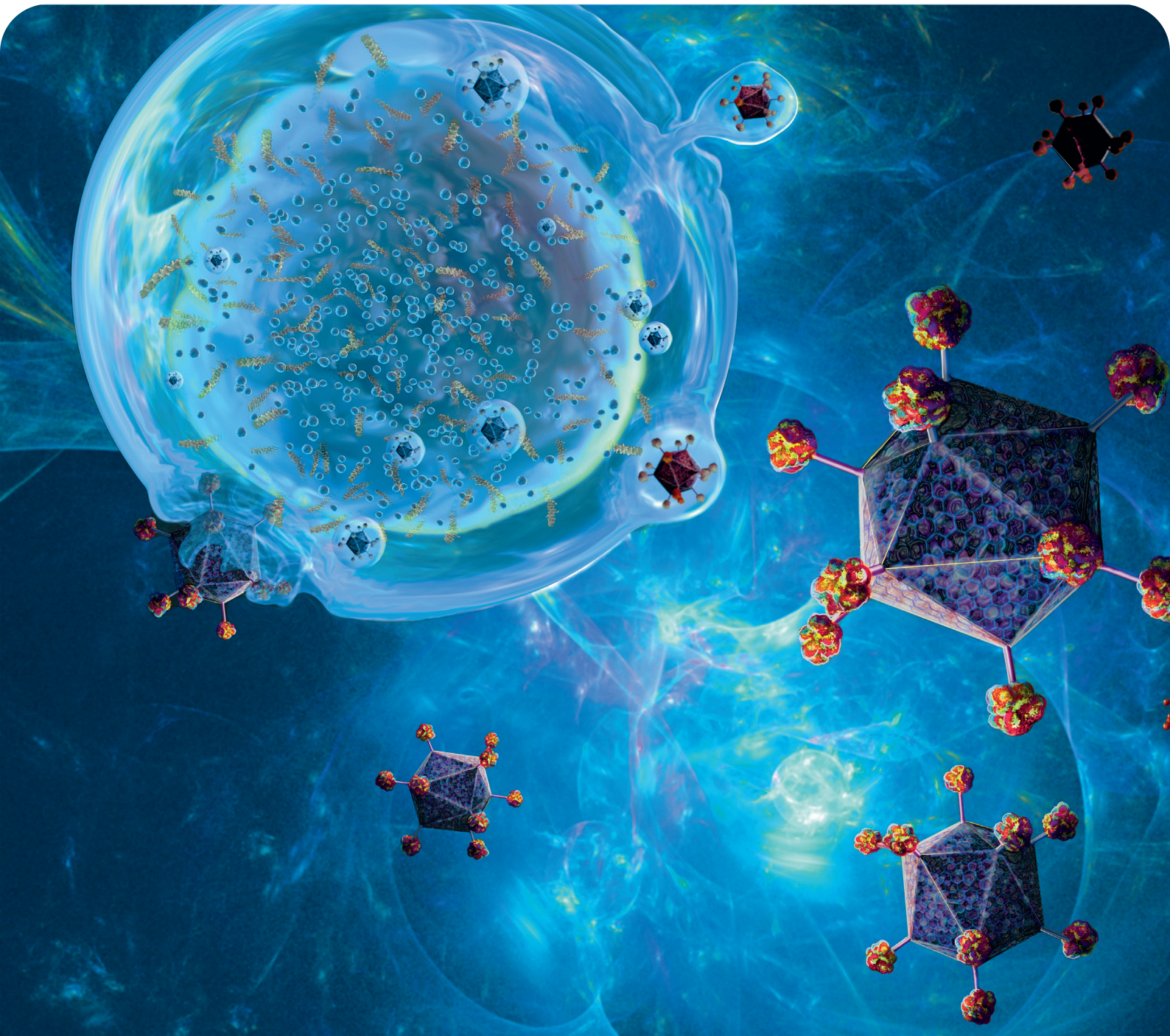
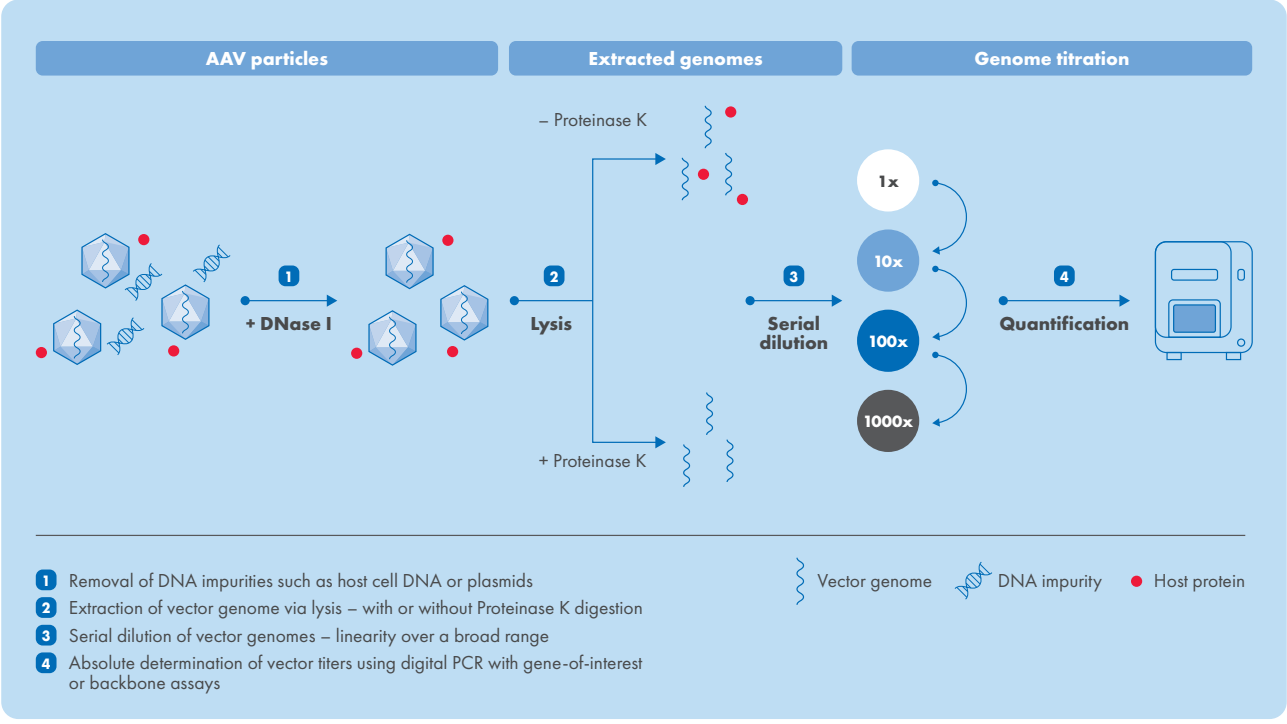
Absolute. Accurate. Verified.

Simplifying AAV quantification to support the next generation of gene therapies



Adeno-associated viruses (AAVs) are widely used viral vectors for gene therapy applications. Viral vector generation and purification require precise quality control to ensure safe and reliable dosing during clinical studies or patient care.

To maximize success, we have developed a standardized digital PCR (dPCR) workflow from capsid lysis to viral titer quantification. It offers significant advantages in accuracy, reproducibility, speed and ease of use, enabling robust vector genome titration critical to manufacturing and determining clinically effective dosages of AAV-based gene therapies.



The complete workflow combining CGT Viral Vector Lysis Kit with QIAcuity® CGT dPCR Assays and QIAcuity Probe PCR Kit on the QIAcuity dPCR platform provides:

- Standardization of AAV lysis with easier SOP implementation and QC of current viral titration workflows
- Consistent and reproducible measurement of viral titers for multiple serotypes
- Accurate quantification of AAV vector genomes down to 0.3 copies/µL
- Robustness of <10% CV between operators and assays
- Ability to run singleplex or multiplex reactions
- Compatibility with in-process and purified samples

The CGT Viral Vector Lysis Kit is compatible with several dPCR platforms

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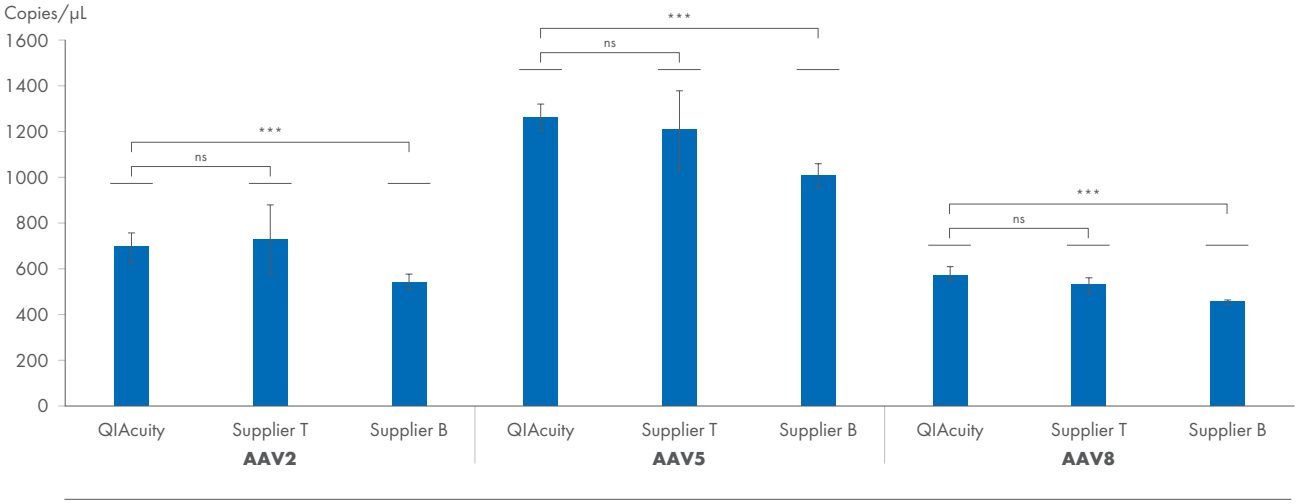

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Samples	QIAcuity CV (%)	Supplier T CV (%)	Supplier B CV (%)
AAV2	8.9	18.6	7.0*
AAV5	4.4	14.1	5.4*
AAV8	4.6	5.8	2.1*

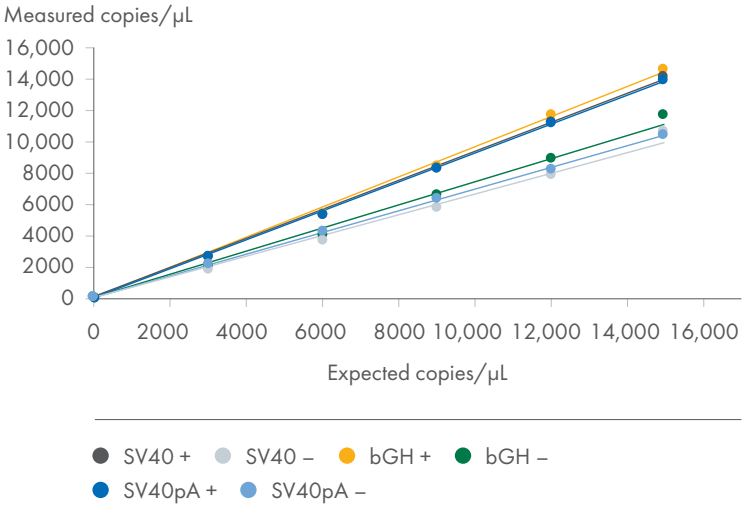
*Based on only 2 targets. Other values are based on 3 targets.

Three purified AAV reference standards (Vector Biolabs) were processed using the CGT Viral Vector Lysis Kit. Three biological replicates per serotype were processed, and all samples were run with four technical replicates each. After processing, the same lysates were used for quantification on the QIAcuity using 8.5k Nanoplates as well as on the digital PCR system from supplier T and supplier B following the manufacturer's protocol.
A. Quantification is shown for the CMV promoter target.
B. Triplex and duplex quantifications were used for statistical analysis.
An unpaired t-test was performed with ns $p > 0.05$, * $p < 0.05$, ** $p < 0.01$, *** $p < 0.005$. Mean coefficients of variations for all assays are shown.

Removal of secondary structures from the inverted terminal repeat (ITR) regions leads to increased quantification of ITR and non-ITR targets

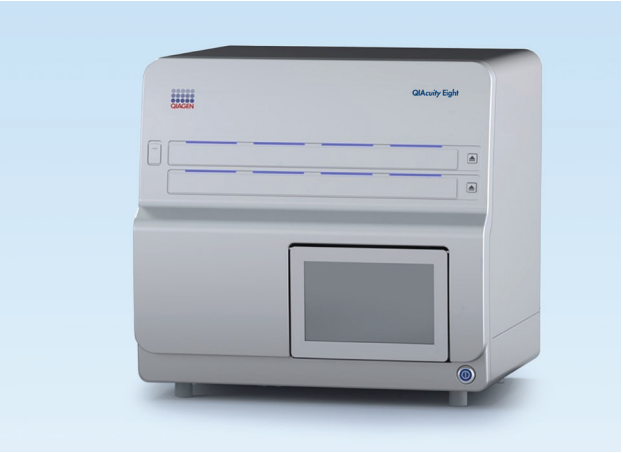
AAV2 in-process samples were processed using the CGT Viral Vector Lysis Kit and quantified on a QIAcuity dPCR system using 8.5k nanoplates and CGT dPCR assays. Half of the samples were digested using HpaII and the other half was left undigested. The samples were serially diluted in six steps from 15,000 cop/µL down to 2.5 cop/µL.

The expected copies are based on an ITR estimate determined by qPCR measurements from the reference standard supplier and are not directly comparable to dPCR measurements.



Maximize AAV workflow efficiency with high-throughput processing on the QIAcuity Eight Digital PCR Platform

Feature	QIAcuity Eight
Plates processed	8
Detection channels (multiplexing)	5
Thermocycler(s)	2
Time to result	First plate under 2 h Every ~40 min a following plate
Throughput (samples processed per 24 hour-shift)	Up to 3264 (96-well)



Ordering Information

Product	Contents	Cat. no.
QIAcuity CGT dPCR Assays	One tube of target sequence dPCR assay, for 500x12 µL reactions in 8.5k Nanoplate	250230–250256
CGT Viral Vector Lysis Kit	Reagents enough for 100 and 1000 samples	250272, 250273
QIAcuity Probe PCR Kit	1 mL or 5 mL master mix for the QIAcuity dPCR instrument; water	250101, 250102

For more information about the new viral vector lysis method, visit **www.qiagen.com/cgt-viral-vector-lysis-kit**

Ordering	**www.qiagen.com/shop**
Technical Support	**www.support.qiagen.com**
Website	**www.qiagen.com**