

Fluorescence Labeled Oligonucleotides

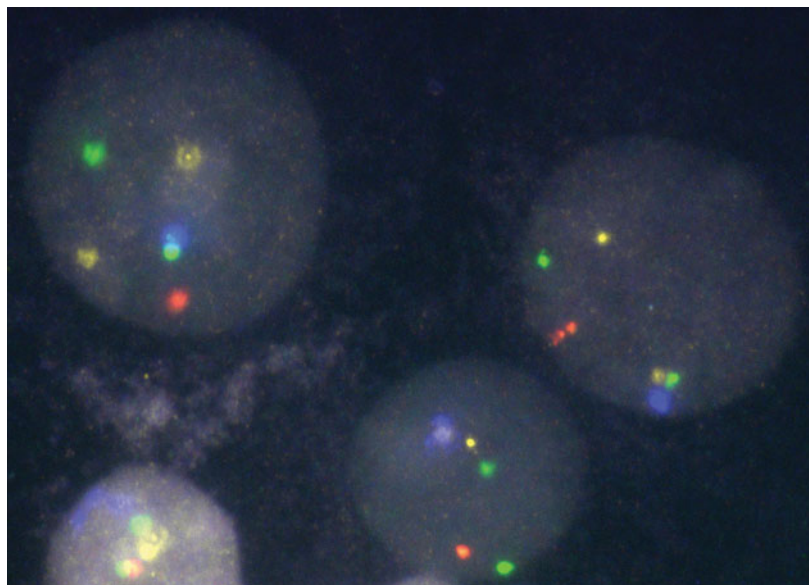
Description

Fluorescence technology is a routinely used tool in life science research. Fluorescence reagents provide the advantages of highest sensitivity and specificity paired with low toxicity. They are today one of the best choices to trace the presence of biomolecules in cells and other biological systems.

Continuous development in the field of fluorescence detection requires a high range of flexibility for dye modified biopolymers. Our portfolio (see table 2) provides a wide range of oligonucleotides single, double or even multiple labeled with classic fluorescence dyes or recently developed alternatives.

Our possibilities for fluorescence modifications allow the application of all common methods, like FISH (see figure 1), *In situ* Hybridization, sequencing and genotyping. Fluorescence and Quencher pairs are available for qualitative and quantitative detection of PCR products (qPCR/Realtime PCR). Furthermore, fluorescence modifications could be combined with non-fluorescence modifications like Phosphate, Amino, Thiol, Biotin (see table 1) or could be attached to oligonucleotides with incorporated special bases like LNA, wobbles, RNA, PTO. The flexibility for many combinations allows the adaptation to your special requirement.

Figure 1) Multicolour FISH with Dyomics Dyes



Advantages:

- High sensitivity
- High specificity
- Low toxicity
- Broad range of classic dyes
- Broad range of alternative dyes, like Dyomics, NuLight, Bodipy, Atto
- Highest combination possibilities with fluorescence dyes, quenchers or other modifications
- Combination with special bases, like LNA, wobbles, RNA, siRNA, PTO
- Probes for qPCR/RealtimePCR assays (Taqman, Molecular Beacons, FRET)
- Routinely HPLC purification and MS check

Applications:

- FISH, *In situ* Hybridization
- qPCR/Realtime PCR
- FRET (Fluorescence Resonance Energy Transfer)
- Sequencing
- Genotyping
- Microarray Studies (see figure 2)

Coupling of fluorescence dyes – the techniques:

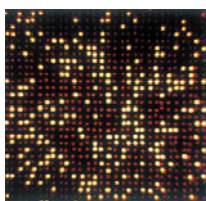
In principle there are two main methods for coupling in chemical synthesis:

1. Modification during solid phase chemical synthesis:

Automatic coupling of modified phosphoramidites (PA) or starting with modified controlled pore glasses (CPG) during solid phase synthesis on the synthesizer.

- 5' modifications that are available as phosphoramidites, i.e. Fam, Hex, Tet, CyDyes, Yakima Yellow
- 3' modifications that are available as pre-modified solid support material (CPG = controlled pore glass) and that are stable enough to pass the synthesis chemistry, i.e. Fam, Tamra, BHQ, Dabcyl, Eclipse Quencher
- all internal modification that are available as modified phosphoramidite bases, i.e. Fluorescein-dT, Tamra-dT, Dabcyl-dT

Figure 2) Microarray using fluorescence Dyes



2. Post-synthetic modification:

Manual labeling of an activated dye with amino or thiol groups of the oligonucleotide.

Modifications available as activated esters (NHS ester) are coupled with amino groups whereas thiol reactive dye reagents, like maleimides, are coupled with thiol groups. In the first production step, an amino- or thiol-modified oligonucleotide is synthesised. After the oligonucleotide has been cleaved from the solid support, the chemical reaction between linking group and activated dye is carried out.

- 5' modifications that are not available as pre-modified phosphoramidite but as activated esters, i.e. Tamra, Joe, Rox, Bodipy, Molecular Probes dyes, Dyomics dyes
- 3' modifications that are not available as pre-modified solid support material (CPG = controlled pore glass), but as activated esters, i.e. Joe, Rox, Bodipy, Molecular Probes dyes, Dyomics dyes
- internal modifications that are not available as a pre-designed dye-bases (i.e. dye-dT) can be coupled as activated esters to internal amino groups. In general, this internal amino group is inserted via Amino-dT (C6)-phosphoramidite during automatic synthesis.

Possible combinations:

Please note that combinations marked in blue are not available!

Table 1)

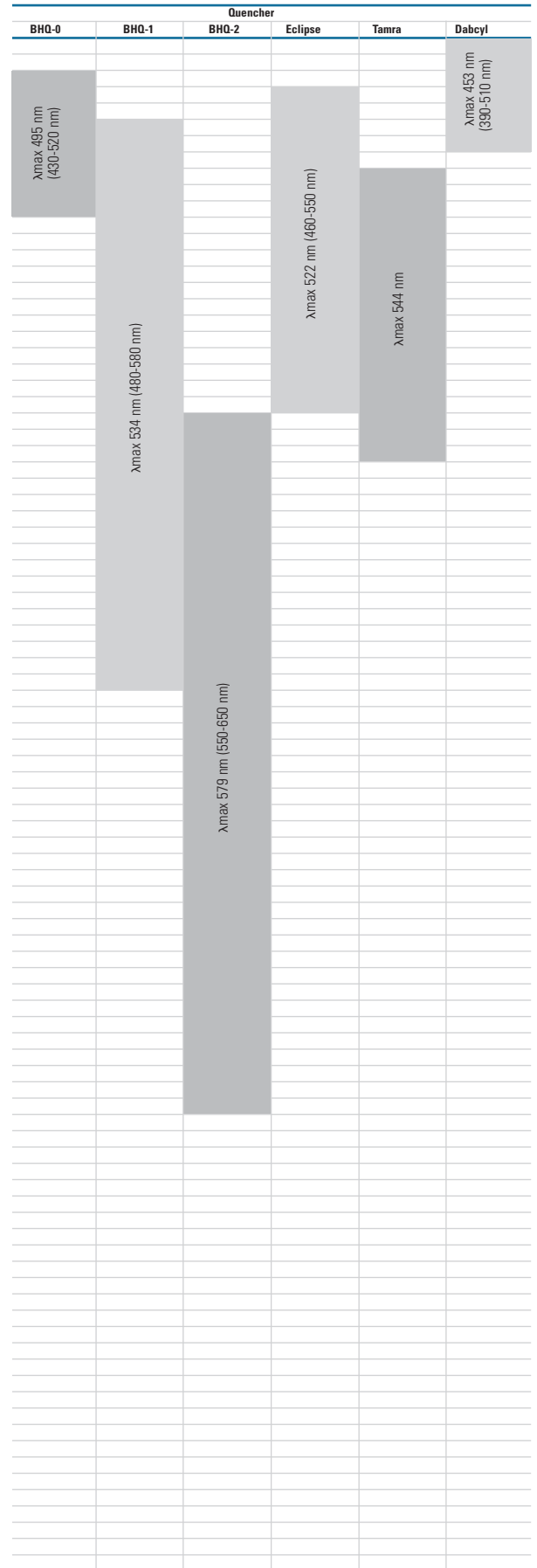
		3'-terminal																					
5'-terminal	Amino C6	Thiol C3	Phosphate	Biotin	Digoxigenin	Dabcyl	BHQ 1 or 2	Inverted end	3'-Block (C3 Spacer)	Fluorescein	6-FAM	TAMRA	JOE	ROX	Cy TM dyes	AMCA	Bodipy TM dyes	Marina Blue	Pacific Blue	Oregon Greens	Rhodamine Green	Rhodamine Red	Texas Red
Amino C6	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•
Thiol C6	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•
Phosphate	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•
Biotin	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•
BHQ 1 or 2	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•
Digoxigenin	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•
Fluorescein	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•
6-FAM	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•
HEX	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•
TET	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•
TAMRA	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•
JOE	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•
ROX	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•
Cy TM dyes	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•
AMCA	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•
Bodipy TM dyes	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•
Marina Blue	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•
Pacific Blue	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•
Oregon Greens	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•
Rhodamine Green	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•
Rhodamine Red	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•
Texas Red	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•
HRP	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•	•

Please inquire for further combinations

Fluorescence Dyes and Quenchers:

Table 2)

"Fluorescence dye"	Absorption maximum [nm]	Emission maximum [nm]	Extinction coefficient [M-1 cm-1]	Emission area
Cascade Blue	400	420	28.000	
AMCA353	422	19.000		
Pacific Blue	416	451	36.000	
Marina Blue	385	460	20.000	
DY-415	418	467	34.000	
Bodipy 493/503	493	504	89.000	
Bodipy FL	504	513	68.000	
Oregon Green 500	499	519	78.000	
6-Fam	495	520	83.000	
Fluorescein	495	520	83.000	
DY-495	495	520	70.000	
ATTO 488	501	523	90.000	
Oregon Green 488	496	524	70.000	
Rhodamine Green	505	527	68.000	
Rhodamine Red	505	527	120.000	
Oregon Green 514	511	530	70.000	
DY-505	505	530	80.000	
Tet	521	536	73.000	
ATTO 520	516	538	110.000	
Bodipy 507/545	508	543	69.000	
Bodipy R6G	528	547	70.000	
Joe	527	548	71.000	
Yakima Yellow	531	549	84.000	
Bodipy 530/550	535	552	62.000	
ATTO 532	532	553	115.000	
Hex	535	556	73.000	
Bodipy 558/588	560	568	74.000	
Bodipy 564/570	563	569	142.000	
Cy3	550	570	150.000	
DY-548	558	572	150.000	
DY-554	551	572	100.000	
DY-555	547	572	100.000	
DY-556	548	573	100.000	
Bodipy TMR	535	574	50.000	
DY-547	557	574	150.000	
NuLight Dy547	557	574	150.000	
DY-549	560	575	150.000	
Tamra	544	576	90.000	
ATTO 550	554	576	120.000	
DY-560	559	578	120.000	
Bodipy 576/589	575	588	83.000	
Bodipy 581/591	581	591	136.000	
ATTO 565	563	592	120.000	
Redmond Red	579	595	52.300	
DY-590	580	599	120.000	
Rox	576	601	82.000	
Texas Red	583	603	116.000	
Cy3.5	588	604	116.000	
Bodipy TR	588	617	45.000	
ATTO 590	594	624	120.000	
DY-610	610	630	80.000	
Bodipy 630/650	632	640	100.000	
DY-615	621	641	200.000	
DY-630	636	657	200.000	
DY-632	637	657	200.000	
DY-633	637	657	200.000	
DY-631	637	658	200.000	
DY-634	635	658	200.000	
Bodipy 650/665	651	660	100.000	
ATTO 647	645	669	120.000	
Cy5	650	670	250.000	
DY-635	647	671	200.000	
DY-636	645	671	200.000	
NuLight Dy647	653	672	250.000	
DY-647	653	672	250.000	
DY-648	653	674	250.000	
DY-650	653	674	220.000	
DY-652	654	675	220.000	
DY-649	655	676	250.000	
DY-651	656	678	220.000	
ATTO 655	663	684	125.000	
Cy5.5	675	694	250.000	
DY-677	673	694	180.000	
DY-675	674	699	180.000	
DY-676	674	699	180.000	
ATTO 680	680	700	125.000	
DY-681	691	708	140.000	
DY-680	690	709	140.000	
DY-682	690	709	140.000	
ATTO 700	700	719	120.000	
DY-700	707	730	140.000	
DY-701	706	731	140.000	
DY-730	732	758	240.000	
DY-732	736	759	240.000	
DY-731	736	760	240.000	
DY-734	734	766	240.000	
Cy7	743	767	200.000	
DY-752	748	772	270.000	
DY-750	747	776	270.000	
DY-751	751	779	270.000	
DY-780	782	800	170.000	
DY-782	783	800	170.000	
DY-776	771	801	240.000	
DY-831	844	-	220.000	



Product offering

Most of our fluorescence dyes are available in 4 standard scales. Prices are indicated in our actual standard price list (www.thermofisher.com/biopolymers)

Classic Fluorescence Dyes

	available at	Synthesis scale			
		0.02 µmol	0.04 µmol	0.2 µmol	1.0 µmol
6-Fam, Hex, Joe, Rox	3' / 5'	•	•	•	•
Tet, Tamra	5'	•	•	•	•
Cy Dyes ¹⁾	3' / 5'	•	•	•	•
Dabcyl ¹⁾	3' / 5'	•	•	•	•
BHQ 1, 2 ¹⁾	3' / 5'	•	•	•	•
Eclipse Dark Quencher ¹⁾	3' / 5'		please inquire		

¹⁾ Please refer to our license information

Alternative Fluorescence Dyes

	available at	Synthesis scale			
		0.02 µmol	0.04 µmol	0.2 µmol	1.0 µmol
BODIPY dyes ¹⁾	3' / 5'	•	•	•	•
AMCA ¹⁾ , Marina Blue ¹⁾ , Pacific Blue ¹⁾ , Texas Red ¹⁾ , Oregon Green ¹⁾ , Rhodamine Red ¹⁾ , Rhodamine Green ¹⁾	3' / 5'	•	•	•	•
Atto Dyes	3' / 5'		please inquire		
Yakima Yellow ¹⁾ , Redmond Red ¹⁾	3' / 5'		please inquire		
Dyomics Dyes	3' / 5'	•	•	•	•
NuLight™ Dy547 ¹⁾ , NuLight™ Dy647 ¹⁾	3' / 5'	•	•	•	•

¹⁾ Please refer to our license information

Yields and length restrictions

Type	Length (bases)	Purification	Minimum OD / scale			
			0.02 µmol	0.04 µmol	0.2 µmol	1.0 µmol
Single modification	16-40	HPLC	1,0	2,0	5,0	15,0
		PAGE or IEC	0,3	0,5	1,5	5,0
	8-15 or 41-100 (3' end max 70)	HPLC	0,7	1,5	3,0	12,0
		PAGE or IEC	0,2	0,3	1,0	4,0
Double modification	16-40	HPLC	0,5	1,0	2,5	7,5
		PAGE or IEC	0,2	0,3	0,5	2,5
	8-15 or 41-100 (3' end max 70)	HPLC	0,3	0,7	2,0	6,0
		PAGE or IEC	0,2	0,2	0,3	2,0

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We can certainly help.

Licences information:

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- Molecular Probes' dyes are licensed from Invitrogen
- Yakima Yellow, Redmond Red and EclipseTMDark Qu. are made and sold under license from EPOCH Biosciences, Inc.
- NuLightTMDYes are sold under the license of Thermo Fisher Scientific
- Dyomics Dyes are sold under the license of Dyomics GmbH

In addition to these offices, Thermo Fisher Scientific maintains a network of representative organizations throughout the world.

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