qPCR or dPCR: Choosing the right solution for your research needs

Roche

Quantitative PCR (qPCR) and digital PCR (dPCR) are used to quantify specific nucleic acids in biological samples. Both techniques are compatible with many applications but differ in a number of ways, from throughput to sensitivity. So, which method is better suited to your laboratory application?

WORKFLOW OVERVIEW

Extraction sample

Prepare PCR reaction mix with intercalating dye or hydrolysis-based probes

qPCR	dPCR
Transfer reaction mix and sample (nucleic acid) to PCR plate	Partition reaction mix and sample (nucleic acid) into many chambers for individual PCR reactions
Perform real-time PCR	Perform endpoint PCR
Quantify against a standard reference curve	Apply Poisson calculation to determine concentration
Results in ~ 1.5 – 2 hours	Results in ~ 2 hours

ADVANTAGES AND DISADVANTAGES

qPCR		dPCR	
Well-established protocols and assays	✓	More complex workflow	✗
Easy to automate and increase throughput	✓	Lower throughput with fewer options for automation	✗
Cost effective	✓	Higher cost	✗
Susceptible to PCR inhibitors	✗	Highly tolerant of PCR inhibitors	✓
Relative quantification against a standard reference curve	✗	Absolute quantification that shows a binary result, without the need for reference standards	✓
Low sensitivity	✗	High sensitivity, which increases further with more partitions	✓
Poor precision	✗	High precision over a wide dynamic range	✓

APPLICATIONS

qPCR	dPCR
	Copy number variation (CNV)* analysis
	Haplotyping
	NGS library quantification
Gene expression analysis in complex samples and single cells	
	Pathogen detection
	Liquid biopsy (cfDNA and ctDNA)
siRNA, miRNA and lncRNA detection	
	Genome editing detection and validation
	GMO detection
Residual host cell DNA quantification	
	Rare mutation detection
	Viral load quantification

Choosing between qPCR and dPCR ultimately depends on your goals, requirements and available resources. **There is no right or wrong answer,** and most applications are amenable to both methods.

*Also known as copy number alteration (CNA)

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