and negeviruses for humans and their interference with arbovirus replication in competent vectors is largely unknown, however, recent studies suggest that the insectspecific viruses could alter the vector competence of the mosquitoes for some arboviruses resulting in superinfection exclusion or by alteration of the vector's immune response.

PIV219 - IN VIVO AND IN VITRO VIRULENCE ANALYSIS OF A BACULOVIRUS ISOLATED FROM CHRYSODEIXIS (=PSEUDOPLUSIA) INCLUDENS, A SOYBEAN PEST IN THE BRAZILIAN CERRADO

de Souza, M.L.; Sanches, M.M.; Sihler, W.; Sosa Gomes, D.R.

1. *EMBRAPA RECURSOS GENÉTICOS E BIOTECNOLOGIA*
2. *EMBRAPA SOJA*

Baculovirus are important biological control agents of Lepidoptera. In February 2014, larvae with symptoms of viral infection were observed in populations of Chrysodeixis (=Pseudoplusia) includens infesting soybean field at BuritisMG (S15o22.2′ W46o50.7′). Observations of larval tissue under optical microscope showed the presence of typical virus particles of Nucleopolyhedrovirus (NPV). The virus was identified as Pseudoplusia includens single nucleopolyhedrovirus (PsinSNPV) by transmission electron microscopy. The present work was carried out in order to investigate the potential of this viral isolate to control this insect pest. Two bioassays were performed. In the first, larvae infected with virus were macerated and incorporated to the artificial diet and offered to 432 C. inludens larvae (third instar). In the second, semipurified virus particles were incorporated to the artificial diet and offered to third instar C. includens larvae. The mortality was verified at 10 and 15 d.p.i. and the lethal concentration LC50 and LC99 were calculated using Probit analysis. To select a good candidate for in vitro multiplication of the virus, analysis was performed with six different lepidopteran cell lines: Bombyx mori (BM5), Lymantria dispar (IPLBLD625Y), two Trichoplusia ni (BTITn5B14 and TN368), and two Spodoptera frugiperda cells (IPLB-SF21AE and Sf9).The cells were seeded at a density of 1x106 per 60mm2 dish. The virus was obtained from infected larvae hemolymph at 4 d.p.i., treated and allowed to be adsorbed by cells during 1 hour. Then, the cells were kept in TNMFH complete medium and incubated at 270C. Morphological analysis was monitored by light microscopy during five days, using an Olympus CK2 optical microscope. The best results in the bioassays were obtained with the semipurified virus. The LC50 obtained was 10,918 polyhedra/ ml artificial diet (pol/ml) and the LC99 was 247,710 pol/ml, at10 d.p.i. Furthermore, at 15 d.p.i the LC50 was 6,709 pol/ml and LC99 was 146,880 pol/ml. In addition, preliminary analysis of the cell lines incubated with this isolate showed typical cytopathic effects as cell rounding, nuclear hypertrophy and the presence of polyhedra inTn5B14 cells at 4 d.p.i. The bioassays showed potential to use this virus isolate as a biopesticide. Moreover, the Tn5B14 cell line demonstrated to be prospective for further in vitro studies.

PIV220 - COMPARISON OF THE INFECTIVITY OF TWO SPODOPTERA FRUGIPERDA CELL LINES TO SFMNPV

de Souza, M.L.; Sihler, W.; Sanches, M.M.; Barros, A.M.R.; Valicente, F.H.

1. *EMBRAPA RECURSOS GENÉTICOS E BIOTECNOLOGIA*
2. *UNIVERSIDADE PAULISTA*
3. *EMBRAPA MILHO E SORGO*

The fall armyworm, Spodoptera frugiperda, is a severe pest in South America causing damage to different crops, especially in maize. The Spodoptera frugiperda Multiple Nucleopolyhedrovirus (SfMNPV), a baculovirus highly pathogenic to this pest, has been largely used as a biocontrol agent. So far the baculovirus production has been done by multiplication of the virus in its insect host in despite of difficulties as intense cuticle lyses and cannibalism behavior. Therefore, optimization of baculovirus in vitro production is essential as an alternative technology. In the present work, the SfMNPV production in IPLBSF21AE and Sf9 cell lines were compared. The polyhedra yield was determined as well as the kinetics of viral protein synthesis. In addition, larval mortality was determined by virulence assays with 3th to 4th instar larvae. Cells seeded at a density of 2X106 in a T25 flasks were incubated with the SfMNPV I19 isolate for 1h adsorption time and kept in TNMFH complete medium at 270C. At 5 dpi the cells were collected by centrifugation at 3000rpm for 5 min. The cell pellet was disrupted by treatment with 1% SDS, for 1h, at