

Sensitivity, Precision, and Efficiency:

How qPCR and dPCR Revolutionize Cell & Gene Therapy Research

Quantitative PCR (qPCR) and digital PCR (dPCR) are powerful tools in the field of cell and gene therapy research.

They aid in the discovery of target genes, lead new molecular entities (NMEs), and biomarkers, which are essential for the development of effective therapies. These techniques enable accurate genetic analysis during the discovery research phase, helping to identify and characterize drug targets and biomarkers.

This infographic is intended for research use only and not for diagnostic procedures.

Benefits of qPCR



High sensitivity

Detects very low levels of gene expression or DNA/ RNA targets



Speed

Results are provided within a few hours, making it a rapid research technique for gene expression analysis



Quantification

Provides a precise measurement of the amount of target present



Versatility

Used for a wide range of research applications for biologics discovery & development, such as gene expression analysis, copy number variation analysis, microbial detection, quantitative genotyping, and environmental monitoring with a wide dynamic range



Accuracy

High accuracy and reproducibility, allowing for reliable and consistent results



Cost-effective

Relatively cost-effective, user-friendly technique when compared to other methods like droplet dPCR (ddPCR), dPCR microarray analysis, or sequencing

Thermo Fisher Scientific qPCR ecosystems



Multiplexing

Can detect and quantify multiple targets simultaneously, enabling the analysis of different genes



Automation

Easily automated, saving time and reducing the risk of human error



Small sample requirement

Only a small amount of starting material is required, making it suitable for precious or limited samples



Standardization

qPCR assays can be standardized and evaluated to facilitate consistency

Benefits of dPCR



Absolute quantification

Enables absolute quantification of DNA or RNA targets without the need for a standard curve, making it highly accurate and reliable



High precision

High precision and lower variability compared to qPCR, particularly when quantifying low target concentrations



Enhanced sensitivity

Detects and quantifies small amounts of target nucleic acids, even in complex samples with high background noise or low target abundance



Improved robustness

Less prone to inhibition from PCR inhibitors present in complex sample matrices, allowing more reliable and consistent results



Copy number variation analysis

Particularly suitable for detecting and quantifying copy number variations (CNVs) or rare genetic mutations, providing valuable insights into genomic alterations



Detecting rare alleles and small-fold changes

Identifies and quantifies rare alleles making it useful for detecting low-frequency mutations or monitoring minimal residual disease



Increased resolution

Can offer improved resolution by partitioning samples into thousands of individual reactions, allowing for absolute quantification of targets with high precision



Accurate detection of target DNA/RNA

High tolerance for PCR inhibitors and is less affected by sample quality or degradation, making it a robust method for detecting and quantifying target DNA or RNA



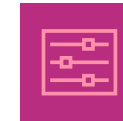
No need for perfect amplification efficiency

Unlike qPCR, dPCR does not require amplification efficiency to be perfect for accurate quantification, making it less sensitive to variations in PCR efficiency



Flexibility in sample types

dPCR can be performed on a wide range of sample types, including challenging samples such as degraded DNA, fragmented RNA, or samples with low target abundance



Better control over PCR bias

dPCR provides better control over PCR bias, as it works on the basis of absolute quantification rather than relying on relative amplification efficiencies, reducing the impact of bias on results



Digital readout

dPCR provides a clear digital readout of positive or negative results, making it easier to interpret and analyze data compared to qPCR, which relies on fluorescence signals and amplification curves

Workflow Stages

Stage 1



Discovery Phase

- New cell and gene therapy approaches are identified and researched
- Underlying mechanisms are investigated
- Novel cells or vectors are developed



Preclinical Research

- The most promising therapy approaches are tested in animal models
- Safety and efficacy evaluation

Stage 2



Pharmacology and Toxicology

- Pharmacokinetic studies (behavior of the drug in the body) are conducted
- The toxicity of the research therapy in different organs and tissues is assessed

Stage 3



Production and Quality Control

- Therapeutics are manufactured on a larger scale
- Quality control measures are implemented to support consistency, purity, and safety of the product

Stage 4



Identify genes of interest (GOI) or targets



Biomarker discovery



Lead identification

Application in Cell and Gene Therapy Research Development

Optimizing plasmid construction

Assessing gene copy number

Testing and characterizing therapy components

Supporting purity and stability of the final product and batches under cGMP production

Used in *in vivo* analysis or bioanalysis (preclinical and clinical studies):

- Biodistribution
- Pharmacokinetics/pharmacodynamics (PK/PD)
- Safety and Efficacy studies

Conclusion



qPCR and dPCR are essential techniques in the field of cell and gene therapy research. They allow for the discovery of causal variants, target genes, and biomarkers, vital components in the development of effective therapies.



By providing accurate genetic analysis during the early stages of research, these techniques help identify and characterize drug targets and biomarkers, thus avoiding costly lead elimination in later stages.



qPCR and dPCR also enable the confirmation and screening of biomarkers, optimization of plasmid construction, assessment of gene copy number, and testing and characterization of various components in cell and gene therapy development. These techniques also play a significant role in ensuring the purity, safety, stability, and identity of therapy products.



qPCR and dPCR are indispensable tools for advancing the field of cell and gene therapy research.