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Eefect of U V Protectants on the Pathogenicity Growth and Spore Production of *Beauveria bassiana* (Bals) Vuil.

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ABSTRACT

Ultra violet(UV) protectants,viz,,charcoal(1%) Indian ink (1%)congo red (1%)robin blue (0.5%),and Yeast extract 2% were evaluated for their effect on the bioefficacy of *Beauveria bassiana* (Bals) Vuill at its LC_{50} (9.4×10^4 spores ml $^{-1}$) against the third instar larvae of *Spodoptera litura* (Fabricius) after exposing them to UV-light (30 minutes, one hour and three hours,. Further, the fungal growth and spore protectants resulted significantly higher larval mortalities (20.61 to 71.20%), fungal growth (1.40 to 7.77 cm diam.) and spore production (8.75 to 617.5 $x10^4$ spores ma $^{-1}$) after 30 minutes to three hours of exposure to UV-light. The LT_{50} values for *B.bassiana* at its LC_{50} with and without any UV-protectants ranged between 91.2 to 194.4 hours after 30 minutes to one hour exposure to UV-light. Whereas, the LT_{50} values of *B.bassiana* at its LC_{50} with charcoal (1%) and Indian ink (1%) were 235.2 and 242.4 hours after three hours of exposure to UV-light, respectively.

Key words : *Beauveria bassiana*, Charcoal, Congo red, Indian ink, Robin blue, *Spodoptera litura*, UV-protectants, Yeast extract.

There is a world wide- renewed interest in the use of entomopathegenic fungi as biological control agents (Khachatourians,1986) and of all the enthomopathogenic fungi as microbial insecticides , *Beauveria bassiana* (Bals) Vuil,is the most successful so far.However ,ultra violet (UV) rays in the natural sun light are one of the major limiting factors against the field persistence of microbial insecticides (Edgington *et al.,* 2000).This has led to the present study, aimed at the tobacco cutworm, *Spodoptera litura* (Fabricius) in the laboratory. Further the effect of the UV-protectants on the growth and spore production of B.bassiana was also evaluated.

MATERIAL AND METHODS

Pure culture of *Beauveria bassiana* (Bals.) Vuil. Obtained from Biological control laboratory, ANGRAU,Rajendra nagar,Hyderabad, was sub-cultured on saboraud dextrose agar(SDA) medium (Delarosa et al.,1997) and maintained at 4^o C for further use .the test insect, *Spodoptera litura* (Fabricius.) was reared in overlapping genarations on *Ricinus communis* Linn.leaves under hygienic conditions at room temparateure($27+-2^o$ C).

The selected UV-protectants, charcoal (1%), congo red (1%),rabbin blue (0.5%) and yeast extract(2%) (M/s S.D.Fine chemical Ltd.,Mumbai) were added to the prefixed LC_{50} ($9.4x10^4$ spores/ ml) of B.bassiana and were exposed to UV-light for specific duration viz,30 minutes,one hour and three hours.Then the third instar larvae of S. litura were treated with the test combinations and the data on larval mortality was recorded at 3,6 and 9 days after treatment (DAT) after 30 minutes,one hour and three hours of UV exposure, respectively. The LT_{50} values were also calculated from the larval mortality data.

Further the LC_{50} value of the fungal spore suspension with the different UV-protectants was exposed to UV-light for the specified durations and was placed (20µl) at the center of the petri plates containing the SDA medium . The test pathogen was assessed for its growth inhibition (diam.in cm) and spore production (spores/ml) at ten days after incubation.

The data on larval mortality from four replications using twenty larvae per replication after arcsine percentage transformation were subjected to analysis of variance technique using completely randomized block design (Gomez,1984). The LT_{50} values were derived from logarithmic calculations (Finney 1984) using multiple linear programme on computer.

RESULTS AND DISCUSSION

Beauveria bassiana (Bals.) Vuill.at $9.4x10^4$ spores/ml (LC_{50}) with charcoal(1%), as UV-protectant after 30 minutes,one hour and three hours of exposure to UV-light resulted in significantly the highest larval mortality (table1) of *Spodopter litura* (FAbricius.) (71.20,47.87,39.17%) followed by *B. bassiana* with Indian ink (1%), congo red(1%) ,robin blue(0.5%) and yeast extract(2%) as against *B.bassiana* without any UV-protectant.

Table 1. Effect of UV-protetants on the efficacy of *Beauveria bassiana* as pure culture at its LC_{50} after different durations of exposure to UV-light on the mortality of the third instar larvae of *Spodoptera litura*.

B.bassiana (9.4×10^4 spores ml^{-1}) + UV-protectants	Per cent larval mortality (DAT)		
	30 minutes	One hour	Three hours
B.bassiana	34.54 (32.50)^{c}	28.71 (24.75)^{d}	18.14 (10.00)^{c}
B.bassiana+Charcoal(1%)	71.20 (88.75)^{a}	47.87 (55.00)^{a}	39.17 (40.00)^{a}
B.bassiana+Indian ink (1%)	66.41 (83.75)^{a}	44.28 (48.75)^{b}	37.71 (37.50)^{a}
B.bassiana+Congo red (1%)	60.91 (76.25)^{b}	35.44 (33.75)^{c}	23.58 (16.25)^{b}
B.bassiana+Robin blue (0.5%)	56.82 (70.00)^{b}	33.05 (33.00)^{cd}	23.58 (16.25)^{b}
B.bassiana+Yeast extract (2%)	36.24 (35.00)^{c}	31.56 (27.5)^{d}	20.61 (12.50)^{bc}
F test	Sig.	Sig.	Sig.
SEm±	3.05	2.79	2.36
C.D (p=0.05)	6.41	5.88	4.96

DAT : Days After Treatment.
Values in parentheses are angular transformed values.
In each column, values having common alphabet do not vary significantly.

The LT_{50} values (table 2) of B. bassiana 9.4×10^4 spores/ml (LC_{50}) against the third instar, S.litura larvae with the UV-protectants were 91.2 and 134.4 hours for charcoal (1%) ,93.6 and 144.0 hours for Indian ink(1%) 105.6and 160.8 hours for congo red (1%) 112.8 and 158.4 hours for robin blue (0.5%) and 156.0 and 187.2 hours for yeast extract (2%) for 30 minutes and one hour exposure to UV-light as against 160.8 and 194.4 hours in B. bassiana without any UV-protectant. Respectively .The LT_{50} values of B. bassiana at 9.4×10^4 spores/ml (LC_{50}) with charcoal(1%) and Indian ink (1%) as UV-protectants for three hours exposure to UV-light were 235.2 and 242.4 hours respectively .B. bassiana with congo red (1%) robin blue (0.5%) and extract (2%)as UV-protectants and without any UV-protectant for three hours exposure to UV-light did not record 50 percent larval mortality. Hence, LT_{50} values were not obtained . The corresponding values of x^2 , regression equations and fudicial limits of the LT_{50} values because of the addition of UV-protectants compared to B. bassiana without any UV-protectant with 30 minutes and one hour of UV-exposure ranged between 43.28 to 30.86 percent for char coal (1%) 41.79 to 25.92 per cent for Indian ink (1%) ,34.32 to17.28 percent for congo red (1%) ,29.85 to 18.51 per cent for robin blue (0.5%) and 2.98 to 3.70 percent for yeast extract (2%), respectively.

The data on the response of B.bassiana at 9.4×10^4 spores/ml (LC_{50}) with and without the UV-protectants exposed for the specified durations to UV-light (Table3) indicated that charcoal (1%) and Indian ink (1%) significantly reduced the lethal effect of UV-light on the diameter of fungal growth. The increase in B. bassiana fungal growth (diam.in cm) because of addition of UV-protectants compared to B. bassiana without any UV- protectants for 30 minutes to three hours UV exposure ranged from 40.76 to 125.00percent for charcoal (1%) ,34.96to 96.42 percent for Indian ink (1%) ,22.64 to 42.85 per cent for congo red (1%) ,16.30 to 30.00 percent for robin blue (0.5%) and 10.50 to 3.57 percent for yeast extract (2%).

Table 2. Medium Lethal Times (LC_{50}), fungal growth and spore production of *Beauveria bassiana* with and without and without the UV-protectants after different durations of exposure to UV-light against the third instar larvae of *Spodoptera litura*.

B.bassiana (9.4×10^4 spores ml^{-1}) + UV-protectants	LC_{50} (Hours)	Fungal Growth (Diam.in cm)	Spore Productivity
	Thirty minutes of exposure to UV-light		
B.bassiana+Charcoal 1%	91.2	7.77^{a}	617.5^{a}
B.bassiana+Indian ink 1%	93.6	7.45^{a}	557.5^{b}
B.bassiana+Congo red 1%	105.6	6.77^{bc}	532.5^{b}
B.bassiana+Robin blue 0.5%	112.8	6.42^{cd}	447.5^{c}
B.bassiana+Yeast extract 2%	156.0	6.10^{d}	430.0^{c}
B.bassiana	160.8	5.52^{d}	422.5^{c}
	One hour of exposure to UV-light		
B.bassiana+Charcoal 1%	134.4	5.90^{a}	222.5^{a}
B.bassiana+Indian ink 1%	144.0	5.82^{a}	170.0^{b}
B.bassiana+Congo red 1%	160.8	4.92^{b}	125.0^{c}
B.bassiana+Robin blue 0.5%	158.4	4.55^{bc}	120.0^{c}
B.bassiana+Yeast extract 2%	187.2	4.55^{bc}	117.5^{c}
B.bassiana	194.4	4.15^{c}	110.5^{c}
	Three hours of exposure to UV-light		
B.bassiana+Charcoal 1%	235.2	3.15^{a}	20.0^{a}
B.bassiana+Indian ink 1%	242.4	2.75^{a}	15.0^{ab}
B.bassiana+Congo red 1%	*	2.00^{b}	11.25^{b}
B.bassiana+Robin blue 0.5%	*	1.82^{b}	8.75^{b}
B.bassiana+Yeast extract 2%	*	1.45^{b}	8.75^{b}
B.bassiana	*	1.40^{b}	8.75^{b}

*LT_{50} could not be calculated as 50 per cent larval mortalities were not resulted

The data on the response of B .bassiana at 9.4×10^4 spores/ml (LC_{50}) with and without the UV-protectants exposed for the specified durations to UV-light indicated that charcoal (1%) and Indian ink (1%) significantly reduced the lethal effect of UV-light on the spore production .The spore production reduced by B. bassiana in Table2. The increase in spore production because of the addition of UV-protectants compared to B. bassiana without any UV-protectants for 30 minutes to three hours UV exposure ranged from 100.79 to 216.66 per cent for charcoal (1%) ,68.25 to66.66 per cent for Indian ink (1%) ,58.73 to 33.33 per cent for congo red (1%) ,28.57 to 33.33 per cent for robin blue (0.5%) ,and 12.69 to 16.66 per cent for yeast extract (2%)

Thus ,it is evident that the lethal effect of UV-light on the growth and spore production was increasing with increase in period of exposure to UV-light ,the LT50 values were not obtained for the either with or without some of the UV-protectants from the three hours of exposure onwords . However ,charcoal (1%) , Indian ink (1%) can be stated as the successful UV-protectants for the entomopathogenic fungus , B. bassiana wich increased the larval mortality and reduced the LT_{50} values through sustained fungul growth and sore production of the respective fungus.

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