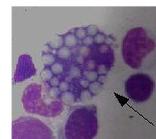


2. LIPSGENE® – GENE EXPRESSION QUANTIFICATION



LIPSGENE® 18S rRNA Kit

Intended use

The LIPSGENE® 18S rRNA Kit is intended for real-time PCR *in vitro* quantification of preformed 18S ribosomal transcript cDNA, in total RNA or mRNA samples prepared from crude or purified cells of human, mice, rat and rabbit origin. The RNA encoding 18S is an extremely abundant molecule. The 18S rRNA levels are reported to be less likely to vary under conditions that affect the expression of mRNAs. They have been shown to be more reliable than other housekeeping genes in rat pancreas, human skin fibroblasts and human and mouse malignant cell lines. However, there are two drawbacks to its use: rRNA cannot be used for normalisation when quantitating targets that have been enriched for mRNA because it is lost during mRNA purification, and rRNA is expressed at much greater levels than the vast majority of target mRNAs.

Kit contents

- ✓ Lyophilized oligonucleotide mix containing 18S rRNA cDNA specific primers and probes, PCR vessels containing stabilized synthetic 18S rRNA standard DNA (ready-to-use reference curves), sample PCR tubes (shipped at room temperature or together with taq polymerase on dry ice).
- ✓ Taq Polymerase, 10x PCR buffer, 50 mM Mg-chloride solution, PCR grade water, 10x ROX (shipped on dry ice).
- ✓ Sufficient to run 120 tests.

Note: Reverse transcription reagents available with a separate kit (M-MLV Reverse Transcription Kit, Cat.no. 1050001XX-050).

Performance assessment

The LIPSGENE® 18S rRNA Kit was evaluated together with total RNA isolated with "Reagent 14" using the Autogen 540 nucleic acids extraction robot (Integrated Separation Systems).

Assessment criterion	Sample type	Performance
Analytical sensitivity	Synthetic 18S rRNA DNA	≥5 copies/run
Linear range	Synthetic 18S rRNA DNA	9 logs

2. LIPSGENE® – GENE EXPRESSION QUANTIFICATION

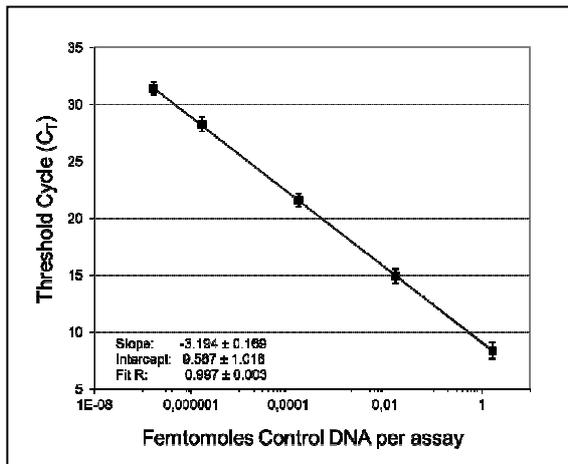
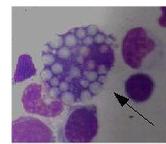


Figure 2.21: Repeatability of *18S rRNA* cDNA quantification assay. The study was performed with synthetic *18S rRNA* DNA specimen and 42 replicates for each concentration (ready-to-use *18S rRNA* DNA standards; 1.66, 1.66×10^{-2} , 1.66×10^{-4} , 1.66×10^{-6} , 1.66×10^{-7} femtomoles per tube) on ABI PRISM SDS. The mean run data are included in the figure.

Typical run results

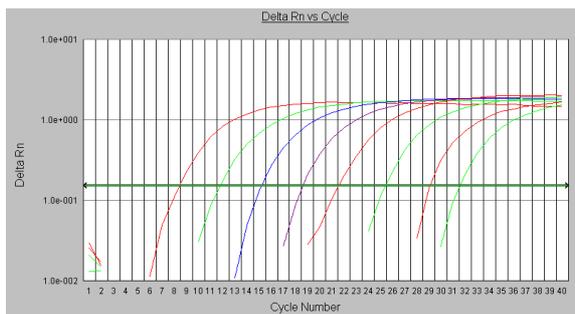


Figure 2.22_ Quantification of *18S rRNA* cDNA using an ABI PRISM SDS. Dynamic range. Amplification of the ready-to-use *18S rRNA* standard DNA (10^9 , 10^8 , 10^7 , 10^6 , 10^5 , 10^4 , 10^3 , 10^2 copies per tube).

Ordering information

Kit version	RP	ST	LP	LC	SC	SP	RS
IvD state	RUO	RUO	RUO	-	-	-	-
120 tests	1030103RP-120	1030103ST-120	1030103LP-120	-	-	-	-

RP = 0.2 ml regular profile 8-well/strip PCR tubes and cap strips (e.g. ABI PRISM® 7000/7300/7700 SDS [Applied Biosystems/Life Technologies], iCycler IQ™; IQ5 [Bio-Rad], MX3000P, Mx3005P [Stratagene]; Mastercycler® ep realplex [Eppendorf]); **ST** = Single 0.2 ml PCR Tube with attached cap (e.g. Rotor-Gene™ 3000/6000, Rotor-Gene Q [Qiagen]; LineGene K [Bioer]); **LP** = 0.1 ml low profile 8-well/strip PCR tubes and cap strips (e.g. , 7500 Fast [Applied Biosystems/Life Technologies], MiniOpticon™, CFX-96 [Bio-Rad]); **LC** = 20 µl LightCycler glass capillaries (LightCycler 2.x, Roche); **SC** = 25 µl SmartCycler tubes (e.g. SmartCycler® [Cepheid]); **SP** = 20 µl DX-12 reaction tubes (Spartan Dx-12); **RS** = 0.1 ml 4-well tube/strips (Rotor-Gene™ 3000/6000, Rotor-Gene Q [Qiagen]).