

RT² NANO PREAMP™ cDNA SYNTHESIS KIT



Enabling the Analysis of 1 Nanogram of RNA with RT² Profiler™ PCR ARRAYS

What Is the RT² Nano PreAMP™ cDNA Synthesis Kit?

RT² Nano PreAMP cDNA Synthesis Kit and Primer Mixes are a breakthrough technology enabling expression analysis using as little as 1 ng of total RNA. It employs a proprietary preamplification process to faithfully increase the amount of targeted cDNA for PCR Array analysis. This technology empowers RT² Profiler PCR Arrays to accurately analyze **nanogram** levels of total RNA.

Samples that can NOW be characterized with real-time PCR Arrays include:

- **Laser Captured Microdissection Samples (LCM)**
- **Fine Needle Aspiration Biopsy (FNAB)**
- **Stem Cell Clusters or Embryoid Bodies**
- **Flow Cytometry / Fluorescent-Activated Cell Sorting (FACS)**

Combined with PCR Arrays, the RT² Nano PreAMP cDNA Synthesis Kit and Primer Mixes create a complete system for the accurate analysis of as little as 1 ng of total RNA.

- **RT² Nano PreAMP cDNA Synthesis Kit:** Proprietary kits include optimized reagents for first strand cDNA synthesis and preamplification from only 1 ng of total RNA.
- **RT² Nano PreAMP cDNA Synthesis Primer Mixes:** Ready-to-use primer mixes for amplifying pathway-specific cDNA templates.

Benefits of RT² Nano PreAMP cDNA Synthesis Technology

- **Robust Performance on Small Samples:** Analyze up to 4 different PCR Arrays with as little as 1 ng of Total RNA.
- **Easy Workflow and Designed for Routine Use:** Simple and quick procedures with minimal hands-on time to preamplify target templates under 2 hours.
- **Superior Sensitivity:** Maximally enhances the sensitivity of RT² Profiler PCR Arrays to analyze limited amounts of RNA.

NANO PREAMP FURTHER INCREASES THE SENSITIVITY OF PCR ARRAYS

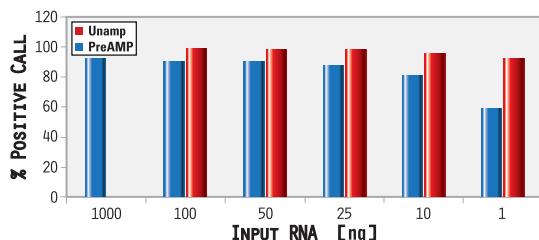


Figure 1. RT² Nano Preamplification Significantly Increases Sensitivity of Detection from 1 ng RNA Samples. Different amounts of human universal RNA were converted to cDNA with (red) or without (blue) RT² Nano preamplification. The unamplified and preamplified samples were then analyzed on the Human Inflammatory Cytokines and Receptors PCR Array (PAHS-011), which contains 84 pathway-specific assays, plus controls, including 5 assays for housekeeping genes. Threshold cycle values (C_t) were obtained and any genes with a C_t < 35 were considered to be present. Results indicate that with Nano preamplification, a 33.7% increase in positive call rate is observed in samples with as little as 1 ng RNA.

How RT² Nano PreAMP cDNA Synthesis Kits Work

Two Simple Steps:



1. cDNA First Strand Synthesis

This kit provides enough reagents for synthesizing first strand cDNA from 12 different samples.

2. Preamplification of cDNA for Pathway Specific Genes

Each first strand cDNA synthesis reaction can be amplified by 4 different sets of PCR Array-specific Primer Mixes, allowing gene expression analysis on as many as 4 different PCR Arrays.

UNBIASED AMPLIFICATION PROCESS COMPARISON OF ΔC_t VALUES BETWEEN PREAMPLIFIED AND UNAMPLIFIED cDNA

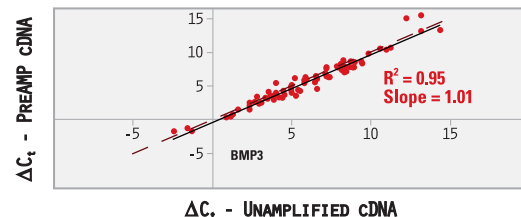


Figure 2. Unbiased Amplification Process - Highly Comparable ΔC_t Values Between Preamplified and Unamplified cDNA from Human Liver Tumor.

First strand cDNA was synthesized from 5 ng of human liver tumor RNA. One-quarter of each RT product was used for preamplification with the RT² Nano PreAMP cDNA Master Mix Kit plus Human Cancer PathwayFinder™ Nano PreAMP Primer Mix. Unamplified cDNA synthesized from 500 ng of the same liver tumor RNA sample was used as the control. PreAMP amplified and unamplified cDNA samples were then analyzed on the Human Cancer PathwayFinder™ PCR Array, and the threshold cycle values (C_t) were obtained. The ΔC_t value for each gene was calculated by subtracting the average C_t value of the five reference genes (B2M, HPRT1, RPL13A, GAPDH, and ACTB) on the PCR Array from the C_t value of each gene of interest. The concordance of the ΔC_t values between preamplified and unamplified samples was evaluated by regression analysis. Data points with C_t ≥ 35 were considered to be absent genes and were excluded from the analysis. The dashed line represents the ideal slope of 1.0. The solid line shows a linear regression fit with the R² and slope indicated. The high correlations between preamplified and unamplified cDNA were also obtained from universal RNA samples (*data not shown*).

RT² Nano PreAMP cDNA Synthesis Products

Product	Catalog #	Price
RT ² Nano PreAMP cDNA Synthesis Kit	C-06	\$ 499
Select RT² Nano PreAMP cDNA Synthesis Primer Mixes		
Human Cancer PathwayFinder™	PBH-2033	\$ 149
Human Apoptosis	PBH-7012	\$ 149
Human Angiogenesis	PBH-4024	\$ 149
Human Mesenchymal Stem Cell	PBH-4082	\$ 149
Human Embryonic Stem Cell	PBH-1081	\$ 149
Human Extracellular Matrix and Adhesion Molecules	PBH-0013	\$ 149
Human Inflammatory Cytokines and Receptors	PBH-4011	\$ 149
Mouse Inflammatory Cytokines and Receptors	PBM-9011	\$ 149
Human Toll-like Receptor Signaling Pathway	PBH-5018	\$ 149
Mouse Toll-like Receptor Signaling Pathway	PBM-0018	\$ 149
RT² Nano PreAMP Primer Mixes for Other PCR Arrays	Various	\$ 149