

# Gene expression assays

## RNA detection without RNA extraction

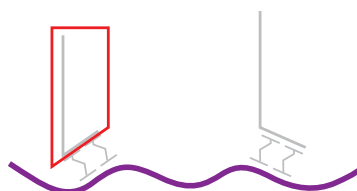
Invitrogen™ gene expression assays based on branched DNA (bDNA) technology are versatile and enable a comprehensive approach to systems biology and translational sciences for verification and quantitation of biomarkers identified by next-generation sequencing or microarray studies, or from published literature.

### Invitrogen™ products based on bDNA technology

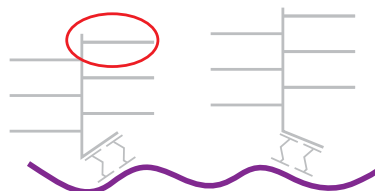
- QuantiGene™ Singleplex assay
- QuantiGene™ Plex assay
- ViewRNA™ ISH tissue assay
- ViewRNA™ ISH cell assay
- ViewRNA™ Cell Plus assay
- PrimeFlow™ RNA assay

### How it works

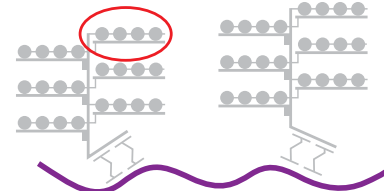
These assays are hybridization-based and utilize bDNA signal amplification technology. It allows for direct quantitation of gene expression transcripts by signal amplification, as seen in Figure 1. A pair of target-specific probe sets, approximately 20 nucleotides in length, hybridizes to contiguous sequences on the target RNA (or DNA). Signal amplification is achieved through successive hybridization of proprietary oligonucleotide sequences to build the bDNA structure, formed by preamplifiers, amplifiers, and labeled probes, resulting in excellent specificity, low background, and high signal-to-noise ratio.



**Step 1:** Preamplifier ("trunk") binds paired probe set



**Step 2:** Amplifier ("branch") binds preamplifier



**Step 3:** Label probes ("leaves") consisting of fluorochrome-conjugated oligos bind to multiple sites on amplifier

Figure 1. bDNA technology.

**bDNA technology–based Invitrogen™ assays at a glance.**

	QuantiGene assays		ViewRNA ISH tissue assays	ViewRNA ISH cell assays		ViewRNA Cell Plus assays	PrimeFlow RNA assays
	Singleplex	Plex		mRNA ISH	High-content ISH		
<b>Target</b>	mRNA, lncRNA, DNA, miRNA	mRNA, lncRNA, DNA	mRNA	mRNA	mRNA	mRNA, miRNA, circular RNA	mRNA, miRNA, lncRNA, viral RNA
<b>Multiplexing</b>	1 mRNA or 1 microRNA or 1 DNA target	Up to 80 RNA targets or up to 77 DNA targets	Up to 2 RNA targets	Up to 4 RNA targets	Up to 4 RNA targets	Up to 3 RNA targets	Up to 4 RNA targets
<b>Simultaneous protein detection</b>	No	No	No	No	No	Yes	Yes
<b>Sample types</b>	Cultured cells, blood, fresh-frozen or FFPE tissues, plants, bacteria, virus	Cultured cells, blood, fresh-frozen or FFPE tissues, plants, bacteria, virus	FFPE and OCT-frozen tissue sections, TMAs, FNAs	Cultured cells (adherent or suspension)	Cultured cells (adherent or suspension)	Cultured cells (adherent or suspension)	Cultured cells, single-cell suspensions; PBMCs, BMCs
<b>Detection</b>	Chemiluminescence	Fluorescence	Chromogenic/fluorescence	Fluorescence	Fluorescence	Fluorescence	Fluorescence
<b>(Substrate)</b>	(Lumigen™ APS-5 substrate)	(Streptavidin-phycoerythrin)	(Fast Red and Fast Blue)	(Invitrogen™ Alexa Fluor™ 488, 546, 647, and 750 dyes)	(Alexa Fluor 488, 546, 647, and 750 dyes)	(Alexa Fluor 488, 546, and 647 dyes)	(Alexa Fluor 488, 568, 647, and 750 dyes)
<b>Limit of detection</b>	≤200 transcripts/well	≤1,000 transcripts/well	Single RNA copy	Single RNA copy	Single RNA copy	Single RNA copy	10–15 RNA copies/cell
<b>Technical requirements</b>	Luminometer	Luminex® 100/200™, MAGPIX®, FLEXMAP 3D® systems	Brightfield or fluorescence microscope or slide scanner	Fluorescence microscope or high-content imaging platform	High-content imaging platform	Fluorescence microscope or high-content imaging platform	Flow cytometer
<b>Compatible instruments</b>	Thermo Scientific™ Fluoroskan™ FL, Luminoskan™, or Varioskan™ LUX instrument	Luminex system	Invitrogen™ EVOS™ imaging system or Thermo Scientific™ CellInsight™ HCA platform	EVOS imaging system or CellInsight HCA platform	CellInsight HCA platform	EVOS imaging system or CellInsight HCA platform	Invitrogen™ Attune™ NxT Flow Cytometer

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