

Random hexamer mix

Catalogue number: MB12901, 500 μL (25 μg)

Primer sequence: 5'- d (NNNNN)-3', where N = G, A, T or C

Description

Random hexamer mix includes oligonucleotides representing all possible hexamer sequences. Random hexamers are commonly used for priming singlestranded DNA or RNA for extension by DNA polymerase or Reverse transcriptase, typically when the template is difficult to copy in its entirely.

During cDNA synthesis, random hexamer will random priming throughout the entire length of the RNA to generate a cDNA pool containing various lengths of cDNA. With this method, all RNAs present in a population constitute templates for the cDNA synthesis experiment.

Concentration

500 μL (25 μg) at 50 ng/μL, 25.25 μM

Applications

- cDNA synthesis using a Reverse Transcriptase (RT reaction)
- DNA synthesis using Klenow Fragment of DNA polymerase I
- DNA probe synthesis

Features

- Ultra-pure grade
- DNase/ RNase free

Storage conditions

Random hexamer mix should be stored at -20 °C, in a constant temperature freezer. Avoid multiple freeze/thaw cycles.

Protocol

Use 1-5 μ L in a 20 μ L reverse transcription (RT) reaction (50-250 ng/reaction).

Note: The sensitivity of cDNA synthesis may be improved when using a mixture of random hexamer and oligo(dT)₁₈ primers.

Quality control assays

Nucleases assays

To test for DNase contamination, 0.2-0.3 μ g of pNZY28 plasmid DNA are incubated with 1 μ L of Random hexamer mix for 14-16 hours at 37 °C. To test for RNase contamination, 1 μ g of RNA is incubated with 1 μ L of Random hexamer mix for 1 hour at 37 °C. Following incubation, the nucleic acids are visualized on a GreenSafe-stained agarose gel. There must be no visible nicking or cutting of the nucleic acids. Similar tests are performed with the reaction buffer.

Functional assay

Random hexamer mix is tested for performance in a RT-qPCR experiment using total RNA from mouse liver. After the first-strand cDNA synthesis with NZY Reverse Transcriptase, the resultant cDNA is then used as template in a quantitative real-time PCR assay using specific primers to amplify the mouse GAPDH gene.

Related products

| Product Name | Cat. No. |
|-----------------------------|----------|
| NZY Reverse Transcriptase | MB124 |
| Oligo (dT)18 primer mix | MB12801 |
| Water for Molecular Biology | MB11101 |

V2201

| Certificate of Analys | is |
|---|--------|
| Assay | Result |
| Nucleases Assay | Pass |
| Functional Assay | Pass |
| Approved by: | |
| Patrícia Ponte Senior Manager, Quality Systems | |

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