

BIOTEKNOLOGIESE INSEK EN/OF AALWURMWEEERSTAND BY SOJABONE

OPSOMMING

Verskeie suiwer Bt isolate, Bt wat vanuit grondmonsters geïsoleer is asook Bt wat vanuit 'n dooie insek geïsoleer is, se toksisiteit is teenoor *Meloidogyne incognita* (ras 2) en teenoor *Ditylenchus destructor* getoets. Die *in vitro* kweking van Bt bakterieë op soliede medium asook in vloeistofkultuur, is gestandaardiseer. Die prosedure vir die isolering van Bt variante vanuit grondmonsters van oor die hele land, is ook gestandaardiseer. Verskeie ander proteïene is ook vanuit 'n verskeidenheid plantweefsels geïsoleer. Daar is ook 'n vinnige en maklike prosedure gevind vir die isolering van redelike suiwer kristalproteïene. 'n Verskeidenheid oplosmiddels is getoets vir effektiewe solubilisering van die kristalproteïene, waarvan 0,015 N NaOH die beste oplosbaarheid, gekoppel aan 'n baie lae toksisiteit (5%) teenoor die aalwurms en 'n baie **hoe** stabiliteit (90%) van die kristalproteïene, gelewer het. Standaardisering van SDS-PAGE van die proteïene is ook afgehandel. Die protoksiene kan ook suksesvol omgeskakel word na toksiene met behulp van tripsien. Die biologiese toetsprosedure is ook gestandaardiseer en vyf Bt variante, naamlik Bt var *israelensis*, Bt var *kurstaki*, Bt var *israelensis new strain* (Wits), Bt geïsoleer uit 'n dooie insek en Bt geïsoleer uit 'n grondmonster se proteïene is reeds op *M.incognita* (ras 2) getoets en al vyf laasgenoemde isolate, asook vyf Bt isolate van MYCOGEN se protoksiene en toksiene is op *Ditylenchus destructor* getoets. Geen toksisiteit is egter oor 'n tydperk van 7 dae waargeneem nie.

SUMMARY

Proteins isolated from several pure Bt variants and Bt isolated from soil and an insect cadaver were tested on *Meloidogyne incognita* (race 2) and *Ditylenchus destructor*. Several proteins were also isolated from different plant tissues and were tested on *Ditylenchus destructor*. The following procedures were also standardised: Isolation of Bt variants from soil, and insect cadavers and the *in vitro* cultivation of Bt bacteria on solid medium and in liquid culture. It was also possible to find a fast and easy method for the isolation of relatively pure crystal proteins. Various solvents were tested for effective solubilization of the crystal proteins. The best solvent proved to be 0, 015 N NaOH with a solubility of **47%** together with a toxicity of only 5% towards the nematodes and a 90% stability of the proteins. SDS-PAGE of proteins was also standardised. It was also possible to metabolise the protoxin to the toxin with the use of trypsin. Standardising work on the bioassay for the nematodes was completed. The proteins of five Bt variants, namely Bt var *israelensis*, Bt var *kurstaki*, Bt var *israelensis* new strain (Wits), Bt isolated from an insect cadaver and Bt isolated from soil were tested on *Meloidogyne incognita* (race 2) and the protoxins and toxins of all five of the above mentioned isolates as well as five isolates from MYCOGEN were tested on *Ditylenchus destructor*. No toxicity was observed over a period of seven days.