pFN11A (BIND) Flexi[®] Vector:

Part No. C934A

Size (units) 20µg

Instructions for use of this product can be found in the CheckMate[™]/Flexi® Vector Mammalian Two-Hybrid System Technical Manual #TM283, available online at: www.promega.com/tbs

Description: The pFN11A (BIND) Flexi® Vector(a-e) is designed for use with the CheckMate™/ Flexi® Vector Mammalian Two-Hybrid System (Cat.# C9360), the Flexi® System, Entry/Transfer (Cat.# C8640) and the Flexi® System, Transfer (Cat.# C8820). The pFN11A (BIND) Flexi® Vector contains the yeast GAL4 DNA-binding domain upstream and in-frame with the cloning site. The vector also expresses the Renilla reniformis luciferase gene under the control of the SV40 promoter, allowing normalization for differences in transfection efficiency. This vector can be used to test putative transcriptional activation domains for protein sequences of interest when co-transfected with the pGL4.31[/uc2P/GAL4UAS/Hygro] Vector (Cat.# C9351). In common with other Flexi® Vectors, the pFN11A (BIND) Flexi® Vector contains a CMV immediate early enhancer/promoter region plus a chimeric intron for mammalian expression, a T7 promoter for in vitro expression of the protein-coding region, a barnase gene for positive selection of the insert, an ampicillin resistance gene for selection of the plasmid, and unique Sgf I and Pme I sites that allow easy insertion and transfer of the insert of interest. Do not use the pFN11A (BIND) Flexi® Vector without an insert as a negative control because the presence of the barnase gene decreases the viability of the transfected cells. The pFN11A (BIND) Flexi® Vector containing a cloned protein-coding region can be used to transfer the insert to other Flexi® Vectors with different expression options using the Flexi® Systems. For more information, see the CheckMate™/Flexi® Vector Mammalian Two-Hybrid System Technical Manual #TM283, and the Flexi® Vector Systems Technical Manual #TM254.

Concentration: 100ng/µl.

GenBank® Accession Number: DQ487212.

Storage Buffer: The pFN11A (BIND) Flexi® Vector is supplied in 10mM Tris-HCl , 1mM EDTA (pH 8.0 at 25°C).

Storage Conditions: See the Product Information Label for storage recommendations. Avoid multiple freeze-thaw cycles and exposure to frequent temperature changes.

Usage Notes: Concentration gradients may form in frozen products and should be dispersed upon thawing. Mix well prior to use.

Quality Control Assays

Nuclease Assay: Following incubation of 1µg of pFN11A (BIND) Flexi® Vector in Restriction Enzyme Buffer B at 37°C for 16 hours, no evidence of nuclease activity is detected by agarose gel electrophoresis.

Physical Purity: $A_{260}/A_{280} \ge 1.80$.

Restriction Digestion: The presence of unique restriction sites for Pmel and Sqfl is confirmed by showing that the vector is linearized and yields the expected fragment sizes after digesting 1µg of vector for 2 hours with 10 units of Pmel, Sqfl and Bgl II.

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Usage Information

pFN11A (BIND) Flexi® Vector Features and Circle Map

The following features are present in the vector based on nucleotide sequence.

CMV immediate/early enhancer/promoter	1–742
Chimeric intron	857–989
T7 RNA polymerase promoter (-17 to +3)	1033–1052
GAL4 fusion protein	1083–1520
IgA linker	1521–1553
Sgfl site	1554-1561
Barnase coding region	1585–1920
Pmel site	1922–1929
SV40 late polyadenylation signal	2081-2302
SV40 enhancer/early promoter	2401–2819
hGH intron	2871–3130
Renilla luciferase coding region	3155-4090
Synthetic polyadenylation signal	4148-4194
β-lactamase (Amp ^r) coding region	4457–5317
ColE1-derived plasmid origin of replication	5472-5508
cer region (site for E. coli XerCD recombinase)	6179–6464

Note: The IgA linker encodes the polypeptide AIPSTPPTPSPAIA.



Figure 1. pFN11A (BIND) Flexi® Vector circle map and sequence reference points.

Maps of all the Flexi® Vectors are available at: www.promega.com/vectors/cloning_vectors.htm

Related Products

Product	Size	Cat.#
Flexi® System, Entry/Transfer	5 entry/20 transfer reactions	C8640
Single Step (KRX) Competent Cells	5 × 200µl	L3001

There are Flexi® Vectors available for many different applications. Visit: www.promega.com/applications/cloning to find out more.

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