pRS314 plasmid in E. coli

77143™

Description

One of a series of pBluescript-based centromere vectors (ATCC 77142 -77145, 77157-77158) differing in the yeast selectable marker gene. YC-type centromere vector permitting visual detection of recombinants and production of ssDNA in *E. coli*. Contains promoters for in vitro RNA synthesis, priming sites useful for sequencing, and encodes the lacZ alpha (lacZ?) peptide. pRSS56, constructed by ligating a PvuI fragment (bp 498-2412) of pBluescript KS+ to a PvuI fragment (bp 2850 -730) of pBS(+), contains the KS MCS from pBluescript KS(+) and the unique NdeI and AatII sites between bla and f1 origin of pBS(+). A genomic HindII/PstI fragment (1.002 kb) containing the TRP1 gene was inserted into the NdeI site and a cassette containing CEN6 and the ARS associated with histone 4 (ARSH4) was inserted into the AatII site of pRSS56. All ends were blunted. An EcoRI site in the TRP1-containing fragment (external to the coding sequence) was destroyed. The order of the major features in this plasmid is: TRP1- f1 ori (NaeI) ? T7 promoter ? lacZ?/MCS ? T3 promoter ? pMB1 ori ? bla ? CEN6 ? ARSH4.

Clone type: Vector
Host: *Escherichia coli* HB101 (ATCC 33694)

Storage Conditions

Product format: Frozen
Storage conditions: -80°C or colder

Intended Use

This product is intended for laboratory research use only. It is not intended for any

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animal or human therapeutic use, any human or animal consumption, or any diagnostic use.

BSL 1

ATCC determines the biosafety level of a material based on our risk assessment as guided by the current edition of *Biosafety in Microbiological and Biomedical Laboratories (BMBL)*, U.S. Department of Health and Human Services. It is your responsibility to understand the hazards associated with the material per your organization's policies and procedures as well as any other applicable regulations as enforced by your local or national agencies.

Certificate of Analysis

For batch-specific test results, refer to the applicable certificate of analysis that can be found at www.atcc.org.

Vector Information

Construct size (kb): 4.785

Intact vector size: 4.785

Vector name: pRS314 (phagemid)

Type of vector: phagemid

Construction: pRSS56 [pBluescript KS+, pBS(+)]

Host range: *Saccharomyces cerevisiae*; *Candida robusta*; *Escherichia coli*

Cloning sites: KpnI; ApaI; XhoI; SalI; ClaI; EcoRI; PstI; SmaI; BamHI; SpeI; EagI

Insert detection: lacZ', <-, 1696-2033

Markers: ampR; TRP1

MCS: SacI...KpnI, ->, 1889-1996

Polylinker sites: KpnI; ApaI; XhoI; SalI; ClaI; HindIII; EcoRV; EcoRI; PstI; SmaI; BamHI

Promoters: *In vitro* transcription T7; lac

Replicon: f1, →, 1463-1562; pMB1, 2451-2451; ARSH4, 4328-4702

Growth Conditions

Medium:
ATCC Medium 1227: LB Medium (ATCC medium 1065) with 50 mcg/ml ampicillin
Temperature: 37°C

Notes

Restriction digests of the clone give the following sizes (kb): EcoRI--4.8;
BamHI--4.8; PvuII--4.2, 0.5.
- ATCC staff

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- Genetics 122: 19-27, 1989

YC-type centromere vector permitting visual detection of recombinants and
production of ssDNA in E. coli. Contains promoters for in vitro RNA synthesis,
priming sites useful for sequencing, and encodes the lacZ alpha (lacZ') peptide.
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pRSS56, constructed by ligating a PvuI fragment (bp 498-2412) of pBluescript KS+
to a PvuI fragment (bp 2850-730) of pBS(+), contains the KS MCS from pBluescript
KS+ and the unique NdeI and AatII sites between bla and f1 origin of pBS(+).
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A genomic HincII/PstI fragment (1.002 kb) containing the TRP1 gene was inserted
into the NdeI site and a cassette containing CEN6 and the ARS associated with
histone 4 (ARSH4) was inserted into the AatII site of pRSS56. All ends were
blunted.

- Genetics 122: 19-27, 1989

An EcoRI site in the TRP1-containing fragment (external to the coding sequence) was destroyed.
- Genetics 122: 19-27, 1989

The order of the major features in this plasmid is: TRP1 - f1 ori (NaeI) - T7 promoter - lacZ'/MCS - T3 promoter - pMB1 ori - bla - CEN6 - ARSH4.
- Genetics 122: 19-27, 1989

Restriction digests of the vector gave the following sizes (in kb): EcoRI 4.8 ; BamHI 4.8 ; PvuII 4.2, 0.5.   -------ATCC staff

Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: pRS314 plasmid in E. coli (ATCC 77143)

References

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

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Revision

This information on this document was last updated on 2024-10-25

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