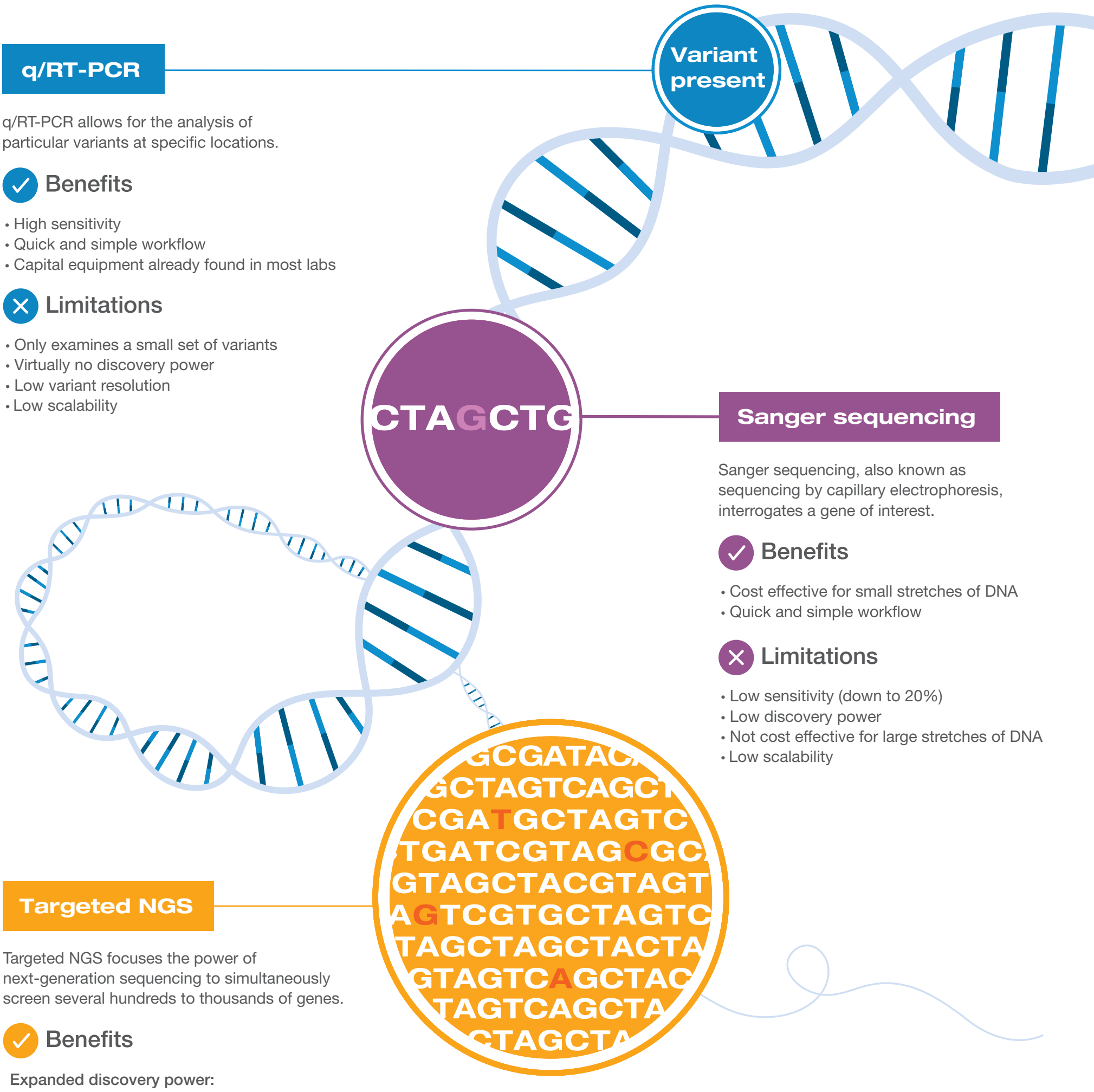
Targeted next-generation sequencing vs qPCR and Sanger sequencing

Technologies used to interrogate DNA and RNA have come a long way. From qPCR to Sanger sequencing to next-generation sequencing (NGS), explore the benefits and the limitations of each to understand which method you should choose.



q/RT-PCR

q/RT-PCR allows for the analysis of particular variants at specific locations.

Benefits

- High sensitivity
- Quick and simple workflow
- Capital equipment already found in most labs

Limitations

- Only examines a small set of variants
- Virtually no discovery power
- Low variant resolution
- Low scalability

Sanger sequencing

Sanger sequencing, also known as sequencing by capillary electrophoresis, interrogates a gene of interest.

Benefits

- Cost effective for small stretches of DNA
- Quick and simple workflow

Limitations

- Low sensitivity (down to 20%)
- Low discovery power
- Not cost effective for large stretches of DNA
- Low scalability

Targeted NGS

Targeted NGS focuses the power of next-generation sequencing to simultaneously screen several hundreds to thousands of genes.

Benefits

Expanded discovery power:

- Comprehensive genomic coverage
- Greater resolution of genomic variants
- More data from smaller DNA amounts
- Higher throughput with sample multiplexing

Limitations

- May be less cost effective when interrogating a low number of samples

Which to choose—and when?

Sanger sequencing and q/RT-PCR are good choices if you need to interrogate a small region of the DNA on a limited number of samples.

Otherwise, targeted NGS is more likely to effectively suit your needs. It allows you to cost effectively screen more samples and detect multiple variants across targeted areas of the genome, which would be a costly and time-consuming effort on the Sanger and q/RT PCR methods.

To learn more about targeted NGS, visit: **www.illumina.com/targeted2**

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