

ASSAY METHODS

MATERIALS

Assay buffer: 50 mM HEPES, pH 7.4,
100 Mm NaCl, 2 mM EDTA, 3 mM DTT.

Prepare 5 X pNPP substrate (10 mM) in
the assay buffer

Enzyme preparation: LAR (0.1 mg/ml)

PROCEDURE

1. Prepare reaction mixtures in a 96-well
plate (keep on ice). Add in the order:

60 μ l of assay buffer

20 μ l of 10 mM pNPP in buffer (final
conc. 2 mM)

10 μ l 1% BSA

10 μ l of 0.1 mg/ml LAR enzyme in the
assay buffer

Mix well and start the reaction at 30 °C
water bath, and incubate for 10 min.

2. Add 100 μ l per well of 2 M K₂CO₃ to stop
the reaction.

3. Read absorbance at 405 nm using a
plate reader.

REFERENCES

1.H. Cho et al. Biochemistry 1991, 30: 6210-6216

2.H. Cho et al. Biochemistry 1992, 31: 133-138

PRODUCT SPECIFIC REFERENCES

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