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LABORATORY EVALUATION OF ANTIOXIDANT EFFECTS ON THE INACTIVATION OF BACILLUS THURINGIENSIS FOR COTTON LEAF WARM BY SIMULATED SUNLIGHT-U V.

A. SHAMS EL DIN
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All representative types of microbial insecticides (i.e. bacteria, fungi, protozoans, viruses) are inactivated by exposure to the ultraviolet-A (UV-A), ultra violet-B (UV-B) spectrum (280-400nm) of sunlight (Ignoffo et al. 1977).

Solar inactivation of Bacillus thuringiensis Berliner and other entomopathogens is a a widely recognized phenomenon. Exposure for short periods (<24h) to wavelenghts below 500 nm inactivates spores, degrades protein structures of virial inclusion bodies, and may inactivate B.thuringiensis crystalline toxins. The half-life for B.thuringiensis has been estimated at 3.8h. when exposed to an ultraviolet (UV) source representative of the UV radiation in natural sunlight (Ignoffo et al. 1977). Numerons attempts have been made to develope protective measures against damaging UV radiation under field conditions, but success has been limited, suggesting that effects of solar radiation on entomopathogens are not sufficiently understood (Raun & Jackson 1966, Ahmed et al 1973, Morris 1983).

Two general hyposeses have been formulated to explain the sunlight inactivation of the microbial insecticides. Sunlight may have a direct effect on microbial DNA by including deleterions cross-linkings, strand breaks or development of labile sites or both (Tyrrell 1973, Tyrrell et al 1974). Although his hypothesis could explain sunlight inactivation of living micro organisms, it would not explain inactivation of the nonliving, crystalline endotoxin of Bacillus thuringiensis. Another hyposthesis that might explain the sunlight inactivation of both types of microbial insecticides is sunlight generation of highly reactive radicals (e.g. peroxides, sigletoxygen, hydroxyls) that would, in turn, degrade the insecticidally active active entity (Yoakum & Eisenstark 1972, Ananthaswamy & Eisenstark 1976, McCormick etal 1976, Ignoffo & Garcia 1992, Pozsgay et al 1987, Ignoffo et al 1989).

The two hypotheses are not mutually exclusive, both may be operational at the same or at different UV-A, UV-B spectra.
If sunlight generation of reactive radicals does occur, it may be possible to increase the persistence of the microbial insecticides using antioxidants that scavenge the degradtion of reactive radicals.
This paper reports the results of studies to determine whether three antioxidants would inhibit the UV inactivation of Bacillus thuringiensis against cotton leafworm Spodoptera littoralis larvae.

MATERIALS AND METHODS

BACILLUS THURINGIENSIS

B.thuringiensis var. entomocidus was propagated on a modified yeast-malt-glucose medium containing yeast extract (0-5%) malt extract (0.5%), tryptone (0.5%), peptone (0.5%), glucose (0.5%), and potassium dihydrogen phosphate (0.1%). Fermentations were done using the method of Nickerson et al (1974), which produced loo g B.t (wet weight) of cell paste containing about 30% spores and crystals by weight after centrifugation. This paste was stored at 2 c until used.

Ultraviolet Source And Exposure Procedure:
A series of BL Fluorescent lamps built into a temperature cabinet were used to stimul ate the ultraviolet - A (320 - 400 nm) and ultra violet - B (280-320nm) spectrum of sunlight (S U V). Water suspension of B.thuringiensis and protectant were placed into UV-transparent quartz cells and exposed to the UV source for 24h.
B.thuringiensis were exposed at (250 ug/ml).
UV protectents: three antioxidants at concentrations ranging from 0,001, 0-01,0.1,1,10 and 100 mg/ml, were tested for ability to retard or prevent the inactivation of B. thuringiensis by UV. The antioxidants were the sodium salt of L-ascorbic acid, phenyl thio-carbomide, and n- propyl gallate.

TREATMENTS AND BIOASSAY

A typical replicate of UV exposure series included the following treatments:

a) B. thuringiensis alone
b) B. thuringiensis pltus an antioxidant at several concentrations,
c) only, antioxidant,
d) larvae receiving no treatment (control)

The activity of B.thuringiensis, after exposure to UV, was determined using 24-h-old larvae of S.littoralis reared on semi-synthetic diet (salama, 1970). Mean percent mortality after 7 days of exposure at 30±1 c based on the use of 100 larvae per treatment and ten replicates per treatment. Percent original activity - remaining (% OAR),when used for a comparison, is the quotient of the mortality.

DATA ANALYSIS:

Percentage mortalities were used for determining LCSO (median lethal concentration for 50 %) by probit analysis values (Finney, 1971). Abbott formula (Abbott, 1925) was used to correct percent mortality.

Table (1) Activity of B.thuringiensis var. entomocidus (250ug/ml) after exposure to ultraviolet against S.littoralis larvae.

HOUR	% MORTALITY
1 (control)	82
4	40
8	34
12	37
16	31
20	39
24	30
control (without B.t)	0

Table (2). Effects of several concentrations of antioxidants on activity of B.thuringiensis exposed for 24-h. to a source of simulated sunlight - UV against neonate larvae of S.littoralis.

TREATMENT	ANTIOXIDANTS		
Antioxidant mg /ml	Ascorbic acid	Phenylthio-carbamide	Propyi/Gallate
	% Mortality		
B.t 250ug/ml not exposed to UV	82		
B.t alone exposed to UV	30		
B.t + 100	79	81	
B.t + 10	78	80	
B.t + 1	43	77	80
B.t + 0.1	32	41	78
B.t + 0.01	30	29	47
B.t + 0.001	30	28	33
Control (without B.t)	0	0	0

Table (3) Original Activity Ratio (OAR) of the antioxidants on B.thuringiensis exposed for 24-h to UV source against neonate larvae of S.littoralis

TREATMENT	% MORTALITY	OAR
1- B.t 250 mg/ml (without UV exposure)	82	
2- B.t (exposed to uv)	30	36%
3- B.t + Ascorbic acid 100 mg/ml	79	94%
4- B.t + Ascorbic acid 10 mg / ml	78	93%
5- B.t + Ascorbic acid 1 mg/ ml	43	52%
6- B.t + Ascorbic acid 0.1 mg / ml	32	39%
7 B-.t + Ascorbic acid 0.01 mg / ml	30	36%
8- B.t + Ascorbic acid 0.001 mg/ ml	30	36%
9- B.t + phenyl this o carbamide 100 mg/ml	81	98 %
10- B.t + phenyl thiocarbamide 10 mg/ml	80	95%
11- B.t + phenyl thiocarbanide 1 mg/ml	77	92%
12- B.t + phenyl thiocarbamide 0.1 mg/ml	41	50%
13- B.t + phenyl thiocarbamide 0.01 mg/ml	29	35%
14- B.t + phenyl thiocarbamide 0.001 mg/ml	28	34%
15- B.t + propyl gallate 1 mg/ml	80	95%
16- B.t + propyl gllate 0.1 mg/ml	78	93%
17- B.t + propyl gallate0.01mg/ml	47	57%
18 - B.t + propyl gallate 0.001 mg/ml	33	40 %

Results and Discussions:

Half- life time of B.thuringiensis after exposure to UV source for different intervals of hours was 4.85 hr (Table 1). A rate of 250 mg/ml of B.thuringiensis var. entomocides was selected for all bioassays because it gave 82 % mortality for S.littoralis neonate larvae and permitted an optimal comparison of differences between the UV - exposed and the non UV exposed B.thurthingiensis samples.

EFFECTS OF ANTIOXIDANTS.

The % OAR of B.thuringiensis without an antioxidant was 30% (Table 2). All three antioxidants provided same level of simulated UV protection of B.thuringiensis (Table 2). Table (3) showed that B.thuringiensis at concentration 250 μg/ml without UV exposure caused 82% against neoate larvae of S.littoralis while the same concentration lost its activity to 36% of the original activity.
Addition of 100, 10, 1, 0.1, 0.01, and o.oo1 mg/ml of ascorbic acid provided % 94, 93, 52, 39, 36 and 36 of original activity ratio respectively. The addition of 100, 10, 1, 0.1, 0.01, and o.oo1 mg/ml of phenyl thiocarbamide provided % 98, 95, 92, 50,35 and 34 of original activity ratio, respectively.
Propyl gallate at concentration 1, o.1, 0.01 and o.oo1 mg/ml provided % 95, 93, 57 and 40 of original activity ratio, respectively.
Propyl gallate, however, provided the most protection. Little or no SUV protection (Less than 50 % OAR) was provided by ascorbic

acid or phenylthiocarbamide at 0.01 mg/ml. At the same concentration, the OAR for propyl gallate was 47 % (Table 3). Concentration of 10 mg//ml ascorbic acid, 1mg/ml of phenylcarbamide and 0.1 mg/ml of propylgallete showed % mortalities of 78, 77 and 78 respectively and provided over 90 % protection of B.thuringiensis activity.

These results demonstrate that antioxidants could inhibit sunlight inactivation of B.thuringiensis and possibly other microbial pesticides. Propyl gallate, because of its apparent effectiveness at low concentrations, its relatively low toxicity to mammals, its low cost, and its common use as a food additive, might be used as an adjuvant or spray - tank additives to provide protection against sunlight - UV.

REFERENCES

1- Abbott, W.S. (1925). A method of computing the effectiveness of insecticide. J. Econ. Entomol. 18, 265 - 267.

2- Ahmed, S.M., M.V. Nagamma and S.I. Majumdar 1973. Studies on granular formulations of Bacillus thuringiensis Berliner. Pestic. Sci. 4: 19-23

3- Ananthaswamy, H.M. and A.Eisenstark. (1976).
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4- Finney, D.J. (1971). Probit analysis (3rd. ed.), Cambridge Univ. press, London, 333pp.

5- Ignoffo, C.M. & C. Garcia, 1992. Combinations of environmental factors and simulated sunlight affecting activity of inclusion bodies of the Heliothis (Lepidoptera: Noctuidae) nucleopolyhedrosis virus. Envviron, Entomol. 21: 210-213.

6- Ignoffo, C.M., D.L. Hostetter, P.P. Sikorowski. G. Sutter & W.M. Brooks. 1977. Inactivation of representative species of entomopathogenic viruses, a bacterium, fungus and protozoan by an ultra-violet light source.Environ. Entomol. 6: 411-415.

7- Ignoffo, C.M., W.C. Rice & A.H. McIntoch. 1989. Inactivation of nonoccluded baculoviruses and baculovirus - DNA exposed to simulated sunlight. Environ. Entomol. 18: 177 - 183

8- Morris, O.N 1983. Protection of Bacillus thuringiensis from inactivation by sunlight. Can. Entomol. 115: 1215 - 1227

9- McCormic, J.P., J.R. Fisher, J.P. Pachlatko & A. Eisenstark.

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10- Nickerson, K.W., G. St. Julian & L.A. Bulla. 1974, Physiology of sporeforming bacteria associated with insects: ratio respirometic survey of carbohydrate metabolism in the 12 serotypes of Bacillus thuringiensis. Appl. Microbiol. 28: 129 - 132

11- Pozgasy, M., P. Fast, H. Kaplan & P.R. Carey. 1987. The effect of sunlight on the protein crystals from Bacillus thuringiensis var. kurstaki HDI and NDR12: a Raman spectroscopic study. J. Invertebr. pathol. 50:246-253

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14- Tyrell, R.M. 1973. Introduction of pyrimidine dimers in bacterial DNA by 365 nm radiation. Phtochem. Photobiol. 17: 69 - 73.

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