Resource Summary Report

Generated by dkNET on Jul 30, 2026

KG#470

RRID:Addgene_110930
Type: Plasmid

Proper Citation

RRID:Addgene_110930

Plasmid Information

URL: http://www.addgene.org/110930

Proper Citation: RRID:Addgene_110930

Insert Name: rab-3 promoter

Organism: Caenorhabditis elegans

Bacterial Resistance: Ampicillin

Defining Citation: PMID:15489510

Vector Backbone Description: Backbone Size:2632; Vector Backbone:pUC; Vector Types:Bacterial Expression; Bacterial Resistance:Ampicillin

Comments: Unique multi-cloning sites: Nhe I, Sbf I, Kpn I/ Age I/ Xho I, Bgl II This vector is the same as KG#59, except the Sal I and Acc I sites of the multi-cloning site have been replaced with a Sbf I site. Synthesized 2 complementary oligos containing Nhe I and Age I sticky ends and Sbf I and Kpn I sites in between. Hybridization of the 2 oligos to each other will produce Nhe I and Age I sticky ends. After hybridization, this synthetic insert (which is non-phosphorylated on its 5' termini) was cloned into Nhe I/ Age I cut KG#59. Transformed into XL1-Blue electrocompetent cells. Miniprepped 6 clones and tested for the Sbf I site by cutting with Sbf I. Chose one clone that has the Sbf I site with correct band size (and only a single band) and made the glycerol stock. Features of the expression construct: This expression construct has a promoter (rab-3) that will drive strong and specific expression (of whatever gene is inserted in the MCS) in the entire nervous system of juvenile and adult animals. The boundaries of the rab-3 promoter sequence were obtained by examining pRabGFPrim3' from Mike Nonet's lab web site and comparing that to the genome sequence. Other features of the expression vector include a "decoy" 5' to the promoter to reduce non-specific expression in the gut (see Fire Lab 1995 Kit documentation for description), the unc-54 3' end including the poly A addition signal, and 3 introns (1 in 5' UTR, 1 just upstream of the unc-54 3' end, and 1 inserted in the unc-54 3' end). These introns help provide more uniform and robust expression, especially if

you are expressing a cDNA with no introns as opposed to a gene with introns. Note: this vector can be converted to a vector that fuses GFP, YFP, or CFP onto the C-terminus of the protein as follows: Remove the ~1000 bp (Kpn I or Age I) + (Spe I or BsiW I [expensive, 55 C cutter with 50% activity at 37 C; heat inactivate 80 C] or Apa I) fragment containing the unc-54 3' control region from this plasmid and replace it with the ~1800 bp like-digested fragment containing 3-intron S65C GFP (see pPD94.81 in Fire Lab plasmids), YFP (see pPD136.64 in Fire Lab plasmids) or CFP (see pPD136.61 in Fire Lab plasmids) + the unc-54 3' UTR and control region. Note if Spe I is used to cut these Fire Lab vectors, you get 3 bands: 1100, 1800, and 4000 (total size 6.9 Kb). The 1800 bp band is the XFP-containing band. For C-terminal fusions, remember to leave the stop codon off of the upstream protein. For the above-mentioned Fire Lab vectors, the reading frame for the downstream XFP relative to the Kpn I and Age I sites is as follows (triplets are the reading frame codons): Kpn I G GTA CCG GT Age I Alternatively, use Gibson Assembly/ NEBuilder to insert any genetically encoded fluorescent protein at the N- or C-terminus of your protein.

Plasmid Name: KG#470

Record Creation Time: 20220422T221540+0000

Record Last Update: 20260725T073503+0000

Ratings and Alerts

No rating or validation information has been found for KG#470.

No alerts have been found for KG#470.

Data and Source Information

Source: Addgene

Usage and Citation Metrics

We have not found any literature mentions for this resource.