

Double- and single-quenched probes for use in 5' nuclease assays

Dyes and quenchers for every experiment

PrimeTime qPCR Probes provide reliable sensitivity even in demanding applications such as multiplexing and digital PCR. PrimeTime qPCR Probes are available in a wide variety of dye-quencher combinations (Figure 1) that are compatible with common qPCR instruments.

Achieve consistent results

All PrimeTime Probes are HPLC purified, and then verified by mass spectrometry, to deliver batch-to-batch consistency and minimize the need for troubleshooting.

| Fluorophore* | | Emission wave- length (nm) | Quencher |
|--------------|--|-------------------------------|--------------------------------|
| | 6-FAM | 520 | |
| | TET™ | 539 | |
| | HEX [™] JOE Yakima Yellow [®] VIC ^{®‡} | 555 555 549 554 | ZEN-Iowa Black® FQ† |
| | Cy® 3 | 564 | |
| | ATTO™ 550§ NED™‡ | 575 575 | |
| | TAMRA ABY®‡ | 583 580 | Iowa Black RQ [∥] |
| | ATTO 565 [§] PET ^{®‡} | 591 595 | |
| | ROX | 608 | |
| | Texas Red®-X JUN ^{®‡} | 617 617 | |
| | ATTO 633§ LIZ®‡ | 657 655 | |
| | ATTO 647§ | 669 | |
| | Су 5 | 668 | TAO-lowa Black RQ ¹ |

ABY and JUN are registered trademarks of Life Technologies, Inc. ATTO is a trademark of ATTO-TEC GmbH. BHQ is a registered trademark of Biosearch Technologies, Inc. Cy is a registered trademark of GE Healthcare. HEX, NED, and PET are trademarks and LIZ, PET, and VIC are registered trademarks of Applied Biosystems, LLC. Texas Red is a registered trademark of Molecular Probes, Inc. Yakima Yellow is a registered trademark of Elitech Group.

www.idtdna.com

benefits

Choose from a wide range of dyes and quenchers, including several license-free combinations

Reduce costs and waste with convenient sizes, starting from 0.5 nmol

Successfully multiplex with ZEN[™] or TAO[™] Double-Quenched Probes for:

- Lower background fluorescence
- Increased endpoint signal
- Reduced crosstalk

Begin your project sooner with rapid shipment for most probes

Discover more at www.idtdna.com/qPCRprobes

Figure 1. Commonly used fluorophores and quenchers.

- * Except where noted, the fluorophores in this chart are free of licensing fees and can be ordered from www.idtdna.com/qPCRprobes.
- † Probes with 6-FAM, TET, HEX, or JOE fluorophores are also available as traditional, single-quenched probes with Black Hole Quencher®-1 (BHQ®-1, additional third-party licenses required for diagnostic use).
- ‡ For reference only. Not available through IDT.
- § Probes with ATTO Dyes can be ordered from www.idtdna.com, Custom DNA Oligos page.
- II Black Hole Quencher-2 (BHQ-2) may also be used as a quencher. However, additional third-party licenses are required for diagnostic use.
- ¶ Cy 5 is also available as a traditional, singlequenched probe with lowa Black RQ (license free) or BHQ-2 (additional third-party licenses required for diagnostic use).



Improve assay sensitivity with double-quenched probes

Reduce background and increase assay sensitivity with ZEN or TAO Double-Quenched Probes. Our exclusive internal quenchers are always 9 bases from the 5' fluorophore and work in combination with the 3' lowa Black quencher for maximum probe performance (Figure 2).

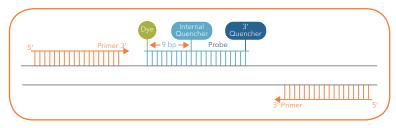
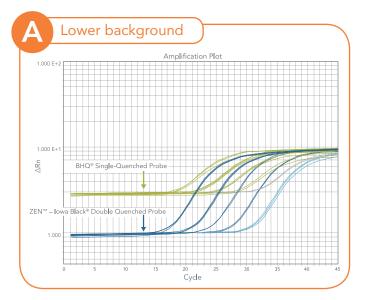


Figure 2. Schematic of a PrimeTime qPCR 5' Nuclease Assay using a doublequenched probe that includes a dye, a ZEN or TAO internal quencher, and a 3' quencher.

With nearly 4 times lower background fluorescence

(Figure 3A) and approximately 30% increased signal (Figure 3B), ZEN Double-Quenched Probes simply perform better. See performance data for TAO Double-Quenched Probes at www.idtdna.com/qPCRprobes.



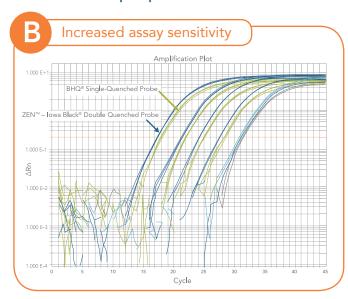


Figure 3. Increased signal detection and assay sensitivity from ZEN Double-Quenched Probes. (A) ZEN Probes (blue) provide greater dye quenching, producing lower background and, therefore, higher signal intensities than standard single-quenched probes (BHQ Probes; green). (B) ZEN Probes increase assay sensitivity, as demonstrated by the earlier C_a values observed compared to standard, BHQ single-quenched probes.

Achieve maximum quenching for long probes

Effective quenching for ZEN Double-Quenched Probes as long as 40 bases means more effective designs, even for AT-rich targets.

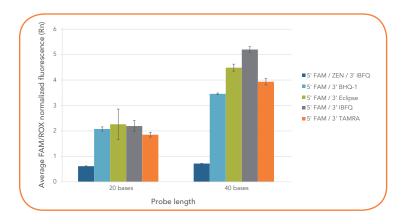


Figure 4. Only ZEN Double-Quenched Probes maintain low background signal with increasing probe length. Probes of 2 lengths (20 or 40 bases) with 5 different quenchers were compared in 10 singleplex qPCRs. Six replicate reactions with each probe type were run with 50 ng of cDNA and the TaqMan® Gene Expression Master Mix (Thermo Fisher) under standard cycling conditions on the Applied Biosystems 7900HT system. Key: IBFQ = Iowa Black FQ Quencher (IDT); BHQ-1 = Black Hole Quencher-1 (Biosearch Technologies); Eclipse® = Eclipse quencher (ELITech Group).

Ordering information

Visit www.idtdna.com/qPCRprobes to enter your sequence and choose modifications.

