This is a set of two vectors permitting expression in both B. subtilis and E. coli from the vegII promoter. One is a shuttle vector, pRB374, ATCC 77374) and one permits the cloning of translational signals based on the production of lacZ (pRB382, ATCC 77378).

**Designation:** Bacillus subtilis/E. coli expression vectors

**ATCC® NUMBER: 77374™**
**Designation:** pRB374 plasmid in E. coli
**Description:** This is a Bacillus subtilis/E. coli shuttle expression vector containing the Bacillus subtilis promoter vegII which can initiate transcription in both strains. Structural stability of the plasmid in B. subtilis can be affected by high levels of protein production. Under these conditions, cell growth and stability may be improved by reducing the antibiotic concentration in the media. It may not be suitable for cloning very strong expression signals. NeoR confers resistance to neomycin and kanamycin and ble confers resistance to bleomycin and phleomycin. - Gene 122: 187-192, 1992.

**Vector type:** plasmid
**Total size of construct (kb):** 5.9
**Markers:** ampR, neoR, bleR
**Cloning sites:** EcoRI SacI KpnI Smal BamHI XbaI SalI AccI PstI SphI HindIII
**Construction:** pUB110, pBR322, vegII
**Promoter:** vegII (B. subtilis)
**Terminator:** rmb
**Replicons:** pMB1, pUB110

**Host range:** Bacillus subtilis ; Escherichia coli
**Distribution host:** Escherichia coli HB101
**Notes:**
Restriction digests of the vector gave the following sizes (in kb): EcoRI - 5.9 ; BamHI - 5.9 ; BglI/BglII - 3.5, 2.2, 2, 1.4.

**Vector Information**

**ATCC® NUMBER: 77378™**
**Designation:** pRB382 plasmid in E. coli
**Description:** This is a Bacillus subtilis/E. coli shuttle expression vector containing the Bacillus subtilis promoter vegII which can initiate transcription in both strains. The 5' truncated lacZ gene, lacking transcriptional and translational initiation signals, is useful in both E. coli and in B. subtilis, since the endogenous beta-galactosidase level in B. subtilis is low. Structural stability of the plasmid in B. subtilis can be affected by high levels of protein production. Under these conditions, cell growth and stability may be improved by reducing the antibiotic concentration in the media. It may not be suitable for cloning very strong expression signals. NeoR confers resistance to neomycin and kanamycin and ble confers resistance to bleomycin and phleomycin. - Gene 122: 187-192, 1992.

**Vector type:** plasmid
**Total size of construct (kb):** 8.9
**Markers:** ampR, neoR, bleR
**Cloning sites:** EcoRI SacI KpnI Smal BamHI XbaI SalI AccI PstI SphI HindIII
**Construction:** pUB110, pBR322, vegII, lacZ
**Promoter:** vegII (B. subtilis)
**Terminator:** rmb
**Replicons:** pMB1, pUB110

**Host range:** Bacillus subtilis ; Escherichia coli
**Distribution host:** Escherichia coli HB101
**Notes:**
Restriction digests of the clone gave the following sizes (in kb): EcoRI - 8.9 ; BamHI - 8.9 ; BglI/BglII - 5.1, 2.2, 1.1, 0.8 ; BglI/BglII - 3.5, 2.2, 2.0, 1.4.

**Propagation**

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+ ampicillin (50-100 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.

References

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

Biosafety Level: 1

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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