

## ddPCR™ Library Quantification Kit for Ion Torrent



The ddPCR library quantification kit provides absolute quantification of Ion AmpliSeq and Ion RNA-Seq next-generation sequencing (NGS) libraries. In combination with the QX100™ or QX200™ Droplet Digital™ PCR system, the ddPCR library quantification kit lets you:

- **Quantify your Ion Torrent libraries** — produces highly precise measurements without the use of standards
- **Visualize the quality of your library** — ddPCR fluorescence amplitude plots highlight well- and poorly formed libraries; these quality metrics are not available when using other methods
- **Balance your sequencing libraries precisely** — enables consistent loading of the Ion Torrent sequencing platforms

For more information, visit [www.bio-rad.com/web/ddPCRIonTorrent](http://www.bio-rad.com/web/ddPCRIonTorrent).

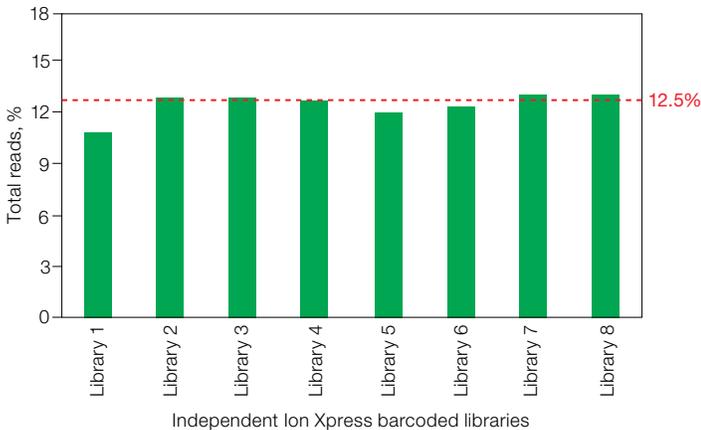
## Absolute Quantification, Quality, and Balance of Ion Torrent Libraries

The kit contains primers and a hydrolysis probe (Figure 1) for detection and absolute quantification of Ion Torrent NGS libraries. A signal from the assay is used to indicate the formation of properly adapted library fragments in a 1-D fluorescence amplitude plot in QuantaSoft™ software.

The kit also enables the user to balance libraries for sequencing (Figure 2). Absolute quantification using the ddPCR library quantification kit is performed without the use of standards. In addition to accurate quantification, the data plots generated by the QX100 and QX200 systems are information rich and provide quality metrics of the library construction that are not available when using other methods.

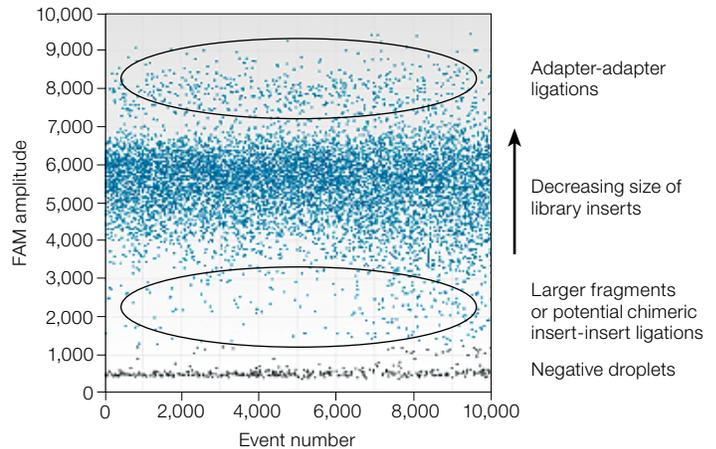


**Fig. 1. Design of the ddPCR library quantification kit assay.** Bio-Rad's ddPCR library quantification kit contains an assay designed to span both the X and P1 (or A and P1) adapter sequences, allowing for quantification of correctly formed library fragments. FP, forward primer; RP, reverse primer.

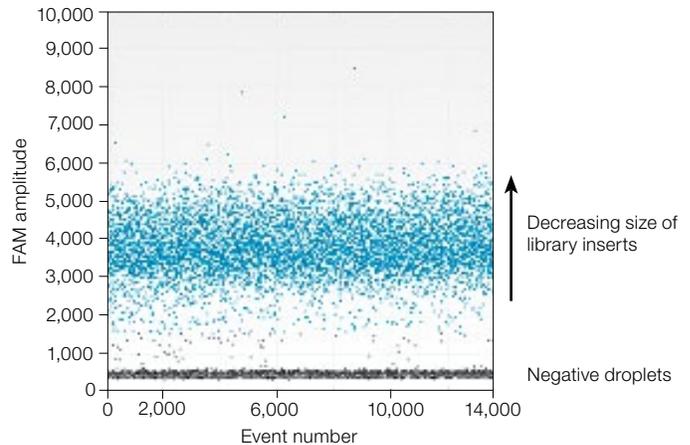


**Fig. 2. Balancing Ion AmpliSeq libraries using the ddPCR library quantification kit assay.** Eight Ion AmpliSeq libraries were quantified by ddPCR and balanced to be equimolar for the sequencing run. Excellent balancing within less than 2% difference was achieved between all eight libraries pooled in the same sequencing run (ideal percentage of total reads for each library is 12.5%).

Figures 3 and 4 are 1-D fluorescence amplitude plots showing library quality.



**Fig. 3. 1-D amplitude plot of an Ion AmpliSeq genomic DNA library demonstrating varying library insert sizes along the y-axis of the FAM fluorescence amplitude.** Larger fragments are observed near the negative droplets at the bottom of the plot. At the highest point in the plot is a population that appears separate from the bulk of the library. This population consists of adapter-adapter ligations with no inserts. Below the insert population are more rare additional adapter or multiple insert ligations. This characteristic pattern allows the quality of the library to be measured.



**Fig. 4. 1-D amplitude plot of an Ion RNA-Seq library demonstrating varying library insert sizes along the y-axis of the FAM fluorescence amplitude.**

### Ordering Information

Catalog # Description

186-3041 **ddPCR Library Quantification Kit for Ion Torrent.**  
200 x 20 µl reactions, includes droplet PCR supermix (2 x 1 ml vials), ddPCR library quantification assay (1 x 200 µl vial), for quantification of Ion Torrent libraries using the QX100 or QX200 system

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