

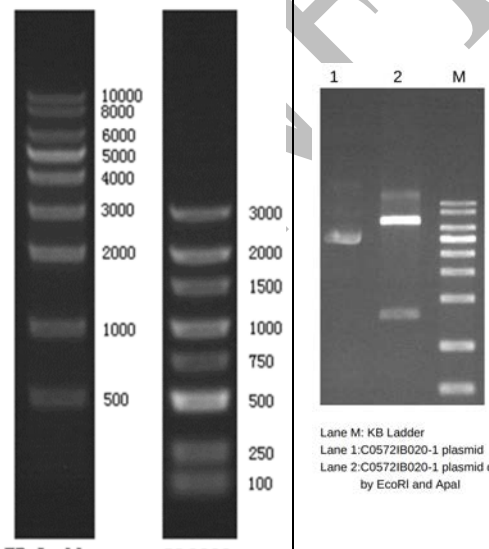
## CERTIFICATE OF ANALYSIS

Vector Name	pCC1FOS	Catalog	V008674
Project/ Lot No.	C0572IB020-1 /PN99239	Size	8,139 bp
Quantity	10 ug	Resistance	Chloramphenicol

### QC Results

Test Items	Specifications	Results
Insert Sequence	Insert sequence results consistent with target	Pass
Vector Sequence	Flanking sequence consistent with expected	Pass
ORF Across Junction	Correct and consistent with target	N/A
Restriction Digest	Expected fragment sizes observed	Pass
PCR Amplification	Correct without non - specific bands	Pass
DNA Quantity/Quality	Actual yield (by A 260 )	5µg/5µg
	Concentration (n/a if lyophilized)	N/A
	Purity (A 260/A280 = 1.8 - 2.0)	Pass
	# of Tubes	2
	Matrix	TE (lyophilized)
Endotoxin Test	Verified, <0.1 EU/µg (Endo-Free Preps Only)	N/A
Appearance	Clear, no visible particles	Pass
Label	Correct and white	Pass
Comments	-	-

### Restriction Digestion Map

 <p>KB Ladder      DL3000</p> <p>10000 8000 6000 5000 4000 3000 2000 1000 500</p> <p>3000 2000 1500 1000 750 500 250 100</p> <p>1    2    M</p> <p>Lane M: KB Ladder Lane 1: C0572IB020-1 plasmid Lane 2: C0572IB020-1 plasmid digested by EcoRI and ApaI</p>		<p><b>Lane 1:</b> Plasmid</p> <p><b>Lane 2:</b> Plasmid    Digested with    BamHI</p> <p><b>Lane M:</b> DNA Marker</p>
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Certified by: Chou Fang

Date: Mar/14/2024

Valid until: Mar/13/2027

### Note

BAC and fosmid clones are highly suitable for modification by recombineering but, because they are present at low (1-2) copies per cell, the DNA is difficult to isolate in high yield and purity. To overcome this limitation vectors, e.g. pCC1BAC/pCC1FOS, have been constructed that contain the additional replication origin, *oriV*, which permits copy-number to be induced transiently when propagated in a suitable host strain, e.g. EPI300, that supplies the cognate trans-replication protein TrfA.

### Protocol for EPI400/EPI300

- Add 4 ml LB media into each test tube. Inoculate each tube with bacterial culture with antibiotic at the proper concentration.
- Incubate the tubes at 37°C, shaking overnight.
- Dilute the starting culture (from step b) 1:10 into antibiotic-supplemented fresh media.
- Supplement induction solution with a ratio of 1:1000, and grow the culture at 37°C for 4 h with vigorous shaking (approx. 250 rpm).
- Isolate DNA from the induced culture cells as per the protocol provided

### Vector Map

