# **Cloning Vector pBR322**

Product Information Sheet # V30302



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SUMMARY

shipped on blue ice; store at -20 °C

For research use only

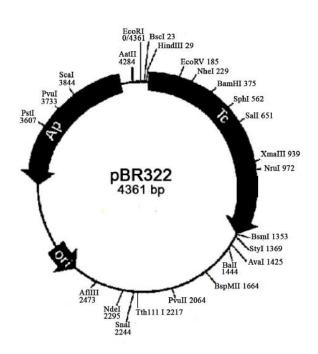
## **Product Description and Application**

pBR322 was one of the first plasmids to be completely sequenced, but the sequence has been revised several times. The last revision and review of the sequence information inclicates that two base pairs in the previously accepted sequence do not exist - hence some sources quote the size as 4363 bp.

Note that the *Bscl* and *HindIII* sites in the promoter region of the tetracycline resistance gene can be used as cloning sites, but may not always cause gene inactivation and loss of tetracycline resistance. The gene is only inactivated if insertion into the promoter region alters the reading frame of the transcribed region. pBR322 can be mobilized because the bom site is present.

The vector DNA is highly purified by ion exchange chromatography, cesium chloride density centrifugation and gel Filtration. Our DNA preparations yield DNA with over 80% supercoiling. Therefore, the plasmid DNA is ready-to-use for enzymatic reactions and transformations.

## **Vector Map**





## **Quality Control & Technical Details**

Protein contamination is monitored by measuring the ratio of absorbance at 260 and 280 nm. All preparations must have  $A_{260}/A_{280}$  greater than 1.8, indicating essentially protein-free DNA.

The absence of nuclease activity is measured by incubating plasmid DNA in restriction buffer for 16 hours. No DNA degradation should be observed.

The DNA's suitability for enzymatic manipulation is tested by restriction with a variety of endonucleases.

The correct banding pattern is confirmed by agarose gel electrophoresis.

The transformation efficiency of the plasmid DNA is measured and antibiotic resistance and blue/white selection is also confirmed.

During storage at 4 °C, plasmid DNA will slowly convert from supercoiled to relaxed circles. Although this will not affect restriction, transformation efficiency will drop.

For long-term storage keep at -20 °C. If multiple freeze-thawing cycles are likely to occur, dispense small volumes into sterile tubes and store at -20 °C.

The plasmid DNA is supplied in TE buffer (10 mM Tris-HCl, pH 8.0, 1 mM EDTA).

## References

Bolivar, F. *et al.*, Gene, 2 (1977) 95 - 113 Sutcliffe, J. G., Cold Spring Horb. Symp. Quant. Biol., 43 (1978) 77 - 90 Watson, N., Gene, 70 (1988) 399 - 403

## Order Information, Shipping and Storage

Order#	Product	Quantity
V30302	pBR322 Vector DNA	25 µg
shipped on blue ice; store at -20 °C		