

DNA oligonucleotides

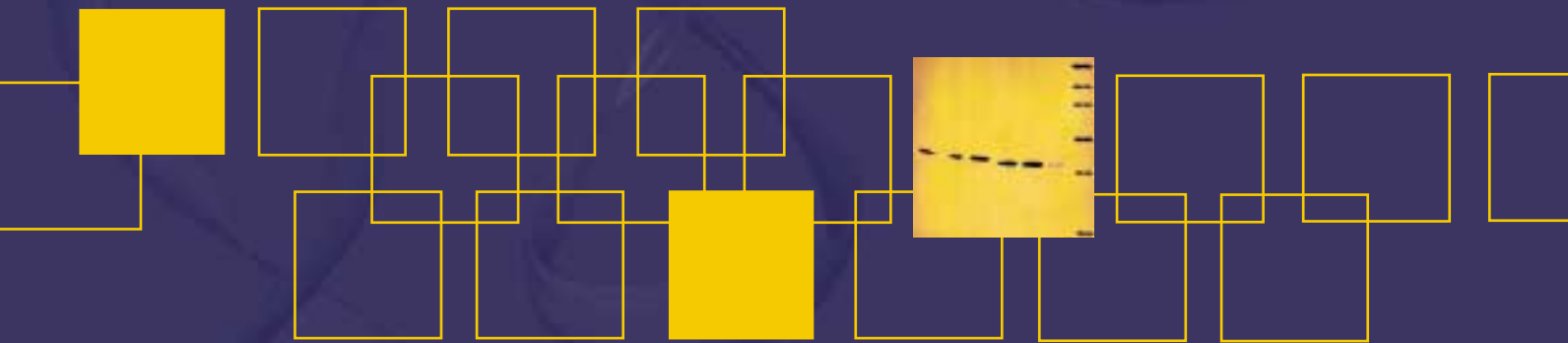
Reporter groups on oligonucleotides

Chimeric oligonucleotides (RNA/DNA hybrids)

RNA oligonucleotides

Purification of oligonucleotides

# Nucleic acid specialties



Oligonucleotides for antisense technology

Reporter groups on oligonucleotides

Modified oligonucleotides

DNA oligonucleotides

Modified nucleoside-triphosphates

Chimeric oligonucleotides (RNA/DNA hybrids)

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## General information

Being a German leader in nucleic acid technology IBA has more than 10 years experience in DNA and RNA synthesis.

We are not only synthesizing standard DNA/RNA oligonucleotides but are also specialized in production and purification of modified nucleosides and oligonucleotides (DNA and RNA). Base, ribose and backbone modifications and reporter groups like dyes and haptens are routinely incorporated into DNA and RNA. The in-house nucleoside and nucleotide chemistry laboratory continuously develops new modifications which are not available elsewhere. Please contact us if the modification you are looking for is not listed in the catalog.

## Quality of products

IBA delivers only highly purified products. We make no compromise in our efforts to reach reproducible highest coupling yields. Also we have developed highly sophisticated purification processes to meet the individual needs of each oligonucleotide and modification.

The following points are strictly followed during synthesis of oligonucleotides:

- Synthesizers from leading manufacturers
- High quality chemicals from selected suppliers
- High concentrations of building blocks
- In-house developed and optimized synthesis cycles
- Prolonged coupling steps optimized for certain building blocks
- Purification of oligonucleotides either by HPLC and/or PAGE
- Two-step purification process for fluorescent probes
- Exact yield determination by measuring UV-absorption and adjustment of the concentration
- Routine use of our products for in-house applications like cDNA synthesis, PCR, *in vitro* transcription, DNA sequencing and gene synthesis

## Service

IBA offers free technical assistance provided by nucleic acid chemists and molecular biologists. Take advantage of more than ten years experience in the field of nucleoside-, nucleotide- and oligonucleotide synthesis and application.

### Hotline for oligonucleotides

Tel. +49 (0) 551 50672-140

Fax +49 (0) 551 50672-183

E-mail [oligo@iba-go.com](mailto:oligo@iba-go.com)

You can reach us from 8 am - 10 pm (CET)

For our local customers in Göttingen we offer a free-of-charge delivery service directly to your lab!

# DNA oligonucleotides

## Standard DNA oligonucleotides

- All prices include a Molecular biology grade purification (page 77)
- Yields are exactly determined by measuring UV-absorption at 260 nm
- Oligonucleotides are provided in a final concentration of 100 pmol/ $\mu$ l
- Oligonucleotides shorter than 8 bases are only lyophilized and not desalted (for desalting DS-grade purification is essential, see page 77)

For more information on available purification grades see pages 76-78.

Standard DNA oligonucleotides	scale	delivery amount [OD]**	cat. no.
2- 35 bases	0.01 $\mu$ mol	0.5 -1	5-0110-011
2- 50 bases	0.05 $\mu$ mol	2.0 - 4	5-0110-012
2-100 bases	0.20 $\mu$ mol	10.0-20	5-0110-013
2-100 bases	1.00 $\mu$ mol	50.0-100	5-0110-014
2- 40 bases	15.00 $\mu$ mol	600.0-1200	5-0110-015

\*\* For a 20mer with uniform base composition

## Set up for 3'- or special wobbles

Standard wobbles (wobbles are equimolar mixtures of 2 to 4 bases) are automatically coupled on the synthesizers without additional charge. For 3'-wobbles or non-equimolar mixtures (special wobbles) the building blocks will be exactly pre-mixed at the requested ratio. An additional set-up will be charged for each different mixture.

product	cat. no.
set-up for 3'- or special wobbles	5-0110-020

## Long range DNA oligonucleotides

The synthesis of premium quality oligonucleotides or long oligonucleotides (100-200 bases) is performed with special cycles and high concentrations of building blocks. In addition IBA uses a special solid support for the synthesis of oligonucleotides that are longer than 100 bases. However, a minimum yield cannot be guaranteed. IBA strongly recommends purification of such products by HPLC or preferably by PAGE (see pages 77, 78).

product	synthesis scale	delivery amount [OD]	cat. no.
DNA oligonucleotide	0.20 $\mu$ mol	depends on sequence and length	5-0120-013
>100 bases	1.00 $\mu$ mol		5-0120-014

## Large scale oligonucleotide synthesis

We offer a large scale synthesis service for oligonucleotides up to 1 mmol. Please inquire for detailed information.

## Ready-to-use oligos!

## Nucleotide ambiguity code

Code	Represents	Complement
A	A	T
C	C	G
G	G	C
T	T	A
M	A or C	K
R	A or G	Y
W	A or T	W
S	C or G	S
Y	C or T	R
K	G or T	M
V	A or C or G	B
H	A or C or T	D
D	A or G or T	H
B	C or G or T	V
X/N	A or C or G or T	X/N
•	not A, C, G or T	-

## Purification of oligonucleotides

Modern oligonucleotide synthesis via phosphoramidites is a complex multi-step procedure. One elongation cycle consists of the following steps:

- Deprotection of the 5'-hydroxy group
- Coupling of the appropriate building block
- Capping of unreacted 5'-hydroxy groups
- Oxidation of the phosphitriester group to phosphatetriester

Even if all steps work optimally, a significant amount of incomplete sequences, called trityl-off failure sequences, will accumulate. The amount of these failure sequences depends on the chain length. The figure shows the amount of full length product depending on oligonucleotide length and three different coupling efficiencies (98.0%, 98.5%, 99.0%).

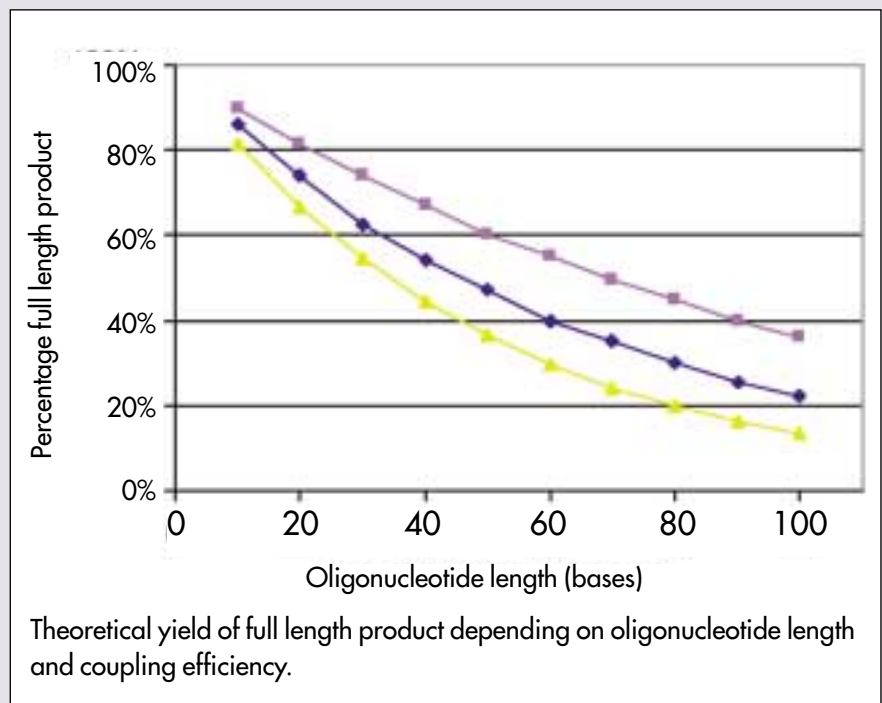
As the oligonucleotide length increases, the amount of full length product decreases. Furthermore, in cases where chain length is greater than 50 bases the growing chains interact with one another, causing the flow of chemicals through the column to slow down.

Trityl-on failure sequences will be present caused by inefficient detritylation, capping, oxidation or by depurination resulting in post synthesis strand scissure during ammonia deprotection.

In addition to trityl-off and trityl-on failure sequences, the base protecting groups in the form of their corresponding amides will be present within the crude reaction mixture. To fulfill the demand of purity for different applications IBA offers a broad spectrum of standard and special purification methods.

- Coupling efficiency 99%
- ◆ Coupling efficiency 98.5%
- ▲ Coupling efficiency 98%

**We recommend HPLC purification for oligonucleotides above 30-40 bases**



## Molecular biology grade purification

Salts, base protecting groups and short failure sequences up to 6 bases are removed efficiently. The amount of full-length product depends on chain length and coupling yield.

A typical 20mer with an average coupling yield of 98.5 % is composed to 70 % of full-length product. This purification is recommended for oligonucleotides with a chain length up to 30 bases for standard molecular biology applications such as PCR, sequencing, etc.

product	synthesis scale	amounts [OD]*
Molecular biology grade purification	0.01 µmol	0.5-1.0
	0.05 µmol	2.0-4.0
	0.20 µmol	10.0-20.0
	1.00 µmol	50.0-100.0
	15.00 µmol	600.0-1200.0

\* delivery amounts for a 20mer with an average base composition of 5xA, 5xG, 5xC and 5xT

## HPLC grade purification

Apart from salts and protecting groups, the trityl-on HPLC step efficiently removes trityl-off failure sequences. The purity of the product depends on the nature and length of the sequence. IBA recommends this purification for all applications where full-length or high reproducibility are essential, including: cloning, PCR, mutagenesis, probes, modified oligonucleotides and oligonucleotides longer than 40 bases. This method is not able to remove trityl-on failure sequences.

Double HPLC purification available as well

product	synthesis scale	delivery amount	cat. no.
HPLC grade purification	0.01 µmol	1 - 2 nmol	5-1020-001
	0.05 µmol	5 - 15 nmol	5-1020-002
	0.20 µmol	25 - 50 nmol	5-1020-003
	1.00 µmol	100 - 200 nmol	5-1020-004
	15.00 µmol	1500 - 3000 nmol	5-1020-005

## RNA HPLC grade purification and RNA purification

For certain applications RNA HPLC grade purification may be sufficient (but generally PAGE purification is recommended; see page 78).

product	synthesis scale	cat. no.
RNA HPLC grade purification	1 µmol	5-1021-004
RNA double HPLC	1 µmol	5-1024-004
RNA cell culture grade	1 µmol	5-1023-004
RNA desalting	1 µmol	5-1041-004

## DS (desalting) grade purification

This method was developed especially for ultra-short oligonucleotides with a chain length between 1 and 7 bases. It consists of an in-house developed 2 x HPLC purification and a desalting step.

product	synthesis scale	delivery amount	cat. no.
DS grade purification	0.05 µmol	1 -3 nmol	5-1040-002
	0.20 µmol	5 -15 nmol	5-1040-003
	1.00 µmol	25 -80 nmol	5-1040-004
	15.00 µmol	500 -1000 nmol	5-1040-005

## Cell culture grade purification

This unique in-house development is a multi-step purification method for all oligonucleotides used in cell culture. It consists of two HPLC purification steps and one sterile filtration step.

product	synthesis scale	delivery amount	cat. no.
Cell culture grade purification	0.05 µmol	1 -3 nmol	5-1050-002
	0.20 µmol	5 -15 nmol	5-1050-003
	1.00 µmol	25 -80 nmol	5-1050-004
	15.0 µmol	500 -1000 nmol	5-1050-005

### PAGE grade/FCS grade purification

PAGE is the most powerful method completely removing all types of failure sequences and intercalating dyes like TAMRA. Due to the lower recovery rate IBA can not guarantee delivery amounts.

Recommended for the following products:

- RNA
- Oligonucleotides for biophysical applications
- FCS-Spectroscopy

product	cat. no.
PAGE purification	5-1060-001

### Reverse phase C18 HPLC purification

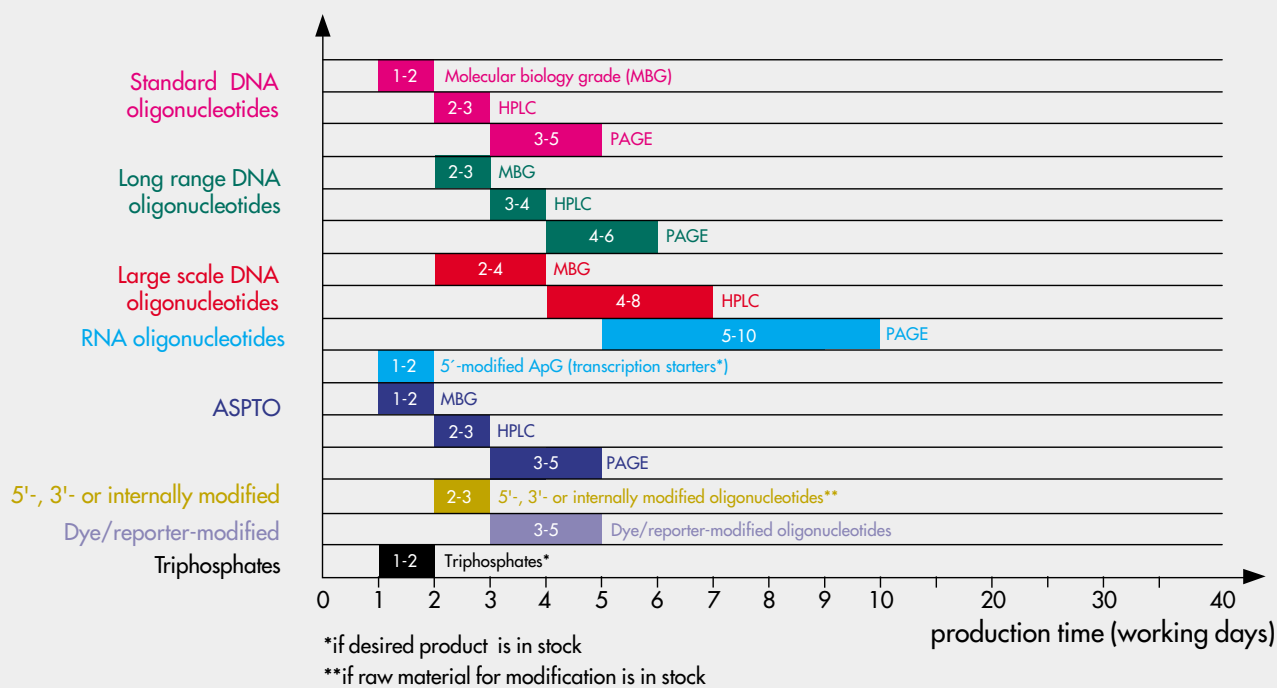
For oligonucleotides requiring more than standard reverse phase purification, such as double dye labeled oligonucleotides.

product	synthesis scale	cat. no.
Reverse phase C18 HPLC purification	1 µmol	5-1022-004

### Documentation

product	cat. no.
PAGE documentation	5-1070-000
HPLC documentation	5-1070-010

### Guidelines for production times of oligonucleotides



## Chimeric oligonucleotides (RNA/DNA hybrids)

Several new applications have recently been described in the literature involving chimeric oligonucleotides (e.g. 1-4). Responding to the increasing demand of our customers we are offering a wide variety of combinations (e.g. DNA/RNA, RNA/2'-O-Me-RNA, 2'-F-RNA/DNA). Our extensive range of fluorescent and non-fluorescent markers (see pages 91-106) can of course be incorporated as well.

### Features

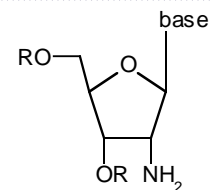
- Chemically synthesized
- Specific labeling with fluorescent markers and other non-radioactive reporter groups
- Easy introduction of modified bases
- Wide variety of combinations available

### Possible combinations

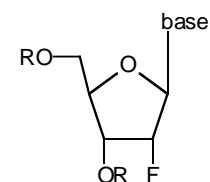
	DNA	RNA	2'-O-Me-RNA	2'-F-RNA	2'-NH <sub>2</sub> -RNA
DNA		x	x	x	x
RNA	x		x	x	x
2'-O-Me-RNA	x	x		x	x
2'-F-RNA	x	x	x		x
2'-NH <sub>2</sub> -RNA	x	x	x	x	

product	scale	cat. no.
DNA	1.00 µmol	5-0110-014
RNA	1.00 µmol	5-0510-014
2'-NH <sub>2</sub> -RNA	1.00 µmol	5-0511-014
2'-F-RNA	1.00 µmol	5-0512-014
2'-O-Me-RNA	1.00 µmol	5-0513-014
HPLC grade purification	1.00 µmol	5-1020-004
PAGE purification	Any scale	5-1060-001

2'-NH<sub>2</sub>-RNA, 2'-F-RNA and 2'-O-Me-RNA are only available with HPLC grade purification or PAGE purification. Further scales available.

2'-NH<sub>2</sub>-RNA

2'-F-RNA

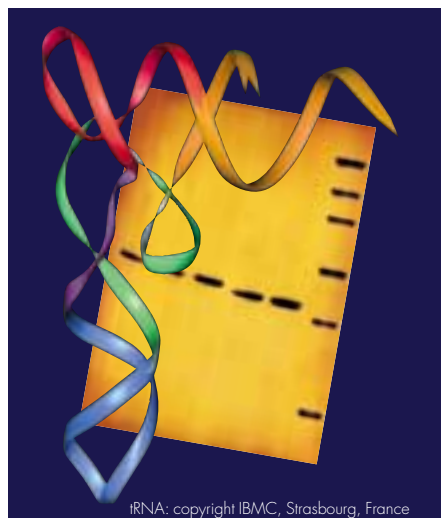


For quotations for chimeric oligonucleotides please contact our nucleic acid department at [oligo@iba-go.com](mailto:oligo@iba-go.com).

### References:

- 1 Wu XS, Liu DP, Liang CC, 2001: J Biomed Sci, Nov/Dec;8(6):439-45.
- 2 Liu L, Rice MC, Kmiec EB, 2001: Nucleic Acids Res Oct 15;29(20):4238-50.
- 3 Kmiec EB, Johnson C, May GD, 2001: Plant J Aug;27(3):267-74.
- 4 Gamper HB, Parekh H, Rice MC, Bruner M, Youkey H, Kmiec EB, 2000: Nucleic Acids Res, Nov 1;28(21):4332-9.

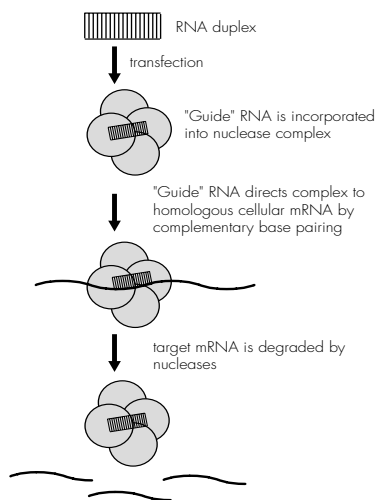
For reference titles see pages 110 ff. or [www.iba-go.com](http://www.iba-go.com).



## References:

- 1 Amarzguioui M, Prydz H, 1998: Cell Mol Life Sci 54:1175-1202.
- 2 Famulok M, Jenne A, 1998: Curr Opin Chem Biol 2:320-327.
- 3 Vaish NK, Kore AR, Eckstein F, 1998: Nucleic Acids Res 26:5237-5242.
- 4 Putz J, Florentz C, Benseler F, Giege R, 1994: Nat Struct Biol 1:580-582.
- 5 Felden B, Florentz C, Westhof E, Giege R, 1993: Biochimie 75:1143-1157.
- 6 Famulok M, Mayer G, 1999: Curr Top Microbiol Immunol 243:123-136.

## Gene Silencing



## References:

- 1 Hunter T, Hunt T, Jackson RJ, Robertson HD, 1975: J Biol Chem Jan 25;250(2):409-17.
- 2 Clemens MJ, Safer B, Merrick WC, Anderson WF, London IM, 1975: Proc Natl Acad Sci U S A Apr;72(4):1286-90.
- 3 Caplen NJ, Parrish S, Imani F, Fire A, Morgan AR, 2001: Proc Natl Acad Sci U S A. 2001, Aug.14;98(17):9742-7.
- 4 Hamilton AJ, Baulcombe DC, 1999: Science 1999 Oct 29;286(5441):950-2.
- 5 Dijkeng A, Shi H, Tschudi C, Ullu E, 2001: RNA 2001, Nov;7(11):1522-30
- 6 Sharp PA, 1999: Genes & Development 13:139-141.

For reference titles see pages 110 ff. or [www.iba-go.com](http://www.iba-go.com).

## RNA oligonucleotides

### Take advantage of more than 10 years experience in RNA synthesis!

Already in 1991, IBA's nucleic acid division (at that time called "NAPS") has established RNA synthesis. In the following years new synthesis methods were developed to include long RNA, labeled RNA, 2'- and base-modified RNA, triphosphates, modified dinucleotides for 5' transcript labeling as well as double stranded RNA (dsRNA).

Our expertise enables us to provide high quality products with excellent purity and high biological activity.

### Some of the most important applications for RNA are

- dsRNA for gene silencing (RNA interference, inhibition of protein synthesis) (see below)
- Chimeric DNA/RNA for targeted gene repair in gene therapy (see page 79)
- Ribozymes for specific modulation of gene expression (1)
- Ribozymes for diagnostics (2)
- Investigation of ribozyme-substrate interactions (3)
- Investigation of tRNA function (4)
- RNA footprinting for the study of RNA-protein interactions (5)
- Aptamers for specific inhibition of protein function (6)
- Synthesis of RNA for *in vitro* translation experiments

IBA offers different methods for synthesis of RNA:

- Chemical synthesis
- *In vitro* transcription

Modified triphosphates for *in vitro* transcription see pages 107-110.

### Double stranded RNA for RNA interference studies

Since 1975 it is known that double stranded RNA (dsRNA) can inhibit protein synthesis (1,2). Several recent applications described the use of short dsRNA oligonucleotides for gene silencing, i.e. the inhibition of protein synthesis (3-5).

Being a specialist for RNA synthesis we are offering the RNA tools you require for these studies.

Short RNA duplexes are offered ready-to-use, i.e. PAGE purified, deprotected and annealed. A negative control sense and antisense RNA are included. There are no additional charges for the number of base pairs.

For longer RNA duplexes the costs are based on a price per base pair plus a fixed fee for annealing and purification.

product	synthesis scale	delivery amount	cat. no.
Ready-to-use RNA duplexes, PAGE purified, deprotected and annealed, plus negative control		50-100 nmol dsRNA; 10-20 nmol ssRNA as control	5-0515-114
dsRNA (long)	1.0 µmol		5-0516-014
dsRNA annealing & purification	1.0 µmol	50-100 nmol	5-1060-004

dsRNA is only available in combination with dsRNA annealing and purification.

## Standard RNA oligonucleotides

- High biological activity
- Up to 50-70 bases depending on sequence and scale
- Easy introduction of modified bases
- Synthesis of larger quantities possible
- Specific labeling with fluorescent markers and other non-radioactive reporter groups
- Totally deprotected, HPLC and PAGE analyzed products
- Chemically synthesized

product	synthesis scale	cat. no.
Standard RNA	1.0/15.0 µmol	5-0510-014/5
RNA set-up for oligonucleotides containing less than 5 couplings		5-0510-000

The delivered amount depends on sequence and length. IBA strongly recommends PAGE purification, see page 78.

## Modified RNA oligonucleotides

### 2'-Amino-modified RNA oligonucleotides (2'-Amino-RNA)

- Stabilization against nucleases
- 2'-Amino-rU and 2'-Amino-rC available
- Number of couplings are limited due to poor coupling efficiency

product	synthesis scale	cat. no.
2'-Amino-RNA	1.0 µmol	5-0511-014

### 2'-Fluoro-modified RNA oligonucleotides (2'-F-RNA)

- Stabilization against nucleases
- Increase of melting temperature by 2 °C per residue
- 2'-F-RNA/RNA duplexes are no substrates for RNase H
- Only 2'-F-rU and 2'-F-rC available

product	synthesis scale	cat. no.
2'-F-RNA	1.0 / 15.0 µmol	5-0512-014/5

### 2'-O-Methyl-modified RNA oligonucleotides (2'-O-Me-RNA)

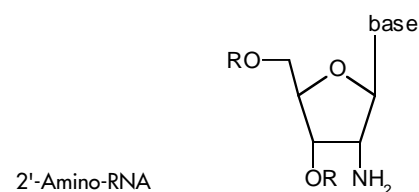
- Provides stabilization against nucleases
- Increases melting temperature by 1.5 °C per residue
- 2'-O-Methyl-RNA/RNA hybrids are not recognized by RNase H
- Important for antisense applications

product	synthesis scale	cat. no.
2'-O-Me-RNA	0.05/0.20/1.00/15.00 µmol	5-0513-012/3/4/5
0.05 and 0.2 µmol scale not available in combination with RNA.		

For purification of oligonucleotides see pages 76 - 78.

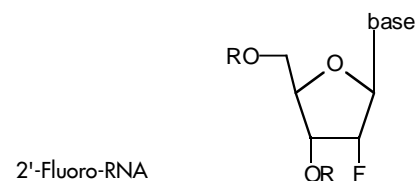
References listing IBA as supplier:

- 1 Kurreck J, Wyszko E, Gillen C, Erdmann VA, 2002: Nucleic Acid Res 30, 9: 1911-1918.
- 2 Zabrorowska Z, Fürste PJ, Erdmann VA, Kurreck J, 2002: J Biol Chem 277, 43: 40617-40622.
- 3 Kurreck J, Bieber B, Jahn R, Erdmann VA, 2001: J Biol Chem 277, 9: 7099-7107.



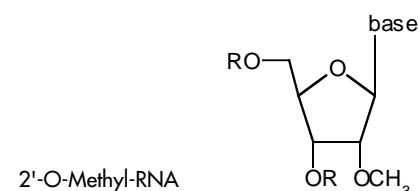
Reference:

- Heidenreich O, Benseler F, Fahrenholz A, Eckstein F, 1994: J Biol Chem 269:2131-2138.



References:

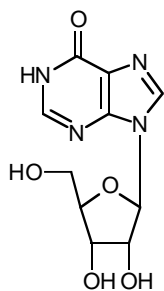
- 1 Pieken WA, Olson DB, Benseler F, Aurrup H, Eckstein F, 1991: Science 253:314-317.
- 2 Heidenreich O, Eckstein F, 1992: J Biol Chem 267:1904-1909.



Reference:

- 1 Inoue H, Hayase Y, Imura A, Iwai S, Miura K, Ohtsuka E, 1987: Nucleic Acids Res 15:6131-6148.
- 2 Majlessi M, Nelson NC, Becker MM, 1998: Nucleic Acids Research 26, 2224-2229.
- 3 Lamond AI, Sproat BS, 1993: FEBS Lett Jun 28; 325(1-2): 123-7.

For reference titles see pages 110 ff. or [www.iba-go.com](http://www.iba-go.com).



Ribo-inosine

Reference:  
Ludwig J, Blaschke M, Sproat BS, 1998:  
Nucleic Acids Res 26:2279-2285.

### Inosine modified RNA oligonucleotides (In-RNA)

- Special application in ribozyme technology
- Changes ribozyme cleavage specificity from NUH to NCH

product	synthesis scale	cat. no.
In-RNA	1.0/15.0 µmol	5-0514-014/5

All DNA modifications (see pages 84 - 90) can also be introduced in RNA. Please contact us if the modification you are looking for is not listed in the catalog.

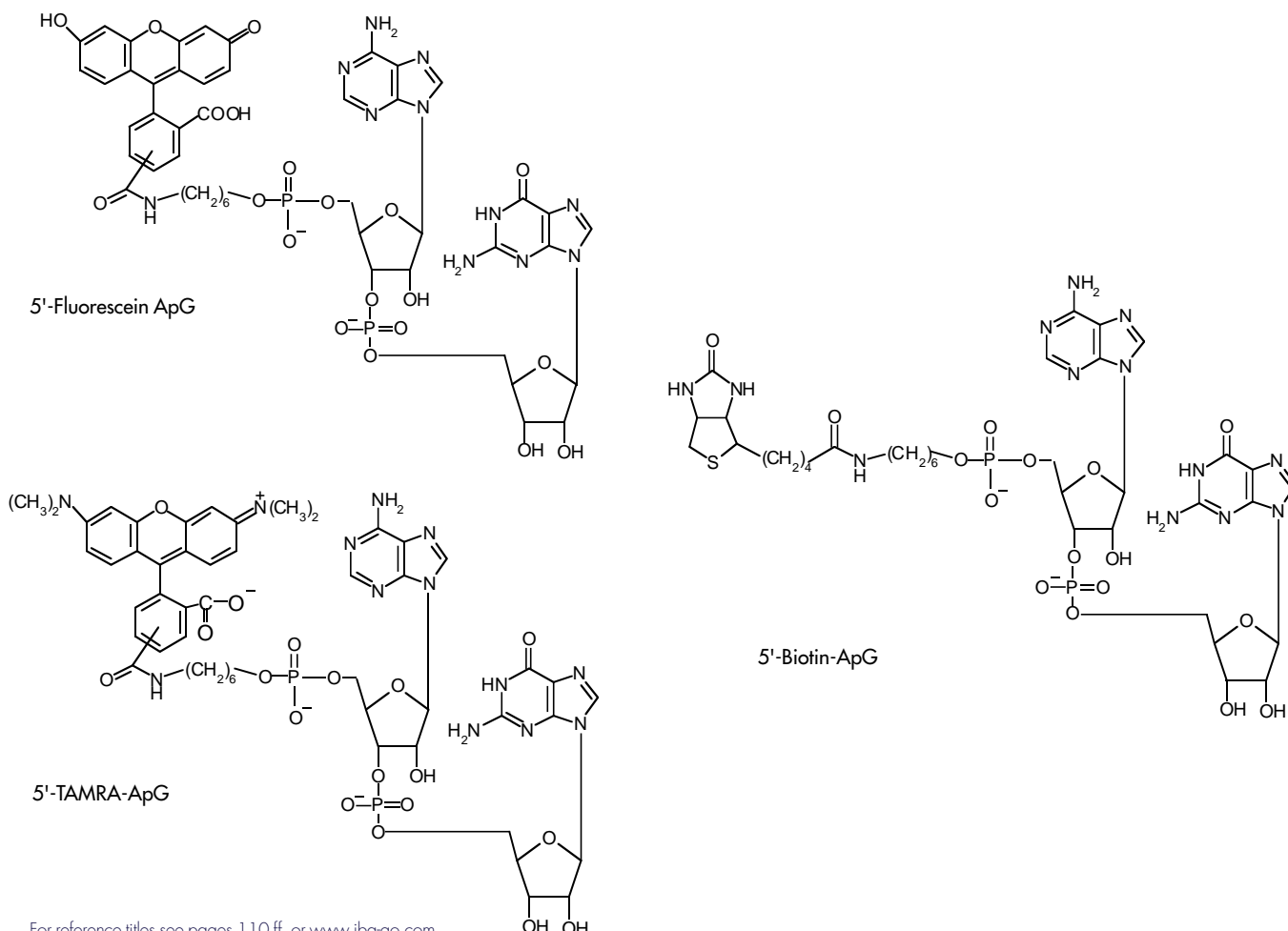
### Transcription starters for 5'-labeled transcripts

5'-Modified ApG can be used in *in vitro* transcription experiments. It is incorporated only once at the 5'-end. By using this compound it is easy to either introduce fluorescent markers to make transcripts visible, or non-radioactive markers like biotin or digoxigenin to isolate transcription products. In principle all of the available dyes and non-radioactive reporter groups can be introduced. Please inquire for more information.

product	delivery amount	cat. no.
5'-Biotin-ApG	0.5/1.0/5.0/10.0 µmol	5-0700-003/4/6/7
5'-Fluorescein-ApG	0.5/1.0/5.0/10.0 µmol	5-0700-103/4/6/7
5'-TAMRA-ApG	0.5/1.0/5.0/10.0 µmol	5-0700-203/4/6/7

Reference:  
Pitulle C, Kleinedam RG, Sproat B and Krupp G,  
1992: Gene 112: 101-105.

Other dinucleotides on request. For further dyes see page 91.



For reference titles see pages 110 ff. or [www.iba-go.com](http://www.iba-go.com).

## Oligonucleotides for antisense technology

Antisense technology is the use of oligonucleotides to inhibit gene expression through sequence specific hybridization. This process has gained increasing acceptance in the study of gene function and may also lead to new therapies for many human diseases.

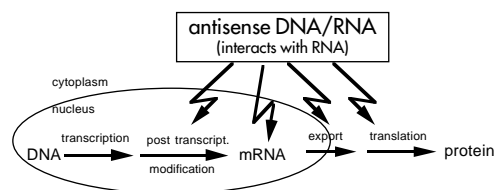
Inhibition of gene expression by antisense oligonucleotides is based on the ability of an oligonucleotide to bind a complementary messenger RNA sequence thus preventing translation of the messenger RNA. Several mechanisms have been shown to mediate the inhibitory functions of oligonucleotides in gene expression. The most important seems to be the physical inhibition of the translation on ribosomes and the destruction of mRNA in RNA/DNA-duplexes by RNase H. An ideal antisense oligonucleotide should bind mRNA specifically and with high affinity. Furthermore, it should be able to penetrate cell membranes and have sufficient resistance to extra- and intracellular nucleases. In these terms, the most successfully used antisense oligonucleotides are:

- Phosphorothioates (see below)
- 2'- Modified oligoribonucleotides (see page 81)
- Double stranded RNA (see page 80)

### Phosphorothioate modified oligonucleotides

Phosphorothioate analogs in which a non-bridging oxygen has been replaced by sulfur have widespread use in molecular biology. The increased resistance of phosphorothioates against nuclease digestion has prompted the consideration of these molecules for medical purposes<sup>1-3</sup>. In addition to their value as antisense probes, phosphorothioates have also been used for the site specific attachment of reporter groups at the DNA/RNA backbone<sup>4</sup> and for preventing degradation of oligonucleotides by 3' → 5'-exonuclease activities of proof reading DNA polymerases such as Vent and Pfu<sup>5</sup>. Phosphorothioate-substituted RNA has been used to study specific RNA-Protein interactions<sup>6</sup>.

product	synthesis scale	delivery amount [OD]	cat. no.
	0.01 µmol	0.5 -1	5-0410-011
Antisense phosphorothioate coupling of	0.05 µmol	2 -4	5-0410-012
DNA oligonucleotides	0.20 µmol	10 -20	5-0410-013
	1.00 µmol	50 - 100	5-0410-014
	15.00 µmol	600 -1200	5-0410-015
Antisense phosphorothioate coupling of	1.00 µmol	50 -100	5-0411-014
RNA oligonucleotides			
Set-up for orders containing less than 5 ASPTO couplings			5-0410-010



Targets for antisense strategies.

Reference:

Stein CA, Cohen JS, 1988: Cancer Res 48:2659-68.

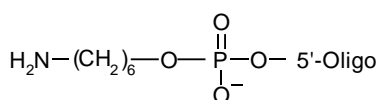
References:

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- 3 Stein CA, Tonkinson JL, Yakubov L, 1991: Pharmacol Ther 52:365-84.
- 4 Conway NE, Fidanza J, McLaughlin LW, 1989: Nucleic Acids Symp Ser 21:43-44.
- 5 Skerra A, 1992: Nucleic Acids Res 20:3551-3554.
- 6 Dertinger D, Behlen LS, Uhlenbeck OC, 2000: Biochemistry 39:55-63.

For reference titles see pages 110 ff. or [www.iba-go.com](http://www.iba-go.com).

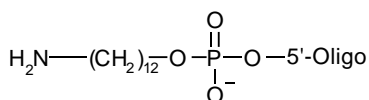
## Modifications available in RNA and DNA

## Price includes HPLC purification

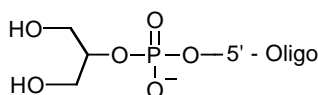


5'-Amino-C6

Reference:  
Agrawal S, Christodoulou C, Gait MJ, 1986:  
Nucleic Acids Res 14:6227-6245.



5'-Amino-C12



5'-Branching

Reference:  
Horn T, Urdea MS, 1989:  
Nucleic Acids Res 17:6959-6967.

## Modified oligonucleotides

IBA offers a wide variety of modified DNA oligonucleotides. The modifications are separated into three groups, depending on the position of the modification introduced:

- 5'-modifications:** Non-nucleosidic compounds such as amino- or thiolinker, phosphate
- 3'-modifications:** Non-nucleosidic compounds such as amino-linker, phosphate
- Internal modifications:** All nucleosidic compounds, even if they are introduced at the 3'- or 5'-end

Oligonucleotides that are modified with dyes and/or other non-radioactive reporter groups are described in the chapter "Reporter groups on oligonucleotides" (see page 91).

Backbone and 2'-sugar-modified oligonucleotides are listed and explained in the chapters "RNA oligonucleotides" (page 80) and "Oligonucleotides for antisense technology" (page 83).

### 5'-Modified oligonucleotides

All modified DNA oligonucleotides are HPLC purified at no additional charge. All modifications can also be introduced in RNA.

#### 5'-Amino-C6

- Primary amino group attached to a 6-carbon spacer
- Useful for the attachment of fluorescent reporter groups, biotin, digoxigenin, and enzymes or antibodies
- Used for immobilization of oligonucleotides on solid supports

product	synthesis scale	delivery amount	cat. no.
5'-Amino-C6	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0210-112/3/4

#### 5'-Amino-C12

- Primary amino group attached to a 12-carbon spacer
- Greater spacing between oligonucleotide and reporter groups
- Can reduce intramolecular quenching of fluorescence
- Useful for affinity chromatography

product	synthesis scale	delivery amount	cat. no.
5'-Amino-C12	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0210-212/3/4

#### 5'-Symmetric Branching (5'-Branch)

- Each coupling doubles the amount of sites used for coupling with modifications

product	synthesis scale	delivery amount	cat. no.
5'-Branch	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0210-222/3/4

For reference titles see pages 110 ff. or [www.iba-go.com](http://www.iba-go.com).

**5'-Cholesteryl (5'-Chol)**

- Useful in antisense technology
- The highly lipophilic compound facilitates membrane penetration

product	synthesis scale	delivery amount	cat. no.
5'-Chol	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0210-312/3/4

**5'-Hexaethylenglycol (5'-HEGL)**

- Hydrophilic spacer
- Can also be introduced at internal sites
- In hammerhead ribozymes it can replace the tetra-loop of stem II with essentially no decrease in activity compared with the wild-type ribozyme

product	synthesis scale	delivery amount	cat. no.
5'-HEGL	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0210-412/3/4

**5'-Phosphate (5'-Phos)**

- For ligation of PCR products
- Very high degree of phosphorylation (>95%)
- Superior to enzymatic methods

product	synthesis scale	delivery amount	cat. no.
5'-Phos	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0210-512/3/4

**5'-Psoralen-C2 (5'-PsorC2)**

- For cross-linking double stranded oligonucleotides

product	synthesis scale	delivery amount	cat. no.
5'-PsorC2	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0210-612/3/4

**5'-Psoralen-C6 (5'-PsorC6)**

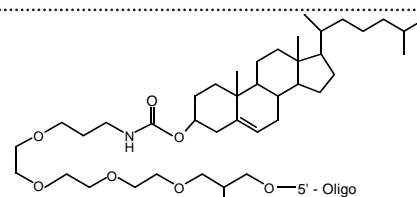
- For cross-linking triple helices

product	synthesis scale	delivery amount	cat. no.
5'-PsorC6	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0210-712/3/4

**5'-Thiol-C6 (5'-SH)**

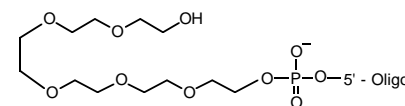
- For coupling oligonucleotides with maleimide or iodoacetate activated antibodies, peptides, enzymes or fluorescent dyes
- Useful for double labelings with different dyes (5'-thiol in combination with internal- or 3'-amino-groups)

product	synthesis scale	delivery amount	cat. no.
5'-SH	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0210-812/3/4

**5'-Cholesteryl**

Reference:

Epa VWR, Rong P, Bartlett PF, Coulson EJ, Barrett GL, 1998: Antisense Nucleic Acid Drug Dev 8:489-498.

**5'-Hexaethylenglycol**

Reference:

Benseler F, Fu DJ, Ludwig J, McLaughlin LW, 1993: J Am Chem Soc 115: 8483-8484.

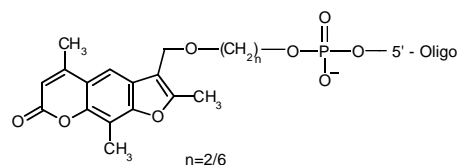
Reference:

Guzaev A, Salo H, Azhayew A, Lönnberg H, 1995: Tetrahedron 51: 9375-9384.

**NEW: Psoralen also available internally!**

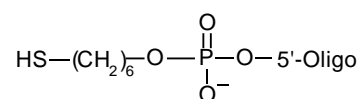
Reference:

Pielas U, Englisch U, 1989: Nucleic Acids Res 17:285-299.

**5'-Psoralen C2/C6**

Reference:

Raha M, Lacroix L, Glazer PM, 1998: Photochem Photobiol 67:289-294.

**5'-Thiol-C6**

Reference:

Connolly BA and Rider P, 1985: Nucleic Acids Res 13:4485-502.

For reference titles see pages 110 ff. or www.iba-go.com.

## Reference:

Benseler F, Fu D, Ludwig J, Mc Laughlin LW, 1993:  
J An Chem Soc 115:8483-8484.

**Price includes  
HPLC purification**

**5'-Triethylenglycol (5'-TriGl)**

- Hydrophilic spacer
- Can also be coupled at internal sites

product	synthesis scale	delivery amount	cat. no.
5'-TriGl	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0210-912/3/4

**3'-Modified oligonucleotides**

All modified DNA oligonucleotides are HPLC purified at no additional charge.  
All modifications can also be introduced in RNA.

**3'-Amino**

- Useful for synthesis of oligonucleotides with multiple reporter groups and markers (see page 92) when modifications are introduced at the 5'- and 3'-end
- No extension by DNA polymerases during PCR

product	synthesis scale	delivery amount	cat. no.
3'-Amino	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0220-012/3/4

**3'-Cholesteryl (3'-Chol)**

- Used in antisense technology
- The highly lipophilic compound mediates the penetration of oligonucleotides through the cell membrane

product	synthesis scale	delivery amount	cat. no.
3'-Chol	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0220-112/3/4

**3'-Phosphate (3'-Phos)**

- Blocks the extension of the 3'-end by polymerases (see also Real-time PCR probes page 93)
- 3'-phosphorylated oligonucleotides cannot be ligated by ligases

product	synthesis scale	delivery amount	cat. no.
3'-Phos	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0220-212/3/4

## Reference:

Corrias MV, Guarnaccia F, Ponzoni MJ, 1997:  
Neurooncol 31:171-180.

## Reference:

Markiewicz WT, Wyrzykiewicz TK, 1989:  
Nucleic Acids Res 17:7149-7158.

## Oligonucleotides with internal modifications

All modified DNA oligonucleotides are HPLC purified at no additional charge (except for dI and dU).

All modifications can also be introduced in RNA.

### Modified adenosines

#### 2-Amino-2'-deoxyadenosine (2-Amino-2'-dA)

- Increases melting temperature by 3°C per 2-Amino-2'-dA/dT base pair

product	synthesis scale	delivery amount	cat. no.
2-Amino-2'-dA	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0230-012/3/4

#### 3'-Deoxyadenosine, Cordycepin (3'-dA)

- For blocking the 3'-end of oligonucleotides from extension or ligation

product	synthesis scale	delivery amount	cat. no.
3'-dA	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0230-212/3/4

#### 7-Deaza-2'-deoxyadenosine (7-Deaza-2'-dA)

- Incorporation of 7-Deaza-2'-dA can reduce secondary structures

product	synthesis scale	delivery amount	cat. no.
7-Deaza-2'-dA	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0230-312/3/4

#### 8-Bromo-2'-deoxyadenosine (8-Br-2'-dA)

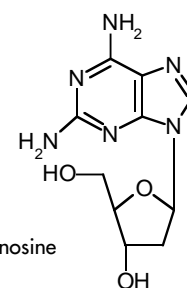
- Halogenated nucleosides are useful for crystallographic studies
- Useful for probing the structure of Protein-RNA, Protein-DNA and DNA-RNA complexes in crosslinking experiments

product	synthesis scale	delivery amount	cat. no.
8-Br-2'-dA	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0230-412/3/4

#### N6-Methyl-2'-deoxyadenosine (N6-Methyl-2'-dA)

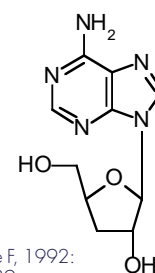
- For studies on methylation effects
- For blocking of restriction enzymes

product	synthesis scale	delivery amount	cat. no.
N6-Me-2'-dA	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0230-512/3/4



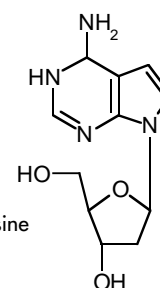
2-Amino-2'-deoxyadenosine

Reference:  
Hoheisel JD, Leirach H, 1990:  
FEBS Lett 274:103-106.



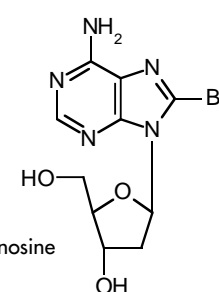
3'-Deoxyadenosine

Reference:  
Austermann S, Kruhoffer M, Grosse F, 1992:  
Biochem Pharmacol 43:2581-2589.

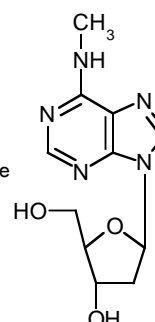


7-Deaza-2'-deoxyadenosine

Reference:  
Jensen MA, Zagursky RJ, Trainor GL, Cocuzza AJ,  
Lee A, Chen EY, 1991: DNA Seq 1:233-239.



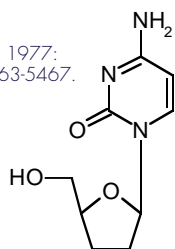
8-Bromo-2'-deoxyadenosine



N6-Methyl-2'-deoxyadenosine

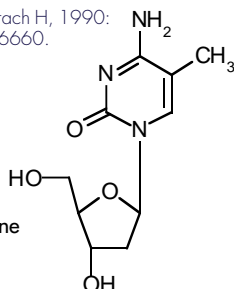
For reference titles see pages 110 ff. or [www.iba-go.com](http://www.iba-go.com).

Reference:

Sanger F, Nicklen S, Coulson AR, 1977:  
Proc Natl Acad Sci U S A 74:5463-5467.

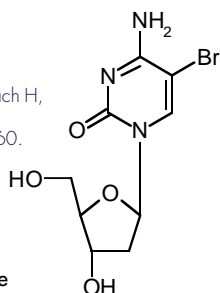
2',3'-Dideoxycytidine

Reference:

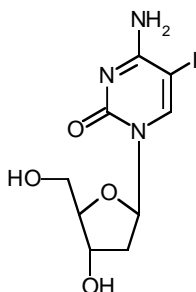
Hoheisel JD, Craig AG, Lehrach H, 1990:  
J Biol Chem 265:16656-16660.

5-Methyl-2'-deoxycytidine

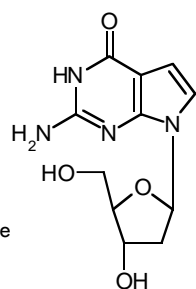
Reference:

Hoheisel JD, Craig AG, Lehrach H,  
1990:  
J Biol Chem 26:16656-16660.

5-Bromo-2'-deoxycytidine

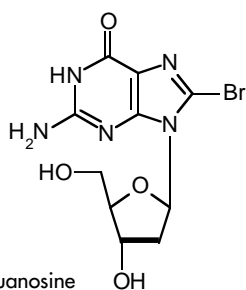


5-Iodo-2'-deoxycytidine



7-Deaza-2'-deoxyguanosine

Reference:

Fernandez-Rachubinski F, Eng B, Murray VVV,  
Blajchman MA, Rachubinski RA, 1990:  
DNA Seq 1:137-140.

8-Bromo-2'-deoxyguanosine

For reference titles see pages 110 ff. or [www.iba-go.com](http://www.iba-go.com).

## Modified cytidines

### 2',3'-Dideoxycytidine (2',3'-ddC)

- Useful for blocking the extension of oligonucleotides e.g. during PCR
- No substrate for RNA- and DNA ligases

product	synthesis scale	delivery amount	cat. no.
2' 3'-ddC	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0231-012/3/4

### 5-Methyl-2'-deoxycytidine (5-Me-2'-dC)

- Increases melting temperature by 1.3°C per 5-Methyl-2'-dC/dG base pair

product	synthesis scale	delivery amount	cat. no.
5-Me-2'-dC	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0231-112/3/4

### 5-Bromo-2'-deoxycytidine (5-Br-2'-dC)

- Halogenated nucleosides are useful for crystallography
- Useful for probing the structure of Protein-RNA, Protein-DNA and DNA-RNA complexes in crosslinking experiments

product	synthesis scale	delivery amount	cat. no.
5-Br-2'-dC	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0231-212/3/4

### 5-Iodo-2'-deoxycytidine (5-I-2'-dC)

- Halogenated nucleosides are useful for crystallography
- Useful for probing the structure of Protein-RNA, Protein-DNA and RNA-RNA complexes in crosslinking experiments

product	synthesis scale	delivery amount	cat. no.
5-I-2'-dC	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0231-312/3/4

## Modified guanosines

### 7-Deaza-2'-deoxyguanosine (7-Deaza-2'-dG)

- Incorporation of 7-Deaza-2'-dG can reduce secondary structures
- Incorporation of 7-Deaza-2'-dG prevents formation of G-tetrads

product	synthesis scale	delivery amount	cat. no.
7-Deaza-2'-dG	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0232-012/3/4

### 8-Bromo-2'-deoxyguanosine (8-Br-2'-dG)

- Halogenated nucleosides are useful for crystallography
- Useful for probing the structure of Protein-RNA, Protein-DNA and DNA-RNA complexes in crosslinking experiments

product	synthesis scale	delivery amount	cat. no.
8-Br-2'-dG	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0232-112/3/4

## Modified thymidines

### 4-Thio-2'-deoxythymidine (4-Thio-dT)

- Useful in cross-linking and affinity-labeling experiments

product	synthesis scale	delivery amount	cat. no.
4-Thio-dT	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0233-012/3/4

### 5-C3-Carboxy-2'-deoxythymidine (5-C3-Carboxy-dT)

- Useful for introduction of molecules with a primary amine such as antibodies and enzymes by standard coupling procedures

product	synthesis scale	delivery amount	cat. no.
5-C3-Carboxy-dT	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0233-112/3/4

### 5-C6-Amino-2'-deoxythymidine (5-C6-Amino-dT)

- Incorporation of internal reporter groups
- Does not interfere with nucleic acid hybridization
- Unique ability to provide functional amino groups at internal sites

product	synthesis scale	delivery amount	cat. no.
5-C6-Amino-dT	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0233-212/3/4

### Inverse-2'-deoxythymidine (Inverse-dT)

- For blocking oligonucleotides from extension or ligation
- Stabilization against 3'-exonucleases

product	synthesis scale	delivery amount	cat. no.
Inverse-dT	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0233-312/3/4

## Other modifications

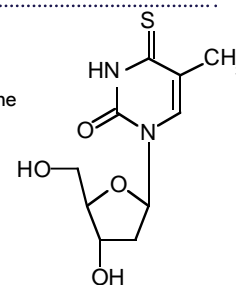
### 2'-Deoxyinosine (2'-di)

- Can form base pairs with A, G, C and T, although the hydrogen bonding to the four bases is low and unequal
- Useful when parts of a target sequence are unknown (e.g. degenerate sequence deduced from peptide sequence) or different mutants of a gene must be targeted

product	synthesis scale	delivery amount	cat. no.
2'-di	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0234-012/3/4

A discount of 50 % for each 2'-di modification is granted if more than 5 modifications are requested in one order, e.g. six 2'-di in one sequence or one 2'-di in six different sequences. HPLC purification is not included, but highly recommended.

4-Thio-2'-deoxythymidine



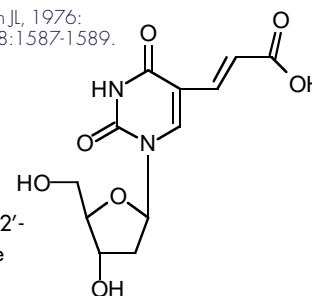
References:

- Warren MA, Murray JB, Connolly BA, 1998: *J Mol Biol* 279:89-100.
- McGregor A, Rao MV, Duckworth G, Stockley PG, Connolly BA, 1996: *Nucleic Acids Res* 24:3173-3180.
- Nikiforov TT, Connolly BA, 1992: *Nucleic Acids Res* 20: 1209-1214.

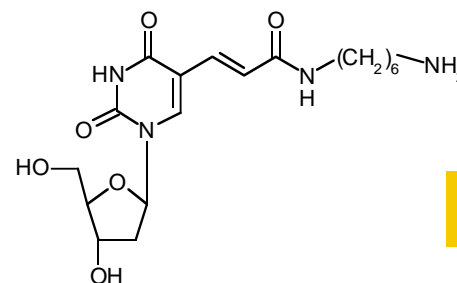
Reference:

- Bergstrom DE, Ruth JL, 1976: *J Am Chem Soc* 98:1587-1589.

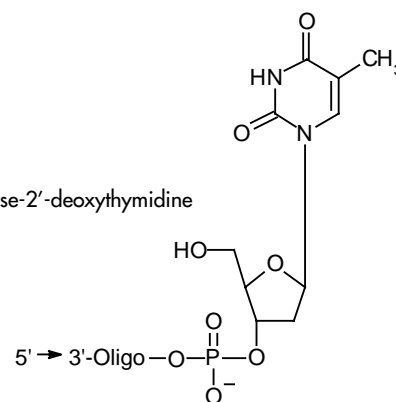
5-C3-Carboxy-2'-deoxythymidine



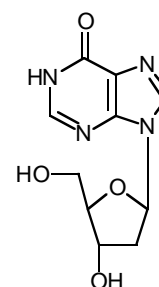
5-C6-Amino-2'-deoxythymidine



Inverse-2'-deoxythymidine



2'-Deoxyinosine

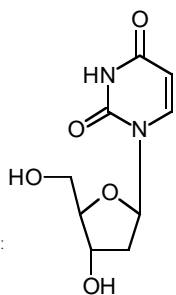


Reference:

- Martin FH, Castro MM, Aboul-ela F, Tinoco I Jr, 1985: *Nucleic Acids Res* 13:8927-8938.

For reference titles see pages 110 ff. or [www.iba-go.com](http://www.iba-go.com).

2'-Deoxyuridine



References:

- 1 Watson DE, Bennett GN, 1997: Biotechniques 23:858-862.
- 2 Germann MW, Kalisch BW, van de Sande JH, 1996: J Biomol Struct Dyn 13:953-962.

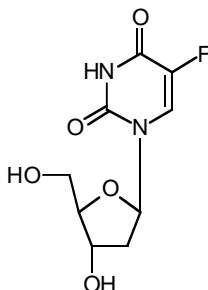
2'-Deoxyuridine (2'-dU)

- Replacement of thymidine with 2'-deoxyuridine results in destabilization of a DNA duplex
- Is used in combination with Uracil-DNA glycosylase to avoid cross-contamination of PCR products
- Useful for cloning PCR products (in combination with Uracil-DNA glycosylase)

product	synthesis scale	delivery amount	cat. no.
2'-dU	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0235-012/3/4

A discount of 50 % for each 2'-dU modification is granted if more than 5 modifications are requested in one order, e.g. six 2'-dU in one sequence or one 2'-dU in six sequences. HPLC purification is not included, but highly recommended.

5-Fluoro-2'-deoxyuridine

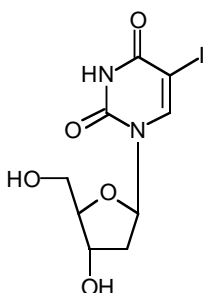


5-Fluoro-2'-deoxyuridine (5-F-2'-dU)

- Halogenated nucleosides are useful for crystallography

product	synthesis scale	delivery amount	cat. no.
5-F-2'-dU	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0235-112/3/4

5-Iodo-2'-deoxyuridine

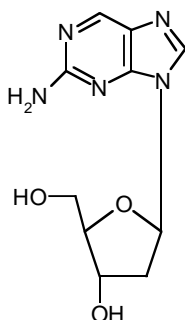


5-Iodo-2'-deoxyuridine (5-Iodo-2'-dU)

- Halogenated nucleosides are useful for crystallography

product	synthesis scale	delivery amount	cat. no.
5-Iodo-2'-dU	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0235-212/3/4

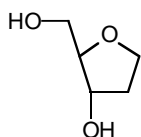
2-Aminopurine



2-Aminopurine and Deoxypurine

product	synthesis scale	delivery amount	cat. no.
2-AminoPurine	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0230-602/3/4
Deoxypurine	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0230-702/3/4

Stable abasic site



Stable abasic site

- One base spacer for oligonucleotides

product	synthesis scale	delivery amount	cat. no.
Stable abasic site	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0230-612/3/4

# Reporter groups on oligonucleotides

## Fluorescent oligonucleotides

IBA is not only a licensed supplier of a broad spectrum of probes labeled with dyes from Molecular Probes or Amersham Pharmacia Biotech, we now also offer an interesting selection of new dyes from Evotec OAI, Dyomics and Atto-Tec. The range of these new dyes includes e.g. Dy 782 which can be excited with infrared light allowing many new applications.

All probes are double HPLC purified\*. Failure sequences are removed during the first HPLC; the second HPLC is performed to remove unlabeled oligonucleotides and excessive dye. As a result only labeled oligonucleotides of excellent quality are obtained.

\*with the exception of the directly coupled dyes where no excessive dye must be eliminated

## We offer

- High quality: double HPLC purification yields labeled oligonucleotides only
- A new selection of dyes including the infrared excitable Dy 782
- Very competitive prices

## Dyes available for oligonucleotide labeling

product	absorption	emission	$\epsilon$ [cm <sup>2</sup> M <sup>-1</sup> ]	page
Pyrene	340 nm	376 nm	43000	102
Dansyl-X	340 nm	520 nm	4200	98
AMCA-X	353 nm	442 nm	19000	94
Hydroxy coumarin (HYCO)	419 nm	447 nm	36000	100
Atto 425	426 nm	470 nm	40000	95
D-AMCA	432 nm	472 nm	56000	98
Dabcyl	453 nm	none	32000	98
Cy2 <sup>TM</sup>	489 nm	506 nm	150000	97
Fluorescein-5-EX	491 nm	515 nm	86000	100
Oregon Green <sup>TM</sup> 488, 6-isomer	492 nm	517 nm	88000	101
FAM, 5-isomer	494 nm	520 nm	78000	100
Oregon Green <sup>TM</sup> 488, 5-isomer	495 nm	521 nm	76000	101
FAM, 6-isomer	496 nm	516 nm	83000	100
FAM Phosphoramidite, mixed isomers	496 nm	516 nm	83000	100
Rhodol Green <sup>TM</sup> , mixed isomers	496 nm	523 nm	63000	103
Oregon Green <sup>TM</sup> 500	499 nm	519 nm	78000	101
BODIPY <sup>®</sup> 493/503	500 nm	509 nm	79000	95/96
BODIPY <sup>®</sup> FL	502 nm	510 nm	82000	95/96
RhodamineGreen <sup>TM</sup> -X, mixed isomers	503 nm	528 nm	74000	103
Rhodamine Green <sup>TM</sup> , mixed isomers	504 nm	532 nm	78000	102
Oregon Green <sup>TM</sup> 514	506 nm	526 nm	85000	101
TET	519 nm	539 nm	**	104
JOE, 6-isomer	520 nm	548 nm	71000	101
Carboxy-rhodamine 6G, 6-isomer	524 nm	550 nm	102000	96
Carboxy-rhodamine 6G, 5-isomer	524 nm	557 nm	108000	96
Atto 520	525 nm	547 nm	105000	95
BODIPY <sup>®</sup> R6G	528 nm	547 nm	70000	95/96
BODIPY <sup>®</sup> 530/550	534 nm	551 nm	77000	95/96
HEX	537 nm	556 nm	**	100
BODIPY <sup>®</sup> TMR-X	544 nm	570 nm	56000	95/96
TAMRA, 5-isomer	546 nm	579 nm	91000	104
TAMRA, 6-isomer	547 nm	573 nm	91000	104
Cy3 <sup>TM</sup>	550 nm	570 nm	150000	97
Oyster 556	556 nm	570 nm	155000	102
BODIPY <sup>®</sup> 558/568	559 nm	568 nm	97000	95/96
Rhodamine Red <sup>TM</sup> -X, 5-isomer	560 nm	580 nm	129000	103
BODIPY <sup>®</sup> 564/570-X	563 nm	569 nm	142000	95/96
Atto 565	566 nm	590 nm	120000	95
Carboxy-X-rhodamine, 5-isomer	574 nm	602 nm	78000	97
BODIPY <sup>®</sup> 576/589	575 nm	588 nm	83000	95/96
Carboxy-X-rhodamine, 6-isomer	575 nm	602 nm	82000	97
Cy3.5 <sup>TM</sup>	581 nm	596 nm	150000	97
BODIPY <sup>®</sup> 581/591-X	582 nm	591 nm	154000	95/96
Texas Red <sup>®</sup> -X, mixed isomers	583 nm	603 nm	116000	104
BODIPY <sup>®</sup> TR-X	588 nm	616 nm	68000	95/96
Atto 590	598 nm	634 nm	120000	95
Atto 610	616 nm	646 nm	150000	95
BODIPY <sup>®</sup> 630/650	625 nm	640 nm	101000	95/96
EVObblue <sup>TM</sup> 30	630 nm	670 nm	100000	99
Dy 630	630 nm	655 nm	120000	99
Dy 635	634 nm	664 nm	120000	99
Oyster 645	645 nm	666 nm	250000	102
BODIPY <sup>®</sup> 650/665	646 nm	660 nm	102000	95/96
Cy5 <sup>TM</sup>	649 nm	670 nm	250000	98
Atto 655	655 nm	680 nm	125000	95
Oyster 656	656 nm	674 nm	220000	102
Methylene Blue	665 nm	690 nm	93000	on request
Atto 680	675 nm	699 nm	125000	95
Cy5.5 <sup>TM</sup>	675 nm	694 nm	250000	98
Cy7 <sup>TM</sup>	748 nm	767 nm	250000	97
Dy 782 (infrared!)	782 nm	800 nm	102000	99

\*\* Please contact PE Biosystems

**Licensed supplier**  
of custom oligos labeled with  
dyes patented by Molecular  
Probes and Amersham  
Pharmacia Biotech

Spectral data have been provided  
by suppliers of the dyes and can vary  
with conjugation and solvent.



References listing IBA as supplier:

- 1 Bernacchi S, Mély Y, 2001: Nucleic Acid Res, 29, 13 e62.
- 2 Foldes-Papp Z, Angerer B, Thyberg P, Hinz M, Wennmalm S, Ankenbauer W, Seliger H, Holmgren A, Rigler R. J Biotechnol 2001 Apr 13;86(3):203-24.

Fluorescently labeled oligonucleotides are used for:

- Non-radioactive sequencing
- Mutation analysis
- Primer extension analysis
- Gene scan analysis
- Fragment analysis
- Multiplex PCR
- Quantitative PCR
- Online monitoring of PCR
- FCS-Spectroscopy
- *In situ* hybridization
- Screening
- Measuring ribozyme kinetics

IBA offers a broad spectrum of dyes for all of these applications.

There are different possibilities for labeling an oligonucleotide:

- All of our dyes (except for HEX and TET) can be attached postsynthetically to the 5'-terminus of an oligonucleotide via an amino- or thiol-linker with a 6-Carbon spacer arm.
- Another possibility for labeling an oligonucleotide at the 5'-terminus is to couple the dye directly during synthesis via its phosphoramidite. This method is available for Cy3™, Cy5™, Cy5.5™, Fluorescein, HEX, TET and Dabcyl.
- Postsynthetic labeling of oligonucleotides at internal sites of the sequence is possible by substituting any thymidine with 5-C6-Amino-2'-deoxythymidine (see page 89). All dyes available for 5'-labeling can also be attached internally.
- It is also possible to label the 3'-end of oligonucleotides postsynthetically via an amino link. This method is available for all dyes. For the 3'-labeling with Fluorescein, Dabcyl, TAMRA and ROX we have special solid supports [CPGs] with the dyes already attached. This is very useful for high quality production of doubly labeled probes.

## Quality of dye labeled oligonucleotides

All dye labeled oligonucleotides are double HPLC purified (with the exception of the directly coupled dyes which have to be purified once only). Failure sequences are removed during the first HPLC purification. The second HPLC is performed to remove unlabeled oligonucleotides and excessive dye. As a result only labeled oligonucleotides are obtained.

## Double and multiple labeling

Labeling oligonucleotides with multiple dyes is also possible. This is used in Fluorescence Resonance Energy Transfer (FRET) applications<sup>1-3</sup>.

In theory, resonance energy transfer between two fluorophores will occur only if the emission spectrum of the donor overlaps the excitation spectrum of the acceptor. It was discovered, however, that "non-overlapping FRET pairs" can also be used, such as FAM/ Cy5™ or a wide range of fluorophores with Dabcyl as quencher. The fluorophores have to be able to reach close vicinity to each other. The excited fluorophore transfers the energy to the acceptor (quencher) without emission of a photon. For most assays fluorescein is used as a donor. Typical acceptor dyes are: Fluorescein, TAMRA, ROX, Cy5™, Carboxy-rhodamine-6G and Dabcyl.

References:

- 1 Hung SC, Mathies RA, Glazer AN, 1997: Anal Biochem 252:78-88.
- 2 Hanne A, Ramanujam MV, Rucker G, 1998: Nucleosides & Nucleotides 17:1835-1850.
- 3 Singh KK, Parwatesh R and Krupp G, 1999: RNA 5:1348-1356.

For reference titles see pages 110 ff. or [www.iba-go.com](http://www.iba-go.com).

## Possible dye combinations

IBA offers a great variety of oligonucleotides with different labels and their combinations. For example: 5'-Fluorescein, 5'-HEX, 5'- Cy3™, 5'- Cy5™, 5'- Cy5.5™, 5'-TET, 3'-Dabcyl, 3'-Fluorescein, 3'-TAMRA and 3'-ROX can be combined with any of the available dyes in varied positions. For double labelings with dyes that are not available as amidites labeling via thiol-reactive compounds is possible.

## Real-time PCR probes

Probes for real-time monitoring of PCR consist of an oligonucleotide complementary to the amplified probe. The probe is labeled at the 5'-end with the fluorescence donor (usually Fluorescein, HEX or TET) and a few bases downstream or on the 3'-end with a quencher, usually TAMRA. When no complementary sequence is available the fluorescence of the donor is quenched. During PCR the labeled oligonucleotide hybridizes to the target sequence and the 5'-dye is removed by 5' → 3'-exonuclease activity of Taq. The fluorescence of the donor is no longer quenched and can be measured.

The fluorescence intensity is proportional to the amount of PCR product formed during the early exponential phase of PCR (threshold value). In the probe design it is important that the melting point of the probe is higher than that of the upstream primer. The 3'-end of the probe must be protected against chain elongation during PCR. For blocking the 3'-end phosphate, cordycepin, 2',3'-dideoxynucleosides, inverseT or the quencher dye itself can be used. Labeling the 3'-end with TAMRA or another quencher can be achieved by using a 3'-amino-linker. Labeling internal positions of the sequence is possible by incorporating a modified thymidine residue which contains an amino-link at the 5-position of the base. This attachment does not interfere with hybridization.

## We offer

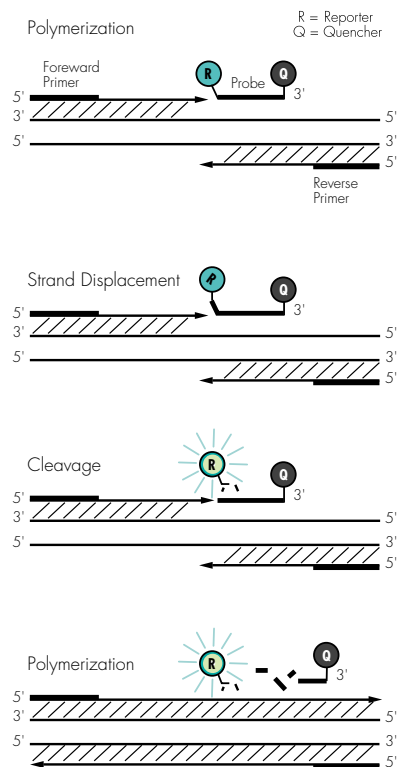
- A great variety of oligonucleotides with different labels and their combinations
- 5'-, 3'- or internal labels
- High purity probes and thorough quality control

Here are the most important dye combinations (further combinations on request):

product	delivery amount	cat. no.
Real-time PCR Probe 5' Fam 3' TAMRA	5/25/100 nmol	5-0901-002/3/4
Real-time PCR Probe 5' HEX 3' TAMRA	5/25/100 nmol	5-0901-012/3/4
Real-time PCR Probe 5' TET 3' TAMRA	5/25/100 nmol	5-0901-022/3/4

	Dye combinations for multiplex real-time PCR								
	FAM	HEX	TET	JOE	Cy3	TAMRA	Texas Red	ROX	Cy5
FAM	***	***	***	***	***	***	***	***	***
HEX	***	***	*	n.r.	n.r.	n.r.	***	***	***
TET	***	*	***	n.r.	n.r.	**	***	***	***
JOE	***	n.r.	n.r.	***	n.r.	*	***	***	***
Cy3	***	n.r.	n.r.	n.r.	***	n.r.	*	*	***
TAMRA	***	n.r.	**	*	n.r.	***	**	**	***
Texas Red	***	***	***	***	n.r.	**	***	n.r.	***
ROX	***	***	***	***	*	**	n.r.	***	***
Cy5	***	***	***	***	***	***	***	***	***

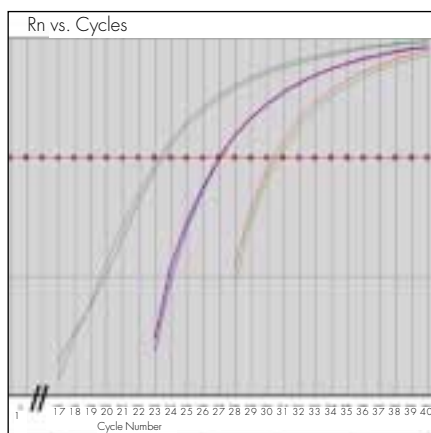
\*\*\* Very good    \*\* good    \* moderate    n.r. not recommended



Principle of real-time PCR probes

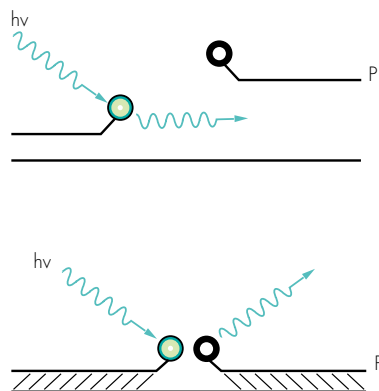
Reference:

Heid CA, Stevens J, Livak KJ, Williams PM, 1996: Genome Res 6:986-994.



Detection of 18sRNA with 5' FAM, 3' TAMRA probe. Amplification plots with (left to right) duplicate samples of 10, 1 and 0.1 ng RNA.

For reference titles see pages 110 ff. or [www.iba-go.com](http://www.iba-go.com).



Principle of FRET probes for real-time PCR applications

## Pair of FRET probes for real-time PCR applications

The FRET principle works with two differently labeled probes. The first oligonucleotide is labeled at the 3'-end (usually with fluorescein) and the second oligonucleotide is labeled at the 5'-end with a FRET acceptor (e. g. Cy5™ or TAMRA). The first oligonucleotide hybridizes to the target in such a way that its 3'-end is separated from the 5'-end of the second oligonucleotide by no more than 1 base. When no complementary sequence is available only the fluorescence of the donor is visible. If the target is present the labeled probes will hybridize with this target and FRET can occur. The fluorescence of the acceptor is mediated by the quencher that emits fluorescence at a longer wavelength than that of the acceptor. The fluorescence intensity is proportional to the amount of PCR product formed during the early exponential phase of PCR (threshold value). The 3'-end of the downstream probes has to be protected against chain elongation during PCR.

We offer the following ready-to-use FRET probes (for Fluorescence Resonance Energy Transfer):

The first oligonucleotide is 5' labeled with Cy5, Cy5.5, Atto 590, Atto 610 or Atto 680 and is 3' blocked. The second oligonucleotide is 3' 6-Fam labeled.

product	delivery amount	cat. no.
Pair of FRET probes	5 nmol of each labeled oligonucleotide	5-0901-102
	25 nmol of each labeled oligonucleotide	5-0901-103

The costs per base pair and HPLC purification are included.

## General ordering information for labeled oligonucleotides

Dedicated to high quality, all labeled oligonucleotides are HPLC purified at no additional costs. Excessive dye and unlabeled oligonucleotides are removed.

The delivery amount is 5 nmol\* for the 0.05 µmol scale, 25 nmol\* for the 0.2 µmol scale, and 100 nmol\* for the 1.0 µmol scale.

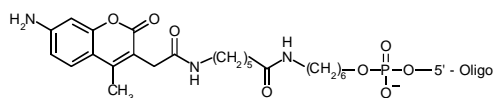
\* for oligonucleotides shorter than 40 bases

## Fluorescent dyes

### AMCA-X

- Useful for multicolor applications because of excitation maximum in the UV range (350 nm)
- Bright blue fluorescence
- Absorption 353 nm, emission 442,  $\epsilon = 19000 \text{ cm}^{-1}\text{M}^{-1}$

product	synthesis scale	delivery amount	cat. no.
5'-AMCA-X	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0310-022/3/4
3'-AMCA-X	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0310-002/3/4
internal AMCA-X	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0310-012/3/4



5'-AMCA-X

Reference:

Delia D, Martinez E, Fontanella E, Aiello A, 1991: Cytometry 12:537-544.

## Atto dyes

- Strong absorption
- High fluorescence quantum yield
- High photostability
- Good water solubility
- Low triplet formation
- Ideally suited for bioanalytical applications
- Produced by ATTO-Tec (see also [www.atto-tec.com](http://www.atto-tec.com))

Atto	absorption	emission	$\epsilon$ [cm <sup>-1</sup> M <sup>-1</sup> ]
425	438 nm	486 nm	40000
520	520 nm	442 nm	105000
565	561 nm	585 nm	120000
590	598 nm	634 nm	120000
610	605 nm	630 nm	150000
655	665 nm	690 nm	125000
680	680 nm	702 nm	125000

product	scale	delivery amount	cat. no.
5'-Atto 425	0.05/0.2/1.0 $\mu$ mol	5/25/100 nmol	5-0311-772/3/4
5'-Atto 520	0.05/0.2/1.0 $\mu$ mol	5/25/100 nmol	5-0311-782/3/4
5'-Atto 565	0.05/0.2/1.0 $\mu$ mol	5/25/100 nmol	5-0311-792/3/4
5'-Atto 590	0.05/0.2/1.0 $\mu$ mol	5/25/100 nmol	5-0311-802/3/4
5'-Atto 610	0.05/0.2/1.0 $\mu$ mol	5/25/100 nmol	5-0311-812/3/4
5'-Atto 655	0.05/0.2/1.0 $\mu$ mol	5/25/100 nmol	5-0311-822/3/4
5'-Atto 680	0.05/0.2/1.0 $\mu$ mol	5/25/100 nmol	5-0311-832/3/4

Also available for 3' and internal labeling

## BODIPY® fluorophores

BODIPY® fluorophores span the complete visible spectrum and have spectral characteristics which are superior over commonly used dyes.

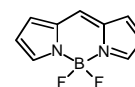
### Advantages

- Higher quantum yields
- Narrower emission bandwidths
- Greater photostability
- Insensitive to pH changes
- Useful for DNA sequencing because the dye exhibits minimal effect on the mobility of the fragment during electrophoresis

## BODIPY® FL

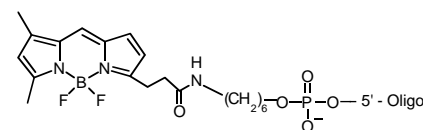
The green fluorescent BODIPY® FL fluorophore has several characteristics potentially providing advantages over fluorescein. These include:

- High extinction coefficient
- High fluorescent quantum yield (often approaching 1.0, even in water)
- Spectral properties that are insensitive to solvent polarity and pH
- Narrow emission bandwidth, resulting in a higher peak intensity than fluorescein
- Little or no spectral overlap with longer wavelength dyes such as tetramethylrhodamine and Texas Red, making it useful for multicolor applications
- Greater photostability than fluorescein in some environments
- Lack of ionic charge

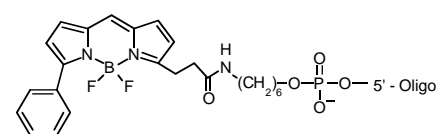


Basic structure of the BODIPY fluorophore

Potentially superior to fluorescein



5'-BODIPY FL



5'-BODIPY R6G

The different BODIPY® dyes are named according to their absorption and emission wavelength or according to the dye they can substitute. The following BODIPY® fluorophores are available:

BODIPY®	absorption	emission	$\epsilon$ [cm <sup>2</sup> M <sup>-1</sup> ]
493/503	500 nm	509 nm	79000
530/550	534 nm	551 nm	77000
558/568	559 nm	568 nm	97000
564/570-X	563 nm	569 nm	142000
576/589	575 nm	588 nm	83000
581/591-X	582 nm	591 nm	154000
630/650	625 nm	640 nm	101000
650/665	646 nm	660 nm	102000
FL (substitute for Fluorescein)	502 nm	510 nm	82000
R6G (substitute for Rhodamine 6G)	528 nm	547 nm	70000
TMR-X (substitute for TAMRA)	544 nm	570 nm	56000
TR-X (substitute for Texas-Red)	588 nm	616 nm	68000

Sold under license from Molecular Probes, Inc.

Reference:

Metzker ML, Lu J, Gibbs RA, 1996:  
Science 271:1420-1422.

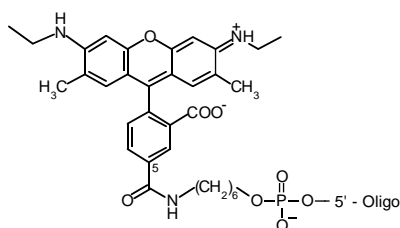
All BODIPY® fluorophores are available in the following scales:

product	synthesis scale	delivery amount
5'-BODIPY®	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol
3'-BODIPY®	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol
Internal BODIPY®	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol

(cat. nos. 5-0310-102 to 5-0310-454)

### Carboxyrhodamine 6G, mixed, 5- or 6-isomers (R6G-5/6, R6G-5 or R6G-6)

- High quantum yield and high photostability
- The absorption maximum is close to the 514 nm spectral line of argon-ion lasers
- Superior electrophoretic and spectroscopic properties to JOE-6 dye
- Used in sequencing
- Available as mixed, 5- and 6-isomer
- R6G, 5-isomer: absorption 524 nm, emission 557 nm,  $\epsilon = 108000$  cm<sup>2</sup>M<sup>-1</sup>
- R6G, 6-isomer: absorption 524 nm, emission 550 nm,  $\epsilon = 102000$  cm<sup>2</sup>M<sup>-1</sup>



5'-Carboxyrhodamine 6G, 5-isomer

Reference:

Hung SC, Ju J, Mathies RA, Glazer AN, 1996:  
Anal Biochem 238:165-170.

product	synthesis scale	delivery amount	cat. no.
5'-R6G-5/6	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol	5-0310-562/3/4
3'-R6G-5/6	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol	5-0310-572/3/4
Internal R6G-5/6	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol	5-0310-582/3/4
5'-R6G-5	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol	5-0310-522/3/4
3'-R6G-5	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol	5-0310-502/3/4
Internal R6G-5	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol	5-0310-512/3/4
5'-R6G-6	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol	5-0310-552/3/4
3'-R6G-6	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol	5-0310-532/3/4
Internal R6G-6	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol	5-0310-542/3/4

## Carboxy-X-rhodamine, mixed, 5-or 6-isomers (ROX-5/6, ROX-5 or ROX-6)

- Widely used for non-radioactive DNA sequencing
- Available as mixed, 5-and 6-isomer
- ROX, 5-isomer: absorption 574 nm, emission 602 nm,  $\epsilon = 78000 \text{ cm}^{-1}\text{M}^{-1}$
- ROX, 6-isomer: absorption 575 nm, emission 602 nm,  $\epsilon = 82000 \text{ cm}^{-1}\text{M}^{-1}$

product	synthesis scale	delivery amount	cat. no.
5'-ROX-5/6	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0310-662/3/4
3'-ROX-5/6	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0310-672/3/4
Internal ROX-5/6	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0310-682/3/4
5'-ROX-5	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0310-622/3/4
3'-ROX-5	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0310-602/3/4
Internal ROX-5	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0310-612/3/4
5'-ROX-6	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0310-652/3/4
3'-ROX-6	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0310-632/3/4
Internal ROX-6	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0310-642/3/4

## Cy™ Dyes

Sold under licence from Amersham Pharmacia Biotech, Inc.

In addition to labeling oligonucleotides during synthesis using Cy amides, we have now also obtained a license from Amersham Pharmacia Biotech, which allows us to use NHS esters of Cy2, Cy3, Cy3.5, Cy5, Cy5.5 and Cy7 for 5', 3'- and internal postsynthetic labelings providing more flexibility and increasing also the quality of the fluorescent probes.

### General order information for postsynthetic Cy™ labeling (Cy2, Cy3, Cy3.5, Cy5, Cy5.5, Cy7)

product	synthesis scale	delivery amount	cat. no.
5'-Cy NHS labeling	0.05/0.2/1.0 $\mu\text{mol}$	5/25/100 nmol	5-0310-722/3/4
3'-Cy NHS labeling	0.05/0.2/1.0 $\mu\text{mol}$	5/25/100 nmol	5-0310-732/3/4
Internal Cy NHS labeling	0.05/0.2/1.0 $\mu\text{mol}$	5/25/100 nmol	5-0310-742/3/4

### Cy2™

- Absorption 489 nm, emission 506 nm,  $\epsilon = 150000 \text{ cm}^{-1}\text{M}^{-1}$

### Cy3.5™

- Absorption 581 nm, emission 596 nm,  $\epsilon = 150000 \text{ cm}^{-1}\text{M}^{-1}$

### Cy7™

- Absorption 748 nm, emission 767 nm,  $\epsilon = 250000 \text{ cm}^{-1}\text{M}^{-1}$

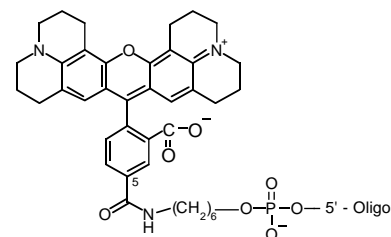
product	synthesis scale	delivery amount	cat. no.
5'-Cy NHS labeling	0.05/0.2/1.0 $\mu\text{mol}$	5/25/100 nmol	5-0310-722/3/4
3'-Cy NHS labeling	0.05/0.2/1.0 $\mu\text{mol}$	5/25/100 nmol	5-0310-732/3/4
Internal Cy NHS labeling	0.05/0.2/1.0 $\mu\text{mol}$	5/25/100 nmol	5-0310-742/3/4

### Cy3™-Amidite

- Excellent spectroscopic properties, such as high quantum yields, high extinction coefficient and great stability of fluorescence signal against changes in pH, various buffers and other conditions
- Absorption 550 nm, emission 570 nm,  $\epsilon = 150000 \text{ cm}^{-1}\text{M}^{-1}$

product	scale	amount	cat. no.
5'-Cy3-Amidite	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0310-702/3/4

Sold under license from Amersham Pharmacia Biotech, Inc. For postsynthetic labeling with NHS esters see above.

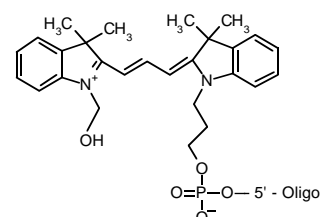


5'-ROX, 5-isomer

#### References:

- 1 Yoshikawa Y, Mukai H, Asada K, Hino F, Kato, 1998: Anal Biochem 256:82-91.
- 2 Lu H, Arriaga E, Chen DY, Dovichi NJ, 1994: J Chromatogr A 680:497-501.

"The cyanine dyes in this product are manufactured on behalf of Amersham Pharmacia Biotech Inc under an exclusive licence from Carnegie Mellon University and are covered by US Patent Number 5 268 486 and other patents pending. Use of the product for commercial purposes is strictly forbidden without written permission from Amersham Pharmacia Biotech Inc."

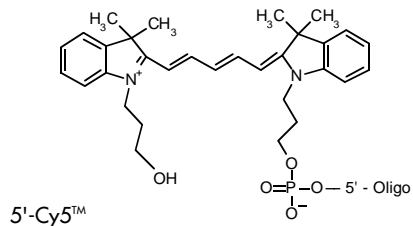


5'-Cy3™

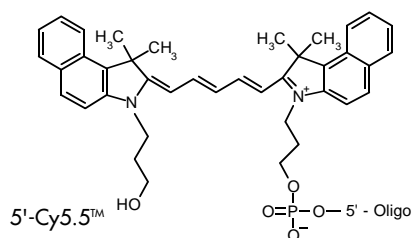
#### Reference:

- Mujumdar RB, Ernst LA, Mujumdar SR, Lewis CJ, Waggoner AS, 1993: Bioconjug Chem 4:105-111.

For reference titles see pages 110 ff. or [www.iba-go.com](http://www.iba-go.com).

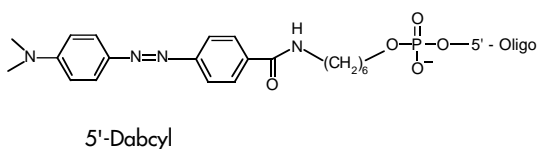


Reference:  
Roman BL, Pham VN, Bennett PE,  
Weinstein BM, 1999: Biotechniques 26:236-238.

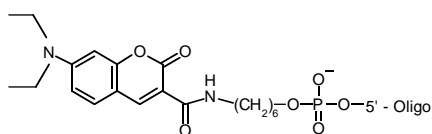


Reference:  
Yager TD, Baron L, Batra R, Bouevitch A, Chan D, Chan K, Darasch S, Gilchrist R, Izmailov A, Lacroix JM, Marchelletta K, Renfrew J, Renfrew J, Rushlow D, Steinbach E, Ton C, Waterhouse P, Zaleski H, Dunn JM, Stevens J, 1999: Electrophoresis 20:1280-1300.

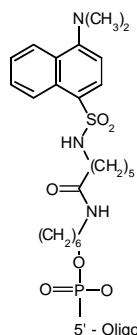
Reference:  
Nazarenko IA, Bhatnagar SK, Hohman RJ, 1997:  
Nucleic Acids Res 25:2516-21.



5'-D-AMCA



5'-Dansyl-X



For reference titles see pages 110 ff. or [www.iba-go.com](http://www.iba-go.com).

## Cy5™-Amidite

- High quantum yields, high extinction coefficient and great signal stability in various buffers and conditions
- One of the best dyes absorbing in the red spectra
- For ALF® sequencer
- STORM FluorImager™ (635 nm excitation light source) is highly sensitive for Cy5™ detection
- Excitation by red light generates extremely low background as most natural fluorophores are excited at shorter wavelengths
- Absorption 649 nm, emission 670 nm,  $\epsilon = 250000 \text{ cm}^{-1}\text{M}^{-1}$

product	synthesis scale	delivery amount	cat. no.
5'-Cy5-Amidite	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0310-822/3/4

Sold under license from Amersham Pharmacia Biotech, Inc. For postsynthetic labeling with NHS esters see page 97.

## Cy5.5™-Amidite

- Absorption in the far red with high quantum yields, a high extinction coefficient and great stability in various buffers and conditions
- Absorption 675 nm, emission 694 nm,  $\epsilon = 250000 \text{ cm}^{-1}\text{M}^{-1}$

product	synthesis scale	delivery amount	cat. no.
5'-Cy5.5-Amidite	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0310-902/3/4

Sold under license from Amersham Pharmacia Biotech, Inc. For postsynthetic labeling with NHS esters see page 97.

## DabcyI

- Universal quencher in combination with FAM, HEX, TET, TAMRA and other dyes
- Does not emit fluorescence and thus avoids overlapping detection signals
- Double labeling possible with 3'-DabcyI and any other dye
- Absorption 453 nm, no emission,  $\epsilon = 32000 \text{ cm}^{-1}\text{M}^{-1}$

product	synthesis scale	delivery amount	cat. no.
5'-DabcyI	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-022/3/4
3'-DabcyI	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-002/3/4
Internal DabcyI	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-012/3/4

## 7-Diethylaminocoumarin-3-carboxylic acid (D-AMCA)

- Short wavelength absorption
- Used in multicolor applications
- Absorption 432 nm, emission 472 nm,  $\epsilon = 56000 \text{ cm}^{-1}\text{M}^{-1}$

product	synthesis scale	delivery amount	cat. no.
5'-D-AMCA	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-122/3/4
3'-D-AMCA	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-102/3/4
Internal D-AMCA	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-112/3/4

## Dansyl-X

- Large Stokes shift
- Environmentally sensitive
- UV-absorption
- Absorption 340 nm, emission 520 nm,  $\epsilon = 4200 \text{ cm}^{-1}\text{M}^{-1}$

product	synthesis scale	delivery amount	cat. no.
5'-Dansyl-X	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-152/3/4
3'-Dansyl-X	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-132/3/4
Internal Dansyl-X	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-142/3/4

**Dy 630**

- Absorption 621 nm, emission 652 nm
- Emission lifetime 0.4 ns
- Molecular weight: 731.92 g x mol<sup>-1</sup>
- Molecular formula: C<sub>40</sub>H<sub>49</sub>N<sub>3</sub>O<sub>8</sub>S

**Dy 635**

- Absorption 634 nm, emission 664 nm
- Emission lifetime 0.4 ns
- Molecular weight: 755.94 g x mol<sup>-1</sup>
- Molecular formula: C<sub>42</sub>H<sub>49</sub>N<sub>3</sub>O<sub>8</sub>S

**Dy 782 (infrared)**

- Absorption 782 nm, emission 800 nm,  $\epsilon = 102000\text{cm}^2\text{M}^{-1}$
- Molecular weight: 660.88 g x mol<sup>-1</sup>

product	scale	delivery amount	cat. no.
5'-Dy 630	0.05/0.2/1.0 $\mu\text{mol}$	5/25/100 nmol	5-0311-742/3/4
5'-Dy 635	0.05/0.2/1.0 $\mu\text{mol}$	5/25/100 nmol	5-0311-752/3/4
5'-Dy 782 (infrared!)	0.05/0.2/1.0 $\mu\text{mol}$	5/25/100 nmol	5-0311-762/3/4

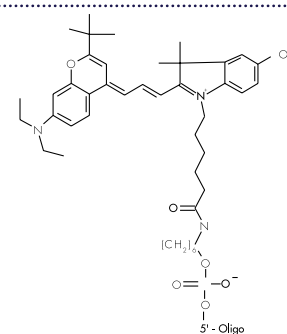
Also available for 3' and internal labeling. For further information see [www.dyomics.com](http://www.dyomics.com).

**EVOblue™ 30**

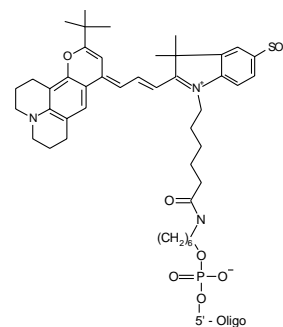
- Superior to UV-dyes like coumarines or dyes excitable with blue or green light like fluoresceines and rhodamines
- The lower excitation energy needed for EVOblue™ 30 causes less photo-destruction and exceeds the lifetime of the fluorescent signal compared to common dyes
- Optimized for an excitation by 635 nm and an emission around 670 nm
- EVOblue™ 30 reveals a high chemical stability
- Absorption 630 nm, emission 670 nm

product	scale	delivery amount	cat. no.
5'-EVOblue 30	0.05/0.2/1.0 $\mu\text{mol}$	5/25/100 nmol	5-0311-732/3/4

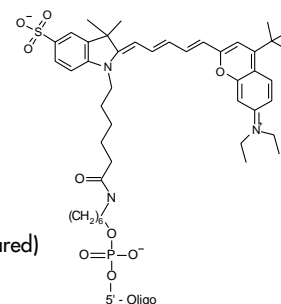
Also available for 3' and internal labeling. For further information see [www.evotecoi.com](http://www.evotecoi.com).

**NEW**

Dy 630



Dy 635



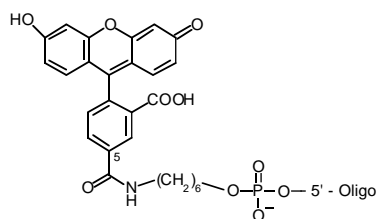
Dy 782 (infrared)

**Further Dyomics dyes  
available for labeling;  
see [www.dyomics.com](http://www.dyomics.com)**

**NEW**

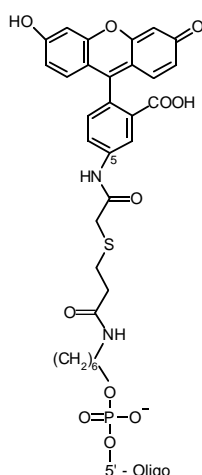
**Excellent new dye  
with red absorption**

For reference titles see pages 110 ff. or [www.iba-go.com](http://www.iba-go.com).



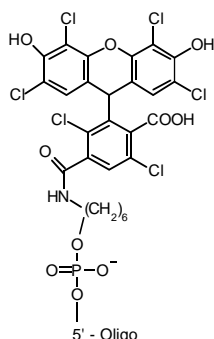
5'-FAM, 5-isomer

Reference:  
Sjoberg R, Nygren J, Kubista M, 1998:  
Biopolymers 46:445-453.



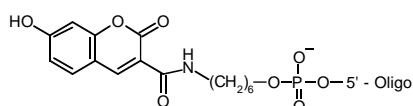
5'-FAM-EX-5

Reference:  
Mann MJ, Gibbons GH, Hutchinson H, Poston RS, Hoyt EG, Robbins RC, Dzau VJ, 1999:  
Proc Natl Acad Sci U S A 96:6411-6416.



5'-HEX

Reference:  
Shin JH, Nolte FS, Holloway BP, Morrison CJ, 1999:  
J Clin Microbiol 37: 165-170.



5'-HYCO

For reference titles see pages 110 ff. or [www.iba-go.com](http://www.iba-go.com).

## Fluorescein, mixed, 5-or 6-isomers (FAM-5/6, FAM-5 or FAM-6)

- High quantum yields
- Excitation maximum in close proximity to the 488 nm spectral line of the Argon-Ion Laser
- Used in applications such as DNA sequencing, GeneScan analysis, *in situ* hybridization, and many others
- Double labelings possible with 3'-or 5'-Fluorescein and any other dye or marker
- Can be introduced as amidite (isomeric mixture) and post synthetically as mixed, 5- or 6-isomer
- FAM, 5-isomer: absorption 494 nm, emission 520 nm,  $\epsilon = 78000 \text{ cm}^2\text{M}^{-1}$
- FAM, 6-isomer: absorption 496 nm, emission 516 nm,  $\epsilon = 83000 \text{ cm}^2\text{M}^{-1}$

product	synthesis scale	delivery amount	cat. no.
5'-FAM-5/6	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-302/3/4
3'-FAM-5/6	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-312/3/4
Internal FAM-5/6	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-322/3/4
5'-FAM-5	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-222/3/4
3'-FAM-5	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-202/3/4
Internal FAM-5	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-212/3/4
5'-FAM-6	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-252/3/4
3'-FAM-6	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-232/3/4
Internal FAM-6	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-242/3/4
5'-FAM Amidite	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-262/3/4

## Fluorescein-EX, 5-isomer (FAM-EX-5)

- Extended linker arm between oligonucleotide and dye
- More hydrophilic than FAM, 5-isomer
- FAM-EX, 5-isomer: absorption 491 nm, emission 515 nm,  $\epsilon = 86000 \text{ cm}^2\text{M}^{-1}$

product	synthesis scale	delivery amount	cat. no.
5'-FAM-EX-5	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-292/3/4
3'-FAM-EX-5	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-272/3/4
Internal FAM-EX-5	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-282/3/4

## HEX-Amidite

- Used for multicolor applications, e.g. the 377 DNA sequencer in Genescan analysis
- Only possible at the 5'-end
- Can be combined with any available dye in varied positions
- Absorption 537 nm, emission 556 nm,  $\epsilon = \text{contact PE Biosystems}$

product	synthesis scale	delivery amount	cat. no.
5'-HEX	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-402/3/4

Also available for 3' and internal labeling.

## 7-Hydroxy coumarin-3-carboxylic acid (HYCO)

- Short wavelength absorption
- Used in multicolor applications
- Absorption 419 nm, emission 447 nm,  $\epsilon = 36000 \text{ cm}^2\text{M}^{-1}$

product	synthesis scale	delivery amount	cat. no.
5'-HYCO	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-522/3/4
3'-HYCO	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-502/3/4
Internal HYCO	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-512/3/4

**JOE, 6-isomer (JOE-6)**

- Routinely used for oligonucleotide labeling and DNA sequencing
- High quantum yields and low pH sensitivity
- Absorption 520 nm, emission 548 nm,  $\epsilon = 71000 \text{ cm}^{-1}\text{M}^{-1}$

product	synthesis scale	delivery amount	cat. no.
5'-JOE-6	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-722/3/4
3'-JOE-6	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-702/3/4
Internal JOE-6	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0311-712/3/4

**Oregon Green™ 488, 5-or 6-isomers**

- An alternative to fluorescein with greater photostability and higher fluorescence
- Available as 5- or 6-isomer
- Oregon Green™ 488, 5-isomer: absorption 495 nm, emission 521 nm,  $\epsilon = 76000 \text{ cm}^{-1}\text{M}^{-1}$
- Oregon Green™ 488, 6-isomer: absorption 492 nm, emission 517 nm,  $\epsilon = 88000 \text{ cm}^{-1}\text{M}^{-1}$

product	synthesis scale	delivery amount	cat. no.
5'-Oregon Green™ 488, 5-isomer	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0312-022/3/4
3'-Oregon Green™ 488, 5-isomer	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0312-002/3/4
Internal Oregon Green™ 488, 5-isomer	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0312-012/3/4
5'-Oregon Green™ 488, 6-isomer	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0312-052/3/4
3'-Oregon Green™ 488, 6-isomer	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0312-032/3/4
Internal Oregon Green™ 488, 6-isomer	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0312-042/3/4

Sold under license from Molecular Probes, Inc.

**Oregon Green™ 500, 5-isomer**

- Absorption maximum only slightly shifted from that of Oregon Green 488 or fluorescein fluorophores
- Greater photostability and higher fluorescence than fluorescein
- Absorption 499 nm, emission 519 nm,  $\epsilon = 78000 \text{ cm}^{-1}\text{M}^{-1}$

product	synthesis scale	delivery amount	cat. no.
5'-Oregon Green™ 500, 5-isomer	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0312-222/3/4
3'-Oregon Green™ 500, 5-isomer	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0312-202/3/4
Internal Oregon Green™ 500, 5-isomer	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0312-212/3/4

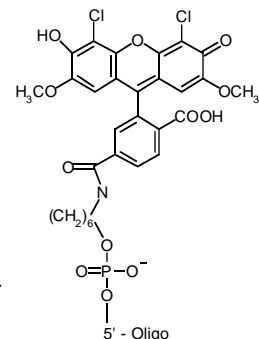
Sold under license from Molecular Probes, Inc.

**Oregon Green™ 514**

- Preferred fluorescein substitute for fluorescence imaging applications due to its high photostability
- 90% single isomer
- Absorption 506 nm, emission 526 nm,  $\epsilon = 85000 \text{ cm}^{-1}\text{M}^{-1}$

product	synthesis scale	delivery amount	cat. no.
5'-Oregon Green™ 514	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0312-422/3/4
3'-Oregon Green™ 514	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0312-402/3/4
Internal Oregon Green™ 514	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0312-412/3/4

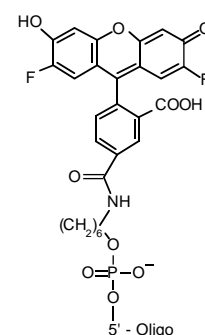
Sold under license from Molecular Probes, Inc.



5'-JOE, 6-isomer

Reference:

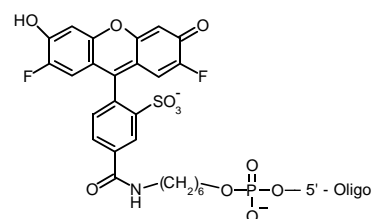
Fausser S, Wissinger B, 1997: Biotechniques 22:964-968.



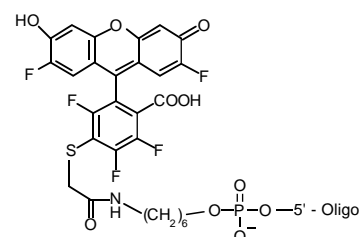
5'-Oregon Green 488, 5-isomer

Reference:

Lin K, Sadee W, Quillan JM, 1999: Biotechniques 26:324-326.



5'-Oregon Green 500, 5-isomer



5'-Oregon Green 514

Reference:

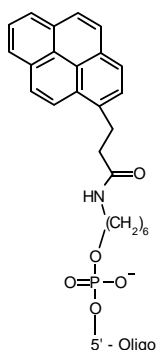
Delmotte C, Delmas A, 1999: Bioorg Med Chem Lett 9:2989-2994.

For reference titles see pages 110 ff. or [www.iba-go.com](http://www.iba-go.com).

NEW



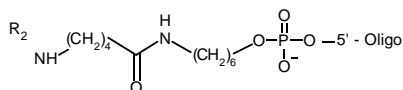
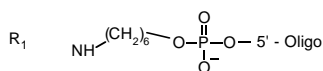
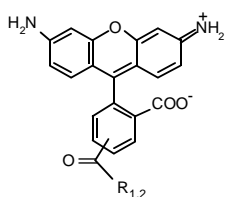
Oyster® 656



5'-Pyrene

References:

- Masuko M, Ohtani H, Ebata K, Shimadzu A, 1998: Nucleic Acids Res 26:5409-5416.
- Rippe K, Fritsch V, Westhof E, Jovin TM, 1992: EMBO J 11:3777-3786.

R<sub>1</sub> = 5'-Rhodamine Green, mixed isomersR<sub>2</sub> = 5'-Rhodamine Green-X, mixed isomers

Reference:

- Rigler R, Foldes-Papp Z, Meyer-Almes FJ, Sammet C, Volcker M, Schnetz A, 1998: J Biotechnol 63:97-109.

## Oyster®

- New class of cyanine dyes
- High photostability
- Bright fluorescence
- Oyster-556 can substitute Cy3: adsorption 560 nm, emission 570 nm,  $\epsilon$  155000
- Oyster-645 can substitute Cy5: adsorption 649 nm, emission 666 nm,  $\epsilon$  250000
- Oyster-656 matches the emission band of a 650 nm laser diode; adsorption 660 nm, emission 674 nm,  $\epsilon$  220000
- produced by De Novo Biolabels (see also [www.denovo.com](http://www.denovo.com))

product	synthesis scale	delivery amount	cat. no.
5'-Oyster-556	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol	5-0315-002/3/4
3'-Oyster-556	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol	5-0315-012/3/4
Internal-Oyster-556	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol	5-0315-022/3/4
5'-Oyster-645	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol	5-0315-032/3/4
3'-Oyster-645	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol	5-0315-042/3/4
Internal-Oyster-645	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol	5-0315-052/3/4
5'-Oyster-656	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol	5-0315-062/3/4
3'-Oyster-656	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol	5-0315-072/3/4
Internal-Oyster-656	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol	5-0315-082/3/4

## Pyrene

- Pyrene is excited by UV light (340 nm) and has an extensive fluorescence lifetime
- Excited-state dimers (excimers) can form with fluorescence emission shifted to longer wavelengths than that of the monomer (this property can be utilized in nucleic acid hybridization experiments).
- Absorption 340 nm, emission 376 nm,  $\epsilon$  = 43000  $\text{cm}^{-1}\text{M}^{-1}$

product	synthesis scale	delivery amount	cat. no.
5'-Pyrene	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol	5-0312-622/3/4
3'-Pyrene	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol	5-0312-602/3/4
Internal Pyrene	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol	5-0312-612/3/4

## Rhodamine Green™, mixed isomers

- Higher photostability than Rhodol Green™ and Oregon Green™ 488
- Fluorescence is completely pH insensitive in the range from pH 4 to pH 9
- Absorption and fluorescence emission maximum is approximately 7 nm shifted from that of fluorescein towards the red spectrum, but remains compatible with standard fluorescein filter sets
- Used in FCS-Spectroscopy
- Absorption 504 nm, emission 532 nm,  $\epsilon$  = 78000  $\text{cm}^{-1}\text{M}^{-1}$

product	synthesis scale	delivery amount	cat. no.
5'-Rhodamine Green™, mixed isomers	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol	5-0313-222/3/4
3'-Rhodamine Green™, mixed isomers	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol	5-0313-202/3/4
Internal Rhodamine Green™, mixed isomers	0.05/0.20/1.00 $\mu$ mol	5/25/100 nmol	5-0313-212/3/4

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## Rhodamine Green™-X, mixed isomers

- Rhodamine Green™ derivative with a longer, more hydrophilic linker arm
- Absorption 503 nm, emission 528 nm,  $\epsilon = 74000 \text{ cm}^{-1}\text{M}^{-1}$

product	synthesis scale	delivery amount	cat. no.
5'-Rhodamine Green™-X, mixed isomers	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0313-252/3/4
3'-Rhodamine Green™-X, mixed isomers	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0313-232/3/4
Internal Rhodamine Green™-X, mixed isomers	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0313-242/3/4

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## Rhodamine Red™-X, 5-isomer

- Absorption located at a wavelength between TAMRA and Texas Red®
- Absorption 560 nm, emission 580 nm,  $\epsilon = 129000 \text{ cm}^{-1}\text{M}^{-1}$

product	synthesis scale	delivery amount	cat. no.
5'-Rhodamine Red™-X, 5-isomer	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0313-422/3/4
3'-Rhodamine Red™-X, 5-isomer	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0313-402/3/4
Internal Rhodamine Red™-X, 5-isomer	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0313-412/3/4

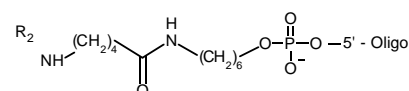
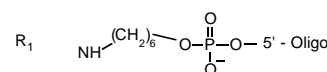
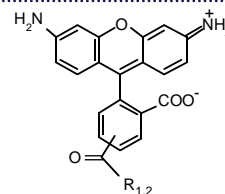
Sold under license from Molecular Probes, Inc.

## Rhodol Green™, mixed isomers

- Rhodol Green™ is an alternative to fluorescein with higher photostability and a higher fluorescence quantum yield
- Absorption 496 nm, emission 523 nm,  $\epsilon = 63000 \text{ cm}^{-1}\text{M}^{-1}$

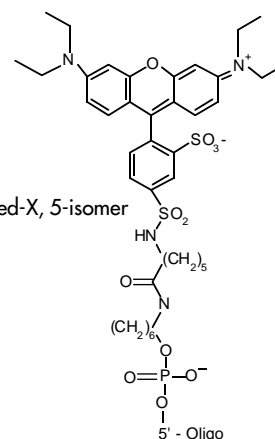
product	synthesis scale	delivery amount	cat. no.
5'-Rhodol Green™, mixed isomers	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0313-622/3/4
3'-Rhodol Green™, mixed isomers	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0313-602/3/4
Internal Rhodol Green™, mixed isomers	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0313-612/3/4

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R<sub>1</sub> = 5'-Rhodamine Green, mixed isomers

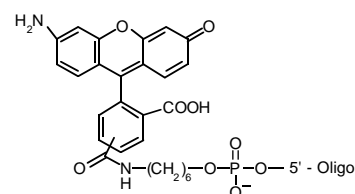
R<sub>2</sub> = 5'-Rhodamine Green-X, mixed isomers



5'-Rhodamine Red-X, 5-isomer

Reference:

Lefevre C, Kang HC, Haugland RP, Malekzadeh N, Arttamangkul S, Haugland RP, 1996: Bioconjug Chem 7:482-489.

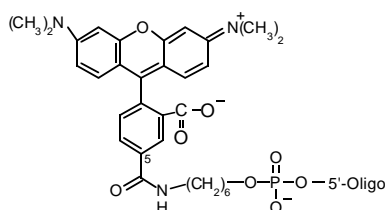


Rhodol Green, mixed isomers

Reference:

Benchabib M, Delorme R, Pluvinage M, Bryon PA, Souchier C, 1996: Histochem Cell Biol 106:253-256.

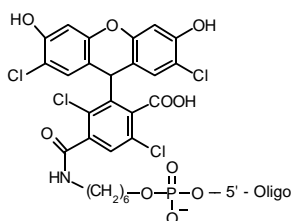
For reference titles see pages 110 ff. or [www.iba-go.com](http://www.iba-go.com).



5'-TAMRA, 5-isomer

Reference:

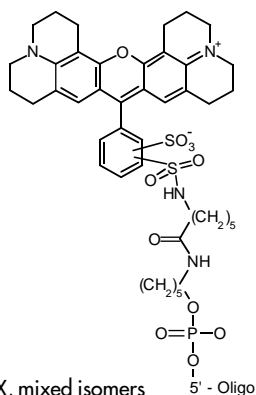
Eggeling C, Fries JR, Brand L, Gunther R, Seidel CA, 1998: Proc Natl Acad Sci U S A 95:1556-1561.



5'-TET

Reference:

Shin JH, Nolte FS, Holloway BP, Morrison CJ, 1999: J Clin Microbiol 37:165-170.



5'-Texas Red®-X, mixed isomers

Reference:

Lefevre C, Kang HC, Haugland RP, Malekzadeh N, Arttamangkul S, Haugland RP, 1996: Bioconjug Chem 7:482-489.

## Tetramethylrhodamine mixed, 5-or 6-isomers (TAMRA-5/6, TAMRA-5 or TAMRA-6)

- Widely used in non-radioactive DNA sequencing
- High photostability
- Used for *in situ* hybridization applications
- Used as a quencher in Fluorescence Resonance Energy Transfer applications (FRET) (see page 94)
- Available as 5- or 6-isomer
- TAMRA, 5-isomer: absorption 546 nm, emission 579 nm,  $\epsilon = 91000 \text{ cm}^2\text{M}^{-1}$
- TAMRA, 6-isomer: absorption 547 nm, emission 573 nm,  $\epsilon = 91000 \text{ cm}^2\text{M}^{-1}$

product	synthesis scale	delivery amount	cat. no.
5'-TAMRA-5/6	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0314-062/3/4
3'-TAMRA-5/6	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0314-072/3/4
Internal TAMRA-5/6	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0314-082/3/4
5'-TAMRA-5	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0314-022/3/4
3'-TAMRA-5	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0314-002/3/4
Internal TAMRA-5	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0314-012/3/4
5'-TAMRA-6	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0314-052/3/4
3'-TAMRA-6	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0314-032/3/4
Internal TAMRA-6	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0314-042/3/4

## TET-Amidite

- Used in multicolor applications such as the 377 DNA sequencer in Genescan analysis
- Only at the 5'-end
- Can be combined with any available dye in varied positions
- Absorption 519 nm, emission 539 nm,  $\epsilon =$  please contact PE Biosystems

product	synthesis scale	delivery amount	cat. no.
5'-TET	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0314-202/3/4

Also available for 3' and internal labeling.

## Texas Red®-X, mixed isomers

- The long wavelength emission property of Texas Red®-X is ideal for multicolor applications
- Extra long spacer between oligonucleotide and dye
- Absorption 583 nm, emission 603 nm,  $\epsilon = 116000 \text{ cm}^2\text{M}^{-1}$

product	synthesis scale	delivery amount	cat. no.
5'-Texas Red®-X, mixed isomers	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0314-422/3/4
3'-Texas Red®-X, mixed isomers	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0314-402/3/4
Internal Texas Red®-X, mixed isomers	0.05/0.20/1.00 $\mu\text{mol}$	5/25/100 nmol	5-0314-412/3/4

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## Biotin, Iminobiotin and Digoxigenin labeled oligonucleotides

Biotin and Digoxigenin offer several advantages over radioactively labeled probes:

- Labeled probes can be stored for a long period without loss of label or activity
- Hybridization solutions can be re-used several times
- The technology is non-hazardous
- A wide range of enzyme conjugates are available for detection and signal amplification (dye, fluorescence, luminescence)

### Biotin

Biotin is a very popular hapten marker for oligonucleotides. An extensive variety of products are available for the detection and immobilization of biotin labeled probes. As biotin has a high affinity to streptavidin, the detection of biotin labeled probes is very sensitive. Examples of this are *in situ* hybridization, cDNA synthesis with biotin oligo-dT, capture of amplified DNA sequences and solid phase sequencing. Biotin can be introduced either at the 3'-end and/or at the 5'-end and at internal sites at any thymidine residue. For best results and highest sensitivity the distance between each biotin in the probe should be at least 15 bases. **Every biotin labeled oligonucleotide is carefully analyzed by HPLC for quantitative labeling.**

product	synthesis scale	delivery amount	cat. no.
5'-Biotin	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0320-032/3/4
3'-Biotin	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0320-012/3/4
Internal Biotin	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0320-022/3/4

### Iminobiotin

2-Iminobiotin, an analogue of biotin, interacts with avidin and streptavidin in a pH-dependent manner allowing recovery of your reporter molecule coupled to iminobiotin. At high pH, the free base form of 2-iminobiotin retains the high-affinity specific binding characteristics of biotin whereas at acidic pH values the salt form of the analogue interacts poorly with avidin or streptavidin.

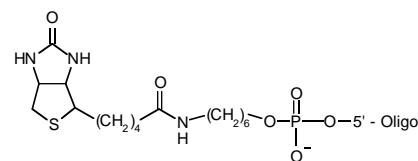
Using these properties for an affinity purification protocol you will be able to load your biomolecule to an avidin matrix at pH 9.5 and elute it at pH 4, thus, overcoming the harsh elution conditions associated with biotin.

#### We offer

- Oligonucleotides labeled with iminobiotin or biotin
- Carefully analyzed by HPLC for quantitative labeling
- Labels introduced at the 3'-end, 5'-end and internally at any thymidine residue

product	synthesis scale	delivery amount	cat. no.
5'-Iminobiotin	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0322-032/3/4
3'-Iminobiotin	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0322-012/3/4
Internal Iminobiotin	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0322-022/3/4

Reference:  
Ward DC, 1990:  
Clin Biochem 23: 307-310.

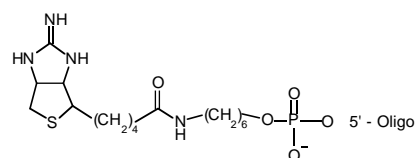


5'-Biotin

References:  
1 Cocuzza AJ, 1989: Tetrahedron Lett 30: 6287-6290.  
2 Nelson PS, Kent M, and Muthini K, 1992:  
Nucleic Acid Res 20: 6253-6259.



Mild purification conditions for active reporter molecules in e.g. DNA/DNA or DNA/protein binding studies



5'-Iminobiotin-Oligo

References:  
1 Fudem-Goldin B, Orr GA, 1990: Methods Enzymol 184: 167-173  
2 Hofman K, Wood SW, Brinton CC, Montibeller JA, Finn FM, 1980: Proc. Natl. Acad. Sci. USA 77: 4666-8

For reference titles see pages 110 ff. or [www.iba-go.com](http://www.iba-go.com).

NEW

Oligo (dT)<sub>20</sub> Probe, biotin-labeled

Probe for immobilization of mRNA on streptavidin-coated solid supports

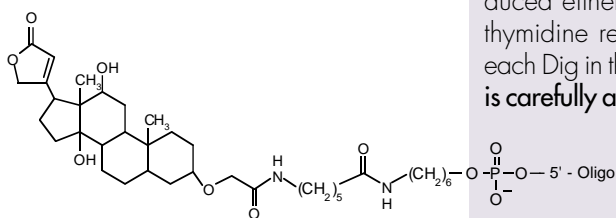
This biotin-labeled oligo (dT) probe can be hybridized with eukaryotic mRNA for subsequent immobilization on streptavidin-coated surfaces, such as PCR tubes or magnetic beads. For further downstream applications like RT-PCR the immobilized mRNA can be isolated or directly processed.

**Other (dT)<sub>x</sub> lengths  
are available**

product	delivery amount	cat. no.
Oligo (dT) <sub>20</sub> Probe, biotin-labeled, HPLC purified	25/100 nmol	5-0000-303/4

## Digoxigenin

Digoxigenin (Dig) is a hapten occurring naturally only in *Digitalis purpurea*. This explains the low endogenous background associated in applications with Dig. A range of conjugated anti-Dig antibodies are commercially available, such as alkaline phosphatase, peroxidase, fluorescein and rhodamine. Dig can be introduced either at the 3'-end and/or the 5'-end as well as at internal sites at any thymidine residue. For best results and highest sensitivity the distance between each Dig in the probe should be at least 15 bases. **Every Dig labeled oligonucleotide is carefully analyzed by HPLC for quantitative labeling.**



## 5'-Digoxigenin

product	synthesis scale	delivery amount	cat. no.
5'-Dig	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0321-032/3/4
3'-Dig	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0321-012/3/4
Internal Dig	0.05/0.20/1.00 µmol	5/25/100 nmol	5-0321-022/3/4

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## References:

- 1 Kessler C, Hölte HJ, Seibl R, Burg J, Mühlegger K, 1990: Biol Chem Hoppe-Seyler 371:917-927.
- 2 Schmitz GG, Walter T, Seibl R, Kessler C, 1991: Analytical Biochem 192:222-231.

## Modified nucleoside-triphosphates

One of the main reasons for the success of IBA in custom oligonucleotide synthesis is the in-house nucleic acid chemistry facility. It enables us to provide modified oligonucleotides in top quality at competitive prices. Consequently, IBA has extensive expertise in:

- Synthesis of modified nucleosides and nucleotides
- Synthesis of special amidites
- Synthesis of non-nucleosidic compounds including linkers, dyes etc.
- Immobilization of nucleic acids, peptides, and proteins on solid phases

IBA has established a nucleoside-triphosphate synthesis facility and offers an extensive range of triphosphates for many applications.

All products are double HPLC purified, desalted and well characterized by HPLC and  $^{31}\text{P}$ -NMR. They are also available as monophosphates and nucleosides.

All triphosphates are also available in the amount of 1  $\mu\text{mol}$ , 5  $\mu\text{mol}$  and 10  $\mu\text{mol}$ .

All  $\alpha$ -Thio nucleoside-5'-triphosphates are also available in the amount of 1  $\mu\text{mol}$ , 5  $\mu\text{mol}$  and 10  $\mu\text{mol}$ .

For prices see separate price list or [www.iba-go.com](http://www.iba-go.com).

### Modified 2'-deoxynucleoside-5'-triphosphates

- Inhibition studies of DNA polymerases
- Studying DNA-protein interactions

product	delivery amount	cat. no.
3'-Amino-2', 3'-ddATP	0.5/1/5 $\mu\text{mol}$	5-0610-013/4/5
3'-Azido-2', 3'-ddATP	0.5/1/5 $\mu\text{mol}$	5-0610-023/4/5
N6-Methyl-2'-dATP	0.5/1/5 $\mu\text{mol}$	5-0612-013/4/5
3'-Amino-2', 3'-ddCTP	0.5/1/5 $\mu\text{mol}$	5-0610-253/4/5
5-Bromo-2'-dCTP	0.5/1/5 $\mu\text{mol}$	5-0612-253/4/5
5-Methyl-2'-dCTP	0.5/1/5 $\mu\text{mol}$	5-0612-263/4/5
3'-Azido-2', 3'-ddGTP	0.5/1/5 $\mu\text{mol}$	5-0610-503/4/5
O6-Methyl-2'-dGTP	0.5/1/5 $\mu\text{mol}$	5-0612-503/4/5
3'-Azido-2', 3'-ddTTP	0.5/1/5 $\mu\text{mol}$	5-0610-753/4/5
5-Bromo-2'-dUTP	0.5/1/5 $\mu\text{mol}$	5-0612-763/4/5
5-Fluoro-2'-dUTP	0.5/1/5 $\mu\text{mol}$	5-0612-773/4/5
2,6-Diaminopurine-2'-deoxyriboside-TP	0.5/1/5 $\mu\text{mol}$	5-0612-783/4/5

### Modified ribonucleoside-5'-triphosphates

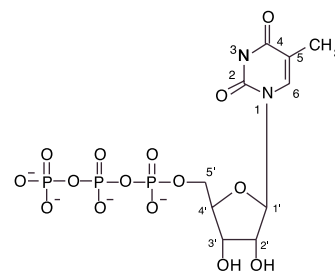
#### Sugar-modified ribonucleoside 5'-triphosphates

- 2'-modified nucleotides to prevent nuclease degradation of RNAs
- 3'-and other modified nucleotides for inhibition of RNA polymerases

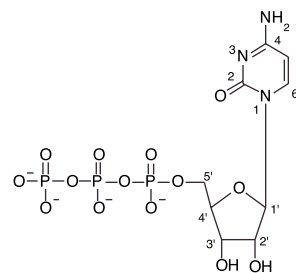
#### Sugar-modified adenosine-5'-triphosphates

product	delivery amount	cat. no.
2'-Amino-2'-dATP	0.5/1/5 $\mu\text{mol}$	5-0611-013/4/5
3'-Amino-3'-dATP	0.5/1/5 $\mu\text{mol}$	5-0611-023/4/5
Ara-ATP	0.5/1/5 $\mu\text{mol}$	5-0611-033/4/5
2'-Azido-2'-dATP	0.5/1/5 $\mu\text{mol}$	5-0611-043/4/5
3'-Azido-3'-dATP	0.5/1/5 $\mu\text{mol}$	5-0611-053/4/5
2', 3'-Epoxy-ATP	0.5/1/5 $\mu\text{mol}$	5-0611-063/4/5
2'-O-Methyl-ATP	0.5/1/5 $\mu\text{mol}$	5-0611-073/4/5
3'-O-Methyl-ATP	0.5/1/5 $\mu\text{mol}$	5-0611-083/4/5
Xylo-ATP	0.5/1/5 $\mu\text{mol}$	5-0611-093/4/5

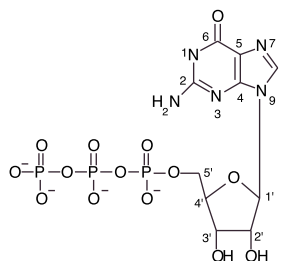
Austermann S, Kruhoffer M, Grasse F, 1992:  
Biochem Pharmacol 43:2581-2589.



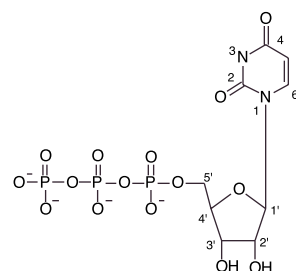
TTP



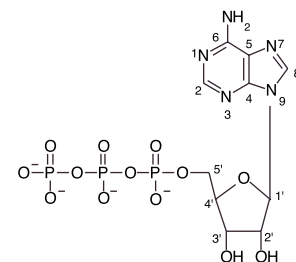
CTP



GTP



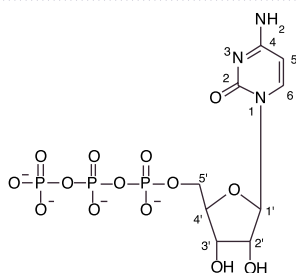
UTP



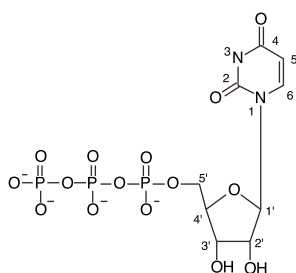
ATP

Reference:  
Kujau MJ, Siebert A, Wolf S, 1997:  
J Biochem Biophys Methods 35:141-151.

For reference titles see pages 110 ff. or [www.iba-go.com](http://www.iba-go.com).



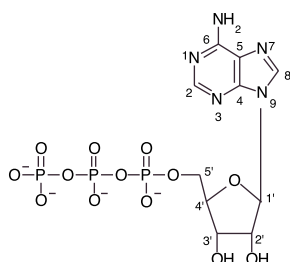
CTP



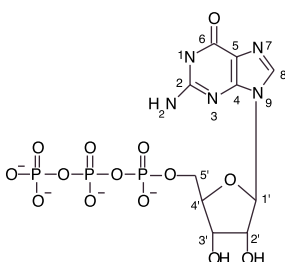
UTP

References:

- Putz J, Florentz C, Benseler F, Giege R, 1994: Nat Struct Biol 1:580-582.
- Mishima Y, Steitz JA, 1995: EMBOJ 14:2679-2687.



ATP



GTP

## Sugar-modified cytidine-5'-triphosphates

product	delivery amount	cat. no.
2'-Amino-2'-dCTP	0.5/1/5 µmol	5-0611-253/4/5
2'-Azido-2'-dCTP	0.5/1/5 µmol	5-0611-263/4/5
2'-O-Methyl-2'-dCTP	0.5/1/5 µmol	5-0611-273/4/5
2'-Fluoro-2'-dCTP	0.5/1/5 µmol	5-0611-283/4/5

## Sugar-modified guanosine-5'-triphosphates

product	delivery amount	cat. no.
2'-O-Methyl-GTP	0.5/1/5 µmol	5-0611-503/4/5

## Sugar-modified uridine-5'-triphosphates

product	delivery amount	cat. no.
2'-Amino-2'-dUTP	0.5/1/5 µmol	5-0611-753/4/5
2'-Azido-2'-dUTP	0.5/1/5 µmol	5-0611-763/4/5
2'-Fluoro-2'-dUTP	0.5/1/5 µmol	5-0611-773/4/5

## Base-modified ribonucleoside-5'-triphosphates

- Studying RNA-protein interactions
- Thiol modified nucleotides for crosslinking experiments

## Base-modified adenosine-5'-triphosphates

product	delivery amount	cat. no.
8-Bromo-ATP	0.5/1/5 µmol	5-0613-013/4/5
7-Deaza-ATP (Tubercidin)	0.5/1/5 µmol	5-0613-033/4/5
N1-Methyl-ATP	0.5/1/5 µmol	5-0613-043/4/5
N6-Methyl-ATP	0.5/1/5 µmol	5-0613-053/4/5

## Base-modified cytidine-5'-triphosphates

product	delivery amount	cat. no.
5-Bromo-CTP	0.5/1/5 µmol	5-0613-253/4/5
5-Iodo-CTP	0.5/1/5 µmol	5-0613-273/4/5
5-Methyl-CTP	0.5/1/5 µmol	5-0613-293/4/5
2-Thio-CTP	0.5/1/5 µmol	5-0613-303/4/5

## Base-modified guanosine-5'-triphosphates

product	delivery amount	cat. no.
8-Bromo-GTP	0.5/1/5 µmol	5-0613-513/4/5
6-Chloro-7-deaza-GTP	0.5/1/5 µmol	5-0613-523/4/5
7-Deaza-GTP	0.5/1/5 µmol	5-0613-533/4/5
6-Mercapto-GTP	0.5/1/5 µmol	5-0613-543/4/5
8-Mercapto-GTP	0.5/1/5 µmol	5-0613-553/4/5
N1-Methyl-GTP	0.5/1/5 µmol	5-0613-573/4/5

## Base-modified uridine-5'-triphosphates

product	delivery amount	cat. no.
5-Bromo-UTP	0.5/1/5 µmol	5-0613-763/4/5
5-Fluoro-UTP	0.5/1/5 µmol	5-0613-773/4/5
5-Methyl-UTP (rTTP)	0.5/1/5 µmol	5-0613-793/4/5
4-Thio-UTP	0.5/1/5 µmol	5-0613-813/4/5

For reference titles see pages 110 ff. or [www.iba-go.com](http://www.iba-go.com).

## Base-modified inosine-5'-triphosphates

product	delivery amount	cat. no.
Inosine-5'-triphosphate (ITP)	0.5/1/5 µmol	5-0613-823/4/5

## Base modified purine-5'-triphosphates

product	delivery amount	cat. no.
2-Aminopurine-riboside-5'-triphosphate	0.5/1/5 µmol	5-0613-843/4/5
2-Amino-6-chloropurine-riboside-5'-triphosphate	0.5/1/5 µmol	5-0613-853/4/5
6-Chloropurine-riboside-5'-triphosphate	0.5/1/5 µmol	5-0613-863/4/5
6-Methyl-mercaptopurine-riboside-5'-triphosphate	0.5/1/5 µmol	5-0613-873/4/5

 $\alpha$ -Thio-nucleoside-5'-triphosphates

All  $\alpha$ -Thio-nucleoside-5'-triphosphates are also available in the amount of 1 µmol, 5 µmol and 10 µmol.

2'-Deoxynucleoside-5'-( $\alpha$ -thio-triphosphates)

- For site-directed mutagenesis<sup>1</sup>
- Stabilizing DNA against nucleases<sup>2</sup>
- X-ray or NMR analysis of enzyme-substrate complexes

product	delivery amount	cat. no.
2'-dATP $\alpha$ S	0.5/1/5 µmol	5-0614-013/4/5
2'-dCTP $\alpha$ S	0.5/1/5 µmol	5-0614-253/4/5
2'-dGTP $\alpha$ S	0.5/1/5 µmol	5-0614-503/4/5
2'-dTTP $\alpha$ S	0.5/1/5 µmol	5-0614-753/4/5
2'-dITP $\alpha$ S	0.5/1/5 µmol	5-0614-763/4/5
2'-dUTP $\alpha$ S	0.5/1/5 µmol	5-0614-773/4/5

Ribonucleoside-5'-( $\alpha$ -thio-triphosphates)

- For RNA-Sequencing<sup>1</sup>
- Introduction of dyes and reporter groups<sup>2</sup>
- High throughput mapping of functionally important groups in RNA (NAIM)<sup>3</sup>
- Stabilizing RNA against nucleases<sup>4</sup>

Modified adenosine-5'-( $\alpha$ -thio-triphosphates)

product	delivery amount	cat. no.
ATP $\alpha$ S	0.5/1/5 µmol	5-0615-013/4/5
7-Deaza-ATP $\alpha$ S (Tubercidin)	0.5/1/5 µmol	5-0615-033/4/5
N1-Methyl-ATP $\alpha$ S	0.5/1/5 µmol	5-0615-043/4/5
N6-Methyl-ATP $\alpha$ S	0.5/1/5 µmol	5-0615-053/4/5

Modified cytidine-5'-( $\alpha$ -thio-triphosphates)

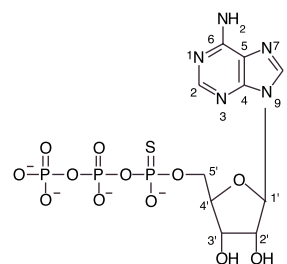
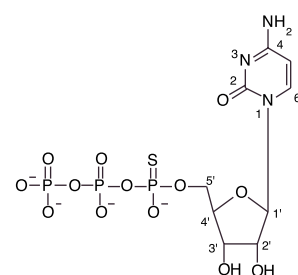
product	delivery amount	cat. no.
CTP $\alpha$ S	0.5/1/5 µmol	5-0615-253/4/5
5-Bromo-CTP $\alpha$ S	0.5/1/5 µmol	5-0615-263/4/5
5-Iodo-CTP $\alpha$ S	0.5/1/5 µmol	5-0615-273/4/5
5-Methyl-CTP $\alpha$ S	0.5/1/5 µmol	5-0615-293/4/5

## References:

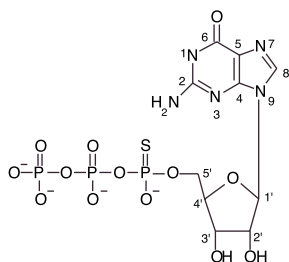
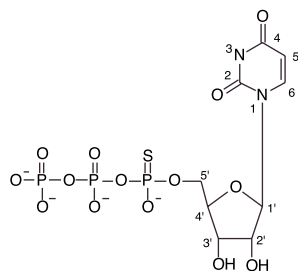
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ATP $\alpha$ SCTP $\alpha$ S

For reference titles see pages 110 ff. or [www.iba-go.com](http://www.iba-go.com).

GTP $\alpha$ SUTP $\alpha$ S

## Modified guanosine-5'-( $\alpha$ -thio-triphosphates)

product	delivery amount	cat. no.
GTP $\alpha$ S	0.5/1/5 $\mu$ mol	5-0615-503/4/5
6-Chloro-7-deaza-GTP $\alpha$ S	0.5/1/5 $\mu$ mol	5-0615-513/4/5
7-Deaza-GTP $\alpha$ S	0.5/1/5 $\mu$ mol	5-0615-523/4/5
6-Mercapto-GTP $\alpha$ S	0.5/1/5 $\mu$ mol	5-0615-533/4/5
N1-Methyl-GTP $\alpha$ S	0.5/1/5 $\mu$ mol	5-0615-553/4/5
N2-Methyl-GTP $\alpha$ S	0.5/1/5 $\mu$ mol	5-0615-543/4/5
N7-Methyl-GTP $\alpha$ S	0.5/1/5 $\mu$ mol	5-0615-563/4/5

## Modified uridine-5'-( $\alpha$ -thio-triphosphates)

product	delivery amount	cat. no.
UTP $\alpha$ S	0.5/1/5 $\mu$ mol	5-0615-753/4/5
5-Bromo-UTP $\alpha$ S	0.5/1/5 $\mu$ mol	5-0615-773/4/5
5-Fluoro-UTP $\alpha$ S	0.5/1/5 $\mu$ mol	5-0615-783/4/5

## Modified purine-5'-( $\alpha$ -thio-triphosphates)

product	delivery amount	cat. no.
Purine-5'-( $\alpha$ -thio-triphosphate)	0.5/1/5 $\mu$ mol	5-0615-823/4/5
2-Aminopurine-5'-( $\alpha$ -thio-triphosphate)	0.5/1/5 $\mu$ mol	5-0615-833/4/5
2-Amino-6-chloropurine-5'-( $\alpha$ -thio-triphosphate)	0.5/1/5 $\mu$ mol	5-0615-843/4/5
6-Chloropurine-5'-( $\alpha$ -thio-triphosphate)	0.5/1/5 $\mu$ mol	5-0615-853/4/5

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