

# Fluorophore and BHQ selection chart

This chart is intended to guide you through the dye selection process for your oligonucleotide. Fluorophore and quencher combinations may be designed to support applications such as probe-based qPCR, SNP genotyping, CNV, presence/absence, and more.

Fluorophore	Alternate dyes	DYE-S <sup>1</sup> - T <sub>10</sub>		Recommended quencher	BHQ™ dye quenching range
		Excitation	Emission		
<b>Biosearch Blue™</b>		352	447	BHQ-1	<b>BHQ-0</b> 430-520 nm
FAM		495	520	BHQ-1	
TET		521	536	BHQ-1	
<b>CAL Fluor™ Gold 540</b>	VIC/TET/JOE	522	544	BHQ-1	<b>BHQ-1</b> 480-580 nm
JOE		529	555	BHQ-1	
VIC®	CIV-550/CAL Fluor Orange 560	530	550	BHQ-1	
<b>CIV-550™</b>	VIC	530	550	BHQ-1	
HEX		535	556	BHQ-1	
<b>CAL Fluor Orange 560</b>	VIC/HEX/JOE/CIV-550	538	559	BHQ-1	
<b>Quasar™ 570</b>	Cy 3	548	566	BHQ-2	
Cy™3		549	566		
NED		546	575		
TAMRA		557	583	BHQ-2	
<b>CAL Fluor Red 590</b>	TAMRA	569	591	BHQ-2	<b>BHQ-2*</b> 559-670 nm
Cy 3.5		581	596		
<b>ROX™</b>		586	610	BHQ-2	
<b>CAL Fluor Red 610</b>	Texas Red/ROX/Alexa Fluor® 594	590	610	BHQ-2	
Texas Red®		597	616		
<b>CAL Fluor Red 635</b>	LC® Red 640	618	637	BHQ-2	
<b>Pulsar™ 650</b>		460	650	BHQ-2	
Cy 5		646	669		
<b>Quasar 670</b>	Cy 5	647	670	BHQ-2*, BHQ-3	
Cy 5.5		675	694		
<b>Quasar 705</b>	Cy 5.5	690	705	BHQ-2*, BHQ-3	<b>BHQ-3</b> 620-730 nm

\* BHQ-2 dye is recommended for **Quasar 670** and **Quasar 705** fluorophores due to static quenching.

○ Indicates Biosearch Technologies' proprietary dyes

Dyes in **BOLDFACE** are available modifications for labeled oligos.

Many dyes are also available in the form of nucleic acid chemistry reagents. Please visit [www.biosearchtech.com](http://www.biosearchtech.com) for current product offerings.

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# Multiplexing recommendations for BHQ Probes and primers



These dye recommendations apply to Biosearch Technologies' Black Hole Quencher™ (BHQ) Probes and may not apply to other probe formats. For tips and guidelines related to qPCR and SNP genotyping, please visit: [www.biosearchtech.com](http://www.biosearchtech.com).

Instrument*	Company	Calibration required?	Dye 1	Dye 2	Dye 3	Dye 4	Dye 5	Dye 6
MX3000P/MX4000	Agilent	No	FAM	CAL Fluor Orange 560	CAL Fluor Red 610	Quasar® 670		
Mx3005P	Agilent	Yes	FAM	CAL Fluor Orange 560	CAL Fluor Red 610	Quasar 570	Quasar 670	
qTOWER	Analytik Jena	Yes	FAM	CAL Fluor Gold 540	CAL Fluor Red 590	CAL Fluor Red 610	Quasar 670	
Mic	Bio Molecular Systems	No	FAM	CAL Fluor Orange 560	CAL Fluor Red 610	Quasar 670		
LineGene 9600 Plus	Bioer Technology	Yes	FAM	CAL Fluor Gold 540	CAL Fluor Red 610	Quasar 670	Quasar 705	
CFX384 Touch™	Bio-Rad Laboratories	Yes	FAM	CAL Fluor Orange 560	CAL Fluor Red 610	Quasar 670		
CFX96 Touch™	Bio-Rad Laboratories	Yes	FAM	CAL Fluor Gold 540	CAL Fluor Red 610	Quasar 670	Quasar 705	
iCycler iQ™	Bio-Rad Laboratories	Yes	FAM	CAL Fluor Orange 560	CAL Fluor Red 610	Quasar 670		
iQ5	Bio-Rad Laboratories	Yes	FAM	CAL Fluor Gold 540	CAL Fluor Red 590	CAL Fluor Red 610	Quasar 670	
QX200 Droplet Digital™	Bio-Rad Laboratories	No	FAM	HEX				
FLUOstar® Omega Microplate Reader	BMG Labtech	No	FAM	HEX				
SmartCycler®	Danaher Corp. (Cepheid)	Yes	FAM	CAL Fluor Orange 560	CAL Fluor Red 635			
SmartCycler II	Danaher Corp. (Cepheid)	Yes	FAM	CAL Fluor Orange 560	CAL Fluor Red 610	Quasar 670		
Mastercycler® ep realplex	Eppendorf	Yes	FAM	CAL Fluor Gold 540				
Biomark HD	Fluidigm	Yes	FAM	**CAL Fluor Gold 540				
Eco™	Illumina	Yes	FAM	CAL Fluor Orange 560	CAL Fluor Red 610	Quasar 670		
MyGo Pro	IT-IS Life Science Ltd.	No	FAM	CAL Fluor Gold 540	CAL Fluor Red 590	CAL Fluor Red 610	Quasar 670	Quasar 705
IntelliQube™	LGC, Biosearch Technologies	Yes	FAM	CAL Fluor Orange 560	TAMRA	SuperROX®	Quasar 670	
Nexar™	LGC, Biosearch Technologies	No	FAM	HEX	CAL Fluor Red 610			
Rotor-Gene Q 5plex	Qiagen	No	FAM	CAL Fluor Orange 560	CAL Fluor Red 610	Quasar 670	Quasar 705	
RainDrop Plus™	RainDance Technologies	No	FAM	***CAL Fluor Orange 560				
cobas® z 480	Roche	Yes	FAM	CAL Fluor Orange 560	CAL Fluor Red 610	Quasar 670		
LightCycler® 2.0	Roche	Yes	FAM	CAL Fluor Red 610	Pulsar 650			
LightCycler 96/LightCycler 480	Roche	Yes	FAM	CAL Fluor Orange 560	CAL Fluor Red 610	Quasar 670		
ABI PRISM® 7000/ABI PRISM 7700/ABI PRISM 7900HT	Thermo Fisher Scientific	Yes	FAM	CAL Fluor Gold 540	SuperROX®			
ABI 7500/7500 Fast	Thermo Fisher Scientific	Yes	FAM	CAL Fluor Orange 560	TAMRA	SuperROX	Quasar 670	
QuantStudio®	Thermo Fisher Scientific	Yes	FAM	CAL Fluor Orange 560	SuperROX	Quasar 670	Quasar 705	
QuantStudio 3 System	Thermo Fisher Scientific	Yes	FAM	CAL Fluor Gold 540	TAMRA	CAL Fluor Red 610		
QuantStudio 3d Digital	Thermo Fisher Scientific	Yes	FAM	CAL Fluor Orange 560	CAL Fluor Red 610			
QuantStudio 5 System/QuantStudio™ 6 Flex/QuantStudio™ 7 Flex	Thermo Fisher Scientific	Yes	FAM	CAL Fluor Gold 540	TAMRA	CAL Fluor Red 610	Quasar 670	
QuantStudio™ 12K Flex	Thermo Fisher Scientific	Yes	FAM	CAL Fluor Gold 540	TAMRA	CAL Fluor Red 610		
StepOne™	Thermo Fisher Scientific	Yes	FAM	CAL Fluor Gold 540				
StepOnePlus™	Thermo Fisher Scientific	Yes	FAM	CAL Fluor Gold 540	TAMRA			
ViiA™ 7	Thermo Fisher Scientific	Yes	FAM	CAL Fluor Orange 560	SuperROX	Quasar 670	Quasar 705	

\* Some instruments may require fluorescence calibration. Please visit our website for more information.

\*\* Fluidigm's BioMark HD System does not support other dyes at this time, however, additional dyes may be run using the following guidelines:

**Excitation wavelengths**  
between 465 and 505 nm  
between 510 and 550 nm

**Emission wavelengths**  
between 500 and 550  
between 540 and 600 nm

\*\*\* Separation not optimal

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