Supplementary Material

Table S1. Sequences used in the P74 phylogeny inference

Species	Isolate	Denomination	GenBank
***Alphabaculovirus* (group I)**			
Antheraea pernyi NPV	Z	AnpeNPV-Z	NC_008035
Anticarsia gemmatalis MNPV	2D	AgMNPV	NC_008520
Autographa californica MNPV	Clone C6	AcMNPV	NC_001623
Bombyx mori NPV	T3	BmNPV	NC_001962
Catopsilia pomona	416	CapoNPV	NC_030240
Choristoneura fumiferana MNPV		CfMNPV	NC_004778
Condylorrhiza vestigialis MNPV		CoveMNPV	NC_026430
Cyclophragma undans NPV	Whiov	CyunNPV-Wh	KT957089
Dasychira pudibunda NPV	ML1	DapuNPV	KP747440
Dendrolimus kikuchii NPV	strain YN	DekiNPV	JX193905
Epiphyas postvittana NPV		EppoNPV	NC_003083
Hyphantria cunea NPV		HycuNPV	NC_007767
Lonomia obliqua MNPV	SP/2000	LoobMNPV	KP763670
Maruca vitrata NPV		MaviNPV	NC_008725
Orgyia pseudotsugata MNPV		OpMNPV	NC_001875
Oxyplax ochracea NPV	435	OxocNPV-435	MF143631
Philosamia cynthia ricini NPV		PhcyNPV	JX404026
Plutella xylostella MNPV	CL3	PlxyMNPV	NC_008349
Rachiplusia ou MNPV		RoMNPV	NC_004323
Samia cynthia NPV	Nagano	SacyNPV-Na	LC375538
Spilosoma obliqua NPV	IIPR	SpobNPV-IIPR	KY550224
Thysanoplusia orichalcea NPV	P2	ThorNPV-P2	NC_019945
***Alphabaculovirus* (group I)**			
Adoxophyes honmai NPV		AdhoNPV	NC_004690
Agrotis ipsilon MNPV		AgipMNPV	NC_011345
Apocheima cinerarium NPV		ApciNPV	NC_018504
Buzura suppressaria NPV	Hubei	BusuNPV	NC_023442
Chrysodeixis chalcites NPV		ChchNPV	NC_007151
Clanis bilineata NPV	DZ1	ClbiNPV	NC_008293
Ectropis obliqua NPV	Strain A1	EcobNPV	NC_008586
Euproctis pseudoconspersa NPV		EupsNPV	NC_012639
Helicoverpa armigera MNPV		HaMNPV	NC_011615
Helicoverpa armigera NPV	Strain G4	HearSNPV-G4	NC_002654
Hemileuca sp NPV		HespNPV	NC_021923
Hyposidra talaca NPV		HytaNPV	MH261376
Lambdina fiscellaria NPV	GR15	LafiNPV-GR15	NC_026922
Leucania separata NPV	Strain AH1	LeseNPV	NC_008348
Lymantria dispar MNPV		LdMNPV	NC_001973
Mamestra brassicae MNPV	K1	MabrMNPV-K1	NC_023681
Operophtera brumata NPV	MA	OpbuNPV-MA	NC_040621
Orgyia leucostigma NPV	CFS77	OrleNPV	NC_010276
Peridroma sp NPV	GR-167	PespNPV	NC_024625

Perigonia lusca SNPV		PeluSNPV	NC_027923
Pseudoplusia includens SNPV	IE	PsinSNPV-IE	NC_026268
Spodoptera exigua MNPV		SeMNPV	NC_002169
Spodoptera frugiperda MNPV	3AP2	SfMNPV-3AP2	NC_009011
Spodoptera littoralis NPV	AN1956	SpliNPV-AN1956	JX454574
Spodoptera litura II MNPV		SpltMNPV-II	NC_011616
Sucra jujuba NPV	473	SujuNPV-473	KJ676450
Trichoplusia ni SNPV		TnSNPV	NC_007383
Urbanus proteus NPV	Southern Brazil	UrprNPV	NC_029997
Betabaculovirus			
Adoxophyes orana GV		AdorGV	NC_005038
Agrotis segetum GV		AgseGV	NC_005839
Choristoneura occidentalis GV		ChocGV	NC_008168
Clostera anachoreta GV	HBHN	ClanGV	NC_015398
Cnapahlocrocis medinalis GV	Strain Enping	CnmeGV-E	NC_029304
Cryptophlebia leucotreta GV		CrleGV	NC_005068
Cydia pomonella GV		CpGV	NC_002816
Diatraea saccharalis GV	Parana-2009	DisaGV-P09	NC_028491
Epinotia aporema GV		EpapGV	NC_018875
Erinnyis ello ello GV	Strain BrS86	ErelGV	NC_025257
Helicoverpa armigera GV		HearGV	NC_010240
Mocis latipes GV	Southern Brazil	MolaGV	NC_029996
Mythimna unipuncta GV	MyunGV#8	MyunGV	NC_033780
Phthorimaea operculella GV		PhopGV	NC_004062
Pseudaletia unipuncta GV	Strain Hawaiin	PsunGV	NC_013772
Pieris rapae GV	Wuhan	PiraGV	NC_013797
Plodia interpunctella GV	Cambridge	PlinGV	NC_032255
Plutella xylostella GV		PxGV	NC_002593
Spodoptera frugiperda GV	VG008	SpfrGV	NC_026511
Spodoptera litura GV	Strain K1	SpltGV	NC_009503
Trichplusia ni GV	LBIV-12	TrniGV-LBIV12	KU752557
Xestia c nigrum GV		XecnGV	NC_002331
Gammabaculovirus			
Neodiprion lecontei NPV		NeleNPV	NC_005906
Neodiprion sertifer NPV		NeseNPV	NC_005905
Neodiprion abietis NPV		NeabNPV	DQ317692
Deltabaculovirus			
Culex nigripalpus NPV	Florida1997	CuniNPV	NC_003084
Nudiviridae			
Gryllus bimaculatus nudivirus		GrBNV	NC_009240.1
Hytrosaviridae			
Musca domestica salivary gland hypertrophy virus		MdSGHV	NC_010671.1
Nimaviridae			
Marsupenaeus japonicus endogenous nimavirus DNA		Nimav-1_LVa	BFCD01000001
White spot syndrome virus strain	MEX2008	WSSV	KU216744

Table S2. P74 Molecular weight and isoelectric point

P74	MW (Da)	PI
AcMNPV (complete)	73,885.48	4.90
SeMNPV (complete)	74,213.85	4.94
HearSNPV (complete)	78,406.81	5.13
AcMNPV (Nt)	51,673.47	6.52
SeMNPV (Nt)	51,606.36	6.48
HearSNPV (Nt)	54,706.64	6.06
AcMNPV (Ct)	22,230.02	3.95
SeMNPV (Ct)	22,625.51	3.99
HearSNPV (Ct)	23,718.18	4.45

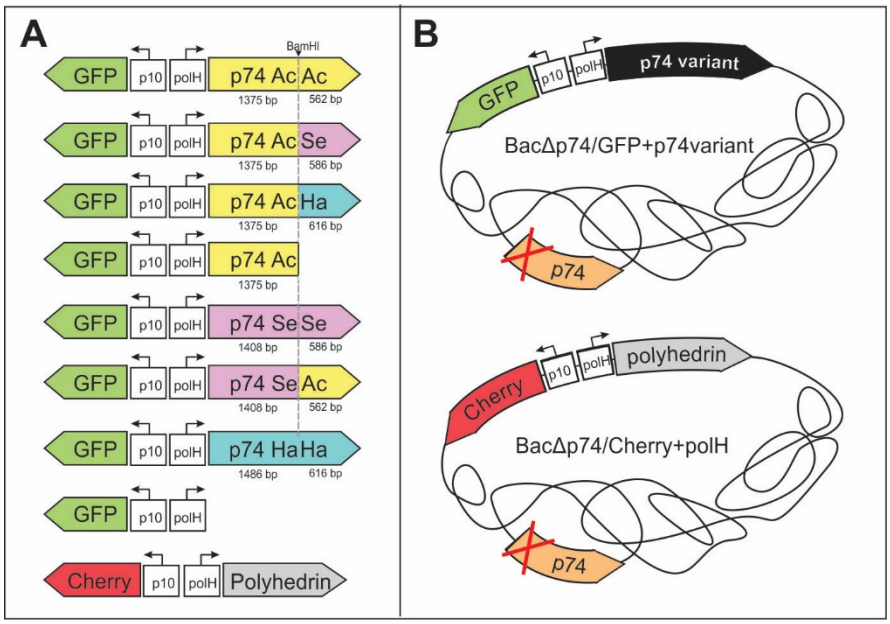

Figure S1. **AcMNPV variants**. The ORF of the *p74* gene of AcMNPV, SeMNPV and HearSNPV were used alone or combining sectors corresponding to the protein amino and carboxyl terminals. **A**. Illustration showing the composition of the different donor plasmids generated containing the *p74* variants and GFP, or polyhedrin and mCherry. Grey dotted line represents the BamHI recognition site between the amino and carboxyl P74 protein domains. **B**. Illustration showing bacmids deficient in the *p74* gene (indel mutation by CRISPR/Cas9 technology and NHEJ) after Bac-to-Bac complementation, one contributing with polyhedrin and the other one with a P74 variant. When both are coinfected in the same cells OBs can be generated with *per os* infective potential.

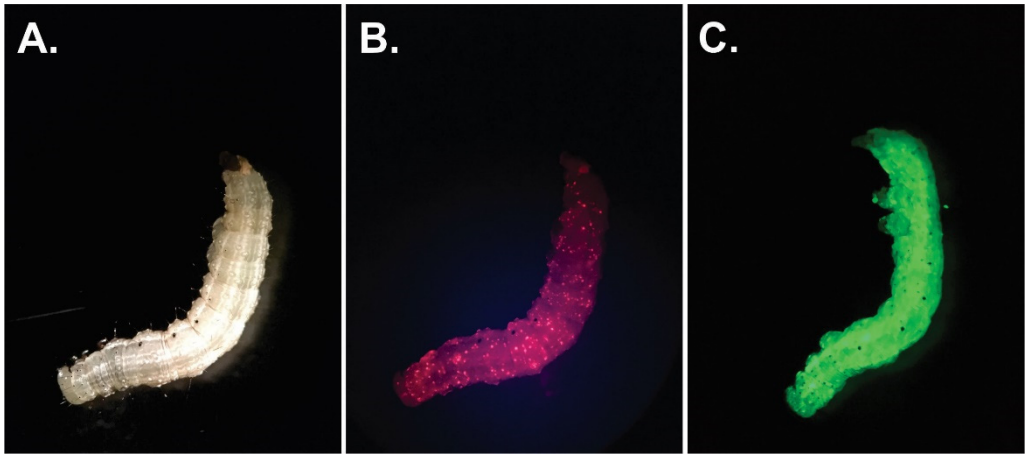

Figure S2. **Infectivity of complemented AcMNPV p74 knockout**. *R. nu* larvae exposed to virions derived from AcMNPV knocked out in *p74* and supplemented with the ORF variant of the same virus carrying the point mutation to introduce the BamHI site. Photographs (0.8X) taken 120 hours after treatment with the virus through *per os* route showing the appearance of a larva in bright field (A.), and the mCherry (B.) and GFP (C.) expression.