**EXPERIMENTAL USE ONLY**

# ABIETIV

Flowable Biological Insecticide

**FOR USE IN FORESTRY**

**NOT FOR SALE**

**NOT FOR DISTRIBUTION TO ANY PERSON OTHER THAN RESEARCHER OR COOPERATOR**

**READ THE LABEL BEFORE USING**

**KEEP OUT OF REACH OF CHILDREN**

GUARANTEE: *Neodiprion abietis Nucleopolyhedrovirus*, NeabNPV (Newfoundland strain): $4 \times 10^9$ polyhedral inclusion bodies (PIBs) per milliliter.

**RESEARCH PERMIT NO: 21-RP-05 PEST CONTROL PRODUCTS ACT**

Net Contents: 40 mL ($1.6 \times 10^{11}$ PIBs)

Natural Resources Canada
Canadian Forest Service – Atlantic Forestry Centre
P. O. Box 4000, 1350 Regent Street
Fredericton, New Brunswick, E3B 5P7

**NOTICE TO USER:** This control product is to be used only in accordance with the directions on this label. It is an offence under the *Pest Control Products Act* to use a control product act under unsafe conditions.
**NATURE OF RESTRICTION:** This product is to be used only in the manner authorized. Consult provincial pesticide regulatory authorities about use permits that may be required.
**RESTRICTED USE:** For use against balsam fir sawfly larvae (*Neodiprion abietis*) in forests.
**DIRECTIONS:** Mix with a 20% aqueous solution of molasses at a rate of 1 mL Abietiv to 10L molasses solution. Aerial application. Apply 1-5 billion ($10^9$) PIBs/hectare of Abietiv in 2.5 L 20% aqueous solution of molasses/hectare. Apply when larvae are young (preferably first and second instars) and are actively feeding. To be effective, larvae must ingest foliage with deposits of Abietiv. Uniform spray deposit coverage of the foliage is essential for optimum control. Recommended droplet size is 100 μm.

**PRECAUTIONS**

**KEEP OUT OF REACH OF CHILDREN**

**POTENTIAL SENSITIZER**

**CAUTION EYE IRRITANT**

PRECAUTIONS: KEEP OUT OF REACH OF CHILDREN. Avoid contact with skin, eyes or clothing. Wear a long-sleeved shirt, long pants, waterproof gloves and eye goggles when handling, mixing/loading or applying the product and during all clean-up/repair activities. Wash thoroughly with soap and water after handling. Remove contaminated clothing and wash before reuse.

**FIRST AID**

IF SWALLOWED - Rinse mouth and throat with copious amounts of water. Do not induce vomiting.
IF ON SKIN/CLOTHING – Take off contaminated clothing. Wash skin with plenty of soap and water.
IF INHALED – Move to fresh air.
IF IN EYES – Hold eye open and rinse slowly and gently with water. Remove contact lenses, if present, and then continue rinsing eye.
GENERAL – Seek medical attention immediately if irritation or signs of toxicity occur and persist or are severe. Take container or experimental label with you when seeking medical attention.

STORAGE: Store in the refrigerator at 4°C. Store container upright and keep tightly closed when not in use. Shake vigorously to resuspend contents immediately prior to addition to molasses solution.

DISPOSAL: Do not reuse container. Follow provincial instructions for any required cleaning of the container prior to its disposal. Make empty container unsuitable for use and dispose in accordance with provincial requirements. **Return any unused, unwanted product to the Canadian Forest Service**.

## NeabNPV Product Profile and Proposed Use Patterns

a) *Neodiprion abietis Nucleopolyhedrovirus* NeabNPV Baculoviridae

b) NeabNPV is a baculovirus within the genus *Nucleopolyhedrovirus* (NPV). NPVs are a large group of viruses with covalently closed, double-stranded DNA genomes of 88-153 kilobases (kb) (NeabNPV genome is approximately 95 kb). In NeabNPV, numerous singly enveloped virions are occluded within polyhedral inclusion bodies (PIBs) that are 0.5–1.0 μm in diameter. Polyhedrin is a 29-kiloDalton protein. Baculoviruses are restricted to arthropods, mostly insects. NPVs have a high degree of host specificity affecting a single insect species or ones that are closely related. Sawfly NPVs are especially host specific and those described to date only seem to infect and replicate in the midgut epithelial cells of a single host species (Wallace and Cunningham 1995). Sawfly NPVs (including NeabNPV) are ingested by host larvae. Polyhedrin is dissolved in the gut releasing the virions. The virions fuse with the microvilli of the midgut epithelial cells and nucleocapsids are transported into the nucleus where the virus uncoats, undergoes replication and morphogenesis. Ultimately, the host cell dies and lyses releasing PIBs into the gut lumen of the host and PIBs pass out with the frass and may then be consumed by other host larvae. Infected host larvae usually die with 7 – 14 days. NeabNPV was isolated from balsam fir sawfly (*Neodiprion abietis*) larvae collected near Corner Brook, Newfoundland in 1997.

c) The recommended application rate of NeabNPV and other sawfly NPVs is 1-5 x $10^9$ PIBs/ha (see also Wallace and Cunningham 1995).

d) Bioinsecticide (larvicide).

e) Restricted for use in forestry.

f) NeabNPV (4 x $10^9$ PIBs/mL) suspended in water. To be diluted 1:2,000 to 1:10,000 with 20% aqueous molasses.

g) *Neodiprion abietis* (Diprionidae: Symphyta), the balsam fir sawfly.

h) Normally 1-5 x $10^9$ PIBs/ha (but, up to 1 x $10^{10}$ PIBs/ha for the purpose of NeabNPV field production) suspended in 20% aqueous molasses and applied at a volume of 2.5L/ha.

i) Single application to coincide with first and second larval instars. NeabNPV must be ingested by feeding larvae to be effective.

j) Fix-winged aircraft or helicopter equipped with Micronair® AU 4000 nozzles or equivalent. May also be applied from the ground using powered ground sprayers or backpack sprayers.

k) Standard procedures for aerial applications (protective clothing, eyewear, etc.) to be followed.

l) Sawfly NPVs are only known to infect and replicate in their specific sawfly host. Use buffer and exclusion zones as designated by federal and provincial environmental agencies.

## Biological Properties of NeabNPV

a) NeabNPV has been reported in populations of balsam fir sawfly from Alberta, Saskatchewan, Manitoba, Ontario, Quebec, Ontario (see Olofsson 1972) and Newfoundland (Campbell et al. 2004). Balsam fir sawfly feed on one-year old and older foliage of balsam fir (*Abietis balsamea*). NeabNPV is the principle cause of balsam fir population declines (Moreau et al. 2004) and would be pervasive in the environment surrounding these declining populations.

b) Balsam fir sawfly, *Neodiprion abietis* (Hymenoptera: Diprionidae). Larval infection is *per os*. NeabNPV virions infect epithelial cells of the larval midgut. Viral replication is only known to occur in the nucleus of midgut epithelial cells of larval sawflies (Federici 1993). The $LD_{50}$ for NeabNPV against balsam fir sawflies at 7 days post-inoculation (95% confidence limits) for second instars was 87.4 PIBs (7.1-304.9) and 2074.8 PIBs (813.1-3999.0) for third instars. Sawfly larvae tend to feed gregariously. This combined with high virulence contribute to sawfly NPVs being highly contagious amongst host larvae.

c) NeabNPV is known to only infect and replicate in the midgut cells of balsam fir sawfly larvae. NeabNPV may cause mortality in other sawfly larvae, specifically *Acantholyda erythrocephala* (pine false webworm, Pamphiliidae), *Diprion similis* (introduced pine sawfly, Diprionidae), *Gilpinia hercyniae* (European spruce sawfly, Diprionidae), *Pristiphora geniculata* (mountain ash sawfly, Tenthredinidae). NeabNPV does not appear to replicate in these other sawfly species.

d) Sawfly NPVs only infect the midgut epithelium of host sawflies (Federici 1993). Following the virus replicative cycle, infected cells, containing PIBs, are sloughed off into the frass and out of the body where they can be ingested and subsequently infect other host insects. Host death normally occurs within a one to two weeks but, during that time, the host produces infective units of the disease.

e) No plasmids or extra chromosomal DNA. NeabNPV is a naturally occurring pathogen of the balsam fir sawfly and was isolated from that host in Newfoundland in 1997. NeabNPV genome is a covalently closed, double stranded DNA approximately 95 kb in size. This virus has not been subjected to any type of nucleic acid recombination.

f) NeabNPV can only be produced *in vivo* in balsam fir sawfly larvae. Currently, there are no tissue culture systems available for the production of NeabNPV. Like most baculoviruses, NeabNPV is probably sensitive to UV radiation.

g) Not known to have any unusual morphological, physiological or biochemical characteristics.

h) Experimental ground applications of NeabNPV were made against balsam fir sawfly in Ontario in the early 1970s (Olofsson 1972) and extensive field trials have been carried by the Canadian Forest Service in Newfoundland in 2000-2003. Other sawfly NPVs have been used successfully in trials against pest sawflies (Wallace and Cunningham 1995). Registration for two sawfly NPVs for use to control sawfly pests have previously been sought in Canada: Sertifervirus (*Neodiprion sertifer Nucleopolyhedrovirus*, NeseNPV) for European spruce sawfly (*Neodiprion sertifer*) and Lecontvirus (*Neodiprion lecontei Nucleopolyhedrovirus*, NeleNPV) for redheaded pine sawfly (*Neodiprion lecontei*). (See Wallace and Cunningham 1995).

i) No known relationship to any pathogen of plants or vertebrates. NeabNPV belongs to the Baculoviridae, a family of viruses known only to infect arthropods, mostly insects. (See, Gröner 1986, 1990, OECD 2002 for reviews of the scientific literature).

j) No know relationship to any known human dermatophyte.

k) NeabNPV is not related to any toxigenic human pathogen. (See Gröner 1986, OECD 2002 for reviews of the scientific literature).

l) No adverse effects by baculoviruses to humans or other vertebrates. (See, Gröner 1986, OECD 2002 for reviews of the scientific literature).

## NeabNPV Product Characterization and Analysis

*Product Manufacture and Formulation*

Natural Resources Canada
Canadian Forest Service – Atlantic Forestry Centre
1350 Regent Street
P.O. Box 4000
Fredericton, New Brunswick, E3B 5P7

*Proposed Trade Name:* Abietiv

## Origin, Derivation and Identification of NeabNPV

a) *Neodiprion abietis Nucleopolyhedrovirus* (NeabNPV, Baculoviridae) (Volkman et al. 1995).
b) In August 1997, balsam fir sawfly larvae were collected from two plots near Corner Brook, Newfoundland (Ecozone 5). These insects were reared in a laboratory at the Canadian Forest Service in Fredericton, New Brunswick and larvae that died in rearing were examined for the presence of NeabNPV. This virus was found in a number of larvae and was isolated. Amplification of NeabNPV was carried out at the Pasadena Field Station in July and August in 1998 and 1999. Here, larvae were reared on balsam fir foliage in 5-L plastic tubs. NeabNPV was applied to the foliage and dead insects were picked from the foliage, by hand, and were frozen. Since 1999, NeabNPV has been mass-produced according to the method described below.
c) Stock isolates of NeabNPV are held at either 4°C or -20°C.

## Manufacturing Methods and Quality Assurance

*Preservation and Maintenance of the Productive Strain*

Balsam fir sawfly larvae infected with NeabNPV from the original collection area have been frozen and stored at -20°C.

*Manufacturing Processes*

Semi-purified NeabNPV PIBs in 20% aqueous molasses (commercial grade) are applied to balsam fir trees infested with balsam fir sawfly larvae using fixed-wing aircraft, helicopters, motorized ground-sprayers and/or backpack sprayers up to the equivalent concentration of 1 x $10^{10}$ PIBs/ha. Collections of larvae begin at the first sign of larval mortality (about 7 days after application) and continue for the next 10 days. Larvae are knocked onto tarpaulins placed under balsam fir trees by beating the tree branches with a 2-m length of pruning pole. Collected larvae are transferred to 50-lb brown paper bags so that the bags are one-third filled with larvae and fir needles. Three, 30-cm, branch tips cut from balsam fir trees are added as a source of food and an additional 2-3 mL of NeabNPV PIBs suspended in water (1 x $10^{7}$ PIBs/mL) is misted onto the foliage. The bag tops are folded over and stapled shut. The larvae are left in the bags to die or finish their development at ambient laboratory temperatures (18-20°C). Following the death of the larvae, the branch tips are removed and the contents of three bags are placed into a single, clean 50-lb brown paper bag. These bags are stapled shut and are stored in the laboratory at ambient temperature (18-20°C). By this time the needles and dead larvae are quite dry. Dead larvae, from these bags, are picked out from the needles,

by hand, and are placed into 50-mL centrifuge tubes and frozen at -20°C. NeabNPV from the dead larvae are purified using the method described below.

*NeabNPV Isolation Protocol - Large Scale.*

1. Thaw and re-hydrate NeabNPV-infected balsam fir larvae in water, overnight.
2. Homogenize larvae in a 1000 mL beaker using a hand held blender.
3. Dilute with water and add 1% SDS to a concentration of 0.3% (final volume approximately 10 times the volume of dead larvae).
4. Add magnetic stirrer bar and stir for 60 min.
5. Filter through plastic mesh, save filtrate (contains NeabNPV).
6. Resuspend solid debris in 0.3% SDS and stir as in step 4 for 5 min.
7. Filter again through plastic mesh and repeat until clear filtrate is obtained.
8. Filter NeabNPV suspension through 8 layers of cheesecloth.
9. Centrifuge for 15 min in a Sorvall RC 28S centrifuge and HS-4 rotor (or equivalent at approximately 2000 x g).
10. Discard supernatant and add more of the NeabNPV suspension to centrifuge tubes, repeat steps 9 and 10 until all the NeabNPV suspension has been used.
11. Resuspend NeabNPV PIB pellets in 0.3% SDS and vortex.
12. Repeat centrifugation and resuspension until a clear supernatant is obtained.
13. Resuspend and pool NeabNPV PIB pellets.
14. Resuspend pellet in 0.5M NaCl. Centrifuge.
15. Resuspend pellet in water (5 – 10 x volume of pellet).

NeabNPV suspensions are stored in water at 4°C to inhibit growth of contaminating bacteria. To further reduce unwanted bacterial propagation, NeabNPV suspensions are only added to the hopper on the aircraft containing the 20% aqueous solution of molasses immediately prior to the aircraft taking-off to the spray site in field applications.

*Quality Assurance*

NeabNPV PIBs are too small (0.5-1.0 μm) to be counted accurately using a hemocytometer. Instead, PIBs are quantified by combining a known volume of an unknown concentration of NeabNPV PIBs with a known volume of a known concentration of latex beads (2.97 μm diameter, SD 0.04). Latex beads and PIBs from 50 fields of view on each of four slides are counted under the 100X oil lens of a compound microscope. The concentration of PIBs is determined as a proportion of the number of latex beads counted. The volume of the supernatant is adjusted to give a final concentration of $1.4 \times 10^9$ PIBs/mL. When a 40 mL volume of NeabNPV at this concentration is place in a 50-mL centrifuge tube, there is sufficient virus to spray 160 ha at an application rate of $1.0 \times 10^9$ PIBs /ha in a mix volume of 2.5L/ha (400 L total mixture volume).

*Potency Estimation*

The potency of NPVs is expressed in terms of the number of PIBs that provide a lethal dose to 50 percent of test insects ($LD_{50}$). The $LD_{50}$ at 7 days post-inoculation (95% confidence limits) for second instars was 87.4 PIBs (7.1-304.9) and 2074.8 PIBs (813.1-3999.0) for third instars. In aerial application trials against lepidopteran larvae, the range has been in the order of $1.6 \times 10^{10}$ to $4.4 \times 10^{12}$ PIBs/ha (Cunningham and Kaupp 1995, Wallace and Cunningham 1995).

For sawfly NPVs the range has been 1.3 x $10^9$ to x 3.9 $10^{11}$ PIBs/ha (Cunningham and Kaupp 1995, Wallace and Cunningham 1995). In field efficacy trials of NeabNPV, we have found that applications of 1–3 X $10^9$ PIBs/ha can result in significant declines in sawfly populations in spray blocks compared to controls, in the year following NeabNPV application. In the year of the application, numbers of balsam fir sawfly larvae/$m^2$ foliage do not necessarily decline to a greater extent in the application versus control blocks but, the levels of NeabNPV infection is generally much higher in the application blocks following the spray. Timing of the applications is important. Earlier instars (first and second) are more susceptible to the virus than are the later instars. Also the stage of the population cycle at the time of the application can also affect NeabNPV efficacy. Generally, it can be said that, applications against first and second instar balsam fir sawfly larvae prior to peak population levels will be more effective than at any later stage.

*Unintentional Ingredients*

Baculoviruses are obligate pathogens of insects and can only be produced in the host species, a susceptible insect usually closely related to the host or in an *in vitro* insect cell line. Sawfly NPVs are highly host specific and no *in vitro* cell line has been developed for any sawfly or sawfly NPV. Thus, sawfly NPVs must be produced in the host sawfly. Sawfly NPVs only replicate in the midgut epithelium of the host species. Lepidopteran NPVs, on the other hand, initially infect the larval midgut but then the infection spreads to other tissues such as the fat body and hemocytes. Many lepidopterans can be reared on artificial diets; this is not true of sawflies. The inability to rear sawfly larvae on artificial diets means that the larvae must be reared on host plant foliage and limited tissue tropism means that the amount of virus produced per larva is lower than what one would get from a lepidopteran larva. The advantage of sawfly NPVs is that they are highly communicable between their hosts and aerial application rates can be one or two orders of magnitude lower than those for lepidopterans (Cunningham and Kaupp 1995, Wallace and Cunningham 1995). To produce sawfly NPVs economically, it must be done in the field (Cunningham and MacPhee 1986). We have reared balsam fir sawfly larvae, on foliage, in the laboratory, intensively for up to six months and have produced sufficient NeabNPV to spray 2,000 ha at a rate of 1.0 x $10^9$ PIBs/ha. Using aerial applications in the field, we have produced this much from collecting NeabNPV-infected larvae in one week from 1 ha. Field production, however, does result in contamination by microbes that are part of the microflora of the environment from which the larvae were collected. Also, since the aircraft used to apply NeabNPV are also used to spray *Bacillus thuringiensis*, this bacterium may also contaminate the final product. The current mixing procedure for NeabNPV is 1 ml of NeabNPV at 4 x $10^9$ PIBs/mL in 2-10 L 20% aqueous molasses. Thus, the dilution factor for the NeabNPV suspension and any contaminating microbes is 2,000 to 10,000 times and 2.5 L of this are applied to a hectare of forest. NeabNPV is only added to the molasses carrier immediately before the aircraft taxis for take-off to the spray block. Waiting to add NeabNPV to the molasses just before take-off reduces the time during which any contaminating bacteria would have to reproduce.

As part of the manufacturing process, filtration and centrifugation are used to remove as much insect tissue and other contaminating debris as possible. However, cells, tissues, fats, proteins, nucleic acids and other materials derived from the insect host may remain in the product following these steps in the manufacturing process. As a precaution against the inadvertent

inclusion of vertebrate pathogenic microbes and/or metabolites, each batch of NeabNPV product is sent to accredited laboratories for bacterial screening and mouse intraperitoneal injection assays.

*Analysis for Microbial Contaminants*
Microbial contaminant analyses have most recently been conducted by IG MicroMed Environmental Inc., Richmond, B.C. This company is accredited by Standards Council of Canada (SCC) for most of the test methods of interest (DACO). We will continue to use them or an equivalent company to test for microbial contaminants. The references for the methods used are listed below.

Total Aerobic Bacteria
MFHPB-18. Determination of the aerobic colony counts in foods. October 2001. Health Products and Food Branch, Ottawa, ON. In: *Compendium of Analytical Methods*. http://www.hc-sc.gc.ca/food-aliment/mh-dm/mhe-dme/compendium/volume_2/ e_mfhpb1801.html

Total Coliforms, Fecal Coliforms, *Escherichia coli*
MFHPB-19. Enumeration of coliforms, faecal coliforms and of *E. coli* in foods using the MPN Method. April 2002. In: *Compendium of Analytical Methods*. Health Products and Food Branch, Ottawa, ON. http://www.hc-sc.gc.ca/food-aliment/mh-dm/mhe-dme/compendium/ volume_2/e_mfhpb1901.html

Fecal *Streptococcus*/*Enterococcus*
Standard Methods for the Examination of Water and Wastewater, 20th Ed. 1998. Section 9230 Fecal *Streptococcus* and *Enterococcus* Groups. Part 9000, Microbiological examination.

*Salmonella* spp.
MFHPB-20. Isolation and identification of *Salmonella* from foods. April 1998. In: *Compendium of Analytical Methods*. Health Protection Branch, Ottawa, ON. http://www.hc-sc.gc.ca/food-aliment/mh-dm/mhe-dme/compendium/volume_2/e_mfhpb2001.html

*Shigella* spp.
BAM Chapter 6, *Shigella*. January 2001. In: *U.S. Food and Drug Administration – Bacteriological Analytical Manual (BAM)*. Center for Food Safety & Applied Nutrition.

Aerobic Sporeformers
MFLP-44. Determination of aerobic and anaerobic sporeformers. April 1998. In: *Compendium of Analytical Methods*. Health Protection Branch, Ottawa, ON. http://www.hc-sc.gc.ca/food-aliment/mh-dm/mhe-dme/compendium/volume_3/e_mflp4401.html

*Bacillus cereus*
MFLP-42. Isolation and enumeration of *Bacillus cereus* in foods. April 2003. In: *Compendium of Analytical Methods*. Health Products and Food Branch, Ottawa, ON. http://www.hc-sc.gc.ca/food-aliment/mh-dm/mhe-dme/compendium/volume_3/e_mflp4201.html

*Staphylococcus aureus*
MFHPB-21. Enumeration of *Staphylococcus aureus* in foods. November 2000. In: *Compendium of Analytical Methods*. Health Protection Branch, Ottawa, ON. http://www.hc-sc.gc.ca/food-aliment/mh-dm/mhe-dme/compendium/volume_2/e_mfhpb21e01.html

*Vibrio* sp. (*cholerae*)
BAM Chapter 9, *Vibrio cholerae*, *V. parahaemolyticus*, *V. vulnificus*, and other *Vibrio* spp. January 2001. In: *U.S. Food and Drug Administration – Bacteriological Analytical Manual (BAM)*. Center for Food Safety & Applied Nutrition. http://www.cfsan.fda.gov/~ebam/bam-toc.html

Molds and Yeast
MFHPB-22. Enumeration of yeasts and moulds in foods. March 2002. In: *Compendium of Analytical Methods*. Health Protection Branch, Ottawa, ON. http://www.hc-sc.gc.ca/food-aliment/mh-dm/mhe-dme/compendium/volume_2/e_mfhpb2201.html

Vertebrate Pathogens
Mouse IP injections are conducted according to Good Laboratory Practices (GLP) at an accredited laboratory. Observation period, seven days. Currently carried out by Alberta Research Council, Hwy 16A & 75 Street, Postal Bag 4000, Vegreville, AB.

## Summary of Physical and Chemical Properties

a) Physical state; solids (NeabNPV PIBs, approximately 0.5-1.0 μm diameter) suspended in water.
b) Density, bulk density of specific gravity; not applicable.
c) Viscosity; water, 1.0
d) Corrosion character; not corrosive
e) Suspendibility; PIBs suspended in water.

## References

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Wallace, D. R. and J. C. Cunningham. 1995. Diprionid sawflies. *In*: J. A. Armstrong and W. G. H. Ives (editors). Forest Pest Insects in Canada. Natural Resources Canada. Ottawa, Ontario. Pp. 193-232.

# DRAFT MATERIAL SAFETY DATA SHEET

*Product name*: Abietiv
*Chemical name*: Balsam fir sawfly (*Neodiprion abietis*) nucleopolyhedrovirus (NeabNPV)
*Formula*: Biological organism, virus, Baculoviridae: Nucleopolyhedrovirus
*Molecular weight*: Not applicable
*Synonyms*: Balsam fir sawfly nuclear polyhedrosis virus
*Chemical family*: Not applicable

## I. Physical data

*Boiling point*: Not applicable
*Freezing point*: Not applicable
*Specific gravity*: 1.0014
*Vapour pressure*: Not applicable
*Vapour density*: Not applicable
*Evaporation rate*: Not applicable
*% Volatiles by volume*: None
*Solubility in water* (% by wt): Insoluble
*Appearance and colour*: brownish suspension, musty odor.

## II. Ingredients

Balsam fir sawfly nucleopolyhedrovirus (NeabNPV) polyhedral inclusion bodies (PIBs) 1.5%
Water 98.5%
*Impurities*: Proteins, fats, cells, tissues, cuticle from host insect (balsam fir sawfly).

This nucleopolyhedrovirus is non-toxic to vertebrate animals. Impurities may cause eye irritation.

## III. Fire and explosion hazard data

*Flash point*: Not applicable
*Flammable limits*: Not applicable
*Extinguishing media*: Water
*Special fire fighting procedures*: None except to avoid inhalation of particulates released by fire.
*Unusual fire and explosion hazards*: Not applicable

## IV. Health hazard data

*Oral*: A single dose of $1 \times 10^8$ PIBs by oral gavage showed no evidence of acute oral toxicity or pathogenicity to Sprague-Dawley® rats (average initial weights 101-124 g).
*Intravenous injection*: A single dose of $1 \times 10^7$ PIBs by intravenous injection showed no evidence of acute injection toxicity or pathogenicity to Sprague-Dawley® rats (average initial weights 112-129 g).
*Inhalation*: A single dose of $1 \times 10^8$ PIBs by intratracheal instillation showed no evidence of acute pulmonary toxicity or pathogenicity to Sprague-Dawley® rats (average initial weights 123-148 g).
*Dermal*: A single topical dose of 2 g NeabNPV/kg body weight showed no evidence of acute

dermal toxicity or pathogenicity to New Zealand white rabbits.

EMERGENCY AND FIRST AID PROCEDURES:

Remove from exposure situation. If in eyes, flush with plenty of water. If irritation persists, get medical attention. If on skin, wash with soap and water.

NOTES TO PHYSICIAN

Prolonged exposure may cause allergies and hypersensitivity in some individuals.

## V. Reactivity data

*Stability*: Stable
*Conditions to avoid*: Do not store in direct sunlight (ultraviolet sensitive) or at temperatures above 30°C.
*Incompatibility*: Not applicable
*Hazardous decomposition products*: May contain bacteria

## VI. Spill or leak procedures

STEPS TO BE TAKEN IF MATERIAL IS RELEASED OR SPILLED

Containment and cleanup by placing in a sealable container for transport to an approved disposal site.

WASTE DISPOSAL METHOD

Triple rinse containers and dispose of them at an approved site. Washing waste from this product may be disposed of on site or at an approved disposal site.

## VII. Special protection information

*Respiratory protection*: Medical face mask as appropriate.
*Ventilation*: Use in areas with good ventilation
*Protective clothing*: Coveralls, gloves, safety goggles and medical facemask are required for mixers.
*Other protective equipment*: Have eyewash, soap and water available at project location.

## VIII. Special precautions

KEEP OUT OF REACH OF CHILDREN.

1. Avoid direct application to bodies of water.
2. Do not contaminate water, food or feed by inappropriate storage and disposal.
3. Only for use as a biological insecticide for balsam fir sawfly control programs limited to forestry.
4. Avoid heat and direct sunlight.
5. Other handling and storage conditions: Wastes from this product may be disposed of on site or at an approved waste disposal facility.
6. Do not reuse empty containers but arrange for disposal in a sanitary landfill or by incineration.