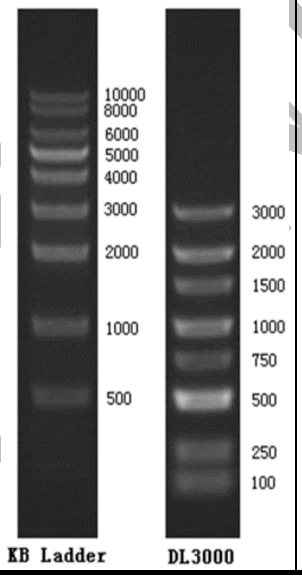
CERTIFICATE OF ANALYSIS

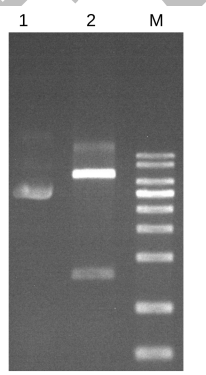
Vector Name	pCC1BAC	**Catalog**	V008675
Project/ Lot No.	C0572IB020-1 /PN99239	**Size**	8,139 bp
Quantity	10 ug	**Resistance**	Chloramphenicol

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





Lane M: KB Ladder
Lane 1:C0572IB020-1 plasmid
Lane 2:C0572IB020-1 plasmid digested by EcoRI and ApaI

Lane 1: Plasmid

Lane 2: Plasmid Digested with BamHI

Lane M: DNA Marker

Certified by: Chou Fang

Date: Mar/14/2023

Valid until: Mar/13/2025

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

a) Add 4 ml LB media into each test tube. Inoculate each tube with bacterial culture with antibiotic at the proper concentration.

b) Incubate the tubes at 37°C, shaking overnight.

c) Dilute the starting culture (from step b) 1:10 into antibiotic-supplemented fresh media.

d) 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).

e) Isolate DNA from the induced culture cells as per the protocol provided

Vector Map