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Rsal is a restriction enzyme purified from *Rhodopseudomonas sphaeroides*.

| <u>Catalogue No</u> | 129-1, 500 U | | |
|---------------------|----------------|--|--|
| | 129-2, 2500 U | | |
| | 129-3, 12500 U | | |
| | | | |

| <u>Concentration</u> | 10-12u/μl and 40- | | |
|--------------------------|----------------------------|--|--|
| | 60u/µl* | | |
| *Add an H to cat.# to or | der the high concentration | | |

Reagents supplied: 10x M and 10x K buffer

Unit substrate: Lambda DNA.

Unit calculation assay conditions: 50 mM NaCl, 10 Tris-HCl (pH 7.9 @ 25°C), 10 mM MgCl₂, 1 mM dithiothreitol, 100 μ g/ml BSA. Incubate at 37°C.

Absence of contaminants: 400 units of Rsa I do not produce any unspecific cleavage products after 16 hrs incubation with 1 µg of λ DNA at 37°C. After 10-fold overdigestion with Rsa I, greater than 95% of the DNA fragments can be ligated and recut with this enzyme.

Storage buffer: 50 mM KCl, 10 mM Tris-HCl (pH 7.4), 0.1 mM EDTA, 1 mM dithiothreitol, 200 μ g/ml BSA and 50% glycerol. Store at -20°C.

Heat inactivation: 65°C for 20 minutes.

Methylation Sensitivity:

dam methylation: Not sensitive dcm methylation: Not sensitive CpG methylation: Blocked by some combinations of overlapping **Note:** Cleaves single-stranded DNA slowly.

| Percent Activity in MINOTECH Buffers | | | | | |
|--------------------------------------|-----|----|-----|-----|-----|
| L | М | Н | SH | А | К |
| 75-100 | 100 | 50 | <10 | <10 | 100 |

General reaction mixture:

| 10U Rsal | 1μl | | | |
|-----------------------------|-------------|--|--|--|
| 10x M or K buffer * | 2µl | | | |
| DNA substrate | <1µg | | | |
| Sterile ultrapure water | Up to 20 µl | | | |
| Incubate for 15 min at 37°C | | | | |

*In the case of M buffer we recommend the addition of BSA to a final concentration of 100 μ g/ml.

Frequency of Cutting

| λ | Ad-2 | Фх174 | pUC18 | M13mp18 | pBR322 |
|-----|------|-------|-------|---------|--------|
| 113 | 83 | 11 | 3 | 19 | 3 |



Lambda DNA 1.4 % agarose

