

The Laboratories for Reproductive Biology

PLASMID INFORMATION FORM

NAME: GST-AR 4-52

DESCRIPTION : Amplify ^{pACT2-}AR 4-52 by PCR with primers 5314 (EcoRI created) and 5381 (XhoI created).
The PCR product was digested with EcoRI and XhoI, then ligated into pGEX-4T-1 (EcoRI XhoI digested)

ORIGINATOR: Suxia Bai

VECTOR: pGEX-4T-1

VECTOR SIZE: 4969 bp

INSERT: AR 4-52

INSERT SIZE: 147 bp

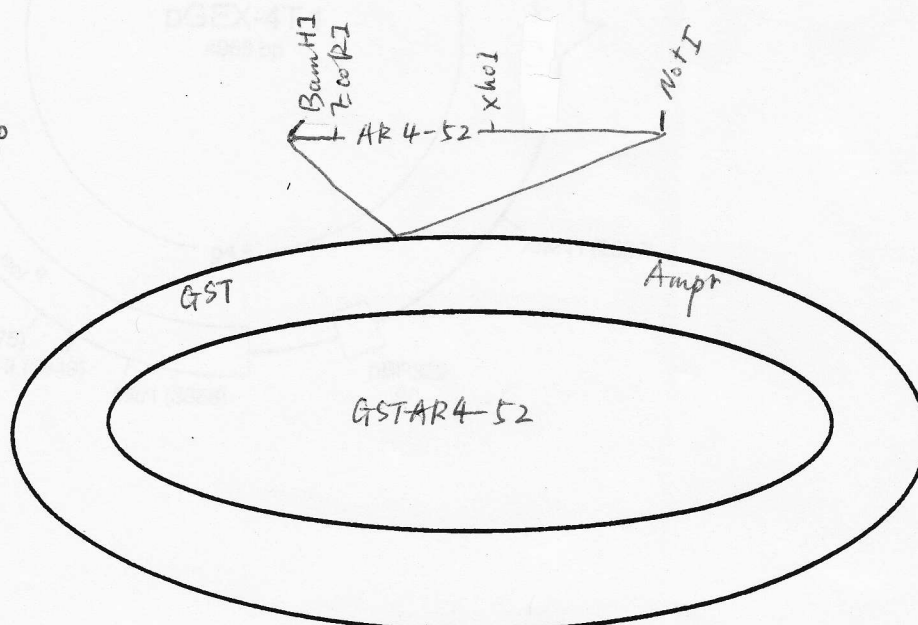
ANTIBIOTIC RESISTANCE: Amp

HOST: XL-1 blue

RELEVANT INFORMATION:

RNA CONCENTRATION:

REFERENCES:



E coli BL21 Cells

5. Structural elements on pGEX-4T-1

Glutathione S-transferase gene region: *tac* promoter: -10: 205-211; -35: 183-188; *lac* operator: 217-237; Ribosome binding site for GST: 244; Start codon (ATG) for GST: 258; Coding region for thrombin cleavage: 918-935; Primer region for double-stranded sequencing: 874-890
MCS: 930-966

β -lactamase gene region: Promoter: -10: 1330-1335; -35: 1307-1312; Start codon (ATG): 1377; Stop codon (TAA): 2235

***lacI^d* gene region:** Start codon (GTG): 3318; Stop codon (TGA): 4398

Plasmid replication region: Site of replication initiation: 2995; Region necessary for replication: 2302-2998.

References

1. Smith, D. B. and Johnson, K. S., *Gene* 67, 31 (1988).
2. Eaton, D., et al., *Biochemistry* 25, 505 (1986).
3. Marston, F. A. O., *Biochem J.* 240, 1 (1986).
4. Schein, C. H. and Noteborn, M. H. M., *Bio/Technology* 6, 291 (1988).
5. Smith, D. B. and Corcoran, L. M., *Current Protocols*, pg. 16.7.1 (1990).

Lambda ZAP[®] is a registered trademark of Stratagene Cloning Systems.

GENOTYPE:

The BL21 strain is F-ompT, *r_B⁻*, *m_B⁻*.

GROWTH CONDITIONS:

The lyophilized culture should be resuspended in 1 ml of L-broth. Grow overnight before plating onto L-broth media plates.

LONG TERM STORAGE:

Mix an equal volume of stationary phase culture (growth in L-broth) with glycerol and store at -70°C. BL21 can be revived by streaking onto L-broth media plates.

RECOMMENDED USAGE:

The BL21 strain is recommended for expression of GST fusion proteins. Since it does not transform well, an alternate strain is recommended for maintenance of the plasmid.

REFERENCE:

Studier, F. W., et al., *Methods in Enzymology*, 185: 60.

Sheet 1 of 3-Back

MAP:

