European Commission

Draft Assessment Report prepared according to the Commission Regulation (EU) N° 1107/2009

Spodoptera exigua multicapsid nucleopolyhedrovirus (SeMNPV)

Product data: SPEXIT

Volume 3 – Annex B.5 Analytical Methods

Rapporteur Member State: Spain

April 2020

Version History

When	What
18/09/2018	Completeness check report of the dossier submitted by the notifier
December 2019	DAR submitted to the Notifier for commenting
February 2020	DAR updated with notifier comments
April 2020	DAR updated after EFSA completeness check

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INTRODUCTION

The company Andermatt Biocontrol GmbH submits the current dossier for the approval of the baculovirus (BV) *Spodoptera exigua* multi nucleopolyhedrovirus (SeMNPV) as a new microbial pest control agent (MPCA) and SPEXIT as its reference microbial pest control product (MPCP) to the European Authorities.

BVs used as MPCA in the EU are regulated as microorganism according to Regulation 1107/2009[1]. Data requirements for the registration of BVs as an active substance and their products are laid down in part B of the regulation documents 283/2013 [2]and 284/2013 [3]and the principles for evaluation and authorization of plant protection products contained microorganism according to regulation 546/2011[4].

BV isolates however, represent a unique case in which the wild type isolates are genetically heterogeneous (mixture of different genotypes or pool of isolates). These variations may influence in some biological properties, such as the virulence, but it has no consequences on the safety towards non-target organisms or the environment. Isolation of a single genotype is difficult and even not appropriated, since genetic variation is needed to account for variation in the target organisms and obtain better efficacy in the control of insect populations. Therefore, the BVs were not necessary evaluated at strain level (Sanco/0253/2008). [5]The high similarity between BVs justifies a general assessment at the level of the family *Baculoviridae*, considering species-specific information where necessary. The proposed procedure to include BVs at species level was adopted by the member states and the European Regulatory Authorities already in 2007, when the first BV species was included in Annex I, and for the REBECA proposal 2008[6], for a simplified inclusion of BVs on the species level into Annex I. Most of the formally required data are published and equal for all BVs, already assessed by MS and EU authorities and therefore, some data on the isolate or species level are not mandatory.

The BVs are included on species level in Annex I of directive 1107/2009 and the different pool of isolates were added after they have been evaluated to a separate list, to be maintained in the Review Report and to be amended by taking note in the Standing Committee (Sanco/0253/2008). This approach has been confirmed by a decision in the Standing Committee on May 15, 2007[7] were *S. exigua* NPV was listed at species level in Annex I. The experience that BVs present no risk for the environment have been confirmed by numerous studies during the last fifty years, since their first use as biocontrol agents. With regard to safety considerations, it is important to note that the whole *Baculoviridae* family are naturally present in our environment and are closely associated with their host occurrence. Therefore, their application in pest control would only produce a non-permanent fluctuation of the virus titre in the biotope of the pest insect. Due to their host specificity, BVs do not affect other organisms like vertebrates, arthropods other than their host species, microorganisms, or plants. BVs do not produce any metabolites at all.

For the BV specie *S. exigua* multicapsid nucleopolyhedrovirus (SeMNPV) a DAR with a reference isolate (Florida isolate SeNPV-F1, the first applied for) was approved in 2006 and the isolate SeNPV-F1 was listed on Annex I. Two new more isolates were further applied for at Member State level: the SeMNPV-SP2, approved in 2008 and

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[1]*Regulation (EC) No 1107/2009 of the European Parliament and of the Council of 21 October 2009 concerning the placing of plant protection products on the market and repealing Council Directives 79/117/EEC and 91/414/EEC. Official Journal of the European Union L 309, 1-50.*

[2] Commission Regulation (EU) No 283/2013 of 1 March 2013 setting out the data requirements for active substances, in accordance with Regulation (EC) No 1107/2009 of the European Parliament and of the Council concerning the placing of plant protection products on the market. Official Journal of the European Union L 93, 1-84.

[3]*Commission Regulation (EU) No 284/2013 of 1 March 2013 setting out the data requirements for plant protection products, in accordance with Regulation (EC) No 1107/2009 of the European Parliament and of the Council concerning the placing of plant protection products on the market. Official Journal of the European Union L 93, 85-152.*

[4]*Commission Regulation (EU) No 546/2011 of 10 June 2011 implementing Regulation (EC) No 1107/2009 of the European Parliament and of the Council as regards uniform principles for evaluation and authorisation of plant protection products. Official Journal of the European Union L155, 127-175.*

[5]*SANCO/0253/2008 rev. 2, 22 January 2008. Guidance Document on the assessment of new isolates of baculovirus species already included in Annex I of Council Directive 91/414/EEC.*

[6]*Ehlers RU., 2011 Regulation of Biological Control Agents and the EU Policy Support Action REBECA. In: Ehlers RU. (eds) Regulation of Biological Control Agents. Springer, Dordrecht.*

[7]*Review report for the active substance Spodoptera exigua nuclear polyhedrosis virus. Finalised in the Standing Committee on the Food Chain and Animal Health at its meeting on 15 May 2007 in view of the inclusion of Spodoptera exigua nuclear polyhedrosis virus in Annex I of Directive 91/414/EEC. Spodoptera exigua NPV SANCO/T14/2007 - rev. final1 12 March 2007.*

the SeNPV-BV0004, approved in 2010. Conversely, the current dossier was based on the data already assessed by the MS and EU authorities:

- The previous DAR document for the approval of a new active substance SeNPV-F1 submitted by Mitsui Agri Science International S.A and evaluated by The Netherlands in 2007.
- The evaluation report of the new isolate of SeMNPV, BV0004 previously submitted by the company Andermatt Biocontrol GmbH and evaluated by the Netherlans in 2010.

Active substances are approved for maximum period of 10 years under Directive 91/414/EEC[8]. The active substance SeMNPV was under programme of renewal Regulation EU 686/2012 (AIR-III programme[9]). According to draft working document AIR III renewal programme SANCO/2012/11284[10], *Spodoptera exigua* nuclear polyhedrosis virus was included in Batch 9" Active substance *Spodoptera exigua* nuclear polyhedrosis virus No application for renewal of approval has been submitted. Previous expiry date 30/11/2017"

Commission implementing regulation (EU) No 844/2012[11] setting out the provisions necessary for the implementation of the renewal procedure for active substances, as provided for in Regulation (EC) No 1107/2009 establishes in its Art 1: "the application for the renewal of an approval of an active substance shall be submitted by a producer of the active substance to the rapporteur Member State, no later than three years before the expiry of the approval"

The application for the renewal of the active substance *Spodoptera exigua* nuclear polyhedrosis virus was not submitted before of three years before the expiry date of the approval of the active substance SeMNPV (30/11/2017).

The applicant then have submitted an application for SeMNPV as a new active substance.

In this RAR, the information submitted regarding *Spodoptera exigua* multicapside nucleopopyhedrovirus (SeMNPV) is evaluated as new active substance, therefore, all information is considered and evaluated as new.

Literature reference included by the applicant comes from a literature search according to EFSA (2011) [12]in order to identify relevant recent published peer reviewed references covering the last 10 years. The RMS has also included relevant studies considered important to support the application for the approval of *Spodoptera exigua* multipolyhedrivirus (SeMNPV) genotype pool BV-0004 and the microbial product SPEXIT.

[8] *Council Directive 91/414/EEC of 15 July 1991 concerning the placing of plant protection products on the market. OJ L 230 of 19.8.1991.C.*
[9] *Programme of renewal Regulation EU 686/2012 (AIR-III programme).*
[10] *SANCO/2012/11284 –rev. 22, December 2018. Draft working document AIR III renewal programme.*
[11] *Commission implementing regulation (EU) No 844/2012, of 18 September 2012. Setting out the provisions necessary for the implementation of the renewal procedure for active substances, as provided for in Regulation (EC) No 1107/2009 of the European Parliament and of the Council concerning the placing of plant protection products on the market.*
[12] *Submission of scientific peer-reviewed open literature for the approval of pesticide active substances under Regulation (EC) No 1107/2009. EFSA Journal 2011;9(2):2092.*

PPP (product name/code):	SPEXIT	Formulation type:	SC
Active Substance: (SeMNPV)	*Spodoptera exigua* multicapsid nucleopolyhedrovirus	Conc. of a.s.:	3.75×10^{12} OBs/L
Applicant:	Andermatt Biocontrol GmbH	professional use	☒
Zone(s):	EU	non professional use	☒
Safener:	n.a.	Conc. of safener:	n.a.
Synergist:	n.a.	Conc. of synergist:	n.a.
Verified by RMS:	yes		

Table MP B.5.1. Summary of critical Good Agricultural Praxis for SPEXIT

1	2	3	4	5	6	7	8	9	10	11	12	13
Use-No.	**Member state(s)**	**Crop and/ or situation (crop destination / purpose of crop)**	**F G or I**	**Pests or Group of pests controlled (additionally: developmental stages of the pest or pest group)**	**Application**			**Application rate**			**PHI (days)**	**Remarks: e.g. g safener/synergist per ha**
					Method / Kind	**Timing / Growth stage of crop & season**	**Max. number (min. interval between applications) a) per use b) per crop/ season**	**L product / ha a) max. rate per appl. b) max. total rate per crop/season**	**OBs/ha a) max. rate per appl. b) max. total rate per crop/season**	**Water L/ha min / max**		
1	EU	Pepper (CPSAN)	F/G	*S. exigua* (LAPHEG)	Spray	At infestation (preferably on early larva instar: L1 and L2). First treatment just before hatching)	a) 18 (6) b) 18 (6)	a) 0.2 b) 3.6	a) 7.5×10^{11} b) 1.35×10^{13}	200 / 1600	-	- 2 to 3 applications per pest generation, up to 6 generations (i.e. max. of 18 app.). -Interval between applications: min. of 6 sunny days; 2 partially sunny days = 1 sunny day
2	EU	Leafy vegetables (lettuce crops) (3LETC)	F/G	*S. exigua* (LAPHEG)	Spray	At infestation (preferably on early larva instar: L1 and L2). First treatment just before hatching)	a) 18 (6) b) 18 (6)	a) 0.2 b) 3.6	a) 7.5×10^{11} b) 1.35×10^{13}	200 / 1600	-	- 2 to 3 applications per pest generation, up to 6 generations. -Interval between applications: min. of 6 sunny days; 2 partially sunny days = 1 sunny day

n.a. Not applicable

B.5.1 Methods for the analysis of the preparation

S. exigua multicapsid nucleopolyhedrovirus (SeMNPV) is a naturally occurring virus. Neither SeMNPV produce nor the end-use product (SPEXIT) contains chemical compounds of critical toxicological, environmental, or ecotoxicological concern. The same applies for the semi-synthetic insect diet which is based on sugar, wheat germ and dry yeast.

As SeMNPV does not produce antibiotics and secondary metabolites of toxicological and/or environmental, or ecotoxicological concern, the analytical methods described in the following are limited to the determination of the microorganism itself.

Methods to differentiate a mutant or genetically modified microorganism form the parent strain are not required, because the used strain of SeMNPV is of natural origin and not a mutant or genetically modified.

The fact that the cultivated strain is without mutants or genetic modifications has been verified by restriction endonuclease analysis of viral DNA. Spontaneous changes will most likely be reflected in a change of infectivity. Thus, any change will be detected in the bioassay that is performed to maintain product quality.

The content of SeMNPV is determined in a biotest. For details, please refer to confidential information, see Volume 3, Annex MA-4.

Regarding methods to identify contaminant microorganisms in the MPCP, please refer confidential information, see Volume 4, Annex C.

BVs do not produce any metabolites. For methods to show control to a specified and acceptable level, of microbial impurities and of any other impurities of toxicological concern, including toxic metabolites, which are known or suspected to be present at any stage of the manufacturing process, please refer confidential information, see Volume 4, Annex C.

For methods to show presence of any human and mammalian pathogens, please refer confidential information; see Volume 4, Annex C.

Please refer to Volume 3 MA B.4.2 regarding methods for determination of residues. Methods described for the active substance are as well applicable for the product.

The methods for determination of contaminants in the formulation are described according to following published methods:

Microbial contaminants and pathogens	**ISO guidelines methods**
Aerobic mesophile count	ISO 4833
Bacillus cereus	ISO 7932
Staphylococcus aureus	ISO 6888-2
Escherichia coli	ISO 16649-2
Salmonella spp.	ISO 6579

Table MP B.5.1-1 Analytical methods for microbial contaminants in SPEXIT

B.5.1.1 Methods for the identification and the determination of the content of the microorganism in the preparation

A method for identification and determination of the microorganism in the preparation is presented in the confidential information, See Volume 4, Annex C.

B.5.1.2 Methods to establish regular control of the preparation to show that it does not contain other organisms than the indicated ones and to establish uniformity

Confidential information, See Volume 4, Annex C.

B.5.1.3 Methods to identify any contaminating microorganisms of the preparation

The same methods as for the active microorganism are sufficient to detect and identify contaminating microorganisms in the formulated preparation. See Volume 4, Annex C.

B.5.1.4 Methods used to determine the storage stability and shelf life of the preparation

The method for determination of the storage stability at 5ºC for 12 months has been submitted under data point B.2.2, Storage stability and shelf-life, Annex B.2, in reference Konrad, R., 2014.

B.5.1.5 Methods for the determination of relevant impurities or metabolites in the manufactured material, if available

Confidential information, See Volume 4, Annex C.

B.5.2 METHODS TO DETERMINE AND QUANTIFY RESIDUES (VIABLE OR NON-VIABLE)

Please refer to Volume 3 MA B.4.2. Methods described for the active substance are as well applicable for the product.

Because no maximum residue level is proposed for *S. exigua* multicapsid nucleopolyhedrovirus (SeMNPV) analytical methods for the determination of residues of SeMNPV in plants, plant products, foodstuffs of plant and animal origin, or in feeding stuffs are not considered necessary.

S. exigua multicapsid nucleopolyhedrovirus is not pathogenic to humans and is not expected to produce toxins that are relevant for human health. In view of those conclusions, the Commission considers that the inclusion of such substance in Annex IV to Regulation (EC) No 396/2005 is appropriate (please refer to Commission Regulation (EU) 2016/439 of 23 March 2016 amending Annex IV to Regulation (EC) No 396/2005 of the European Parliament and of the Council as regards *S.exigua* multicapsid nucleopolyhedrovirus). In consequence, no MRL are required in crops, in foodstuffs and feeding stuffs, in animal and human body tissues and fluids. Additionally, no residue definition is proposed for environmental matrices (soil, water and air). Therefore, methods for the determination and quantification of residues are currently not required.

B.5.3 REFERENCE LIST

Data point	Author(s)	Year	Title Owner, Report No. Source (where different from owner) GLP or GEP status Published or not	Vertebrate study Y/N	Data protection claimed Y/N	Justification if data protection is claimed	Owner
KMP 2.1/01 KMP 2.2.1/01 MP B.5.1.4/01	Konrad, R.	2014	SPEXIT ONE YEAR STORAGE STABILITY AND CORROSION CHARACTERISTICS AT 5 °C Andermatt Biocontrol AG, CH, not available Andermatt Biocontrol AG, Grossdietwil, Switzerland GLP/GEP: no Published: no	N	Y		ABA
MP B.5.2/01	Anonymous	2016	Commission Regulation (EU) 2016/439 of 23 March 2016 amending Annex IV to Regulation (EC) No 396/2005 of the European Parliament and of the Council as regards *S.exigua* multicapsid nucleopolyhedrovirus	N	Y		