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CellsDirect[™] One-Step and Two-Step qRT-PCR Kits

Sensitive results, faster throughput with limited samples

- Increased sensitivity—No RNA isolation steps to contribute to sample loss and PCR inhibition
- Significant savings—Less hands-on time and a lower cost than RNA purification kits
- Robust results—Real-time detection of targets from a single cell to 10,000 cells
- Versatility—Compatible with a wide variety of cell lines and samples from Laser Capture Microdissection (LCM)

CellsDirect[™] qRT-PCR kits deliver highly sensitive and specific, real-time qPCR results directly from cells, without the need for an RNA purification step. CellsDirect[™] technology is designed for maximum sensitivity with small samples (Figure 1). By eliminating costly, time-consuming RNA purification procedures (Figure 2), it is especially valuable for high-throughput applications and is ideal for gene expression and RNAi knockdown experiments.



HeLa cells were lysed and resuspended according to the CellsDirect[™] protocol. Ten-fold serial dilutions of cell lysate were prepared from 10,000 cells to one cell. A low abundance target, RRKCA, was detected using a TaqMan[®] probe with the CellsDirect[™] One-Step qRT-PCR Kit with ROX on an ABI PRISM[®] 7700 instrument. Efficiency as measured from a standard curve (not shown) was 97%, with an R² value of 0.998.

Figure 2—CellsDirect[™] technology increases throughput



CellsDirect[™] qRT-PCR technology provides a significant savings of hands-on time and cost compared to conventional qRT-PCR methods involving expensive and lengthy RNA purification steps.

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CellsDirect[™] One-Step qRT-PCR Kits

CellsDirect[™] One-Step qRT-PCR kits combine sensitivity with the highest level of convenience and throughput. With CellsDirect[™] One-Step Kits, just add your cell lysate directly to your qRT-PCR reaction mixture, place it in your real-time instrument, and go. These kits are ideal for gene expression assays from samples collected using LCM (Figure 3).

Figure 3—CellsDirect[™] One-Step qRT-PCR Kits provide sensitive results with LCM samples



Samples were isolated from an ovine source using the LCM cap method, and cells were lysed using the CellsDirect[™] protocol. A medium-to-low abundant target, MCP-1a, was detected via a TaqMan[®] probe using the CellsDirect[™] One-Step qRT-PCR kit with ROX on the ABI PRISM[®] 7700 instrument. Successful amplification was seen from as few as 10 cells.

CellsDirect[™] Two-Step qRT-PCR Kits

CellsDirect[™] Two-Step qRT-PCR Kits provide the sensitivity of CellsDirect[™] technology (Figure 4) while allowing archiving of cDNA. Two-step kits are available for either probe/primer or SYBR[®] Green detection.



qPCR was performed on 10-fold serial dilutions (undiluted to 1:10,000) of cDNA synthesized from 10,000 HeLa cells using the CellsDirect[™] Two-Step qRT-PCR Kit. Four microliters of each dilution were added to a 50-µl qPCR reaction containing 200 nM each primer and 100 nM TaqMan[®] Probe to GAPDH (Panel A; standard curve: 98% efficiency, R² = 0.9967) or TFRC (Panel B; standard curve: 97% efficiency, R² = 0.9999). Reactions were incubated for 2 min. at 50°C, then 2 min. at 95°C followed by 50 cycles of 95°C for 15 sec; 60°C for 30 sec. using the ABI PRISM[®] 7700.

Ordering information

For sensitive, convenient qRT-PCR from limited samples, order CellsDirect[™] One-Step and Two-Step qRT-PCR Kits today.

Product	Quantity	Cat. no.
Kits for fluorogenic probe/primer detection		
CellsDirect [™] One-Step qRT-PCR Kit with ROX	100 rxns	11754-100
for ABI 7000, 7300, 7700, or 7900(HT)	500 rxns	11754-500
CellsDirect [™] One-Step qRT-PCR Kit	100 rxns	11753-100
for ABI 7500 and all other instruments	500 rxns	11753-500
CellsDirect™ Two-Step qRT-PCR Kit	100 rxns	11737-030
for any instrument	500 rxns	11737-038
Kits for SYBR® Green detection		
CellsDirect [™] SYBR [®] Green Two-Step qRT-PCR Kit	100 rxns	11738-060
for any instrument	500 rxns	11738-068

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