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Evaluation of Some Biocontrol Agents/Antagonistic Microbes Against Pastule Development of Leaf Rust of Wheat Caused by *Puccinia recondita* f. sp. *Triticum* Roberge ex. Desmaz (Erikson and Henn) D.M. Henderson

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Abstract: Out of five biocontrol agents/antagonistic microbes viz., *Verticillium lecanii*, *Paecilomyces fumosoroseus*, *Beauveria bassiana*, *Cladosporium cladosporioides* and *Metarrhizium anisopliae*, *Beauveria bassiana* proved the best in retarding the postulation/rust development in wheats. When the microbes tested alone against leaf rust development, *B. bassiana* proved excellent and in combination with *Verticillium lecanii* and *Paecilomyces fumosoroseus*. Although there was an increase in colony diameter after 2nd week but the pattern of growth was the same as after one week of incubation of bio-control agents application.

Key words: Biocontrol agents, wheat, leaf rust, fungus

Introduction

Wheat (*Triticum aestivum* L.) is a principal staple food crop for the people of Pakistan. It is grown on an area of 8169.8 thousand hectares with a production of 21078 thousand tonnes (Anonymous, 2000). Per acre grain yield is very low as compared to other wheat growing countries. Among several factors responsible for the low grain yield, wheat diseases especially the rusts are very important. Leaf rust caused by *Puccinia recondita* f. sp. *Triticum* Roberge ex Desmaz (Eriks and Henn) D.M. Henderson is the most destructive and devastating disease due to its time of appearance, nature of attack regular occurrence and prolonged season prevalent for the disease development in wheat growing areas of Pakistan. It appears in March April in the form of orange colour pustules, scattering on leaf and leaf sheath. The epidemic of stripe and leaf rusts together in 1977-78 (due to early and large scale development) reduced the total grain production of wheat by 2.2 million tones (Hassan, 1979; Hafeez, 1986). During these epidemic, 50-80% disease severity on most of the commercial cultivars and 30% yield losses were recorded in Punjab (Khan, 1985). In 1973, leaf rust intensity ranged from 40-50% with 100% infection on susceptible varieties but luckily crop escaped from severe losses due to short duration of the favourable season of rust (Hassan, 1973). Wheat rust management strategies include cultivation of resistant varieties and seeds/foliar application of fungicides. Chemical control of leaf rust is not practiced in Pakistan on commercial basis due to higher cost of chemicals, low market price of wheat, risks of health hazard and lack of proper disease forecasting system to apply fungicide economically.

The present studies primarily aim to investigate the effect of different bio-control agents/antagonistic microbes on leaf rust postulation and ultimately to trace out alternative, cheap and environmental safer method for leaf rust management.

Materials and Methods

In the field, ten wheat cultivars (C-591, local white, C-273, Morocco, SA-42, WL-711, SHL-88, CHB-70, Lu-26 and SA-75) were sown at the experimental area of the Department of Plant Pathology, University of Agriculture, Faisalabad in December, 2000. Rust inoculum was sprayed twice i.e. before and after flag leaf stage. In order to maintain crop vigour, normal agronomic practices including recommended doses of fertilizer and irrigation schedule was observed. At proper stage, just with the initiation of rust initials a loopful inoculum (in 100 ml) of actively growing pure

biocontrol agents namely *Verticillium lecanii*, *Paecilomyces fumosoroseus*, *Beauveria bassiana*, *Cladosporium cladosporioides* and *Metarrhizium anisopliae* were sprayed separately on the plants, those were tagged later on. Except *C. cladosporioides*, all other four biocontrol agents were obtained from a research lab of USA and were multiplied on Saboraud dextrose medium for further studies. Data on pustule development were recorded for each treatment after 10 days interval following the modified Cobb's scale, described by Peterson *et al.* (1948).

In order to test the viability of urediospores of *P. recondite* hanging drop technique was used (Duggar, 1901). For the purpose, vauntieghem cells were fixed on to the glass slides and were half filled with sterilized water. To avoid contamination of water, vaseline was applied to the edges. Data of spore germination were recorded and was analyzed statistically.

Results and Discussion

Leaf rust management through biocontrol agents namely *Verticillium*, *Paecilomyces*, *Beauveria*, *Cladosporium* and *Metarrhizium* revealed that in general the disease incidence was low in all the treatments especially in T₃ (*Beauveria bassiana*) as compared to control. Data also indicates that all the biocontrol agents produced pronounced effect on the rust postulation. The response T₁, T₂, T₄ and T₅ was more or less similar but these treatments were also effective against pustule development of leaf rust of wheat and these treatments significantly differed from the control after first application. T₃ (*Beauveria bassiana*) gave best control against leaf rust of wheat. While T₁, T₂, T₄ and T₅ remained statistically at par with each other (Table 1). T₆ (control) gave the minimum control. Similarly T₃ (*Beauveria bassiana*) showed more control against leaf rust of wheat (Table 2). Treatments T₁, T₂, T₄ and T₅ remained statistically at par with each other less control was observed in case of control. Due to risks of health hazards, high cost of chemicals helpful for rust management, non availability of disease forecasting system for the effective and economic use of rusticides biological control of rusts is a modern, advanced and risk free alternative method of rust managements. Through cereals rusts management is in infancy but is being practiced effectively in other countries of the world. Spencer (1980) used an antagonistic microbe (*Verticillium lecanii*) against carnation rust and proved that the rust infection was averted. Spencer and Atkey (1981) used *V. lecanii* against brown rust of wheat and showed that formation of urediniospores was arrested and new colony formation was inhibited because of reduced viability of urediniospore, invaded by *V. lecanii*. Effectiveness of *Beauveria bassiana* may be due to its special mode of action i.e. coagulation and disintegration of cytoplasm of rust spores as described by Sultana (2000) about the similar action of *Verticillium lecanii* and *Cladosporium cladosporioides* in case of soybean rust. The incidence of rust development (after 2nd application) on all the ten susceptible moderately resistant and resistant varieties is invariably low in all the treatments especially in T₃ and similar response was observed even after 3rd application. It may be due to the fact that all the treatments have shown a profound excellency against leaf rust of wheat. It was reported that when in the presence of *Verticillium lecanii*, uredospore of *Puccinia recondite* were applied to wheat leaf, the level of rust was

Sheroze *et al.*: Biocontrol agents, wheat, leaf rust, fungus

Table 1: Comparative effect of biocontrol agents on pustule development (after first application) in field

Treatments	Rust development (percent) on different wheat cultivars										Means
	C-591	Local white	C-273	Morocco	SA-42	WL-711	SHL-88	CHB-70	LU-26	SA-75	
T ₁	1	5	1	5	5	10	5	5	10	5	3.050b
	5	1	1	1	1	1	1	1	1	5	
	5	1	5	5	5	5	1	1	1	1	
T ₂	1	5	1	1	1	1	5	1	5	1	3.275b
	5	1	10	5	5	5	5	1	5	5	
	1	5	5	1	1	10	5	1	1	1	
T ₃	1	5	1	1	1	1	5	1	1	1	1.500c
	5	1	1	5	5	5	1	1	1	10	
	1	1	1	1	1	1	1	5	5	1	
T ₄	1	1	1	1	1	1	1	1	1	1	2.975b
	1	5	1	1	1	1	1	1	1	1	
	10	1	1	5	5	1	1	10	5	5	
T ₅	5	1	1	1	1	1	1	1	1	1	3.825b
	5	1	10	10	10	10	10	5	5	5	
	1	10	1	5	5	1	1	1	1	5	
T ₆	1	5	1	1	1	5	1	5	5	1	6.800a
	5	1	5	5	5	1	5	5	1	1	
	10	10	10	10	10	20	1	10	1	20	
	5	5	5	1	1	5	10	5	10	5	
	10	1	5	10	10	5	10	5	10	10	
	5	5	1	5	5	5	5	1	5	10	

Table 2: Comparative effect of biocontrol agents on pustule development (after second application) in field

Treatments	Rust development (percent) on different wheat cultivars										Means
	C-591	Local white	C-273	Morocco	SA-42	WL-711	SHL-88	CHB-70	LU-26	SA-75	
T ₁	1	10	5	5	10	10	20	5	10	10	6.450b
	5	1	1	5	1	5	5	5	5	20	
	5	1	5	5	5	10	5	1	5	5	
T ₂	5	5	1	1	5	5	10	20	10	10	7.000b
	1	5	10	5	20	20	5	5	5	5	
	1	5	1	1	5	5	10	10	5	20	
T ₃	1	5	1	5	10	10	5	5	10	20	2.600c
	1	1	5	5	5	5	1	1	1	5	
	5	1	1	1	1	1	1	5	1	5	
T ₄	1	5	1	5	5	1	1	1	1	1	6.100b
	10	5	1	5	1	5	5	20	5	10	
	5	1	5	1	10	10	10	10	1	20	
T ₅	1	1	1	5	5	5	5	5	1	5	7.225b
	5	5	5	5	10	10	5	10	5	10	
	1	5	1	5	1	5	20	5	5	5	
T ₆	5	5	10	10	10	10	10	10	10	10	17.00a
	1	10	10	10	10	5	5	5	5	10	
	5	5	5	5	20	5	10	5	20	5	
	20	10	20	20	10	30	10	20	5	20	
	10	5	10	5	20	10	20	10	20	30	
	10	5	5	5	30	5	40	10	30	10	
	5	5	5	5	5	20	60	30	40	50	

T₁ = *Verticillium lecanii*, T₂ = *Paecilomyces fumosoroseus*, T₃ = *Beauveria bassiana*,
T₄ = *Cladosporium cladosporioides*, T₅ = *Metarrhizium anisopliae*, T₆ = Control,

reduced to 10-16% (Spencer and Atkey, 1981). The incidence of rust development was low in all the cultivars. It may be due to the specialized nature of biological agents to grow within the rust pustule. The presence of large number of hyphae between these uredinospores of stripe rusts (Mendgen, 1981).

The findings of these experiments confirm the effectiveness of *Verticillium lecanii*, *Cladosporium* along with the new finding *Beauveria bassiana* (the most effective bio-control agent of leaf rust of wheat), however it is needed to find out the combined

effect of two or more bio-agents against leaf rust and to determine economical doses/concentrations and mass multiplication of these bio-control agents for effective use of these bio-control agents to cut down the use of rusticides for the protection of agro-ecosystem and to change the trend of the scientists to promote biological control of plant pathogens.

It is concluded that T₃ (*Beauveria bassiana*) showed best control against leaf rust of wheat as compared to other biocontrol agents and less control was observed in case of control.

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