This is a cloning vector that allows mobilization into a wide range of Gram- and Gram+ bacteria. After mobilization, the plasmid can be maintained by integration into the host chromosome via homologous recombination. Excision of the intervening plasmid sequence by a double cross-over event can be facilitated by selection on medium containing 10% sucrose. The sacB gene has been modified to eliminate the HindIII and EcoRI sites in the coding region. This vector differs from pK18mobsacB (ATCC# 87097) only in the orientation of the polylinker.


Designation: pK19mobsacB plamid in E. coli SCS110

Distribution Host: Escherichia coli SCS110

1. Open vial according to instructions.
2. Aseptically add 0.3 to 0.4 mL of liquid medium to the freeze-dried pellet and mix well. Transfer 100 µL to a test tube containing 5 mL LB+ kanamycin (50 µg/mL). A loopful of culture can also be streaked on an agar plate of the same. Incubate cultures at 37°C.
3. Isolate DNA using standard plasmid preparation procedures.

Growth Conditions
Temperature: 37°C
Medium
ATCC Medium 1065 (see below) plus kanamycin (50 mcg/ml) ATCC Medium 1065: Tryptone (Difco 0123), 10.0 g Yeast Extract (Difco 0127), 5.0 g NaCl, 10.0 g Distilled water, 1.0 L

Construct size (kb): 5.66
Marker(s): kanR,sacB

Vector type: plasmid
Cloning sites: HindIII SphI PstI XbaI SmaI EcoRI
Construction: pK19, pSUP102 (RP4 mob) sacB; the sacB gene was inserted into the pK19mob vector.
Insert: sacB
Genome: Bacillus subtilis
Gene name: levansucrase
Insert end: Ecl136II (modification: blunt ended)
Insert size (kb): 1.9
Complete coding sequence ?: Y

Vector: pK19mob
Vector size (kb): 3.76
Type of vector: plasmid
Vector ends: AsuII (modification: blunt ended)

Host range: Escherichia coli; Salmonella sp.; Serratia sp.
Features (with orientation and location, if known):
Marker: kanR
Marker: sacB (sucrose sensitivity)
Other: oriT
Other: oriV
Insert detection: lacZ’, □
MCS: HindIII….EcoRI

References
References and other information relating to this product are available online at www.atcc.org.

Restriction digests of the clone gave the following sizes (in kb): EcoRI 5.6; HindIII 5.6; PstI 5.6.

-ATCC Staff
Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the current publication of the *Biosafety in Microbiological and Biomedical Laboratories* from the U.S. Department of Health and Human Services Centers for Disease Control and Prevention and National Institutes for Health.

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