Universal Human Reference RNA

Catalog #740000

Storage Store the Universal Human Reference RNA at –80°C. Store the RNase-free water at –20°C.

INTRODUCTION

Stratagene's Universal Human Reference RNA is composed of total RNA from 10 human cell lines. The reference RNA is designed to be used as a reference for microarray gene-profiling experiments. Since RNA species differ in abundance between cell lines, an ideal reference sample should represent these different RNAs. Equal quantities of DNase-treated total RNA from each cell line were pooled to make the Universal Human Reference RNA. This Universal Reference RNA is suitable for microarray experiments. Stratagene also supplies a QPCR Human Reference Total RNA, suitable for QRT-PCR, which has undergone further DNase treatment.

MATERIALS PROVIDED

Material Provided	Quantity
Reference RNA	2 tubes x 200 µg each
RNase-free water	1.5 ml

Cell Line Derivations	
Adenocarcinoma, mammary gland	Melanoma
Hepatoblastoma, liver	Liposarcoma
Adenocarcinoma, cervix	Histiocytic lymphoma; macrophage; histocyte
Embryonal carcinoma, testis	Lymphoblastic leukemia, T lymphoblast
Glioblastoma, brain	Plasmacytoma; myeloma; B lymphocyte

ADDITIONAL MATERIALS REQUIRED

RNase-free 70% Ethanol

PROTOCOL

Universal Human Reference RNA is provided in a solution of 70% ethanol and 0.1 M sodium acetate. Prepare the Reference RNA for use as follows:

1. Centrifuge the tube at 12,000 × g for 15 minutes at 4°C.
2. Carefully remove the supernatant.
3. Wash the pellet in 70% ethanol.
4. Centrifuge the tube at 12,000 × g for 15 minutes at 4°C.
5. Carefully remove the supernatant and air-dry the pellet at room temperature for 30 minutes to remove retained ethanol.
6. Resuspend the pellet in RNase-free water to the desired concentration.

Proceed with the preparation of labeled cDNA and interrogate the arrays according to the manufacturer's instructions.

QUALITY CONTROL TESTING

The quality of the Universal Human Reference RNA is assessed by observing distinct 28S and 18S ribosomal bands on a 1× MOPS agarose gel under denaturing conditions. The purity of the RNA is assessed by spectrophotometry (A260/A280 ≥1.8). The RNA is then shown to be free of contaminating RNases by incubation in a suitable buffer at 37°C followed by gel analysis against known RNAse-free controls. The RNA is further tested functionally by synthesizing labeled cDNA, which is then hybridized to a microarray to examine gene representation and coverage.

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