

# Data sheet

## Cannabinoid 1 Receptor Mammalian Transfection Kit

Cat. No: G0516

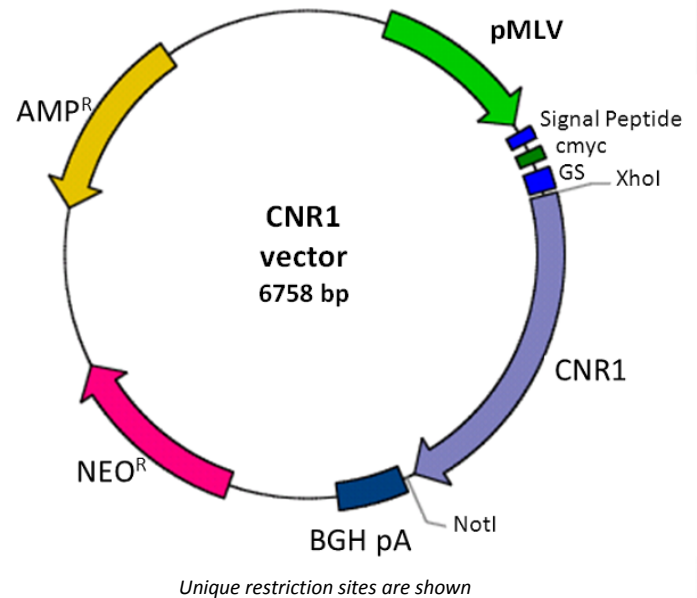
Cat. No: G0516 Plus

### Description

The Cannabinoid 1 receptor (CNR1) is a G protein-coupled cannabinoid receptor primarily located in the brain. It is activated by the endocannabinoid neurotransmitters anandamide and 2-arachidonoyl glyceride and by plant cannabinoids. CNR1 modulates neurotransmitter release when activated in a dose-dependent manner. Most CNR1 receptors are coupled through G  $\alpha$  i/o proteins. This receptor plays a major role in the maintenance of homeostasis in health and disease. Increased receptor expression has been found in human hepatocellular carcinoma tumor and other prostate cancer cells. CNR1 is well known for their cardiovascular activity. Disease relationships for CNR1 gene are obesity, multiple sclerosis, catalepsy, cannabis dependence, liver fibrosis, metabolic disorder, dyslipidemia.

### Kit Components

Components	G0516	G0516 Plus
CNR1 Mammalian Expression Vector (1 $\mu$ g/ $\mu$ L)	15 $\mu$ L	15 $\mu$ L
CANFAST Transfection Reagent (1 mg/ mL)	-	1 mL



Promoter	pMLV
ORF Sequence (GenBank based)	NM_016083.4
Protein Sequence (SwissProt)	P21554
Bacterial Selection Antibiotics	Ampicillin
Mammalian Selection Antibiotic	Neomycin

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## Assay procedure

### Transfection Protocol (stable or transient)

1. For adherent cells, seed the cells 18-24 hours before transfection to obtain 60-80% confluence the day of transfection, according the next table.

For suspension cells, seed the cells the day of transfection. Seed the cells to obtain 60-80% confluence according to the table below. The number of cells to seed depends on cell growth.

Recommended Number of Cells to seed for CANFAST Transfection			
Tissue Culture Vessel	Growth Area (mm <sup>2</sup> )	Cell number/ well	Final volume/ well (mL)
<b>Adherent Cells to Seed</b>			
24 well plate	200	6,0·10 <sup>4</sup> -2,0·10 <sup>5</sup>	0,5
6 well plate	962	2,5-8,0·10 <sup>5</sup>	2
<b>Suspension Cells to Seed</b>			
24 well plate	200	2,0·10 <sup>4</sup> -1,0·10 <sup>5</sup>	0,5
6 well plate	962	1,0-5,0·10 <sup>5</sup>	2

On the day of transfection, it is not necessary to change the medium

2. On the day of transfection, prepare CANFAST and DNA solution. Please use medium without serum to prepare them according to the table below.
3. Prepare the transfection mix adding CANFAST solution drop to drop into DNA solution which is gently stirring at vortex.

Recommended Ratios CANFAST Transfection Reagent / DNA					
Tissue Culture Vessel	DNA Solution		CANFAST solution		Transfection Mix (mL)**
	DNA (µg)	Medium without serum (µL)*	CANFAST Reagent (µL)	Medium without serum (µL)*	
96 well plate	0,15	7,5	0,4-1	7,5	15
48 well plate	0,3	15	1-1,8	15	30
24 well plate	0,6	30	2-4	30	60
12 well plate	1	50	2-6	50	100
6 well plate	1-2	100	6-12	100	200
35 mm plate	1-2	100	6-12	100	200
60 mm plate	3-6	300	18-36	300	600
100 mm plate	8-16	800	48-96	800	1600

\* Final volume after DNA or CANFAST Reagent addition  
 \*\* Transfection mix is the addition of volumes from 3<sup>rd</sup> and 5<sup>th</sup> columns.

4. Incubate transfection mix 15-20 minutes at room temperature.
5. For transfection, add transfection mix into each well by leaking. Gentle shake the plate and incubate it 24 – 72 hours. Some cell lines are more sensitive and require change the culture medium 1 – 16 hours after adding the transfection mixture to avoid toxicity.

### PRODUCT USE LIMITATION

This product is developed, designed and sold exclusively for research purposes and *in vitro* use only. The product was not tested for use in diagnostics or for drug development, nor is it suitable for administration to humans or animals. Please refer to [www.canvaxbiotech.com](http://www.canvaxbiotech.com) for Material Safety Data Sheet of the product.