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NUCLEOTIDES

GENERAL PARAMETERS

- ✓ Available as ready-to-use set or mix, for contamination-free applications
- ✓ Purity ≥98 % (HPLC-tested)
- ✓ Tested for 'long-run-PCR' of up to 30 kb
- ✓ DNAse-, RNAse-, Protease- and Phosphatase-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

PCR, Light-cycling, cDNA synthesis, labelling and primer extension, mutagenesis assays, sequencing and *in vitro* transcription.

All Roth nucleotides are manufactured from highest-quality reagents and are most thoroughly tested for quality. This testing procedure not only includes standard-PCR (fig. 1) but also 'long-range-PCR' of up to 30 kb, repeated quantitative light-cycling reactions (fig. 2), and tests for physical stability (fig. 3).

*of dATP, dGTP, dCTP and dUTP. dTTP is synthesized chemically.

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 mixtures are also provided **adjusted to pH 7.0** (dNTP-Set 1 (pH 7), Art. No. 0178 and Roti[®]-Mix PCR 3 (pH 7), Art. No 0179).

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 oder -Mix	0178, 0179	7.0 ±0.1	Water
NTP- solutions	K045-K048	8.0 ±0.2	20 mM Tris-HCI*
NTP-sets	K049	8.0 ±0.2	20 mM Tris-HCI*
ddNTP- solutions	K040-K043	8.0 ±0.2	Water
ddNTP-sets	K044	8.0 ±0.2	Water
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 periodes 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.

Carl Roth GmbH + Co. KG

Schoemperlenstraße 3-5 76185 Karlsruhe Postfach 100121 76231 Karlsruhe Telefon: +49 (0) 721/ 5606-0 Telefax: +49 (0) 721/ 5606-149 E-Mail: info@carlroth.de Internet: www.carlroth.de

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