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PATHOTYPE DELINEATION IN *RHIZOCTONIA SOLANI* ANASTOMOSIS GROUP ONE CAUSING FOLIAR BLIGHTS IN MUNGBEAN

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ABSTRACT
Web blight caused by *Rhizoctonia solani* anastomosis group 1, intraspecific group IB was the major pathotype in Eastern Uttar Pradesh and causing an average yield loss up to 40% in both the years. Web blight pathotype differs from aerial blight which was initially thought to be misnomer. Isolates causing web blight of mungbean fused only with tester isolates AG-1IA and AG-1IB but not with AG-1IC. All isolates identified in samples from various locations were *R. solani* AG-1. Isolates of *R. solani* collected from mungbean causing web blight symptoms, exhibited C2 reaction (imperfect fusion or killing reaction) with AG-1IA and AG-1IB tester isolates and C1 type reaction with AG-1IC. Therefore, the web blight isolates were identified to belong to AG-1IB where as aerial blight isolates belong to AG -1-IA. Sneh *et al.*(1996), reported that *R. solani* AG-1IC causes damping off and crown root rot in Buckwheat, Carrot, Soybean, Flax and Pine, hence, it is clearly indicated that aerial blight isolates of mungbean have no possibility to belong to AG-1IC.

INTRODUCTION

Mungbean (*Vigna radiata* L.) is an important pulse crop, cultivated as a sole or mixed crop across the country. The major impediment in the north India against successful cultivation of green gram is the severe occurrence of stem and leaves blight caused by a soil borne common and wide spread fungal pathogen i.e. *Rhizoctonia solani* (Kühn).The host range of the organism is very high which encompasses almost all the plant families and more than three hundred plant genera. As a matter of fact that *R solani* is consists of various intra specific groups which are otherwise identical but differs in their pathogenicity potential & symptomatic behavior. The web blight and aerial blight symptoms which were initially thought to be misnomer are now differentiated as two distinct pathotypes. In a study O'Neill *et al.* (1977), concluded that aerial blight type characterized by the production of sasakii type sclerotia on diseased tissue or in the culture while, web blight is characterized by the production of abundant microsclerotia on diseased tissue during growing season. On the basis of symptoms Rhizoctonia foliar blight had been divided into two types. The first one is aerial blight caused by *R. solani* AG-1IA and the other one is web blight caused by *R. solani* AG-1IB. Ogoshi (1987) suggested that based on pathogenicity, AG-1 can be divided into at least three subgroups IA, IB and IC. Isolates of AG-1IB (the Web blight fungus) are also virulent on rice but causes different type of symptom on it than do the isolates of AG-1IA *Rhizoctonia solani,* the casual agent of Rice sheath blight. Classification of different types of Rhizoctonia foliar blights of mungbean has not been studied till now in India. Dwivedi and Saksena (1974) reported for the first time that the disease is caused by *Rhizoconia solani,* but so far no efforts were made to determine the prevalence or disease distribution of web blight type. Furthermore, it is not clear that which one of the two intraspecific groups, AG-1IB or AG-1IC, is the primary causal agent of mungbean in India. It is necessary therefore, to clarify the type and the causal agent(s) of Rhizoctonia foliar blights in India. Owing to which the objective of present investigation is to establish the pathotype variability prevalent in Rhizoctonia foliar blight of mungbean, on the basis of cultural characteristics and anastomosis grouping which is supposed to be a very objective parameter.

MATERIALS AND METHODS

Survey were conducted through the methods described by Singh *et al.* (2003), during II[nd] to IV[th] week of August, 2007 and 2008 for incidence of different type of Rhizoctonia foliar blight of mungbean, at twenty five locations of five districts in Eastern Uttar Pradesh viz., Azamgarh, Ghazipur, Jaunpur, Sant Ravidas Nagar and Varanasi, where mungbean is a major crop and Rhizoctonia foliar blight was supposed to be the major problem. At each location at least three fields were sampled. Because web blight can be readily identified by the presence of microsclerotia on diseased tissue(Yang *et al.*, 1990), the

web blight (WB) and aerial blight (AB) samples were collected separately and brought to the laboratory for isolation and identification of fungi.

Morphological identification was conducted following the description of *R. solani* (Parmeter and Whiteny, 1969). Foliar blight isolate of *R. solani* were grown on 2% water agar (WA) and examined that if microsclertia were produced, it was identified as the web blight pathogen. If the isolate did not produced microsclerotia, the causal agent was considered to be the aerial blight pathogen (Yang *et al.*, 1990). The frequency of web blight and aerial blight in each field was calculated by the number of disease foci of each type by the total number of disease foci observed.

Anastomosis test were conducted following the procedure described by Parmeter *et al.*(1969) . If an isolates anstomosed a tester (known) isolates of *R. solani* AG-1 (IA, IB and IC)? Five slides for each of the anastomosis test was examined for different type of anastomosis reaction (C 0 to C 3 type) as given by Mac-Nish *et al.*(1993) and Cubeta and Vilgalys (1997).

Reaction	Phenotype	Nature of genetic relationship between isolates
C0- No interaction	Hyphae grow and pass each other no recognition	Isolates have no genetic relationship and belong to different AG
C1- Hyphal contact only	No evidence of wall or membrane contact, reaction may or may not be accompanied by cell death	Isolates have a distant genetic relationship and belong to either the same or different AG
C2-Killing	Wall fusion (anastomosis) evident, with cell death of anastomosis and adjacent cells, somatic incompatibility response often macroscopically visible	Isolates represent genetically distinct individual that belong to the same AG
C 3 - Perfect fusion	Wall and membrane fusion evident, point of anastomosis not clearly visible, cell death absent	Isolates are genetically identical or closely related, individuals belong to the same AG and may represent clones

In *R. solani*, branching habit indicates the direction of growth of the fungus and therefore, hyphae could easily be traced back to their origin to ensure that anastomosis had occurred between paired isolates and not within the mycelium of the same isolates. Hyphae were considered to be compatible when at least 5 point on each slide showed C 2 type (imperfect fusion) of anastomosis reaction (Kuramae *et al.*, 2003).The whole experiment was performed three times for reproducibility of the findings.

RESULTS AND DISCUSSION

The incidence of Rhizoctonia blights of mungbean recorded during IIIrd to IVth week of August, 2007 & 2008 reveled that this was the major problems for mungbean grown in Eastern Uttar Pradesh. The table 01 elucidates that Rhizoctonia aerial blight was encountered at each and every location of survey with average incidence ranged from 1 to 27 per cent. Results also elucidate that the web blight disease was also encountered each and every place of survey except Pali in Jaunpur, Gyanpur in Sant Ravidas Nagar and Pindra in Varanasi during both the years. Rhizoctonia aerial blight of mungbean was more frequently observed than the web blight in Eastern Uttar Pradesh. Maximum aerial and web blight incidence were recorded from Budhanpur in Azamgarh and B.H.U. Agricultural Farm in Varanasi during both the year. The present findings are corroborated with the findings of Singh *et al.* (2003). Variation in the incidence of Rhizoctonia aerial and web blight in different years at the same location could be attributed to seed and soil borne nature of the pathogen. During the growing season, the air borne nature of microsclerotia function as the air borne propogule causing secondary infection for rapid spread of pathogen in the field (Yang *et al.,* 1990).Besides factors such as cultivar selection (Singh *et al.,* 2008 and Pasuvaraji *et al.,* 2013), row spacing and dates of sowing (Singh *et al.,* 2012) reported to have accelerated the expression of Rhizoctonia aerial and web blight.

During the investigation it is evident that foci of Rhizoctonia aerial blight have sasakii type sclerotia on diseased tissue where as web blight foci often having microsclerotia on large number of diseased leaves. Similar finding was reported by Yang *et al.* (1990). Because of the production of microsclerotia, web blight has a significant air borne phase which is different from the leaf borne nature of aerial blight.

Isolates from the 25 location surveyed in both the years, were morphologically similar to the description of sharewood type 1 and 2 (= AG-1IB & AG-1IA). No isolates fitting sharewood type 3 were observed. Isolates from the tissue of the host plant with microsclerotia, produced abundant microscleroia and few sasakii type sclerotia on 2 per cent water agar medium. On PDA, isolates of *R. solani* AG-1IB did not produced microsclerotia. Isolates of AG-1IA produced only typical sasakii type sclerotia on PDA and water agar medium. Two intraspecific groups were classified primarily according to the pathogenicity of causal agents, disease signs and the cultural characteristics of isolates on PDA.DNA hybridization among anastomosis groups of *R. solani* has indicated the possibility of genetic differences between AG-1IA and AG-1IB (Vilgalys, 1988). The present findings are in accordance with the findings of Yang *et al.* (1990), in soybean .The initiation and development of microsclerotia are of a later type different from loose sasakii type slerotia, indicated that microsclerotia may be related to sexual stage of fungi (Yang *et al.*,1989).

All the isolates identified from various locations were *R. solani* AG-1. Isolates of *R. solani* collected from mungbean causing web blight symptoms, exhibited C 2 reaction (imperfect fusion or killing reaction)with AG-1IA and AG-1IB tester isolates and C 1 type reaction with AG-1IC except for occasional (1or 2 imperfect fusion / 5 slides)C 2 reaction (Table-2). However, isolates collected from aerial blight symptoms were exhibited C 2 type reaction with all three tester isolates of AG-1(Table-02). From the observations it is evident that web blight isolates did not belong to AG-1IC due to C 1 type of reaction between them. Similarly since the web blight isolates did not fuse very well with AG-1IC, they may not belong to AG-1IA which known to fuse very well with AG-1IB and AG-1IC. Therefore, the web blight isolates were tentatively identified to belong to AG-1IB. Ogoshi (1987) reported that his isolates belonging to AG-1IA fuse very well with AG-1IB and AG-1IC also. Kuramae *et al.*(2003), reported his local isolates of *R. solani* from lettuce belonging to AG-1IB anastomosed with tester isolates AG-1IA and AG-1IB. Grosch *et al.*(2004), reported that his 94 isolates

Table 1: Disease incidence in two successive years *i.e.* 2007 and 2008

Districts	Villages surveyed	Average Incidence (%)			
		2007		2008	
		Aerial blight	Web blight	Aerial blight	Web blight
Azamgarh	Tarawa	10.3	13.3	12.6	14.97
	Chandesher	15.0	10.7	17.28	13.33
	Budhanpur	27.0	25.08	18.66	14.7
	Bhilihali	18.0	14.9	10.33	14.33
	Devgoan	15.05	9.8	12.0	0.28
Ghazipur	Saidpur	7.7	5.0	5.78	4.33
	Jhoria	13.3	10.6	11.34	15.28
	Narayanpur	19.0	14.7	13.33	9.66
	Aharakhpur	13.0	14.7	17.52	11.34
	Badhupur	9.30	11.3	15.28	13.33
Jaunpur	Pali	2.50	0.0	4.5	0.0
	Sikarara	8.4	5.1	14.28	10.03
	Shahganj	12.03	12.33	21.14	15.0
	Birbhanpur	9.66	8.55	14.0	10.03
	Patarahi	10.0	8.41	16.60	15.0
Sant Ravidas Nagar	Newada	18.90	7.80	11.42	8.90
	Suriyavan	12.0	11.50	12.5	18.33
	Gyanpur	9.50	0.0	8.9	0.0
	Maharajpur	12.50	13.02	17.28	19.99
	Jangiganj	4.6	3.2	0.66	0.0
Varanasi	Danganj	10.10	9.66	10.33	12.0
	Pindra	5.6	0.0	1.0	0.0
	B.H.U., Agrill.farm	20.0	22.0	26.66	25.0
	Jansa	12.50	13.83	14.33	18.88
	Chaubeypur	14.30	17.60	15.0	22.22

Table 2: Anastomosis reaction of aerial and web blight isolates of Mungbean with tester isolates.

Location from isolates Collected	Reaction types with tester isolates					
	Aerial blight isolate				Web blight isolate	
	AG-1IA	AG-1IB	AG-1IC	AG-1IA	AG-1IB	AG-1IC
Tarawa	C2	C2	C2	C2	C2	C1
Chandesher	C2	C2	C2	C2	C2	C1
Budhanpur	C2	C2	C2	C2	C2	C1
Bhilihali	C2	C2	C2	C2	C2	C1
Devgoan	C2	C2	C2	C2	C2	C1
Saidpur	C2	C2	C2	C2	C2	C1
Jhoria	C2	C2	C2	C2	C2	C1
Narayanpur	C2	C2	C2	C2	C2	C1
Aharakhpur	C2	C2	C2	C2	C2	C1
Badhupur	C2	C2	C2	C2	C2	C1
Pali	C2	C2	C2	C2	C2	C1
Sikarara	C2	C2	C2	C2	C2	C1
Shahganj	C2	C2	C2	C2	C2	C1
Birbhanpur	C2	C2	C2	C2	C2	C1
Patarahi	C2	C2	C2	C2	C2	C1
Newada	C2	C2	C2	C2	C2	C1
Suriyava	C2	C2	C2	C2	C2	C1
Gyanpur	C2	C2	C2	C2	C2	C1
Maharajpur	C2	C2	C2	C2	C2	C1
Jangiganj	C2	C2	C2	C2	C2	C1
Danganj	C2	C2	C2	C2	C2	C1
Pindra	C2	C2	C2	C2	C2	C1
B.H.U., Agrill.farm	C2	C2	C2	C2	C2	C1
Jansa	C2	C2	C2	C2	C2	C1
Chaubeypur	C2	C2	C2	C2	C2	C1

from lettuce anastomosed with tester isolates AG-1IA, AG-1IB and AG-1IC, it is already well established that isolates AG-1IA fuse very well with tester isolates AG-1IB and AG-1IC (Ogoshi,1987). In the present study isolates causing web blight of mungbean fused only with tester isolates AG-1IA and AG-1IB but not with AG-1IC. Therefore, the web blight isolates

were identified to belong to AG-1IB whereas, aerial blight isolates belongs to AG -1IA. Sneh *et al.* (1996), reported that *R. solani* AG-1IC causes damping off and crown root rot in Buckwheat, Carrot, Soybean, Flax and Pine hence, it is clearly indicated that aerial blight isolates of mungbean has no possibilities to be belonged AG-1IC and belongs to AG -1IA .

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