

DNA quantitation in plasma, blood, tissues, paraffin tissues, and biological fluids

Set Quant DNA Real time

Cat. n. 1.440RT

The DNA quantitation in plasma, blood, tissues, paraffin tissues and in biological fluids is performed by Quantitative Real Time PCR technique. The quantitation of plasma circulating DNA, that allows quantitation of pico-grams (pg) of DNA, is a useful marker for the early diagnosis of lung cancer. It allows, furthermore, the monitoring of the disease, detecting an early relapse and the presence of tumor metastasis in the patients follow-up. The medium concentration of circulating DNA in lung patients plasma is 8 fold higher than healthy subjects. The hTERT has been used as target gene, mapped on 5p.15.33 chromosome and the amplified fragment is of 98bp (from 13059 to 13156, GeneBank accession number AF128893).

This kit allows to quantify circulating and genomic DNA in plasma, blood, tissues, paraffin tissues and in biological fluids using quantitative REAL TIME PCR technique. It is based on the detection and quantification of a fluorescence reporter engendering a signal proportional increasing to the yield of PCR products. The fluorescence reporter is linked to the end of a specific probe for the target gene and it is turned off by the quencher linked to the opposite site of the probe. This quencher links to the target gene and, when PCR begins, the probe is degraded by DNA polymerase. The digestion of the probe causes the distancing of quencher and reporter and the reporter may give out fluorescence. The PCR technique is based on two primers and a fluorescent probe specific for the target fragment. The probe is linked at 5' with JOE (reporter) and at 3' with TAMRA (quencher).

Sensitivity: < 15pg DNA

Specificity: 99%

Principle of method: A) isolation of DNA B) Real-Time PCR.

Applicability: Su fc DNA isolated and purified by plasma, blood, fresh or paraffin tissues, biological fluids.

Number of reaction: 100

KIT CONTAINS AND STORAGE

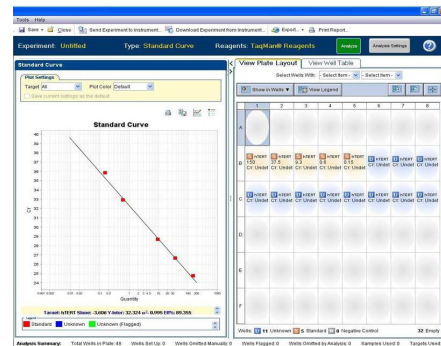
AMPLIFICATION

PCR mix 2X Primer/Probe	4°C
mix 20 X H ₂ O sterile	-20°C
Standard 1 (150 ng)	-20°C
Standard 2 (37.5 ng)	+ 4°C
Standard 3 (9.3 ng)	+ 4°C
Standard 4 (0.6 ng)	+ 4°C
Standard 5 (0.15 ng)	+ 4°C

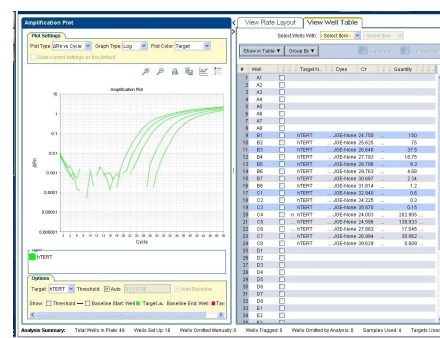
Stability: over 12 months if correctly stored.

ANALYSIS OF RESULTS

Standard Curve



Amplification Plots



References:

- Clin Chem* (2006); 52: 1833-42
- J Clin Oncol* (2003); 21: 3902-8.
- Mutat Res* (2007); 635: 105-17.