

Shuttle Vector

An Expression Vector for Gram(+) and Gram(-) Prokaryotes

Invention

The heterologous expression of genes in prokaryotes can be challenging, especially if the genes originate from a distant host or if the source is uncertain, such as a metagenomic expression library. Many vectors have been developed based on broad host range origins of replication, but these tend to focus on either gram(+) or gram(-) prokaryotes.

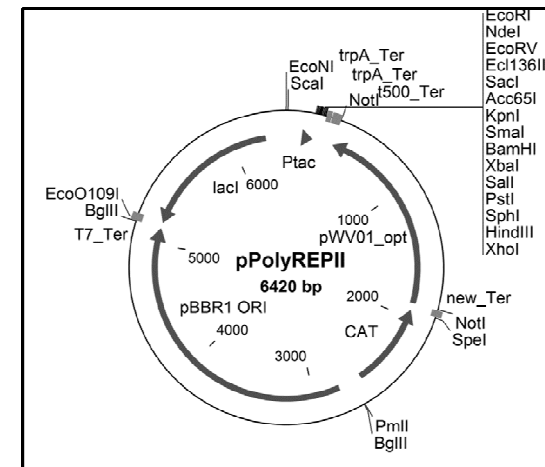


Fig.: pPolyREPII

E. coli DH5 α , *Pseudomonas putida* KT2440 and *Bacillus subtilis* 168. The presence of GFP was confirmed by western blot analysis using antibodies against His₆ tag.

This novel broad host range expression vector can be equally well used in gram(+) and gram(-) hosts. Thus, this new tool may be used to establish environmental DNA (eDNA) expression libraries and to screen for desired activities in different gram(+) as well as gram(-) hosts at the same time. The vector may allow finding novel biocatalysts, which are not functional in the limited number of vector compatible hosts so far.

Commercial Opportunities

The present invention provides a novel broad host range expression vector for establishing expression libraries with unknown DNA sequences like eDNA libraries.

Current Status

PROvendis offers access to rights for commercial use. In case of interest we will be pleased to inform you about the current patent status.

Relevant Publication

Paper submitted.

An invention of the Westfaelische Wilhelms-University of Muenster (UniMuenster).

Competitive Advantages

- **Broad host range vector enables to use gram(+) and gram(-) hosts at the same time for expression**
- **Suitable for the establishment of expression libraries with unknown DNA (e.g. environmental DNA)**
- **Might be a new way to find novel biocatalysts**

This invention regards the construction of a completely synthetic expression vector, namely pPolyREPII, based on both the pBBR1 (for gram-positive) and pWV01 (for gram-negative) origin of replication, allowing replication in both types of organisms.

The expression of pPolyREPII was assessed by cloning the *gfp* gene with a His₆ tag sequence. A strong *gfp* expression could be observed in various prokaryotes, such as

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