

Transfer Vectors Quick Start Guides

OET's range of baculovirus transfer vectors are designed for high level expression of foreign genes in both insect and mammalian cell lines. Our pOET transfer plasmids are ideal for use with the flashBAC™ and BacPAK6 baculovirus expression systems but are also fully compatible with other homologous recombination technologies



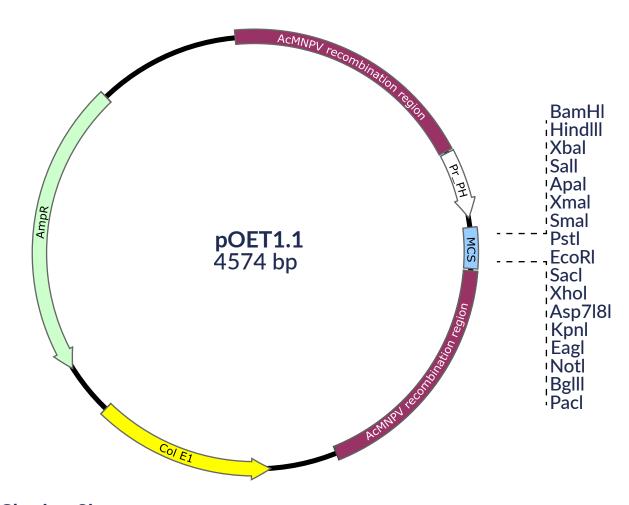




QUICK START GUIDE to pOET1.1

Catalogue Number	200101
Storage	Tightly capped at -20°C
Product Guarantee	1Year from the date of purchase, when properly stored and handled

pOET1.1 is a baculovirus transfer vector designed for high level expression of foreign genes under the powerful AcMNPV polyhedrin (polh) promoter (Pr_PH). The vector is smaller than other available transfer vectors (4574bp), which greatly facilitates the cloning steps. It has a bacterial origin of replication (ColE1) and an ampicillin resistance gene (AmpR) for selection in E. coli. The polh sequences have been replaced by a multiple cloning site (MCS) containing 17 unique restriction enzyme sites for insertion of the foreign gene in the correct orientation. The AcMNPV sequences flanking the gene in the transfer vector's MCS allow recombination with the viral DNA to insert the expression cassette into the polh locus. pOET1.1 is compatible with any baculovirus system that utilizes



Multiple Cloning Site



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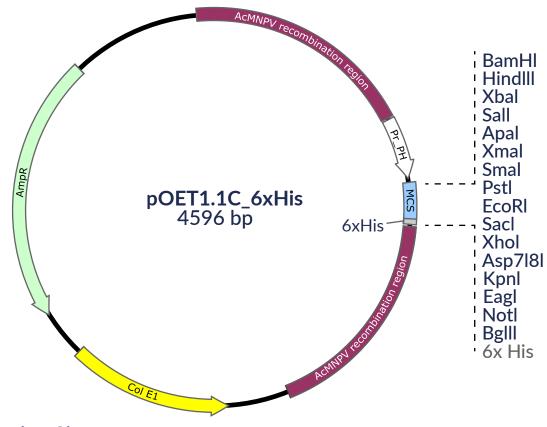




QUICK START GUIDE to pOET1.1C_6xHis

Catalogue Number	2001012
Storage	Tightly capped at -20°C
Product Guarantee	1Year from the date of purchase, when properly stored and handled

pOET1.1C 6xHis is a baculovirus transfer vector designed for high level expression of foreign genes under the powerful AcMNPV polyhedrin (polh) promoter (Pr_PH). The vector encodes an optional C-terminal 6xHis-Tag® fusion sequence that may be utilised. This greatly eases the purification of the recombinant protein since the 6xHis-containing fusion proteins bind with high affinity to Ni-NTA Agarose. pOET1.1C is smaller than other available transfer vectors (4596 bp) which greatly facilitates the cloning steps. It has a Col E1 origin of replication and an ampicillin resistance gene for selection in E. coli. The polh sequences have been replaced by a multiple cloning site (MCS) containing unique restriction enzyme sites for insertion of the foreign gene in the correct orientation. The AcMNPV sequences flanking the gene in the transfer vector's MCS allow recombination with the viral DNA to insert the expression cassette into the polh locus. pOET1.1C is compatible with any baculovirus system that utilises homologous recombination in insect cells.



Multiple Cloning Site

Xmal Asp7l8l Eagl **Smal** BamHI HindIII Xbal Apal Sall Pstl EcoRl Sacl Xhol **Kpnl** Notl BgIII GGATCCAAGCTTCTAGAGTCGACGGGCCCCCGGGCTGCAGAATTCGAGCTCTCGAGGTACCGCGGCCGCAGATCT

CATCATCACCACCATCAC

6xHis



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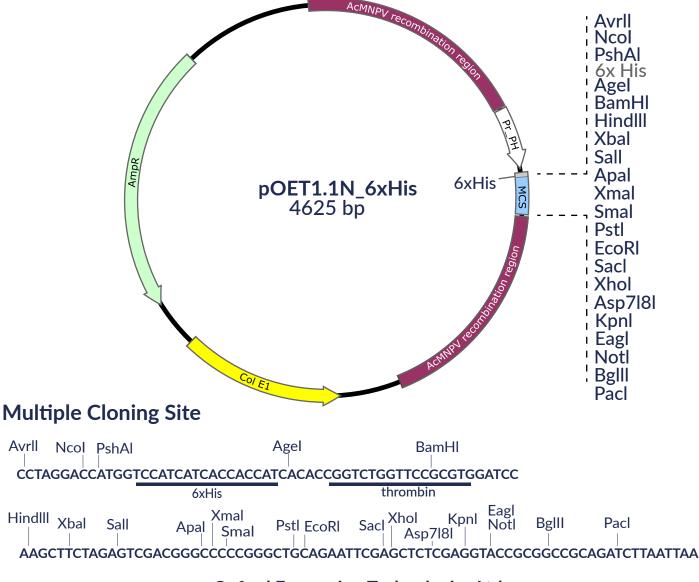




QUICK START GUIDE to pOET1.1N_6xHis

Catalogue Number	2001011
Storage	Tightly capped at -20°C
Product Guarantee	1Year from the date of purchase, when properly stored and handled

pOET1.1N 6xHis is a baculovirus transfer vector designed for high level expression of foreign genes under the powerful AcMNPV polyhedrin (polh) promoter (Pr_PH). The vector encodes an optional N-terminal 6xHis-Tag® fusion sequence that may be utilised if the insert allows read-through in the correct reading frame. This greatly eases the purification of the recombinant protein since the 6xHis-containing fusion proteins bind with high affinity to Ni-NTA Agarose. If required, the 6xHis-Tag® can be removed by incubating the fusion protein in the presence of the proteinase cleavage enzyme Thrombin. pOET1.1N is smaller than other available transfer vectors (4625 bp) which greatly facilitates the cloning steps. It has a Col E1 origin of replication and an ampicillin resistance gene for selection in E. coli. The polh sequences have been replaced by a multiple cloning site (MCS) containing unique restriction enzyme sites for insertion of the foreign gene in the correct orientation. The AcMNPV sequences flanking the gene in the transfer vector's MCS allow recombination with the viral DNA to insert the expression cassette into the polh locus. pOET1.1N is compatible with any baculovirus system that utilises homologous recombination in insect cells.





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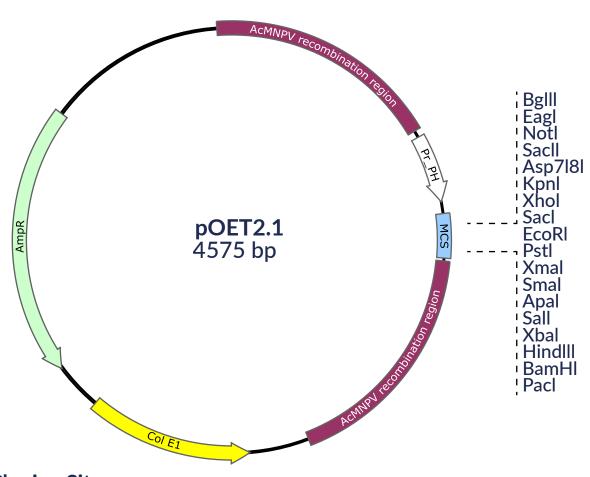




QUICK START GUIDE to pOET2.1

Catalogue Number	200103
Storage	Tightly capped at -20°C
Product Guarantee	1Year from the date of purchase, when properly stored and handled

pOET2.1 is a baculovirus transfer vector designed for high level expression of foreign genes under the powerful AcMNPV polyhedrin (polh) promoter (Pr_PH). The vector is smaller than other available transfer vectors (4575bp) which greatly facilitates the cloning steps. It has a bacterial origin of replication (ColE1) and an ampicillin resistance gene (AmpR) for selection in E. coli. The polh sequences have been replaced by a multiple cloning site (MCS) in the reverse orientation to pOET1, containing unique restriction sites for insertion of the foreign gene. The coding strand of the MCS as transcribed from the polh promoter is shown below the circular map. The AcMNPV sequences flanking the gene in the transfer vector's MCS allow recombination with the viral DNA to insert the expression cassette into the polh locus. pOET2.1 is compatible with any baculovirus system that utilizes homologous recombination in insect cells.



Multiple Cloning Site



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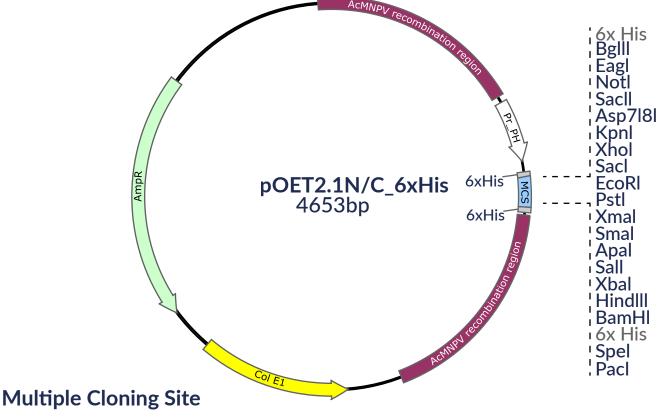




QUICK START GUIDE to pOET2.1 N/C_6xHis

Catalogue Number	2001031
Storage	Tightly capped at -20°C
Product Guarantee	1Year from the date of purchase, when properly stored and handled

pOET2N/C 6xHis is a baculovirus transfer vector designed for high level expression of foreign genes under the powerful AcMNPV polyhedrin (polh) promoter (Pr_PH). The vector encodes an N-terminal 6xHis-Tag® fusion sequence that may be utilised if the insert includes a stop codon. This greatly eases the purification of the recombinant protein since the 6xHis-containing fusion proteins bind with high affinity to Ni-NTA Agarose. If required, the 6xHis-Tag® can be removed by incubating the fusion protein in the presence of the proteinase cleavage enzyme Thrombin. There is also a 6xHis-Tag®for C-terminal fusions where the insert's own start codon can be used to replace the start codon supplied in pOET2N/C. It has a Col E1 origin of replication and an ampicillin resistance gene for selection in E. coli. The polh sequences have been replaced by a multiple cloning site (MCS) containing unique restriction sites for insertion of the foreign gene in the correct orientation. The AcMNPV sequences flanking the gene in the transfer vector's MCS allow recombination with the viral DNA to insert the expression cassette into the polh locus. pOET2N/C is compatible with any baculovirus system that utilises homologous recombination in insect cells.



CATCATCACCACCATCACACCGGTCTGGTTCCGCGTGGATCA thrombin

CATCATCACCACCATCACTAGTTAATTAA



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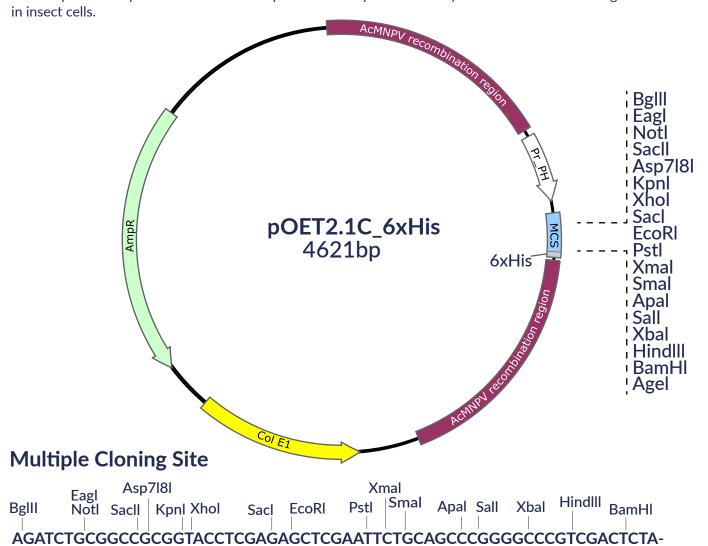




QUICK START GUIDE to pOET2.1C_6xHis

Catalogue Number	2001032
Storage	Tightly capped at -20°C
Product Guarantee	1Year from the date of purchase, when properly stored and handled

pOET2.1C 6xHis is a baculovirus transfer vector designed for high level expression of foreign genes under the powerful AcMNPV polyhedrin (polh) promoter (Pr PH). The vector encodes an optional C-terminal 6xHis-Tag® fusion sequence that may be utilized. This greatly eases the purification of the recombinant protein since the 6xHis-containing fusion proteins bind with high affinity to Ni-NTA Agarose. pOET2.1C 6xHis is smaller than other available transfer vectors (4621 bp) which greatly facilitates the cloning steps. pOET2.1C 6xHis has a Col E1 origin of replication and an ampicillin resistance gene for selection in E. coli. The polh sequences have been replaced by a multiple cloning site (MCS) containing unique restriction enzyme sites for insertion of the foreign gene in the correct orientation. The AcMNPV sequences flanking the gene in the transfer vector's MCS allow recombination with the viral DNA to insert the expression cassette into the polh locus. pOET2.1C 6xHis is compatible with any baculovirus system that utilizes homologous recombination in insect cells.



TCTGGTTCCGCGTGGATCAACCGGTCACCATCAT-6xHis

Agel



thrombin

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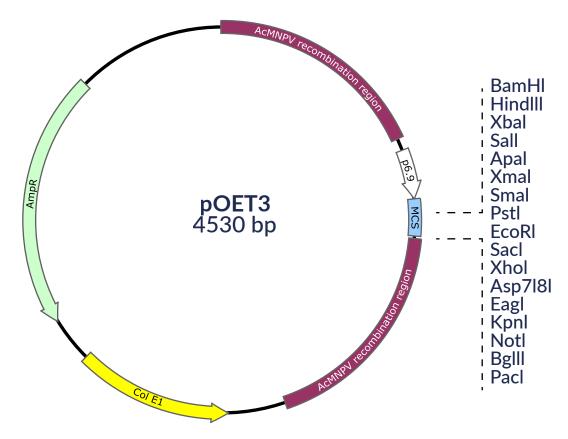




QUICK START GUIDE to pOET3

Catalogue Number	200104
Storage	Tightly capped at -20°C
Product Guarantee	1Year from the date of purchase, when properly stored and handled

pOET3 is a baculovirus transfer vector designed for high level expression of foreign genes under the late AcMNPV basic (p6.9) promoter. Using this promoter will provide earlier expression compared to the polyhedrin promoter. This has been shown to be beneficial when expressing proteins which require extensive post translational modifications i.e. glycosylation. The vector is smaller than other available transfer vectors (4530bp) which greatly facilitates the cloning steps. It has a bacterial origin of replication (Col E1) and an ampicillin resistance gene (AmpR) for selection in E. coli. The polyhedrin sequences have been replaced by a multiple cloning site (MCS) containing unique restriction sites for insertion of the foreign gene in the correct orientation. The AcMNPV sequences flanking the gene in the transfer vector's MCS allow recombination with the viral DNA to insert the expression cassette into the polyhedrin locus. pOET3 is compatible with any baculovirus system that utilizes homologous recombination in insect cells.



Multiple Cloning Site



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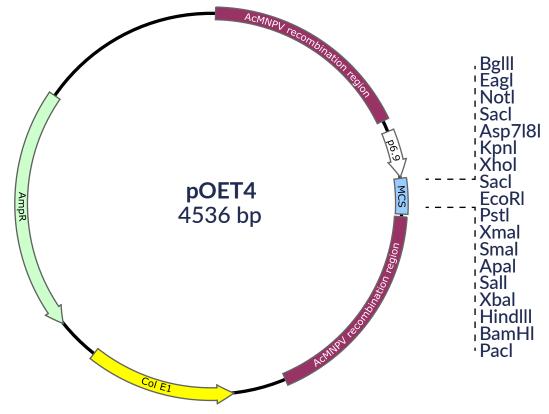




QUICK START GUIDE to pOET4

Catalogue Number	200105
Storage	Tightly capped at -20°C
Product Guarantee	1Year from the date of purchase, when properly stored and handled

pOET4 is a baculovirus transfer vector designed for high level expression of foreign genes under the late AcMNPV basic (p6.9) promoter. Using this promoter will provide earlier expression compared to the polyhedrin promoter. This has been shown to be beneficial when expressing proteins which require extensive post-translational modifications i.e. glycosylation. The vector is smaller than other available transfervectors (4536bp) which greatly facilitate the cloning steps. It has a bacterial origin of replication (Col E1) and an ampicillin resistance gene (AmpR) for selection in E. coli. The coding strand of the MCS as transcribed from the polh promoter is shown below the circular map. The AcMNPV sequences flanking the gene in the transfer vector's MCS allow recombination with the viral DNA to insert the expression cassette into the polyhedrin locus. The polyhedrin sequences have been replaced by a multiple cloning sitecontaining unique restriction sites for insertion of the foreign gene in the correct orientation. pOET4 is compatible with any baculovirus system that utilizes homologous recombination in insect cells.



Multiple Cloning Site

Asp718I **Xmal** Eagl HindIII BamHI Smal Apal Sall Xbal **BgIII Pacl** Sacll Kpnl Xhol **EcoRI** Sacl



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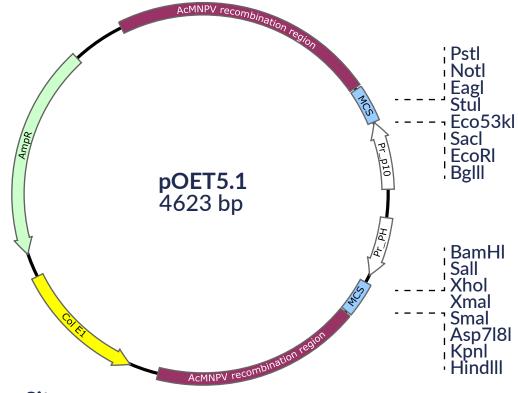




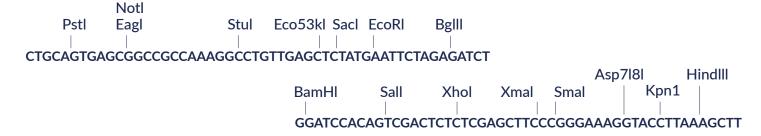
QUICK START GUIDE to pOET5.1

Catalogue Number	200106
Storage	Tightly capped at -20°C
Product Guarantee	1Year from the date of purchase, when properly stored and handled

pOET5.1 is a dual promoter baculovirus transfer vector designed for high level expression of two foreign genes simultaneously under the powerful AcMNPV polyhedrin (polh) promoter (Pr_PH) and the very late p10 promoter (Pr_p10). The promoters are in opposite orientations to minimize recombination. The vector is smaller than other available transfer vectors (4623bp), which greatly facilitates the cloning steps. It has a bacterial origin of replication (Col E1) and an ampicillin resistance gene (AmpR) for selection in E. coli. The polh sequences have been replaced by two multiple cloning sites containing unique restriction sites for insertion of the foreign genes in the correct orientation. The AcMNPV sequences flanking the gene in the transfer vector's MCS allow recombination with the viral DNA to insert the expression cassette into the polh locus. pOET5.1 is compatible with any baculovirus system that utilizes homologous recombination in insect cells.



Multiple Cloning Sites





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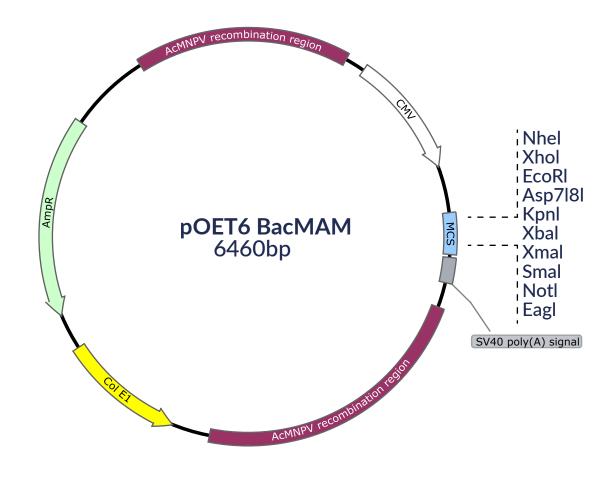




QUICK START GUIDE to pOET6 BacMAM

Catalogue Number	200107
Storage	Tightly capped at -20°C
Product Guarantee	1Year from the date of purchase, when properly stored and handled

pOET6 is a baculovirus transfer vector designed for expression of foreign genes in mammalian cells under the cytomegalovirus immediate early gene promoter (CMV). It has a bacterial origin of replication (Col E1) and an ampicillin resistance gene (AmpR) for selection in E. coli. The polh sequences have been replaced by a multiple cloning site (MCS) containing 10 unique restriction enzyme sites for insertion of the foreign gene in the correct orientation. The coding strand of the MCS as transcribed from the polh promoter is shown below the circular map. pOET6 is compatible with any baculovirus system that utilizes homologous recombination in insect cells.







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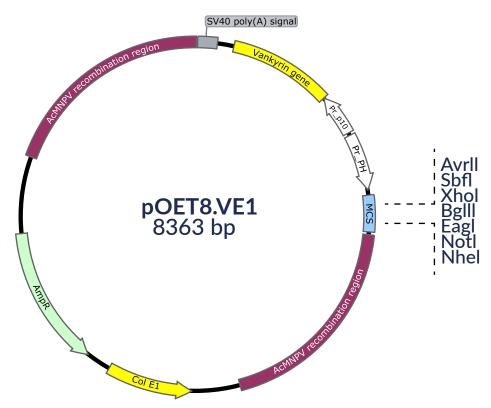
QUICK START GUIDE to pOET8 VE1

Catalogue Number	200121
Storage	Tightly capped at -20°C
Product Guarantee	1Year from the date of purchase, when properly stored and handled

pOET8.VE1 is a baculovirus transfer vector designed for high level expression of foreign genes under the powerful AcMNPV polyhedrin gene (polh) promoter (Pr_PH). Derived from the pUC57 vector, it contains a vankyrin expression cassette, P-vank-11, which encodes an anti-apoptotic protein to help delay cell death following virus infection. It has a bacterial origin of replication (Col E1) and an ampicillin resistance gene (AmpR) for selection in E. coli whilst the polh coding sequences have been replaced by a multiple cloning site (MCS) containing unique restriction enzyme sites for insertion of the foreign gene in the correct orientation (see circular map below). pOET8.VE1 is compatible with any baculovirus expression system that utilizes homologous recombination in insect cells.

Reference

¹Fath-Goodin, A., Kroemer, J.A., Martin, S.B., Reeves, K., and Webb, B.A. (2006). Polydnavirus genes that



Multiple Cloning Site

Eagl Notl Sbfl Xhol Nhel Avrll BgIII



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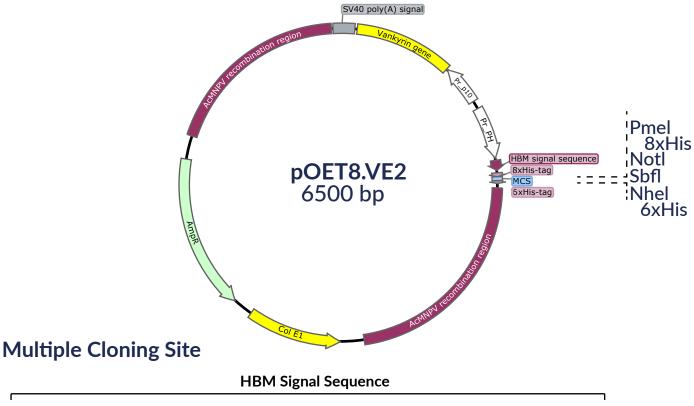
QUICK START GUIDE to pOET8 VE2

Catalogue Number	200122
Storage	Tightly capped at -20°C
Product Guarantee	1Year from the date of purchase, when properly stored and handled

pOET8.VE2 is a baculovirus transfer vector designed for high level expression of foreign genes under the powerful AcMNPV polyhedrin gene (polh) promoter (pr_PH). Derived from the pUC57 vector, it contains a vankyrin expression cassette, P-vank-11, which encodes an anti-apoptotic protein to help delay cell death following virus infection. It has a bacterial origin of replication (Col E1) and an ampicillin resistance gene (AmpR) for selection in E. coli whilst the polh coding sequences have been replaced by a multiple cloning site (MCS) containing unique restriction enzyme sites for insertion of the foreign gene in the correct orientation (see circular map below). The pOET8.VE2 vector features an additional Honey Bee Melittin (HBM) signal sequence to target protein secretion and an N-terminal 8xHis-tag fusion sequence to facilitate protein purification. pOET8.VE2 is compatible with any baculovirus expression system that utilizes homologous recombination in insect cells.

Reference

¹Fath-Goodin, A., Kroemer, J.A., Martin, S.B., Reeves, K., and Webb, B.A. (2006). Polydnavirus genes that enhance the Baculovirus Expression Vector System. Advances in Virus Research, vol. 68, pp. 75-90.



. ATGAAATTTCTAGTAAACGTTGCCTTAGTCTTTATGGTGGTTTACATATCTTATATC-

Pmel Notl Sbfl Nhel
| | | | | | |
FCCGAGTTTAAACCACCACCATCACCACCATCACGGGGCCGCACCTGCAGGGCTAGCA

TGATCCGAGTTTAAACCACCACCATCACCACCATCATCACGCGGCCGCACCTGCAGGGCTAGCA

8X His-Tag



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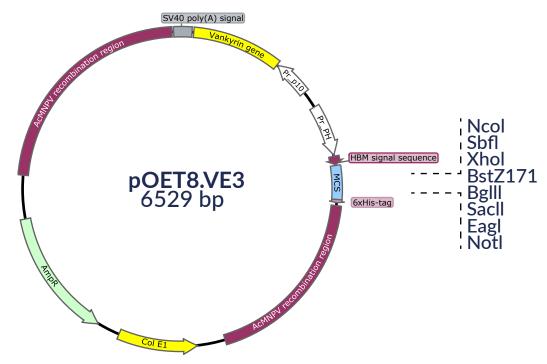
QUICK START GUIDE to pOET8 VE3

Catalogue Number	200123
Storage	Tightly capped at -20°C
Product Guarantee	1Year from the date of purchase, when properly stored and handled

pOET8.VE3 is a baculovirus transfer vector designed for high level expression of foreign genes under the powerful AcMNPV polyhedrin gene (polh) promoter (Pr_PH). Derived from the pUC57 vector, it contains a vankyrin expression cassette, P-vank-11, which encodes an anti-apoptotic protein to help delay cell death following virus infection. It has a bacterial origin of replication (Col E1) and an ampicillin resistance gene (AmpR) for selection in E. coli whilst the polh sequences have been replaced by a multiple cloning site (MCS) containing unique restriction enzyme sites for insertion of the foreign gene in the correct orientation (see circular map below). The pOET8.VE3 vector features an additional Honey Bee Melittin (HBM) signal sequence to target protein secretion and a C-terminal 6xHis-tag fusion sequence to facilitate protein purification. pOET8.VE3 is compatible with any baculovirus expression system that utilizes homologous recombination in insect cells.

Reference

¹Fath-Goodin, A., Kroemer, J.A., Martin, S.B., Reeves, K., and Webb, B.A. (2006). Polydnavirus genes that enhance the Baculovirus Expression Vector System. Advances in Virus Research, vol. 68, pp. 75-90.



Multiple Cloning Site

HBM Signal Sequence

ATGAAATTTCTAGTAAACGTTGCCTTAGTCTTTATGGTGGTTTACATATCTTATATCTATGCTGATCCGCCTGG

Ncol Sbfl Xhol BstZ171 Bglll Sacll Notl

TCCATGGCCCTGCAGGCAACTCGAAGTTCGAAAGTATACAAAGATCTCAAGTACCGCGGTCGAAGCGGCCG

6X His-Tag
CTCTATCTAGCTGCACATCACCATCACCATCAC



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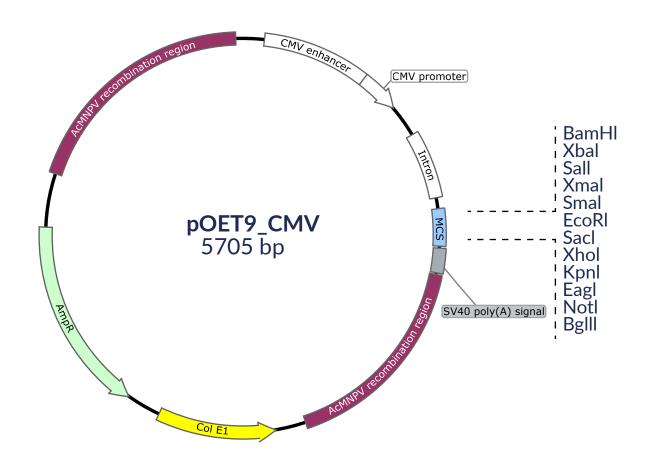




QUICK START GUIDE to pOET9^{CMV}

Catalogue Number	200133
Storage	Tightly capped at -20°C
Product Guarantee	1Year from the date of purchase, when properly stored and handled

pOET9^{CMV} is a baculovirus transfer vector designed for expression of foreign genes in mammalian cells under the Cytomegalovirus (CMV) gene promoter. The vector is smaller (5705bp) than other available transfer vectors, which greatly facilitates the cloning steps. It has a bacterial origin of replication (Col E1) and an ampicillin resistance gene (AmpR) for selection in E. coli. The polh sequences have been replaced by a multiple cloning site (MCS) containing unique restriction enzyme sites for insertion of the foreign gene in the correct orientation. pOET9^{CMV} is compatible with any baculovirus system that utilizes homologous recombination in insect cells.







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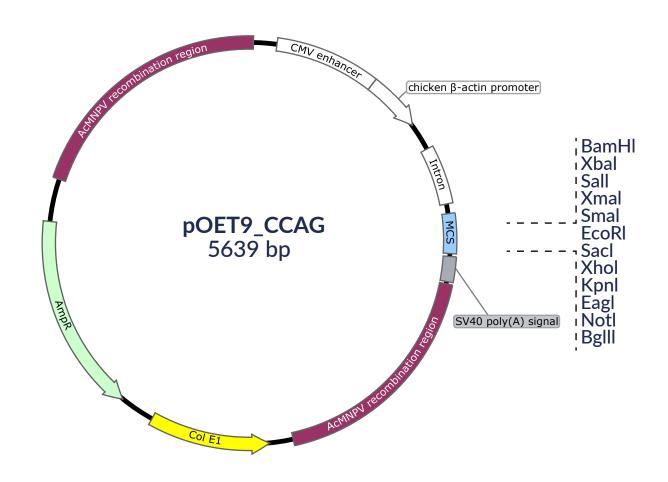




QUICK START GUIDE to pOET9^{CCAG}

Catalogue Number	200132
Storage	Tightly capped at -20°C
Product Guarantee	1Year from the date of purchase, when properly stored and handled

pOET9^{CCAG} is a baculovirus transfer vector designed for expression of foreign genes in mammalian cells under the Chicken Beta-Actin gene promoter (CCAG). The vector is smaller (5639bp) than other available transfer vectors, which greatly facilitates the cloning steps. It has a bacterial origin of replication (Col E1) and an ampicillin resistance gene (AmpR) for selection in E. coli. The polh sequences have been replaced by a multiple cloning site (MCS) containing unique restriction enzyme sites for insertion of the foreign gene in the correct orientation. pOET9^{CCAG} is compatible with any baculovirus system that utilizes homologous recombination in insect cells.







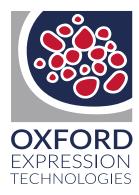
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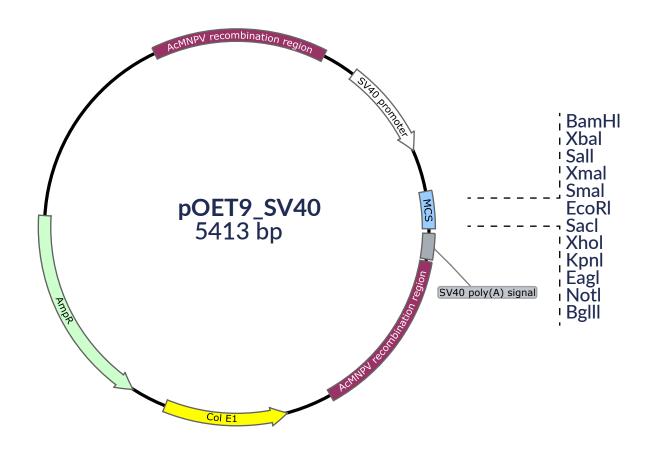




QUICK START GUIDE to pOET9^{SV40}

Catalogue Number	200134
Storage	Tightly capped at -20°C
Product Guarantee	1Year from the date of purchase, when properly stored and handled

pOET9^{SV40} is a baculovirus transfer vector designed for high level expression of foreign genes mammalian cells under the Simian Virus 40 (SV40) gene promoter. The vector is smaller (5413bp) than other available transfer vectors, which greatly facilitates the cloning steps. It has a bacterial origin of replication (Col E1) and an ampicillin resistance gene (AmpR) for selection in E. coli. The polh sequences have been replaced by a multiple cloning site (MCS) containing unique restriction enzyme sites for insertion of the foreign gene in the correct orientation. pOET9^{SV40} is compatible with any baculovirus system that utilizes homologous recombination in insect cells.



Multiple Cloning Site

BamHI Xbal Sall | Smal | EcoRI SacI Xhol Kpnl Notl Bglll | GGATCCAGCTTCTAGAGTCGACGGCCCCCGGGCTGAGAATTCGAGCTCTCGAGGTVACCGCGGCCGCAGATCT



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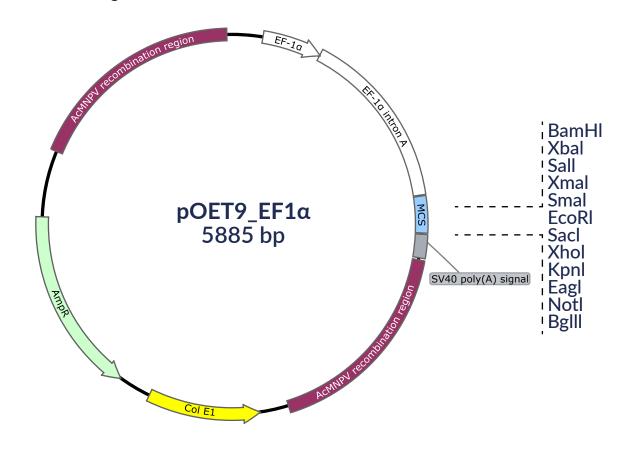




QUICK START GUIDE to pOET9^{EF1a}

Catalogue Number	200131
Storage	Tightly capped at -20°C
Product Guarantee	1Year from the date of purchase, when properly stored and handled

pOET9^{EF1 α} is a baculovirus transfer vector designed for high level expression of foreign genes mammalian cells under the Human Elongation Factor-1 Alpha (EF1 α) gene promoter. The vector is smaller (5885bp) than other available transfer vectors, which greatly facilitates the cloning steps. It has a bacterial origin of replication (Col E1) and an ampicillin resistance gene (AmpR) for selection in E. coli. The polh sequences have been replaced by a multiple cloning site (MCS) containing unique restriction enzyme sites for insertion of the foreign gene in the correct orientation. pOET9^{EF1 α} is compatible with any baculovirus system that utilizes homologous recombination in insect cells.







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