

Economic Importance

Unless the area affected and the severity of the disease are carefully assessed, it is dif-

difficult to estimate the extent of losses caused by the country as a whole. The damage estimated in Maharashtra on a three-year basis was 22.7 per cent, amounting to Rs. 541,510 (Anonymous, 1964). During the Monsoon season of 1965, yield trials of Taichung Native-1 at Maruteru in Andhra Pradesh provided an opportunity for assessing the damage due to the disease. Depending upon the severity and stage of infection, losses in grain yield were estimated to range from 6 to 60 per cent (Srivastava et al, 1966).

Epidemiology

The most important aspect of epidemiology which determines the success of prophylactic and chemical control measures against any disease, is the manner of perpetuation of pathogen from one season to another; in other words the sources which provide the primary inoculum. One or more of such agencies as seed, debris, weeds, soil and insects may serve this role. From the Japanese literature (Tagami and Mizukami, 1962), it is now established beyond doubt that the weed hostes *Leersia sayanuka* and *L. oryzoides* constitute the chief source of primary inoculum in Japan. Infected seeds, straw (stored in warehouses) and stubbles could also harbour the bacterium in viable form to the next season, but were not considered important. The bacterium in the soil was not found to remain active till the next season. Nothing seems to be known about the role of insects.

Before considering the role of the above agencies under Indian conditions, it will be pertinent to mention one of the important aspects of rice culture under diverse climatic conditions. In North and Central India, only one rainy season crop is taken, except for negligible areas where irrigation facilities and temperature conditions permit a second winter crop. On the other hand, in South India such as Kerala, Madras, Mysore and Andhra Pradesh and Parts of Orissa and Maharashtra, considerable area grows two crops. In the double cropped areas, the pathogen has been observed to perpetuate from season to season on the host itself. During October, 1965 the author observed in Maruteru, Andhra Pradesh a nursery of the winter crop growing in a corner of a field from which a severely blighted crop was harvested a fortnight earlier and, all around, severely infected crop was in the process of harvesting. Therefore, the perpetuation of the bacterium in the double cropped areas hardly needs any discussion.

The question of perpetuation of the pathogen in the vast stretches of the single cropped areas in North and Central India is an important one. Neither species of *Leersia* have been reported to occur in these areas. After rice is harvested by October or November, the fields remain exposed to severe winter between December and February and later the hot summer during May and June until the nurseries are sown with the onset of rains towards the end of June. During the last three years, investigations on the perpetuation of the bacterium through seeds, infected straw, stubbles, soil and weeds have been in progress at Delhi (Srivastava and Rao, 1967). Artificial epiphytic of the disease was reproduced by inoculating cultures of the Shahabad isolate on a 8-week crop of rice variety T.90 12'×8' plot during the monsoon season of 1964. The straw and the stubbles were left in the plot through the following winter and summer. Seeds of the same variety treated by the method of Srivastava and Rao (1964b) were sown in the same plot and watched for infection of bacterial blight throughout the season, but there was no appearance of the disease, suggesting that the infected debris and the soil did not perpetuate the pathogen. This, however, could not be taken as final, since only one isolate of the bacterium and only one variety of the host were employed in the experiment.

This experiment is being repeated this year from the debris of 128 rice varieties screened at Delhi under field conditions to 5 isolates of the bacterium in 1966.

Seeds from infected areas immediately after harvest were found to be heavily infected with the bacterium (Srivastava and Rao, 1964a). When pieces of husk from such seeds were

examined microscopically, bacterial streaming from the vascular strands was obvious. Sections of endosperms from such seeds on microscopic examination also showed bacterial streaming, suggesting that the infection was deep seated. Typical *Xanthomonas* from infected husk were isolated from several seed samples, but most of such isolates were either weakly pathogenic or avirulent. With ageing of the seed, the bacterial ooze from the husk was greatly reduced and from the endosperm it was not detectable. During the monsoon season of 1966, in two small pots well separated to prevent secondary infection and located far away from any rice field, 100 treated and 100 untreated seeds of Taichung Native-1 obtained from an infected field of the previous season in South India were seeded on July 1, 1966. Both plots received 60 lb each of Nitrogen and P_2O_5 in the form of ammonium sulphate and superphosphate. Irrigation was provided during dry spells of the monsoon. By September end, 10 plants raised from the untreated seeds showed mild infection of a few leaves each as against none in the treated one. The infected leaves showed typical bacterial streaming from the vascular ends when examined microscopically, but neither the bacterium could be isolated from such leaves nor the symptoms advanced to assume severe proportion. While the result of the above experiment suggested infected seeds as carriers of the bacterium, it would not be conclusive to state that infected seeds alone could lead to full fledged epidemic.

The experiment is being repeated this season with infected seeds of Taichung Native-1 collected from both the October as well as May harvests from South India. Investigations are also in progress to determine if the suckers and rhizomes of graminaceous weeds commonly found in paddy fields are capable of perpetuating the bacterium from one season to another.

According to the Maharashtra report (Anonymous, 1964), seeds carried deep seated endospermic infection and the bacterium could be isolated upto one year from such seeds as also from infected stubbles. No mention is made of the pathogenicity of the cultures so isolated. The bacterium could not be isolated from infected soil and none of the 30 grass hosts inoculated were found to be susceptible.

Chemical Control

In the light of the pattern of rice culture in the double cropped areas of South India, parts of Orissa and Maharashtra, any attempts to control the disease by seed treatment would be fruitless, since the active inoculum is always present on the host. Even in the single cropped areas of North and Central India, unless it is fully established that infected debris, soil and weed hosts do not play any role in the perpetuation of the bacterium, seed treatment would have little justification.

From the Japanese literature (Tagami and Mizukami, 1962), it is apparent that spraying of chemical including antibiotic at nursery stage is helpful in reducing the disease only when the nurseries are likely to harbour severe infection and secondary spread of the pathogen in the field is mild. The spraying of chemicals has been suggested to give better results in high concentrations once at the critical time just before manifestation of the disease, than frequently in dilute formulations at earlier stages. According to the Maharashtra report (Anonymous, 1964), copper fungicide containing 50 per cent metallic copper (copper oxychloride) was superior to other fungicides in reducing the disease significantly, the increase in grain yield being from 12 to 20 per cent. Jain et al (1966), working at the Central Rice Research Institute in Cuttack reported that dipping the seeds for 8 hours in 0.1 per cent Ceresan wet+Streptocyclin at 3.0 g in 25 gallons of water had significant effect in controlling the initial infection. They also reported a reduction of incidence of bacterial blight from 44.2 per cent in the control to 17.5 per cent by spraying copper, but copper was toxic to the variety used. Further, the spread of secondary infection could be checked to a large extent by spraying streptocycline (3.0 g in 25 gallons of water) in the variety CRT-141, while in Taichung Native-1 the second-

ary spread was checked only to a limited extent by antibiotic spray. As regards yield, the differences between the sprayed and unsprayed crops were not significant. Except for the above two references, there is no other report on the control of the disease by seed treatment or chemical spray in India.

In a recent article (Srivastava, 1966), the possibility of prophylactic and chemical control of the disease under Indian conditions has been critically reviewed. Theoretically, it is possible to reduce the incidence of the disease if in the single cropped areas the nursery bed is sterilised with formalin and treated seeds are employed to raise the nursery. However, it has to be borne in mind that India cultivates nearly 88 million acres of rice in the rainy season. The technical knowledge and resources of the farmers and the area involved become limiting factors in the practical application of such measures. The disease is a typical vascular wilt. Its control by spraying chemicals at field level would not constitute a logical approach. Further, during the monsoon season most of the areas in India have frequent heavy rains which can wash and drain away expensive sprays. Most of such areas get flooded and become physically inaccessible.

Control through Host Resistance

It hardly needs to be emphasized that a satisfactory control of bacterial blight is possible only through host resistance. Although, a good deal of work has been done in Japan (Tagami and Mizukami, 1962) on various aspects of host resistance, including screening of varieties, there is no evidence yet of resistance incorporated in commercial varieties either in Japan or elsewhere. According to recent result of large scale varietal screening at the IRRI (Anonymous, 1965), 2060 varieties out of 3275 tested (approximately 60 per cent) reacted as resistant. Twenty three varieties are listed resistant at both the seedling and flowering stages, besides another 21 varieties tested only at flowering stage and found to be resistant. The screening was done by inoculating cultures of the most virulent isolate available there or flag leaves injured by needle punctures. The rating of the reaction was based on a new scale extending from 0 to 9, 0 being immune and 9 the maximum limit of the disease.

During the monsoon of 1966, 128 varieties were screened against a mixed inoculum of 5 isolates of the bacterium under field conditions at Delhi by spray inoculation one month after seeding (Srivastava et al, 1967). Two months later, 104 varieties were rated as highly susceptible to susceptible by this method. The remainder 24, which took either traces of infection on leaf edges or failed to take infection were reinoculated on flag leaves injured by pin pricks. Of these, 13 reacted as resistant and 11 as moderately resistant. All the 39 commercial Indian varieties included in the test were found to be highly susceptible. Two exotic japonica varieties viz., Tainan-3 and Chianung-242, occupying some area in the South, reacted as resistant. Taichung-65 grown to some extent in Mysore State was moderately resistant. Two IRRI accessions 69/469 and 70/470 which reacted as resistant, have grains of the indica type while the grains of the remaining 11 resistant varieties, all from the IRRI are of the japonica type. The rating of the reaction was as follows:

- (1) Highly susceptible:
 - (a) Complete wilting of the plant (Kresiek phase).
 - (b) Complete blighting of the leaves without the Kresiek phase.
- (2) Susceptible: Partial blighting of the leaves.
- (3) Moderately resistant: Average lesion below the point of inoculation between 2 and 5 cm.
- (4) Resistant: Lesions confined to within 2 cm of the point of inoculation.

1 and 2 were rated on the basis of spray inoculation and 3 and 4 by inoculating individual leaves injured by pin pricks.

The fact that 81 per cent of the varieties reacted as susceptible to spray inoculation

shown that mass screening of varieties is possible at Delhi during the monsoon season (July to September), provided the rainfall is normal. From the practical angle, susceptible varieties can be easily scored out by this method and those showing any degree of resistance and be reevaluated by the more drastic method of inoculation on injured leaves within the same season.

Encouraged by last year's screening results, 1,500 varieties which include most of the commercial ones grown in India, are being screened in the current season at Delhi.

India's emergent need for host resistance against blight is complicated by its complete absence in the much preferred indica varieties now under cultivation. Under the leadership of Dr. M. S. Swaminathan, Director of the Indian Agricultural Research Institute, a large volume of irradiated indica material is being examined for blight resistance as a result of mutation.

From the screening work done in Japan, the Philippines and India, it is apparent that high degree of resistance is available in a large number of varieties, including some indica types. The incorporation of resistance in the agronomically desirable types is currently receiving attention of rice breeders. However, before proceeding further in this programme, it is imperative to examine the pathogen both in respect to variation in the virulence of isolates and specialised pathogenic races. From the Japanese literature (Tagami and Mizukami, 1962) and the work of Goto (1965) and Shekhawat (1966) in the Philippines and India, respectively, it is seen that isolates of the bacterium differ considerably in their degree of virulence. Goto has shown that the most virulent Japanese isolate when compared with the Philippine isolates was much less virulent. Therefore, the results of varietal screening in Japan may not hold good for the tropics having more virulent forms of the bacterium.

There is no published information yet from any country on the existence or otherwise of specialised pathogenic races in the bacterium. In a preliminary study (Srivastava et al, 1967), the pathogenicity of 5 isolates collected from different parts of India was separately assessed on each of the 5 varieties viz., B. R. 34, T. 90, Taichung Native-1, Taichung-65 and Tainnan-3 by inoculating on injured leaves, but no indication of any degree of host specialization in these isolates was available. Testing of larger number of isolates on a wider range of varieties would be necessary to come to any definite conclusion on this aspect. According to the Japanese literature (Tagami and Mizukami, 1962), none of the Japanese rice varieties are resistant to the most virulent isolates of Japan (A-Group of National Institute of Agricultural Sciences, A-1 Group of National Institute of Agricultural Sciences and 1-Group of Kyushu Agricultural Experiment Station). On the other hand 2060 rice varieties were found resistant at the IRRI when tested against isolate B-72 which is more virulent than any of the Japanese isolates. This is somewhat perplexing and leads one to suspect the possible existence of specialized pathogenic races in the bacterium between Japan and the Philippines. It becomes necessary, therefore, to quickly test a world collection of isolates for specialized pathogenic races. Cooperation of the interested countries will be necessary in pooling the isolates and selecting one or two isolated locations, preferably islands, for this programme.

Whatever be the race situation, the method of screening and evaluation of varieties needs to be standardized with respect to the virulence of the isolate employed, level of fertility at which the host has been grown, age of the host, technique of inoculation, period after which observation is taken and the scale of rating etc. This may enable a better comparison of results from different sources.

Discussion

K. Goto, Japan: Did you examine grasses of *Leersia* spp. as common host of this pathogen?

Answer: So far as I am aware, neither *Leersia oryziodes* nor *L. sayanuka* are reported to occur in the endemic areas in India.

N. Murata, Japan: Does fertilizer response of varieties in general sense correlate with non-severeness of the disease upon application of nitrogen?

Answer: If I correctly understand your question, you want to know if nitrogen fertilizers aggravate the disease regardless of the variety. My answer is yes.

Y. Tagami, Japan: I want you to make a little correction about our experimental results concerning rice stubbles in Dr. Srivastava's lecture.

Answer: I thank you for suggesting the correction that rice stubbles in Japan over-winter the bacterium. This will be incorporated in the text of the paper.

S. Yoshimura, Japan: (1) You have mentioned, *X. oryzae* was isolated from endosperm. Was it from starchy endosperm or embryo? And in this case did the invasion of the bacteria occur from the wound part of the grain or vascular bundle of pedicel? (2) In your test of inoculation against seed, why was infection not shown in the treated seed plot? (3) Could you let me know the method of your seed inoculation correctly? In such case, what is the relation to *X. oryzicola*?

Answers: (1) The infection of the endosperm was of the starchy endosperm, but the embryonic tissues were not examined critically for infection. They could also be infected. Whether this infection was the result of systemic invasion through the vascular tissues of the pedicel or direct infection of the seed before fertilization was not investigated. (2) I believe you want to know why the plot sown with treated seeds in our experiment on field transmission of the disease through infected seeds did not show the bacterial blight. My answer is that the bacterium in the treated seeds was inactivated as a result of the treatment and hence the disease did not develop in the plot sown with such seeds. (3) I do not understand what you mean by the first of the third question. As discussed earlier, the seeds get infected either systemically by the bacteria coursing through the vascular bundles of the pedicel or the bacteria present externally in the water film on glumes gain access into the seed before fertilization. Both possibilities are there. If your question refers to the method of detecting seed infection, this was done primarily by examining pieces of the husk and sections of the endosperm under the microscope for bacterial streaming from the vascular fibrils. As regards infection of the seed by the leaf streak pathogen, *X. oryzicola*, we do not yet know whether this is a seed borne bacterium. In fact nothing is known about its disease cycle.

E. C. Cada, Philippines: At what stage or stages of growth of the plant is leaf blight observed seriously?

Answer: Severe manifestation of the disease under Indian conditions generally takes place around the booting or flowering stages.

S. Wakimoto, Japan: How do you evaluate the phage method or multi-pin prick method for the detection of causal bacteria in your country?

Answer: Until recently we were using inoculation of leaves with macerated suspension of infected tissues or isolated cultures by the pin prick method for detection of bacteria in infected plant parts. We have established several strains of bacteriophages recently which are also helpful in detecting the presence of bacteria in infected plant parts or seeds.

Comments from S. Yoshimura, Japan: The possibility of seed transmission is high, although it is not thoroughly witnessed in Japan. It is said that the bacteria were isolated from the rice seed in India. In this regard, it is necessary to clarify if the bacteria were isolated from Lemma or Palae or from the inside of the grain, such as Embryo, Endosperm and Aleurone layer. It is still more necessary to make it clear that if the bacteria penetrated through wounds or through the vascular tube of Pedicel into embryo or endosperm. If so, they should

be traced.

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