

ABSTRACT

Hassan Mohamed Ebrahim Mohamed El-Saadany. Pesticidal and Immunological Studies of Some *Bacillus thuringiensis* Isolates. Unpublished Ph. D. Dissertation, Plant Protection Dept., Faculty of Agric., Ain Shams Univ., 2007

Bacillus thuringiensis subsps. *kurstaki* and *entomocidus* (*Btk* and *Bte*) were tested to estimate the toxicity against 1st, 2nd and 4th instars larvae of *S. littoralis* and the safety on mammals. Bioassay of *Btk* and *Bte* were studied at concentrations 10^8 , 10^7 , 10^6 , 10^5 and 10^4 spore/ml. Results revealed that the two tested strains had a great efficacy against larvae of *S. littoralis*, but *Btk* was more effective than *Bte* on the tested larvae. The LC₅₀ values of *Btk* and *Bte* on the first instar larvae were 2.17×10^5 and 7.83×10^6 spore/ml, respectively, while on the 2nd instar were 4.69×10^6 and 3.179×10^9 spore/ml, respectively. The highest LC₅₀ values were recorded for the 4th instar larvae as 1.09×10^{11} and 2.77×10^{13} spore/ml for the same strains, respectively. *Btk* and *Bte* were injected into albino rabbits at 10^6 spore per rabbit to study immune response by using ELISA technique. The most antisera exhibited moderate titers of *Btk* and *Bte*. The inhibition activity were tested to determined antibody sensitivity. *Btk* antibody was higher sensitivity than *Bte* antibody in detection of antigen. Regarding the cross reactivity, using different antibodies raised against *Btk* and *Bte*. *Bte* spore gave cross reaction with antisera from *Btk*, likewise, *Btk* spores gave cross reaction with *Bte* antiserum. Subchronic toxicity tests were carried out on rats with *Btk* and *Bte* by oral administration in daily drinking water at 10^8 , 10^7 and 10^6 spore/ml for 90 days. All animals were observed frequently during the test period for mortality and clinical signs of disease or toxicity. No mortality, no difference in the behavior or the morphological examinations were found in the male and female rats treated with *Btk* and *Bte* at all concentrations. There were slight differences in body weight gained between treated and untreated male and female rats

at some period of the test. The weights of liver, kidney, spleen, heart, lung, brain and testis in treated male rats, were not affected at all tested rates of both strains of *Bt*, except a significant decrease in liver, kidney and heart weight at different concentrations of *Bte*. Data on treated female rats, indicated that the weights of liver, kidney, spleen, heart, lung and brain were not affected with *Bt* strains. On the other hand a significant increase was found in spleen weight at all rats of *Bte*. According to clinco-biochemical aspects such as, liver functions (e.g., ALT, AST, ALP, TP and ALB), ChE, kidney functions (e.g., Urea and Creatinine) and thyroid function (e.g., T₄ and T₃), *Btk* and *Bte* produced a significant differences (increase or decrease) at some period of treatment and some concentrations of *Bt*. Specimens from liver, kidney and spleen of treated male and female rats treated were taken for examination by light microscopy and photographed. *Btk* and *Bte* showed mild effect and cellular immunity (activation of Kupffer cells) on liver. In addition, in the kidney, adverse effect (thickening of glomerular capillary basement membrane), albuminous material in Bowman's space and mild effect (atrophied glomerular capillary tubes) were recorded. Also, in the female rats treated with *Btk*, spleen tissue was less affected (the follicular blood vessels showed thickening of the wall). Lymphocytic depletion in the lymphoid follicles in the spleen of females was noticed with *Bte*. In addition, thickening of the blood vessel wall of follicular vessel and lymphocytic depletion in the lymphoid follicle were found in the spleen of male rats treated with *Bte*.

Key Words: *Bacillus thuringiensis*, *S. littoralis* toxicity, ELISA, pathogenicity, rats.

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LIST OF ABBREVIATION

Ab	Antibody
Abs	Antibodies
ALB	Albumin
ALP	Alkaline phosphatase
ALT	Alanine aminotransferase
AST	Aspartate aminotransferase
<i>Bt</i>	<i>Bacillus thuringiensis</i>
<i>Bte</i>	<i>Bacillus thuringiensis</i> subsp. <i>entomocidus</i>
<i>Bti</i>	<i>Bacillus thuringiensis</i> subsp. <i>israelensis</i>
<i>Btk</i>	<i>Bacillus thuringiensis</i> subsp. <i>kurstaki</i>
CFU	Colony forming units
ChE	Cholinesterase
EPA	Environmental protection agency
H&E	Hematoxilen and eosin
I	Inhibition
<i>Ma</i>	<i>Mitarizium anisoblie</i>
MPCA	Microbial pest control agents
PBS	Phosphate buffer saline
PBSTA	Phosphate buffer saline tween azide
T ₃	Triiodothyronine
T ₄	Thyroxine
TNF- α	Tumor necrosis factor
TP	Total protein