This form should be used for all taxonomic proposals. Please complete all those modules that are applicable (and then delete the unwanted sections). For guidance, see the notes written in blue and the separate document "Help with completing a taxonomic proposal"

Please try to keep related proposals within a single document; you can copy the modules to create more than one genus within a new family, for example.

MODULE 1: TITLE, AUTHORS, etc

Code assigned:	***2015.007aD***	(to be completed by ICTV officers)

Short title: One new species *Chrysodeixis includens nucleopolyhedrovirus*, **in the genus** ***Alphabaculovirus***
(e.g. 6 new species in the genus *Zetavirus*)

Modules attached (modules 1 and 10 are required)	**1** ☒	**2** ☒	**3** ☐	**4** ☐	**5** ☐
	6 ☐	**7** ☐	**8** ☐	**9** ☐	**10** ☒

Author(s):

Robert L. Harrison, on behalf of the Baculoviridae/Nudiviridae Study Group

Corresponding author with e-mail address:

Robert L. Harrison (Robert.L.Harrison@ars.usda.gov)

List the ICTV study group(s) that have seen this proposal:

A list of study groups and contacts is provided at http://www.ictvonline.org/subcommittees.asp . If in doubt, contact the appropriate subcommittee chair (fungal, invertebrate, plant, prokaryote or vertebrate viruses)	Baculoviridae/Nudiviridae Study Group

ICTV Study Group comments (if any) and response of the proposer:

Date first submitted to ICTV:	5/29/15
Date of this revision (if different to above):	9/23/15

ICTV-EC comments and response of the proposer:

9/22/2015: Figure 1 was modified to show that the new species of this proposal is different and distinct from the other two new *Alphabaculovirus* species discussed at EC47, *Agrotis segetum nucleopolyhedrovirus B* and *Sucra jujuba nucleopolyhedrovirus*.

MODULE 2: **NEW SPECIES**

creating and naming one or more new species.

If more than one, they should be a group of related species belonging to the same genus. All new species must be placed in a higher taxon. This is usually a genus although it is also permissible for species to be "unassigned" within a subfamily or family. Wherever possible, provide sequence accession number(s) for **one** isolate of each new species proposed.

Code	***2015.007aD***	(assigned by ICTV officers)

To create new species within:

Genus:	***Alphabaculovirus***
Subfamily:	
Family:	***Baculoviridae***
Order:	

Fill in all that apply.
- If the higher taxon has yet to be created (in a later module, below) write "**(new)**" after its proposed name.
- If no genus is specified, enter "**unassigned**" in the genus box.

Name of new species:	**Representative isolate: (only 1 per species please)**	**GenBank sequence accession number(s)**
Chrysodeixis includens nucleopolyhedrovirus	Pseudoplusia includens single nucleopolyhedrovirus-IE (PsinNPV-IE)	KJ631622

Reasons to justify the creation and assignment of the new species:
- Explain how the proposed species differ(s) from all existing species.
    - If species demarcation criteria (see module 3) have previously been defined for the genus, **explain how the new species meet these criteria**.
    - If criteria for demarcating species need to be defined (because there will now be more than one species in the genus), please state the proposed criteria.
- Further material in support of this proposal may be presented in the Appendix, Module 9

Research with virus isolates from the insect host *Chrysodeixis* (formerly *Pseudoplusia*) *includens* indicates that they constitute a new baculovirus species, *Chrysodeixis includens nucleopolyhedrovirus*. The following lines of evidence place *Chrysodeixis includens nucleopolyhedrovirus* in the genus *Alphabaculovirus* of the family *Baculoviridae*:

- Isolates of viruses in the proposed species establish lethal infections of a host insect of the order Lepidoptera (Alexandre et al., 2010).
- The isolates form occlusion bodies approximately 1 µM in diameter that contain many rod-shaped, enveloped virions that are characteristic of baculoviruses (Alexandre et al., 2010; Herniou et al., 2011)
- The genome consists of a circular double-stranded DNA molecule of a size (139,132 bp) that is within the range described for the genus *Alphabaculovirus* (110–170 kbp) (Craveiro et al., 2015).
- Gene content includes the 37 core genes identified for species in the family *Baculoviridae* (Craveiro et al., 2015; Garavaglia et al., 2012)
- Phylogenetic inference based on alignment of a subset of core genes places viruses of the proposed species in the *Alphabaculovirus* clade (Craveiro et al., 2013; Craveiro et al., 2015; Figure 1)

Species demarcation criteria for baculoviruses have been proposed that rely upon pairwise nucleotide distances estimated with the Kimura-2-parameter substitution model from partial sequences of three conserved baculovirus genes: *lef-8* and *lef-9* (encoding viral RNA polymerase subunits), and *polh* (encoding the viral occlusion body matrix protein) (Jehle et al., 2006). If nucleotide distances between two viruses are less than 0.015 substitutions/site, the two baculoviruses are considered to be the same species. If nucleotide distances between two viruses are greater than 0.05 substitutions/site, the viruses are considered to be different species. If the nucleotide distances lie between 0.015 and 0.050 substitutions/site, additional characteristics of the two viruses (*i.e.* host range) must be considered to make a decision about their taxonomic status. The proposed criterion was originally based on an alignment of sequences from 117 separate baculovirus isolates and the phylogeny inferred from this alignment. Researchers have applied this criterion to other isolates to identify potential new baculovirus species and variants of currently recognized species.

Phylogenetic inference from amino acid alignments of 34 core genes places *Chrysodeixis includens nucleopolyhedrovirus* in a clade with reference isolates of species *Trichoplusia ni single nucleopolyhedrovirus* and *Chrysodeixis chalcites nucleopolyhedrovirus*. Kimura-2-parameter nucleotide distances between *Chrysodeixis includens nucleopolyhedrovirus* and the other viruses in this clade ranged from 0.095 substitutions/site in the *lef-9* locus to 0.185 substitutions/site in the *lef-8* locus (Tables 1, 2). These values exceed the 0.050 substitutions/site threshold for declaring two baculoviruses to belong to different species. Hence, *Chrysodeixis includens nucleopolyhedrovirus* is a species distinct from *Trichoplusia ni single nucleopolyhedrovirus* and *Chrysodeixis chalcites nucleopolyhedrovirus* and, by extension, from the other currently recognized species in family *Baculoviridae*.

This new species is also different and distinct from the two other *Alphabaculovirus* species proposed in 2015, *Agrotis segetum nucleopolyhedrovirus B* and *Sucra jujuba nucleopolyhedrovirus* (Figure 1).

MODULE 10: **APPENDIX**: supporting material

additional material in support of this proposal

References:

Alexandre, T.M., Ribeiro, Z.M., Craveiro, S.R., Cunha, F., Fonseca, I.C., Moscardi, F., Castro, M.E., 2010. Evaluation of seven viral isolates as potential biocontrol agents against *Pseudoplusia includens* (Lepidoptera: Noctuidae) caterpillars. J Invertebr Pathol 105, 98-104.

Craveiro, S.R., Inglis, P.W., Togawa, R.C., Grynberg, P., Melo, F.L., Ribeiro, Z.M., Ribeiro, B.M., Bao, S.N., Castro, M.E., 2015. The genome sequence of Pseudoplusia includens single nucleopolyhedrovirus and an analysis of p26 gene evolution in the baculoviruses. BMC Genomics 16, 127.

Craveiro, S.R., Melo, F.L., Ribeiro, Z.M., Ribeiro, B.M., Bao, S.N., Inglis, P.W., Castro, M.E., 2013. Pseudoplusia includens single nucleopolyhedrovirus: genetic diversity, phylogeny and hypervariability of the pif-2 gene. J Invertebr Pathol 114, 258-267.

Garavaglia, M.J., Miele, S.A., Iserte, J.A., Belaich, M.N., Ghiringhelli, P.D., 2012. The ac53, ac78, ac101, and ac103 genes are newly discovered core genes in the family *Baculoviridae*. J Virol 86, 12069-12079.

Herniou, E.A., Arif, B.M., Becnel, J.J., Blissard, G.W., Bonning, B., Harrison, R.L., Jehle, J.A., Theilmann, D.A., Vlak, J.M., 2011. Baculoviridae, in: King, A.M.Q., Adams, M.J., Carstens, E.B., Lefkowitz, E.J. (Eds.), Virus Taxonomy: Ninth Report of the International Committee on Taxonomy of Viruses. Elsevier, Oxford, pp. 163-174.

Jehle, J.A., Lange, M., Wang, H., Hu, Z., Wang, Y., Hauschild, R., 2006. Molecular identification and phylogenetic analysis of baculoviruses from Lepidoptera. Virology 346, 180-193.

Annex:

Include as much information as necessary to support the proposal, including diagrams comparing the old and new taxonomic orders. The use of Figures and Tables is strongly recommended but direct pasting of content from publications will require permission from the copyright holder together with appropriate acknowledgement as this proposal will be placed on a public web site. For phylogenetic analysis, try to provide a tree where branch length is related to genetic distance.

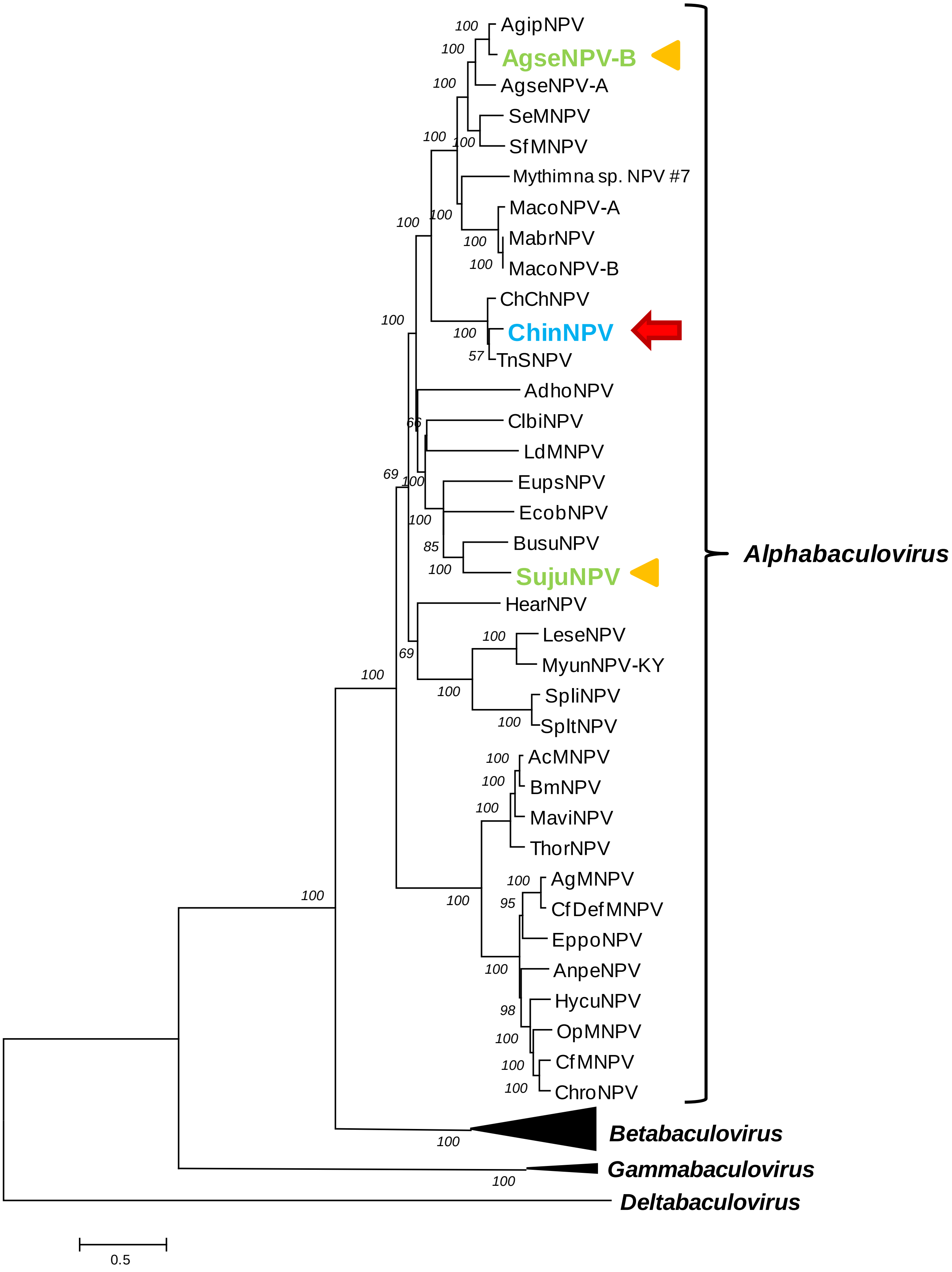

Figure 1. Relationships of baculovirus species and other isolates, inferred from the predicted amino acid sequences of baculovirus core genes. The phylogenetic tree was constructed from the concatenated alignments of 34 baculovirus core gene amino acid sequences using the minimum-evolution method using the James-Taylor-Thornton substitution model and a gamma parameter of 0.85 (estimated from the *dnapol*

alignment). Bootstrap values are shown for each node. Clades corresponding to the four genera of *Baculoviridae* are indicated. The proposed species is listed in blue font and denoted with a red arrow. Additional new *Alphabaculovirus* species discussed at EC47 are listed in green font and denoted with an orange arrowhead. *Alphabaculovirus* taxa include the reference isolates (abbreviations for the corresponding viruses are in brackets) for species *Adoxophyes honmai nucleopolyhedrovirus* (AdhoNPV), *Agrotis ipsilon multiple nucleopolyhedrovirus* (AgipNPV), *Agrotis segetum nucleopolyhedrovirus A* (AgseNPV-A), *Agrotis segetum nucleopolyhedrovirus B* (AgseNPV-B), *Antheraea pernyi nucleopolyhedrovirus* (AnpeNPV), *Anticarsia gemmatalis multiple nucleopolyhedrovirus* (AgMNPV), *Autographa californica multiple nucleopolyhedrovirus* (AcMNPV), *Bombyx mori nucleopolyhedrovirus* (BmNPV), *Buzura suppressaria nucleopolyhedrovirus* (BusuNPV), *Choristoneura fumiferana multiple nucleopolyhedrovirus* (CfMNPV), *Choristoneura fumiferana DEF multiple nucleopolyhedrovirus* (CfDEFNPV), *Chrysodeixis chalcites nucleopolyhedrovirus* (ChchNPV), *Chrysodeixis includens nucleopolyhedrovirus* (ChinNPV), *Clanis bilineata nucleopolyhedrovirus* (ClbiNPV), *Choristoneura rosaceana nucleopolyhedrovirus* (ChroNPV), *Ecotropis obliqua nucleopolyhedrovirus* (EcobNPV), *Epiphyas postvittana nucleopolyhedrovirus* (EppoNPV), *Euproctis pseudoconspersa nucleopolyhedrovirus* (EupsNPV), *Helicoverpa armigera nucleopolyhedrovirus* (HearNPV), *Hyphantria cunea nucleopolyhedrovirus* (HycuNPV), *Leucania separata nucleopolyhedrovirus* (LeseNPV), *Lymantria dispar multiple nucleopolyhedrovirus* (LdMNPV), *Mamestra configurata nucleopolyhedroviruses A* and *B* (MacoNPV-A and -B), *Maruca vitrata nucleopolyhedrovirus* (MaviNPV), *Spodoptera exigua multiple nucleopolyhedrovirus* (SeMNPV), *Spodoptera frugiperda multiple nucleopolyhedrovirus* (SfMNPV), *Spodoptera litura nucleopolyhedrovirus* (SpltNPV), and *Sucra jujuba nucleopolyhedrovirus* (SujuNPV), *Thysanoplusia orichalcea nucleopolyhedrovirus* (ThorNPV), and *Trichoplusia ni single nucleopolyhedrovirus* (TnSNPV), as well as isolates Mythimna spp. nucleopolyhedrovirus #7 (Mythimna sp. NPV #7) , and Mythimna unipuncta nucleopolyhedrovirus (MyunNPV-KY).

Table 1. Pairwise nucleotide distances calculated from partial *lef-8* and *lef-9* sequence alignments*

lef-9 *lef-8*	ChchNPV	ChinNPV	TnSNPV
ChchNPV		***0.095***	0.091
ChinNPV	***0.185***		***0.116***
TnSNPV	0.188	***0.188***	

*The numbers of base substitutions per site between pairs of sequences are shown. Abbreviations of the taxa are as listed for Figure 1. Analyses were conducted using the Kimura 2-parameter model in MEGA6, with gamma parameter values estimated separately for each set of aligned sequences. Pairwise distances for *lef-8* (Table 1) and *polh* (Table 2) sequences are listed below the diagonal lines, and *lef-9* (Table 1) and the concatenated *lef-8/lef-9/polh* (Table 2) distances are listed above the diagonal lines. Values for the proposed species, *Chrysodeixis includens nucleopolyhedrovirus*, are in bold italic type.

Table 2. Pairwise nucleotide distances calculated from partial polh sequence alignments and partial lef-8, lef-9, and polh sequence alignments that have been concatenated together*

lef-8/lef-9/polh *polh*	ChchNPV	ChinNPV	TnSNPV
ChchNPV		***0.138***	0.154
ChinNPV	***0.100***		***0.162***
TnSNPV	0.139	***0.144***	

* See footnote for Table 1.