Applications:
- Highly efficient degradation of ATP to AMP.
- Removal of deoxynucleotides in DNA pyrosequencing between cycles (3).
- Conversion of 5’ triphosphorylated RNA to ligatable monophosphorylated form that can be used for 5’ RNA adaptor ligation.
- Conversion of 5’ triphosphorylated RNA to 5’ exonuclease XRN-1 (NEB #M0338) sensitive monophosphorylated RNA.
- As a metal-dependent enzyme Apyrase can be inhibited by EGTA and EDTA.
- The activity of Apyrase is approximately 30% in NEBuffers 1.1, 2.1, 3.1 and CutSmart™ Buffer.
- Apyrase does not remove 5’ caps from eukaryotic mRNA.

Source:
- Isolated from a strain of E. coli that carries the coding sequence for potato S. tuberosum apyrase (4).

Specific Activity: 3,000 units/mg
- Apyrase has a higher ratio of activity for ATP:ADP (14:1).
- Apyrase is a calcium-activated enzyme. It is approximately 50% active when Mg2+ substitutes Ca2+ in Apyrase Reaction Buffer.

Quality Control Assays
- Phosphatase Activity (PNPP Assay): Incubation of a 50 μl reaction in 1X Apyrase Reaction Buffer containing 10 mM p-Nitrophenyl Phosphate (PNPP) and a minimum of 5 units of Apyrase for 4 hours at 37°C results in < 0.1% substrate dephosphorylation as determined by spectrophotometric analysis.

Notes:
- Apyrase has a higher ratio of activity for ATP:ADP (14:1).
- Apyrase is a calcium-activated enzyme. It is approximately 50% active when Mg2+ substitutes Ca2+ in Apyrase Reaction Buffer.

Heat Inactivation: 65°C for 20 minutes
- Apyrase is a calcium-activated enzyme. It is approximately 50% active when Mg2+ substitutes Ca2+ in Apyrase Reaction Buffer.

Quality Control Assays
- Physical Purity: Purified to > 95% homogeneity as determined by SDS-PAGE analysis using Coomassie Blue detection.

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Single Stranded DNase Activity (FAM-Labeled Oligo): A 50 μl reaction in CutSmart Reaction Buffer containing a 20 nM solution of a fluorescent labeled double-stranded oligonucleotide containing a 3’ extension and a minimum of 2.5 units of Apyrase incubated for 16 hours at 37°C yields < 5% degradation as determined by capillary electrophoresis.

DNase Activity (Labeled Oligo, 3’ extension): A 50 μl reaction in CutSmart Reaction Buffer containing a 20 nM solution of a fluorescent labeled double-stranded oligonucleotide containing a 3’ extension and a minimum of 2.5 units of Apyrase incubated for 16 hours at 37°C yields < 5% degradation as determined by capillary electrophoresis.

DNase Activity (Labeled Oligo, 5’ extension): A 50 μl reaction in CutSmart Reaction Buffer containing a 20 nM solution of a fluorescent labeled double-stranded oligonucleotide containing a 5’ extension and a minimum of 2.5 units of Apyrase incubated for 16 hours at 37°C yields < 5% degradation as determined by capillary electrophoresis.

DNase Activity (Labeled Oligo, blunt end): A 50 μl reaction in CutSmart Reaction Buffer containing a 20 nM solution of a fluorescent labeled double-stranded oligonucleotide containing a blunt end and a minimum of 2.5 units of Apyrase incubated for 16 hours at 37°C yields < 5% degradation as determined by capillary electrophoresis.

References:

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