

occurs followed by desiccation and death of the above ground portion. In case of severe infection even the underground stem dies resulting in thin crop stand and considerable decrease in yield. The main effect of the disease is evident on underground stolons. The symptoms on the underground stolons consist of pinkish-brown lesions in the earlier stage of the disease which gradually turn into dark brown-black patches. These lesions increase in size finally resulting into soft decay of the entire branch of the affected stolons.

A large number of isolations were made from infected stolons and wilted plants. Isolations from young lesions and infected plants mostly yielded *Rhizoctonia* whereas old and soft lesions gave a mixture of *Rhizoctonia* and species of *Fusarium*.

In order to test the pathogenicity, both dormant stolons as well as the intact potted plants were inoculated with various isolates. Stolons were inoculated by placing a drop of spore suspension or mycelial bit on a small wound made by a sterile scalpel. To test the pathogenicity on intact plants, stolons were planted in steam sterilized soil. After about 8 weeks the plants were inoculated by pouring a suspension of mycelium and spores near the base of the plants, stolons of which were injured slightly with a scalpel.

Three different species of *Fusarium* isolated from decaying dormant stolons proved non-pathogenic both to dormant stolons as well as to potted plants. All the isolates of *Rhizoctonia* produced characteristic lesions on the stolons and also caused wilting of potted plants (Fig. 1). The fungus was re-isolated from all the inoculated stolons. Inoculation of intact potted plants during the month of July resulted in yellowing and wilting of the plants six days after inoculation. When the infected plants were removed from the pots and the soil washed away from the roots, most of the stolons and roots were found to be decayed by the fungus. Isolation from these rotten stolons gave a pure culture of *Rhizoctonia*. The isolate of *Rhizoctonia* which has been found to be pathogenic has been identified as *Macrophomina phaseoli* (Maubl) Ashby on the basis of pycnidial characters.

These observations prove that the stolon rot of Japanese mint in India is caused by *Rhizoctonia bataticola* (Taub) Butler [*Macrophomina phaseoli* (Maubl) Ashby]. A similar disease of peppermint (*Mentha piperita*) and spear mint (*Mentha spicata*) has been reported by Green¹ from United States. He isolated species of

Rhizoctonia and *Fusarium* from the diseased stolons. However, the pathogenicity of these organisms was not established, and this is the first report of the establishment of the casual organism of stolon rot of Japanese mint.



FIG. 1. Healthy and wilted mint plants inoculated with *Macrophomina phaseoli* (Maubl) Ashby.

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Regional Research

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BACTERIZATION OF RICE AND OKRA SEEDS WITH *AZOTOBACTER CHROOCOCCUM* AND ESTABLISHMENT OF THE BACTERIUM IN THE RHIZOSPHERES

BACTERIZATION of seeds of some plant species with *Azotobacter* and its establishment in the rhizosphere have been studied by some workers.¹⁻⁶ The results obtained so far are not conclusive on the role of *Azotobacter* in plant rhizosphere. Studies were made by the authors to examine the possibility of establishing *A. chroococcum* in the rhizospheres of rice and

Okra (*Hibiscus esculentus*) by pre-treating the seeds with the bacterial culture and the results are reported here.

An isolate of *A. chroococcum* obtained from the Culture Collections of the Department of Agriculture, Annamalai University, was used in these studies. Surface sterilized rice and okra seeds were soaked for 18 hr. in a thick suspension in sterile distilled water of the bacterial cells. The suspension contained 82 million bacterial cells/ml. and after treatment the rice seeds carried 7,000 cells/gm. of seed on dry weight basis and the okra seeds 16,000 cells/gm. The treated seeds were dried in shade and sown in sterile or unsterile soil contained in 12 inch

pots. The soil was a clayey loam, with pH 7.2, collected from the University Experimental Farm. Both the treated and untreated seeds were sown in different sets of soils and the plants allowed to grow under identical conditions. The rhizosphere samples from each treatment were obtained at periodical intervals and the microbial population estimated following the procedure of Timonin.⁷ The *Azotobacter* population was estimated using the nitrogen-free medium No. 77 (Allen⁸). The data on the *Azotobacter* population in rice rhizosphere are given in Table I and those of okra in Table II.

The results indicate that *A. chroococcum* could establish and multiply in sterile and

TABLE I
Azotobacter population in the rhizosphere of rice crop arising from seeds pre-treated with *A. chroococcum*
(Population expressed as 10^5 /gm. of moisture-free soil)

Treatment	Age of the crop in days											
	10		20		30		40		50		60	
	P	R:S	P	R:S	P	R:S	P	R:S	P	R:S	P	R:S
Treated seeds sown in sterile soil:												
Rhizosphere	9.8	6.1	26.0	13.7	115.0	38.3	180.0	45.0	200.0	48.9	130.0	43.3
Soil	1.6		1.9		3.0		4.0		4.1		3.0	
Treated seeds sown in unsterile soil:												
Rhizosphere	2.5	2.1	20.5	11.4	85.0	34.0	96.0	36.9	123.0	41.0	88.0	35.2
Soil	1.2		1.8		2.5		2.6		3.0		2.5	
Untreated seeds sown in unsterile soil:												
Rhizosphere	1.8	6.0	10.8	27.0	18.5	30.8	23.0	33.0	28.0	35.0	30.6	33.3
Soil	0.3		0.4		0.6		0.7		0.8		0.9	

P = Population
R : S = Rhizosphere-Soil ratio

TABLE II
Azotobacter population in the rhizosphere of okra crop arising from seeds pre-treated with *A. chroococcum*
(Population expressed as 10^6 /gm. of moisture free soil)

Treatment	Age of the crop in days											
	10		20		30		40		50		60	
	P	R:S	P	R:S	P	R:S	P	R:S	P	R:S	P	R:S
Treated seeds sown in sterile soil:												
Rhizosphere	12.0	12.0	64.0	35.6	220.0	55.0	215.0	52.4	167.0	47.7	75.0	37.5
Soil	1.0		1.8		4.1		4.1		3.5		2.0	
Treated seeds sown in unsterile soil:												
Rhizosphere	3.0	6.0	15.0	12.5	92.0	46.0	108.0	45.0	95.0	43.2	42.0	35.0
Soil	0.5		1.2		2.0		2.4		2.2		1.2	
Untreated seeds sown in unsterile soil:												
Rhizosphere	2.1	7.0	10.4	26.0	17.3	34.6	22.5	37.5	28.0	40.0	25.0	35.7
Soil	0.3		0.4		0.5		0.6		0.7		0.7	

P = Population
R : S = Rhizosphere soil ratio

unsterile soils. It could also establish in rhizospheres of rice and okra, growing in sterile and unsterile soils. In the two rhizospheres *Azotobacter* population increased in the early stages and dropped after 50 days in rice and after 40 days in okra. In the untreated check plants there were fewer *Azotobacter* populations, which did not decline much with age of the plants. Also the *Azotobacter* population got more readily established in sterile soil and in the rhizosphere of plants growing in sterile soil than in the unsterile soil.

These results indicate that *A. chroococcum* could be established in plant rhizosphere through the seed. Being present in the rhizosphere region in considerable numbers, it could influence the plant growth either directly or indirectly. It is interesting to note, however, that the normal *Azotobacter* flora of the region did not reduce with plant age, whereas the introduced *A. chroococcum* reduced with age. These results appear to support the findings of Tribunskaya.³ According to Clark⁴ the *Azotobacter* population in tomato rhizosphere declined rapidly, whereas Daste⁶ found that *Azotobacter* multiplied for sometime in the rhizosphere followed by a progressive decline. Zinoveva¹⁰ and Nalivaiko and Romeiko¹¹ obtained results to support the establishment of *Azotobacter* in the rhizosphere of some plants. According to Federov and Tepper² the failure of *Azotobacter* inoculations in the rhizosphere is due to either insufficient excretion of carbonaceous materials from the roots or due to other unfavourable conditions for the growth of the organisms, the chief determining factors being the activity of the plant root system and the bacterial physiology. Studies on the influence of plant root excretions on *Azotobacter* population are needed for a better understanding.

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GALL INDUCTION ON *CYNODON* *DACTYLON* BY *USTILAGO*

Cynodon dactylon (Dubgrass) is the common lawn grass of Delhi. Generally its inflorescence comprises four digitate fascicles which are slender and smooth. In this area it is frequently found infected by *Ustilago cynodontis* (see also Chona et al.²). The fungus is systemic in the host but produces smut spores mostly in the inflorescence¹ which shows various degrees of transformation. Of the four fascicles only one, two or three may be attacked by the fungus. Occasionally only a part of the fascicle shows smut spores and the remaining part bears normal flowers (see Mehta³). The smut sori frequently extend to the stalk of the inflorescence and the flag leaf. In short, the reaction of the host to this fungus is extremely variable.

To this variety of responses, I wish to add a hitherto unrecorded response the formation of green galls in the inflorescence. Only the fascicles are involved in gall formation and the inflorescence axis and the flag leaf remain unaffected. Galls were noticed in a small patch of grass in the Delhi University Campus, during the rainy season of 1964. The specimens represented in Figs. 1-6 were collected on August 20, 1964. Figure 1 shows an infected inflorescence with the usual symptoms. All the four fascicles have retained their slender appearance and bear smut sori. In Fig. 2 only one of the fascicles is seen turned into a gall while the other three are underdeveloped and uninfected. Generally all the four fascicles are transformed into short and thick galls (Figs. 3, 6). Occasionally the tip of the inflorescence axis bears only three galls (Fig. 4). A gall is an oval structure (1-2 cm. long and 3-5 mm. in diameter) consisting of a few thick glumes enclosing a mass of teliospores. The glumes are leaf-like and are even differentiated into the sheath and the lamina (Fig. 3). The covering glumes separate from each other or degenerate at places (Fig. 6) thus exposing the spore mass for dispersal by wind. The gall represented in Fig. 5 is exceptional in that the fascicles and the glumes cannot be made out and the entire inflorescence is represented by