**pVITRO1-neo-mcs**

A multigenic plasmid for high levels of expression

Catalog code: pvitro1-nmcs

[https://www.invivogen.com/pvitro1-mcs](https://www.invivogen.com/pvitro1-mcs)

For research use only

Version 19L17-MM

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**PRODUCT INFORMATION**

Contents
- 20 µg of pVITRO1-neo-mcs provided as lyophilized DNA

Storage and stability
- Products are shipped at room temperature.
- Lyophilized DNA should be resuspended upon receipt and stored at -20°C. Lyophilized DNA is stable for 3 months at -20°C. Resuspended DNA is stable for at least one year at -20°C.

Quality control
- Plasmid construct has been confirmed by restriction analysis and sequencing.
- Plasmid DNA was purified by ion exchange chromatography and lyophilized.

**GENERAL PRODUCT USE**
pVITRO is a family of plasmids developed mainly for in vitro studies. They allow the ubiquitous and constitutive co-expression of two genes of interest. pVITRO plasmids can be stably transfected in mammalian cells and the genes of interest are expressed at high levels. Each pVITRO plasmid is available with either two multiple cloning sites or two reporter genes.

**pVITRO1-neo-mcs** plasmid is selectable with kanamycin in E. coli and G418 in mammalian cells. It contains two multiple cloning sites (MCS) for the convenient cloning of two cDNAs.

**PLASMID FEATURES**

- **rEF1 and mEF1 promoters**: pVITRO1-neo-mcs plasmid carries two elongation factor 1 alpha (EF-1α) promoters, from rat and mouse origins. Similarly to their human counterpart, both promoters display a strong activity that yield similar levels of expression. EF-1α promoters are expressed at high levels in all cell cycles and lower levels during G0 phase. EF-1α promoters are also non-tissue specific; they are highly expressed in all cell types.
- **SV40 enhancer**: which is comprised of a 72-base-pair repeat allows the enhancement of gene expression in a large host range. The enhancement varies from 2-fold in non-permissive cells to 20-fold in permissive cells.
- **CMV enhancer**: The major immediate early enhancer of the human cytomegalovirus (HCMV), located between nucleotides -118 and -524, is composed of unique and repeated sequence motifs. The HCMV enhancer can substitute for the 72-bp repeats of SV40 and is several fold more active than the SV40 enhancer.
- **SV40 pAn**: the simian Virus 40 late polyadenylation signal enables efficient cleavage and polyadenylation reactions resulting in high levels of steady-state mRNA. The efficiency of this signal was first described by Carswell et al.
- **pMB1 ori**: a minimal E. coli origin of replication to limit vector size, but with the same activity as the longer Ori.
- **FMDV IRES**: The internal ribosome entry site of the Foot and Mouth Disease Virus enables the translation of two open reading frames from one mRNA with high levels of expression.
- **EM7**: is a bacterial promoter that enables the constitutive expression of the antibiotic resistance gene in E. coli.
- **Neo**: The neo gene from Tn5 confers resistance to Kanamycin in E. coli and G418 in mammalian cells. In bacteria, neo is expressed from the constitutive E. coli EM7 promoter. In mammalian cells, neo is transcribed from the rat EF-1α promoter as a polycistronic mRNA and translated via the FMDV IRES.
- **EF1 pAn** is a strong polyadenylation signal. InvivoGen uses a sequence starting after the stop codon of the EF1 cDNA and finishing after a bent structure rich in GT.
- **MC51 and MC52**: To facilitate cloning each multiple cloning site contains several restriction sites that are compatible with many other enzymes.

**MC51** contains Bsp EI, Bst 1107I, Bam HI, Bsi WI and Avr II.
- Bsp EI is compatible with Age I and Sgr Al.
- Bst 1107I (blunt-end restriction enzyme)
- Bam HI is compatible with Bgl II, Bst YI and Bci I.
- Bsi WI is compatible with Acc 65I, Ban I and Bsr GI.
- Avr II is compatible with Xba I, Spe I and Nhe I.

**MC52** contains Age I, Eco RV, Bgl II, Bsr GI, and Nhe I.
- Age I is compatible with Bsp EI and Sgr Al.
- Eco RV (blunt-end restriction enzyme)
- Bgl II is compatible with Bam HI, Bst YI and Bci I.
- Bsr GI is compatible with Acc 65I, Ban I and Bsr WI.
- Nhe I is compatible with Xba I, Spe I and Avr II.


**METHODS**

**Plasmid resuspension**

Quickly spin the tube containing the lyophilized plasmid to pellet the DNA. To obtain a plasmid solution at 1 µg/µl, resuspend the DNA in 20 µl of sterile H2O. Store resuspended plasmid at -20°C.

**Plasmid amplification and cloning**

Plasmid amplification and cloning can be performed in E.coli GT116 or other commonly used laboratory E. coli strains, such as DH5α.

**Bacterial antibiotic selection**

Kanamycin (not provided) is normally used for E. coli at a final concentration of 50 µg/ml in liquid or solid media.

**Mammalian antibiotic selection**

G418 is normally used at a concentration of 400 µg/ml. However, the optimal concentration needs to be determined for your cells.

**RELATED PRODUCTS**

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<td>gt116-11</td>
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