Technical Info

Well advised with Roth.

Nucleotides

All Carl Roth nucleotides are manufactured from highest-quality reagents and are most thoroughly tested for quality. This testing procedure not only includes standard-PCR but also 'long-range-PCR' of up to 18 kb, repeated quantitative light-cycling reactions and tests for physical stability.

General Parameters

- Available as ready-to-use set or mix, for contamination-free applications
- Purity ≥90 % (HPLC-tested)
- Tested for 'long-run-PCR' of up to 18 kb
- DNase-, RNase- and Protease-free
- Free of PCR inhibitors like modified bases and tetra-pyrophosphate
- Adjusted to pH 8.5 for superior stability even during larger numbers of freeze-and-thaw cycles
- Also available adjusted to pH 7.0 for special applications (see below)
- Highly efficient enzymatic fabrication

Suitable for

- Common PCR reactions
- Light cycling
- cDNA synthesis
- Labelling and Primer extension
- Mutagenesis assays
- Sequencing
- *In vitro* transcription

pH-Values and Solvents of the Nucleotides

Thorough and extensive evaluation showed that the adjustment of solubilised nucleotides to a higher pH value considerably improves the stability of nucleotides as well as the sensitivity of quantitative light cycling-PCR, particularly when the nucleotides are exposed to repeated freeze-and-thaw cycles. Therefore we increased all standard nucleotides, nucleotide-sets, and -mixtures to a **pH of 8.5±0.1**.

Since, furthermore, few Reverse Transcriptases (e.g. Superscript RT from Invitrogen) require nucleotides of a neutral pH for efficient enzymatic activity, the most important nucleotide mixture is also provided **adjusted to pH 7.0** (Roti®-Mix PCR 3 (pH 7), Art. No 0179).

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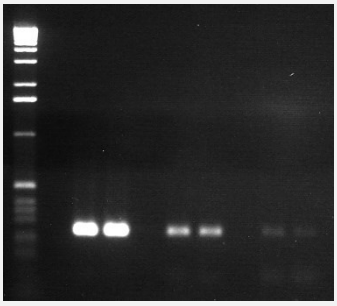
Nucleotides	Art. No.	pH-value	Solvent
dNTP-solutions	K035-K038, L539, P732	8.5 ±0.1	Water
dNTP-sets	K039, L540, P731	8.5 ±0.1	Water
dNTP-mixtures	L541, L542, L785, L786	8.5 ±0.1	Water
dNTP-Set	0179	7.0 ±0.1	Water
NTP- solutions	K045-K048	8.0 ±0.2	20 mM Tris-HCl*
NTP-sets	K049	8.0 ±0.2	20 mM Tris-HCl*
Labelled nucleotides	1047, 1048, 1049	7.5 ±0.2	Water

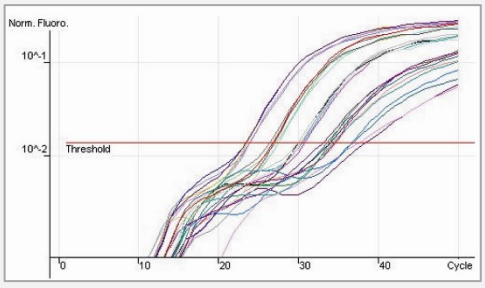
*For high efficient Reverse Transcription

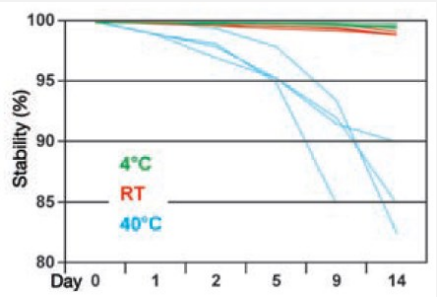
Quality Monitoring



Sensitivity assay I: Amplification of a 260 bp fragment of human genomic DNA with (left to right) 250 ng (A), 25 ng (B) and 2.5 ng (C) DNA as template (two replicates).

Sensitivity assay II: Quantitative light-cycling-PCR on recombinant DNA with (left to right) 10 ng, 1 ng, 100 pg, 10 pg, 1 pg DNA as template (six replicates).





Stability testing: HPLC analysis of all four dNTPs following incubation periods of 1-14 days at different temperatures (4 °C - green; room temp.
-red; 40 °C - blue). Stability of nucleotides is >99 % even after 14 days incubation at room temp. Even after an
incubation period of 9 days at 40 °C, 85 % of all nucleotide molecules are still intact.

LHa 06/2023

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