2006

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chemistry

Reagents and unique building blocks for oligonucleotide synthesis

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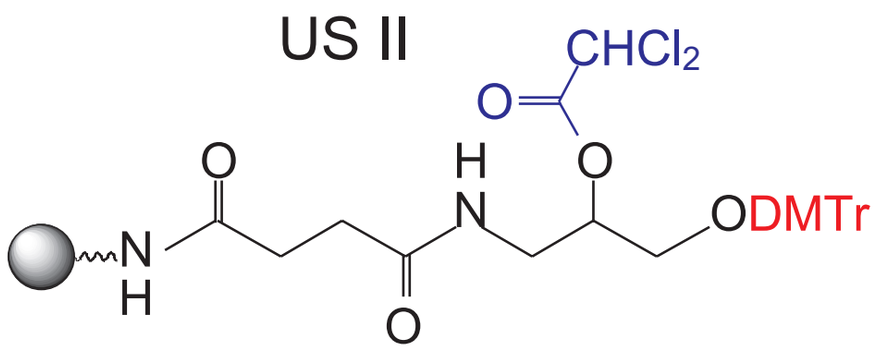
Reagents for RNA and DNA synthesis

UNIVERSAL SOLID SUPPORT

The universal support strategy offers the following clear advantages: eliminates the possibility of errors in parallel synthesis applications where up to 384 wells may contain different supports; eliminates the need for at least four supports for DNA synthesis and four for RNA synthesis; simplifies the preparation of oligonucleotides with modified or unusual nucleosides at the 3'-terminus.

The USII would be appropriate for the production of DNA oligos, long and short, as well as those requiring mild deprotection. It is also compatible with the synthesis of RNA and siRNA. The reagent used for the cleavage/dephosphorylation step is commercially available and the procedures described are fully compatible with high-throughput synthesis.



USII - Truly Universal Solid Support for high throughput oligonucleotide synthesis of DNA, RNA and siRNA.

Catalogue number: 103-00

Description: chemically modified 500Å or 1000Å long chain alkylamino controlled pore glass white to off-white powder

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Reagents for RNA and DNA synthesis

UNIVERSAL SOLID SUPPORT

Storage of dry compound: min. 1 year at +4ºC

Loading: USII-CPG has 20-40 µmol/g loading. Please enquire for custom loading.

Oligo synthesis on CPG: Perform oligonucleotide assembly, using standard protocols, recommended by your synthesizer manufacturer. Upon the completion of synthesis wash the oligonucleotide bound support with pure acetonitrile. Do not perform any washing steps with solvents, containing basic reagents (diethylamine, triethylamine, dimethylamine, etc.) or water.

Cleavage: Cleave the oligo from the support using 3.5N – 4.5N ammonia in methanol (dilute cold 7N ammonia in methanol, Aldrich Cat. No 499145-100ML, with cold anhydrous methanol) at room temperature for 30 minutes. Do not use aqueous ammonium hydroxide and/or mixtures of ammonium hydroxide and methanol for cleavage!

Deprotection AFTER Cleavage:
Standard: After Cleavage, add 1 volume of **3.5N – 4.5N ammonia in methanol**, seal and deprotect for 8-15 hours at 60 °C for removal of the protecting groups on the nucleobases.
or
Alternatively: add 1 volume of 30% ammonium hydroxide, seal and deprotect using the conditions appropriate for removal of the protecting groups on the nucleobases (e.g. at 55ºC for 5 hours).

Polymer beads
We are developing Truly Universal Solid Support for high throughput synthesis on small scale (10-40 µmol/g) as well as for oligonucleotide synthesis on large scale (170-250 µmol/g) loading on variety of polymer beads. Please contact us for additional information and evaluation samples. These types of Truly Universal Solid Support can be prepared and tailored for you upon special request.

Please Note: This product is covered by US Patent No.: 6,770,754 and European Patent No.: 1404695.

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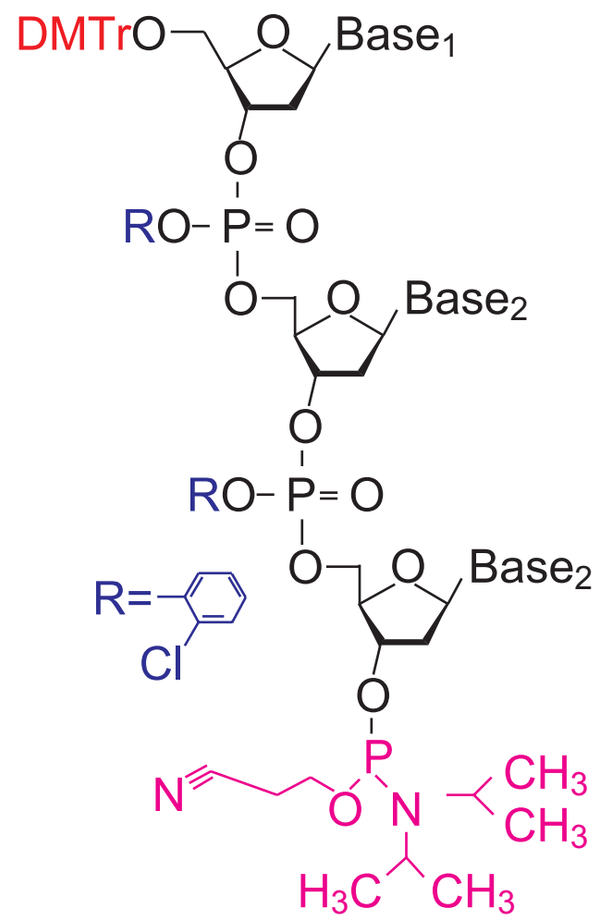
Reagents for RNA and DNA synthesis

TRINUCLEOTIDE (TRIMER CODON) PHOSPHORAMIDITIES

Oligonucleotide-directed mutagenesis - the most popular approach for the preparation of proteins with variations at specific sites. This protein engineering technique uses randomized oligonucleotides to generate libraries of proteins for screening potential improvements in specific biological function.

Traditional approach employs mixtures of monomers to generate codon mixtures. However, the degeneracy of the genetic code guarantees that redundancies and stop codons will be also generated.



Catalogue number: 103-20

Description: white to off-white powder

Storage of dry compound: 1 year at -20°C

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Reagents for RNA and DNA synthesis

TRINUCLEOTIDE (TIMER CODON) PHOSPHORAMIDITIES

There are three significant problems with the traditional approach:

1. The odds of just making a single change of a residue in a protein can be quite small. For instance, the probability of changing a particular Tyr (TAC) to Asn (AAC) in a 100 residue protein is only 1/900 ($1.1x10^{-4}$). If the mutation requires three consecutive nucleosides to be changed - i.e. Tyr (TAC) to Met (ATG), this number is cubed - $(1/900)^3$ - $1.37\ x10^{-9}$. So even a large library of 10^8 clones may not contain the desired mutant!

2. Traditional mutagenic techniques lack the ability to adequately screen for multiple mutations that act cooperatively. For instance, let's assume either mutation Tyr to Lys (TAC to AAA) or Pro to Asp (CCG to GAC) may be insignificant to a particular property individually. However, when both mutations are present within the protein, they act cooperatively to improve the fitness of the protein. The chances of finding both mutations in a single protein are vanishingly small (1.97×10^{-18})!

3. There are 3 out of 64 trimer sequences that are stop codons - TAA, TAG and TGA. There is approximately a 5% chance that a stop codon will be introduced into the sequence with each mutated codon added! Though not an issue with short regions of mutagenesis, the presence of stop codons becomes problematic quickly: mutating a 20 codon region leads to only 36% chance of generating the random codons without a stop codon being present!

The use of Trimer mixes helps these situations considerably:

1. Consider the first case of a Tyr to Met mutation. In a 100 residue protein, the chances of that mutation occurring with a trimer mix of all 20 codons is 5.26×10^{-4} - 5 orders of magnitude better than using the traditional method of degenerate bases!

2. The dual mutations of Tyr to Lys and Pro to Asp, are now more likely to be found within a large library - $2.77\ x10^{-7}$ - an improvement of 11 orders of magnitude over the standard mutagenic methods (1.97×10^{-18})!

3. The use of trinucleotide synthons TP avoids the incorporation of undesired stop codons in the oligonucleotide.

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Reagents for RNA and DNA synthesis

TRIMER (CODON) PHOSPHORAMIDITES SIMPLIFY LIBRARY PREPARATION

Oligonucleotide-directed mutagenesis is probably the most popular approach for the preparation of proteins with variations at specific sites. This protein engineering technique uses oligonucleotides of mixed sequences to generate libraries of proteins for screening potential improvements in specific biological function. It is certainly possible to produce the mixed oligonucleotide sequences by opening the synthesis columns, splitting the supports, and recombining the supports after coupling. This procedure is surely labor-intensive and coupling efficiency is always affected by the splitting and recombination process. The technique is also limited in that the complexity desired may be greater than the number of particles of support in the columns. Another technique is to use mixtures of monomers to generate codon mixtures but the degeneracy of the genetic code guarantees that redundancies and stop codons will be generated. Mutagenesis generating substoichiometric amounts of codons at specific positions has been described[1,2] using a mixture of trimer and monomer phosphoramidites. A further refinement of this strategy has been described[3] using two sets of monomers, one set with 5'-DMT protection and one set with base-labile 5'-Fmoc protected monomers.

The simplest approach for oligonucleotide-directed mutagenesis would be the use of trimer phosphoramidites. Of the 64 possible combinations of codons, only 20 codons would be required to cover the 20 amino acids, although, in practice, several codons will likely be duplicated depending on the organism. Several reports describing[4-6] the synthesis of trimer phosphoramidites have been published. The approach described[7-9] by Kayushin et al seemed to be attractive. However, recently Metkinen Chemistry have developed the procedure ensuring the preparation of trimers of highest quality. This procedure is used for the preparation of all 20 trimers phosphoramidites of Metkinen Chemistry.

Quality control of trimer phosphoramidites is very challenging. We normally use RP HPLC for purity and identity determination of our regular phosphoramidites. However, trimer phosphoramidites have chiral centers at all three phosphorus positions. There are, therefore, $2^3 = 8$ diastereomers in each phosphoramidite, which are at least partially separated on RP HPLC, rendering the technique questionable for purity and identity determination. There is also the concern that the sequence of the trimers has to be verified. For example, CAT coding for histidine, has to be differentiated from TAC, coding for tyrosine. These two trimers have virtually identical lipophilicity and their identity cannot be clearly confirmed by HPLC. This problem has been solved[10] using HPLC electrospray mass spectrometric analysis of the trimers, which provides data confirming molecular weight and sequence.

Reagents for RNA and DNA synthesis

TRIMER (CODON) PHOSPHORAMIDITES SIMPLIFY LIBRARY PREPARATION

In Table 1, the trimers, their coding amino acid and their reaction factor (RF) are listed. The reaction factor is critical since the trimers will likely be mixed and they have differing reactivity in the coupling reaction. RF for AAC is 1.0 and for TAC is 1.6. Therefore, 1.6 equivalents of TAC are needed for every 1.0 equivalent of AAC for equal coupling. Mixtures can easily be made using equimolar solutions or the molecular weight of each trimer has to be used to generate the appropriate weights of each trimer to use if mixing by weight. An example of the preparation of a mixture of all 20 trimers is shown in the right column of Table 1 and completed in the footnotes.

TABLE 1: TRIMER CODING AND PHYSICAL PARAMETERS

Trimer	Amino Acid	MW	RF	MWxRF	mg/10µmol (adjusted for RF)
AAA	Lys	1911.5	1.1	2102.65	21.0 (11)
AAC	Asn	1887.5	1.0	1887.50	18.9 (10)
ACT	Thr	1774.5	1.3	2306.85	23.1 (13)
ATC	Ile	1774.5	1.2	2129.40	21.3 (12)
ATG	Met	1780.5	1.3	2314.65	23.1 (13)
CAG	Gln	1869.5	2.0	3739.00	37.4 (20)
CAT	His	1774.5	1.3	2306.85	23.1 (13)
CCG	Pro	1845.5	1.8	3321.90	33.2 (18)
CGT	Arg	1756.5	1.1	1932.15	19.3 (11)
CTG	Leu	1756.5	1.2	2107.80	21.1 (12)
GAA	Glu	1893.5	1.9	3597.65	36.0 (19)
GAC	Asp	1869.5	1.3	2430.35	24.3 (13)
GCT	Ala	1756.5	1.5	2634.75	26.3 (15)
GGT	Gly	1762.5	1.1	1938.75	19.4 (11)
GTT	Val	1667.5	1.9	3168.25	31.7 (19)
TAC	Tyr	1774.5	1.6	2839.20	28.4 (16)
TCT	Ser	1661.4	1.3	2159.82	21.6 (13)
TGC	Cys	1756.5	1.5	2634.75	26.3 (15)
TGG	Try	1762.5	5.7	10046.25	100.5 (57)
TTC	Phe	1661.4	2.2	3655.08	36.6 (22)
					=592.6 mg

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TRIMER (CODON) PHOSPHORAMIDITES SIMPLIFY LIBRARY PREPARATION

Example of Preparation of Trimer Mixture
Prepare 592.6 mg of the trimer mix, taking the amount (mg) for each trimer from the right column. Dissolve the trimer mix in dichloromethane (highest grade possible; acid-free). Evaporate to dryness to produce a homogenous mixture of all 20 trimers.

Example of Preparation of Trimer Mixture for the Synthesizer
Dissolve 592.6 mg, which is equivalent to 20x10 µmoles (normalized for RF) of the trimer mix in 2.0 mL of acetonitrile-dichloromethane mixture, 1:3 v/v to produce a 0.10N solution of trimers, ready for use in a synthesizer.

All of the trimers are now available individually so that researchers can prepare custom mixtures. A mixture of all 20 trimers designed to produce equal coupling of all 20 is also available.

References:

(1) J. Sondek and D. Shortle, Proc Natl Acad Sci U S A, 1992, 89, 3581-3585.
(2) P. Gaytan, J. Yanez, F. Sanchez, H. Mackie, and X. Soberon, Chem Biol, 1998, 5, 519-527.
(3) P. Gaytan, J. Yanez, F. Sanchez, and X. Soberon, Nucleic Acids Res, 2001, 29, E9.
(4) B. Virnekas, L.M. Ge, A. Pluckthun, K.C. Schneider, G. Wellnhofer, and S.E. Moroney, Nucleic Acids Research, 1994, 22, 5600-5607.
(5) M.H. Lyttle, E.W. Napolitano, B.L. Calio, and L.M. Kauvar, Biotechniques, 1995, 19, 274.
(6) A. Ono, A. Matsuda, J. Zhao, and D.V. Santi, Nucleic Acids Research, 1995, 23, 4677-4682.
(7) A.L. Kayushin, M.D. Korosteleva, A.I. Miroshnikov, W. Kosch, D. Zubov, and N. Piel, Nucleic Acids Research, 1996, 24, 3748-3755.
(8) A. Kayushin, et al., Nucleos Nucleot, 1999, 18, 1531-1533.
(9) A. Kayushin, M. Korosteleva, and A. Miroshnikov, Nucleos Nucleot Nucleic Acids, 2000, 19, 1967-1976.
(10) T. Mauriala, S. Auriola, A. Azhayev, A. Kayushin, M. Korosteleva, M., A. Miroshnikov J. Pharm.Biomed. Anal., 2004, 34, 199-206

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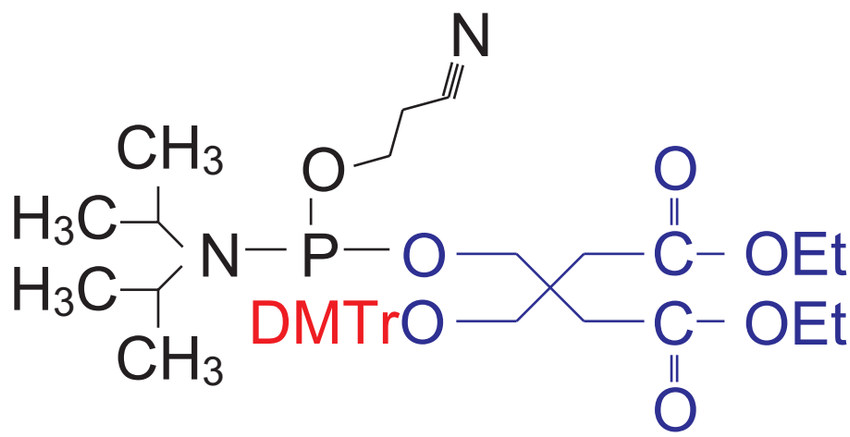
Reagents for RNA and DNA synthesis

CHEMICAL PHOSPHORYLATION REAGENT

Chemical Phosphorylation Reagent (**CPR II**) contains a DMT group which can be left on the oligonucleotide and used for rapid purification of oligonucleotide 5'-phosphates by the popular DMTr-on technique, which employs disposable RP cartridges or "Trityl-on" RP HPLC purification. The DMTr group is removed with aqueous acid (e.g., 2%TFA in the case of Cartridge Purification) and the remaining linker is then eliminated after brief treatment with aqueous ammonium hydroxide (12 -15% ammonium hydroxide at room temperature for 15 minutes) to yield the 5'-phosphate.



[3-(4,4'-Dimethoxytrityloxy)-2,2-dicarboxyethyl]propyl-(2-cyanoethyl)-(N,N-diisopropyl)-phosphoramidite

Catalogue number: 103-10

Description: amorphous colorless glass

Storage of dry compound: 1 year at -20°C

Coupling conditions: 6 minute coupling time.

Omit the capping step after the addition of this reagent!

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Purine Modified Nucleosides

3'- Amino-2',3'-dideoxyadenosine

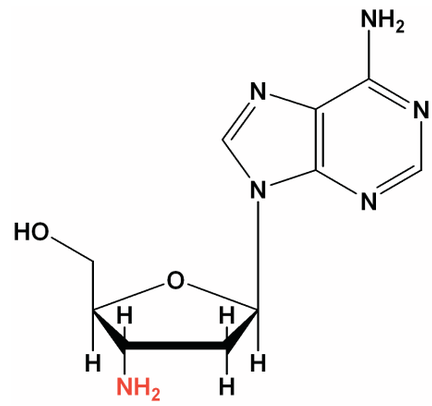
9-(3-amino-2,3-dideoxy- β-D-ribofuranosyl) adenine

Catalogue number: 203-11

Description: white to off-white crystals

Storage of dry compound: 3 years at +4°C

Price:	10 g	600 euro	60 euro/g
	100 g	4 000 euro	40 euro/g
	500 g	15 000 euro	30 euro/g
	1000 g	20 000 euro	20 euro/g



3'-Amino-2',3'-dideoxy-2,6-diaminopurineriboside

9-(3-amino-2,3-dideoxy- β-D-ribofuranosyl)-2,6-diaminopurine

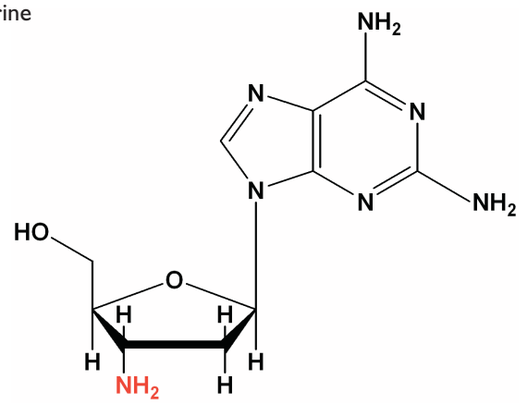
Catalogue number: 203-12

Description: white to off-white crystals

Storage of dry compound: 3 years at +4°C

Price:	10 g	700 euro
	100 g	5 000 euro

For **bulk quantities please enquire**



3'- Amino-2',3'-dideoxyguanosine

9-(3-amino-2,3-dideoxy- β-D-ribofuranosyl) guanine

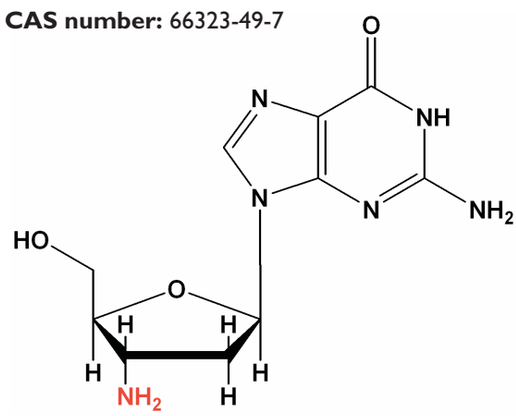
CAS number: 66323-49-7

Catalogue number: 203-10

Description: white to off-white crystals

Storage of dry compound: 3 years at +4°C

Price:	10 g	800 euro	80 euro/g
	100 g	6 000 euro	60 euro/g
	500 g	16 500 euro	33 euro/g
	1000 g	22 000 euro	22 euro/g



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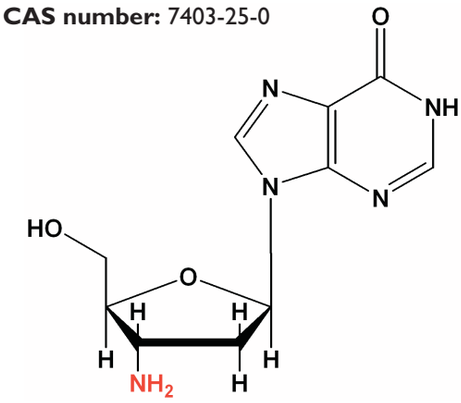
Purine Modified Nucleosides

3'- Amino-2',3'-dideoxyinosine

9-(3-amino-2,3-dideoxy- β-D-ribofuranosyl) hypoxanthine **CAS number:** 7403-25-0



Catalogue number: 203-09

Description: white to off-white crystals

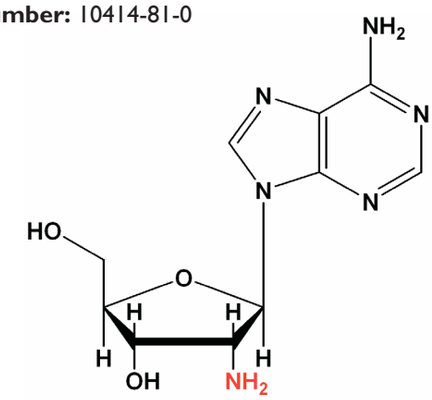
Storage of dry compound: 3 years at +4°C

Price:	10 g	1 000 euro
	100 g	6 000 euro

For **bulk quantities please enquire**

2'-Amino-2'-deoxyadenosine

9-(2-amino-2-deoxy-β-D-ribofuranosyl)-adenine **CAS number:** 10414-81-0



Catalogue number: 203-27

Description: white to off-white crystals

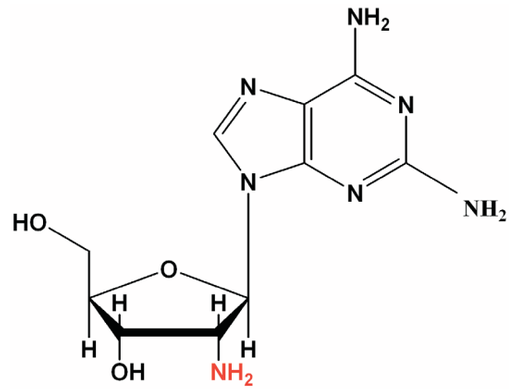
Storage of dry compound: 3 years at +4°C

Price:	1 g	350 euro
	10 g	2 500 euro
	100 g	18 000 euro

For **bulk quantities please enquire**

2'-Amino-2'-deoxy-2,6-diaminopurineriboside

9-(2-amino-2-deoxy-β-D-ribofuranosyl)-2,6-diaminopurine



Catalogue number: 203-28

Description: white to off-white crystals

Storage of dry compound: 3 years at +4°C

Price:	1 g	350 euro
	10 g	2 500 euro
	100 g	18 000 euro

For **bulk quantities please enquire**

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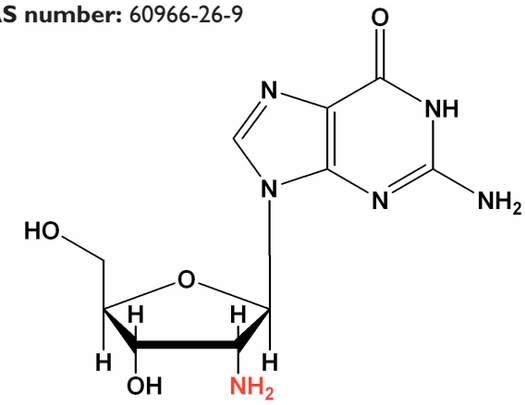
Purine Modified Nucleosides

2'-Amino-2'-deoxyguanosine

9-(2-amino-2-deoxy-β-D-ribofuranosyl)-guanine **CAS number:** 60966-26-9



Catalogue number: 203-29

Description: white to off-white crystals

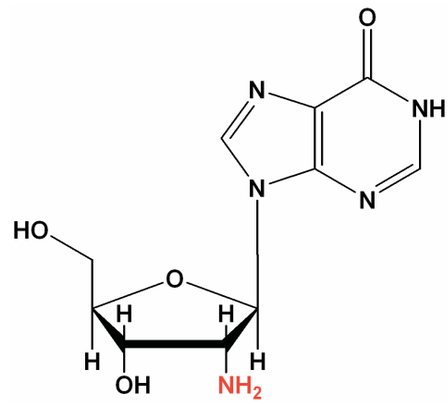
Storage of dry compound: 3 years at +4°C

Price:	1 g	400 euro
	10 g	3 000 euro
	100 g	20 000 euro

For **bulk quantities please enquire**

2'-Amino-2'-deoxyinosine

9-(2-amino-2-deoxy-β-D-ribofuranosyl)-hypoxanthine



Catalogue number: 203-30

Description: white to off-white crystals

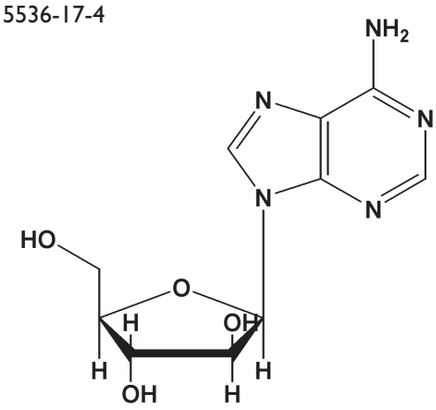
Storage of dry compound: 3 years at +4°C

Price:	1 g	400 euro
	10 g	3 000 euro
	100 g	20 000 euro

For **bulk quantities please enquire**

Arabinofuranosyl-adenine

9-(β-D-arabinoribofuranosyl) adenine **CAS number:** 5536-17-4



Catalogue number: 203-05

Description: white to off-white crystals

Storage of dry compound: 3 years at +4°C

Price:	10 g	400 euro
	100 g	2 000 euro

For **bulk quantities please enquire**

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Purine Modified Nucleosides

Arabinofuranosyl-6-Benzylaminopurine

9-(β-D-arabinoribofuranosyl)-6-Benzylaminopurine

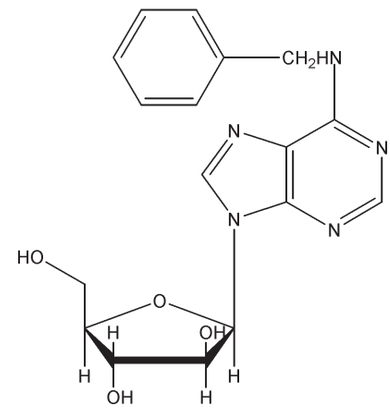
Catalogue number: 203-20

Description: white to off-white crystals

Storage of dry compound: 3 years at +4°C

Price:	1 g	300 euro
	10 g	2 000 euro
	100 g	12 000 euro

For **bulk quantities please enquire**



Arabinofuranosyl-2,6-diaminopurine

9-(β-D-arabinoribofuranosyl)-2,6-diaminopurine

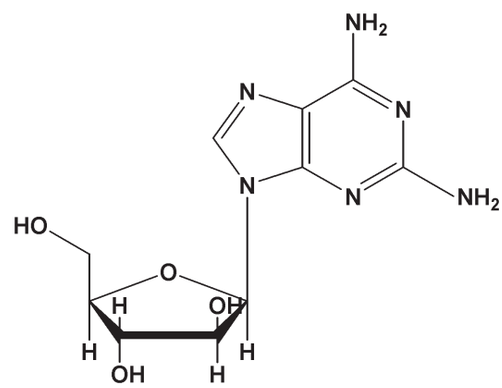
Catalogue number: 203-06

Description: white to off-white crystals

Storage of dry compound: 3 years at +4°C

Price:	10 g	350 euro
	100 g	2 500 euro

For **bulk quantities please enquire**



Arabinofuranosyl-guanine

9-(β-D-arabinoribofuranosyl)-guanine **CAS number:** 38819-10-2

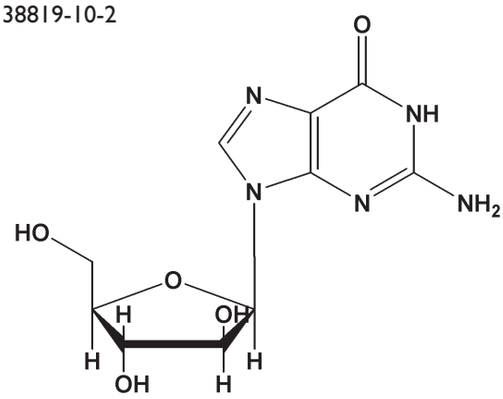
Catalogue number: 203-03

Description: white to off-white crystals

Storage of dry compound: 3 years at +4°C

Price:	10 g	500 euro
	100 g	3 000 euro

For **bulk quantities please enquire**



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Purine Modified Nucleosides

Arabinofuranosyl-hypoxanthine

9-(β-D-arabinoribofuranosyl)-hypoxanthine

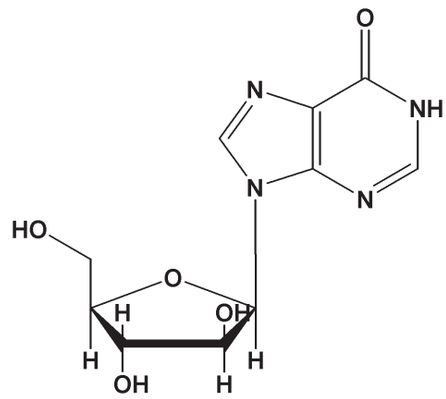
Catalogue number: 203-02

Description: white to off-white crystals

Storage of dry compound: 3 years at +4°C

Price:	10 g	500 euro
	100 g	3 000 euro

For **bulk quantities please enquire**



6-Benzylaminopurine 2'-deoxyriboside

9-(2-deoxy- β-D-ribofuranosyl)-6-Benzylaminopurine

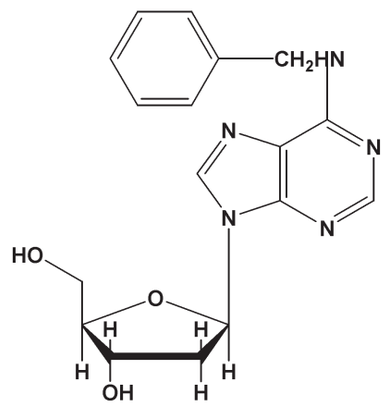
Catalogue number: 203-21

Description: white to off-white crystals

Storage of dry compound: 3 years at +4°C

Price:	1 g	300 euro
	10 g	2 000 euro
	100 g	12 000 euro

For **bulk quantities please enquire**



6-Benzylaminopurine riboside

9-(β-D-ribofuranosyl)-6-Benzylaminopurine **CAS number:** 4294-16-0

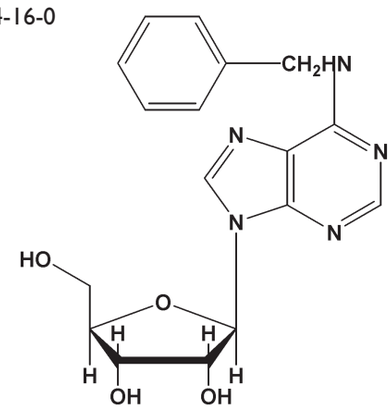
Catalogue number: 203-22

Description: white to off-white crystals

Storage of dry compound: 3 years at +4°C

Price:	10 g	500 euro
	100 g	3 200 euro

For **bulk quantities please enquire**



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Purine Modified Nucleosides

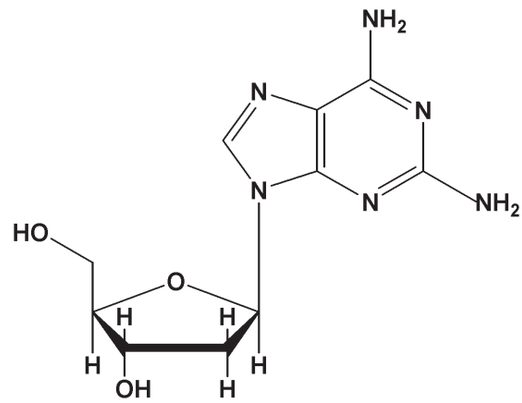
2,6-Diaminopurine 2'-deoxyriboside

9-(2-deoxy-β-D-ribofuranosyl)-2,6-Diaminopurine

Catalogue number: 203-23
Description: white to off-white crystals
Storage of dry compound: 3 years at +4°C

Price:	1 g	300 euro
	10 g	1 500 euro
	100 g	5 000 euro

For **bulk quantities please enquire**



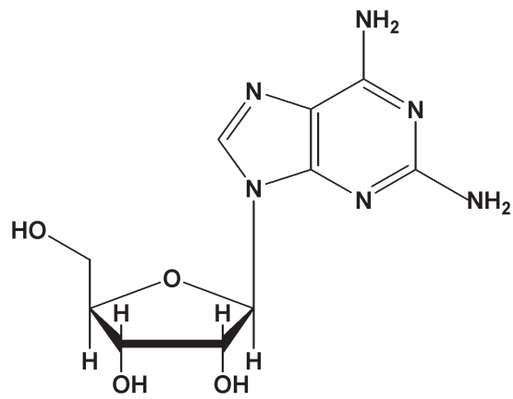
2,6-Diaminopurine riboside

9-(β-D-ribofuranosyl)-2,6-Diaminopurine

Catalogue number: 203-24
Description: white to off-white crystals
Storage of dry compound: 3 years at +4°C

Price:	10 g	400 euro
	100 g	1 200 euro

For **bulk quantities please enquire**



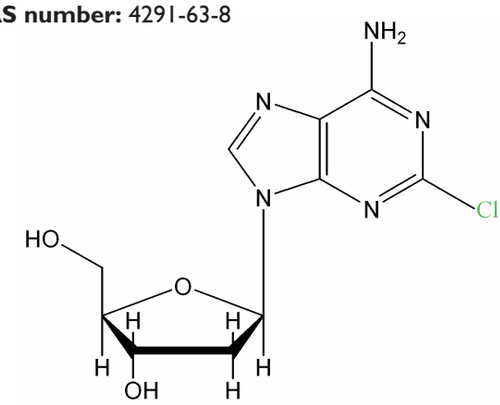
2-Chloro-2'-deoxyadenosine (2-CdA, cladribine)

9-(2-deoxy-β-D-ribofuranosyl)-2-chloro-adenine **CAS number:** 4291-63-8

Catalogue number: 203-25
Description: white to off-white crystals
Storage of dry compound: 3 years at +4°C

Price:	1 g	500 euro
	10 g	3 500 euro
	100 g	21 000 euro

For **bulk quantities please enquire**

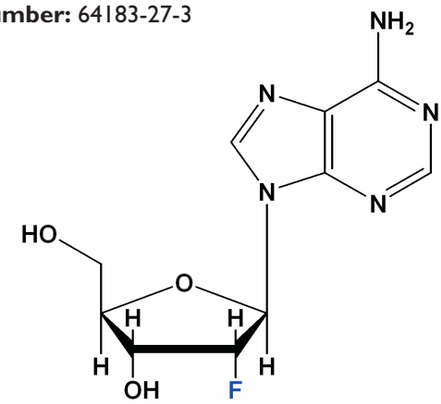


METKINEN
chemistry

Purine Modified Nucleosides

2'-Fluoro-2'-deoxyadenosine

9-(2-fluoro-2-deoxy-β-D-ribofuranosyl)-adenine **CAS number:** 64183-27-3

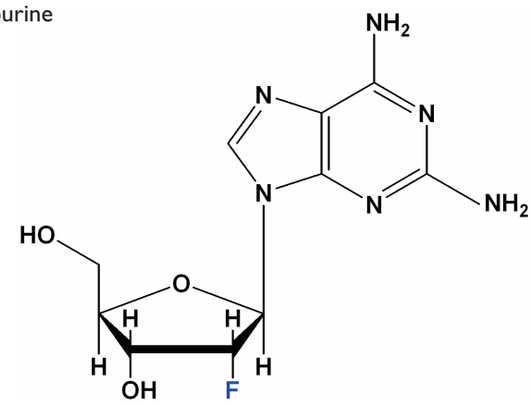


Catalogue number: 203-17
Description: white to off-white crystals
Storage of dry compound: 3 years at +4°C

Price:	1 g	380 euro	380 euro/g
	10 g	1 800 euro	180 euro/g
	100 g	9 000 euro	90 euro/g
	500 g	27 000 euro	54 euro/g
	1000 g	45 000 euro	45 euro/g

2'-Fluoro-2'-deoxy-2,6-diaminopurineriboside

9-(2-fluoro-2-deoxy-β-D-ribofuranosyl)-2,6-diaminopurine



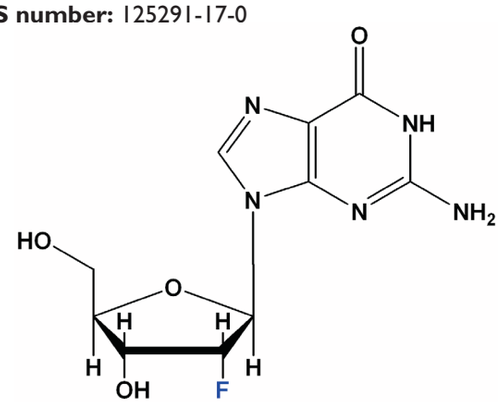
Catalogue number: 203-18
Description: white to off-white crystals
Storage of dry compound: 3 years at +4°C

Price:	1 g	450 euro
	10 g	2 300 euro
	100 g	11 000 euro

For **bulk quantities please enquire**

2'-Fluoro-2'-deoxyguanosine

9-(2-fluoro-2-deoxy-β-D-ribofuranosyl)-guanine **CAS number:** 125291-17-0



Catalogue number: 203-15
Description: white to off-white crystals
Storage of dry compound: 3 years at +4°C

Price:	1 g	500 euro	500 euro/g
	10 g	2 500 euro	250 euro/g
	100 g	12 000 euro	120 euro/g
	500 g	32 500 euro	65 euro/g
	1000 g	55 000 euro	55 euro/g

METKINEN
chemistry

Purine Modified Nucleosides

2'-Fluoro-2'-deoxyinosine

9-(2-fluoro-2-deoxy-β-D-ribofuranosyl)-hypoxanthine

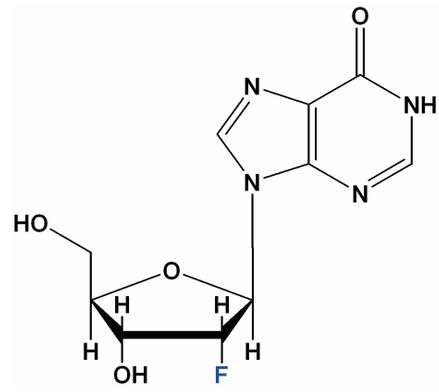
Catalogue number: 203-14

Description: white to off-white crystals

Storage of dry compound: 3 years at +4°C

Price:	1 g	500 euro
	10 g	2 500 euro
	100 g	12 000 euro

For **bulk quantities please enquire**



Kinetin riboside

9-(β-D-ribofuranosyl)-6-furfurylaminopurine **CAS number:** 4338-47-0

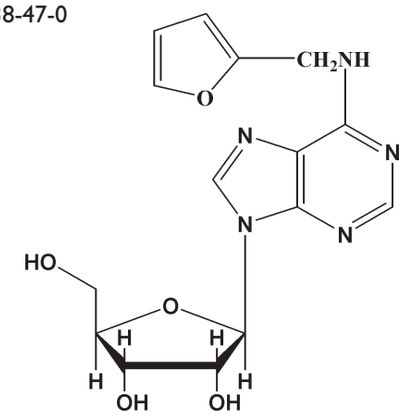
Catalogue number: 203-26

Description: white to off-white crystals

Storage of dry compound: 3 years at +4°C

Price:	10 g	500 euro
	100 g	3 200 euro

For **bulk quantities please enquire**



METKINEN
chemistry

Custom synthesis

2'-FLUORO-RNA

The 2'-F-RNA oligonucleotide adopts an A-form helix on hybridization to a target. Whereas a hydroxyl group of RNA is a hydrogen bond donor, fluorine of 2'-F-RNA appears to be a weak acceptor. These features of 2'-F-RNA oligonucleotides lead to certain interesting properties. For example, it was demonstrated that oligonucleotides hybridize to a RNA oligonucleotide in the following order of increasing stability: DNA < RNA < 2'-OMe-RNA < 2'-F-RNA.[1]

The Aptamers, composed of 2'-F–RNA bind targets with higher affinities and are more resistant to nucleases, compared to RNA aptamers.[2] In addition to that, 2'-F-RNA can be effectively used in siRNA applications. It was clearly demonstrated that siRNA synthesized with 2'-F pyrimidine nucleosides were more inhibitory and showed considerably increased stability in human plasma compared to siRNA.[3] They were absolutely functional in both, cell culture and in vivo.

Today it appears obvious that 2'-F-RNA as well as other chemically modified small oligonucleotides will definitely find a number applications in research and new drug development, especially in **RNA interference** for specific silencing of genes in cells and in vivo.[4]

Metkinen Chemistry is making available a **custom synthesis of RNA oligonucleotides, incorporating both pyrimidine and purine 2'-F-nucleosides, as well as natural ribonucleosides and some nucleoside analogs.** All our modified RNAs are purified by HPLC and characterized with Electrospray Ionization Mass Spectra.

References:

1. A.M. Kawasaki, et al., *J. Med. Chem.*, **1993**, *36*, 831-841.
2. M. Khati, M. Schuman, J. Ibrahim, Q. Sattentau, S. Gordon, and W. James, *J. Virol.*, **2003**, *77*, 12692-12698.
3. J.M. Layzer, A.P. McCaffrey, A.K. Tanner, Z. Huang, M.A. Kay, and B.A. Sullenger, *RNA*, **2004**, *10*, 766-771.
4. M. Manoharan, *Current Opinion in Chem. Biol.*, **2004**, *8*, 570 – 579

METKINEN
chemistry

Custom synthesis

GENERATION OF OLIGONUCLEOTIDE LIBRARIES

Large peptide libraries for drug discovery programs can be created using phage display methods. Oligonucleotide-directed mutagenesis is the favored method for preparing these libraries. For this purpose randomized oligonucleotides are usually prepared using mixtures of nucleotides at each step of synthesis. However, this strategy often results in incorporation of unwanted amino acids, or stop codons in to the sequence. A significant solution to this problem has been introduction of trimeric nucleotide building blocks, which correspond to desired codons.[1] **Metkinen Chemistry** is ready to perform a custom synthesis of randomized oligonucleotide libraries, employing our thoroughly characterized (electrospray mass spectrometry[2]) trimer (codon) phosphoramidites.

References:

1. A.L. Kayushin, M.D Korosteleva, A.I. Miroshnikov, W. Kosch, D. Zubov and N. Piel, *Nucleic Acid Research*, **1996**, *24*, 3748-3755.
2. T. Mauriala, S. Auriola, A. Azhayev, A. Kayushin, M. Korosteleva, M., A. Miroshnikov *J. Pharm.Biomed. Anal.*, **2004**, *34*, 199-206.

SPECIALTY PHOSPHORAMIDITES

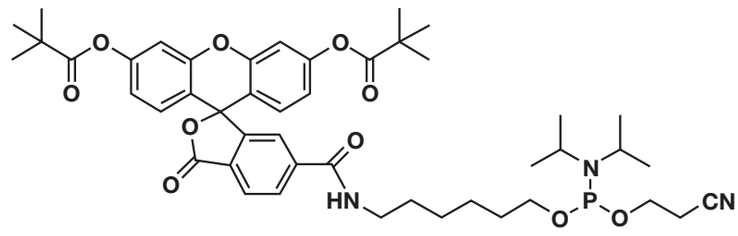
Specialty phosphoramidites derived from modified purine nucleosides listed above are available upon special request. For prices and terms of delivery please inquire.

METKINEN
chemistry

FAM Phosphoramidities

5'-Fluorescein phosphoramidite (I)

[(3',6'-dipivaloylfluoresceinyl)-6-carboxamidohexyl]-1-O-(2-cyanoethyl)-(N,N-diisopropyl)-phosphoramidite



Catalogue Number: 303-01

Diluent: Anhydrous Acetonitrile

Coupling: 3 minute coupling time recommended

Deprotection: No changes needed from standard method recommended by synthesizer manufacturer.

Storage: Freezer storage, -10 to -30°C, dry.

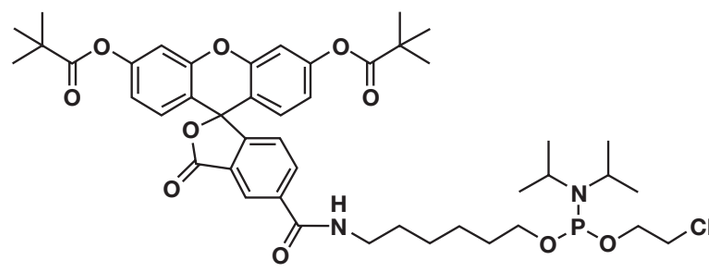
Stability in Solution: 2-3 days, <90% efficient after 4 days

Price:	1 g	900 euro
	10 g	5 600 euro
	100 g	22 500 euro

Store labeled oligo in the dark, either dry or in a neutral aqueous media at -20°C.
Do not store crude fluorescently labeled oligonucleotides in ammonia.

5'-Fluorescein phosphoramidite (II)

[(3',6'-dipivaloylfluoresceinyl)-5-carboxamidohexyl]-1-O-(2-cyanoethyl)-(N,N-diisopropyl)-phosphoramidite



Catalogue Number: 303-02

Diluent: Anhydrous Acetonitrile

Coupling: 3 minute coupling time recommended

Deprotection: No changes needed from standard method recommended by synthesizer manufacturer.

Storage: Freezer storage, -10 to -30°C, dry.

Stability in Solution: 2-3 days, <90% efficient after 4 days

Price:	1 g	900 euro
	10 g	5 600 euro
	100 g	22 500 euro

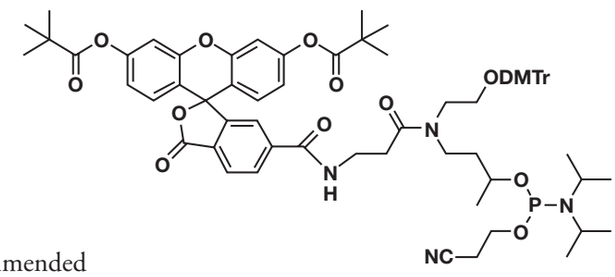
Store labeled oligo in the dark, either dry or in a neutral aqueous media at -20°C.
Do not store crude fluorescently labeled oligonucleotides in ammonia.

METKINEN
chemistry

FAM Phosphoramidities

Fluorescein phosphoramidite (I)

1-Dimethoxytrityloxy-3-aza-3-[1-oxo-3-{(di-O-pivaloyl-fluorescein)-6-carboxamido}propyl]heptyl-6-O-(2-cyanoethyl)-(N,N-diisopropyl)-phosphoramidite



Catalogue Number: 303-03

Diluent: Anhydrous Acetonitrile

Coupling: 10 minutes coupling time recommended

Deprotection: No changes needed from standard method recommended by synthesizer manufacturer.

Storage: Freezer storage, -10 to -30°C, dry.

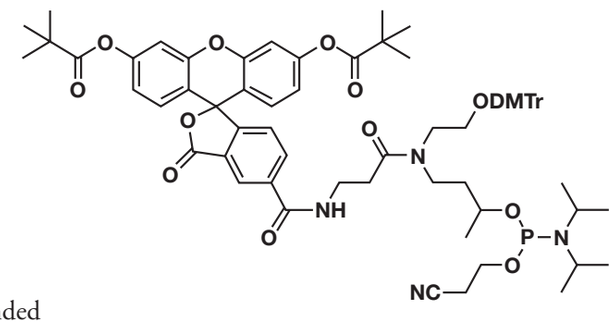
Stability in Solution: 7-10 days, <90% efficient after 14 days

Price:	1 g	900 euro
	10 g	5 600 euro
	100 g	22 500 euro

Store labeled oligo in the dark, either dry or in a neutral aqueous media at -20°C.
Do not store crude fluorescently labeled oligonucleotides in ammonia.

Fluorescein phosphoramidite (II)

1-Dimethoxytrityloxy-3-aza-3-[1-oxo-3-{(di-O-pivaloyl-fluorescein)-5-carboxamido}propyl]heptyl-6-O-(2-cyanoethyl)-(N,N-diisopropyl)-phosphoramidite



Catalogue Number: 303-04

Diluent: Anhydrous Acetonitrile

Coupling: 10 minutes coupling time recommended

Deprotection: No changes needed from standard method recommended by synthesizer manufacturer.

Storage: Freezer storage, -10 to -30°C, dry.

Stability in Solution: 7-10 days, <90% efficient after 14 days

Price:	1 g	900 euro
	10 g	5 600 euro
	100 g	22 500 euro

Store labeled oligo in the dark, either dry or in a neutral aqueous media at -20°C.
Do not store crude fluorescently labeled oligonucleotides in ammonia.

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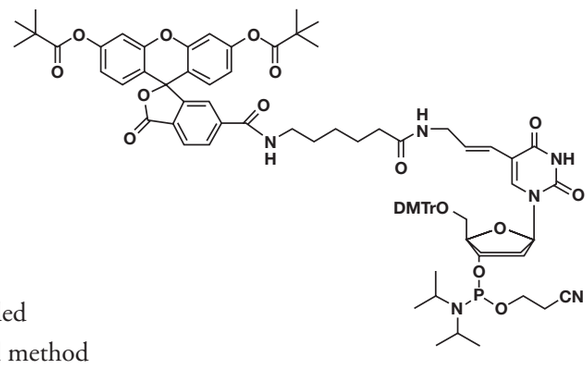
chemistry

FAM Phosphoramidities

6-Fluorescein dT phosphoramidite

5'-Dimethoxytrityl-5-[3-(6-(di-O-pivaloyl-fluoresceinyl-6-carboxamido)hexanoylamido)-propenyl]uridine, 3'-(2-cyanoethyl)-(N,N-diisopropyl)-phosphoramidite



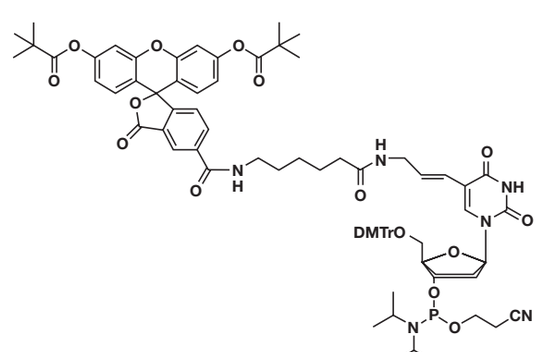
Catalogue Number: 303-05
Diluent: Anhydrous Acetonitrile
Coupling: 10 minutes coupling time recommended
Deprotection: No changes needed from standard method recommended by synthesizer manufacturer.
Storage: Freezer storage, -10 to -30°C, dry.
Stability in Solution: 7-10 days, <90% efficient after 14 days

Price:	1 g	1 000 euro
	10 g	6 000 euro

Store labeled oligo in the dark, either dry or in a neutral aqueous media at -20°C.
Do not store crude fluorescently labeled oligonucleotides in ammonia.

5-Fluorescein dT phosphoramidite

5'-Dimethoxytrityl-5-[3-(6-(di-O-pivaloyl-fluoresceinyl-5-carboxamido)hexanoylamido)-propenyl]uridine, 3'-(2-cyanoethyl)-(N,N-diisopropyl)-phosphoramidite



Catalogue Number: 303-06
Diluent: Anhydrous Acetonitrile
Coupling: 10 minutes coupling time recommended
Deprotection: No changes needed from standard method recommended by synthesizer manufacturer.
Storage: Freezer storage, -10 to -30°C, dry.
Stability in Solution: 7-10 days, <90% efficient after 14 days

Price:	1 g	1 000 euro
	10 g	6 000 euro

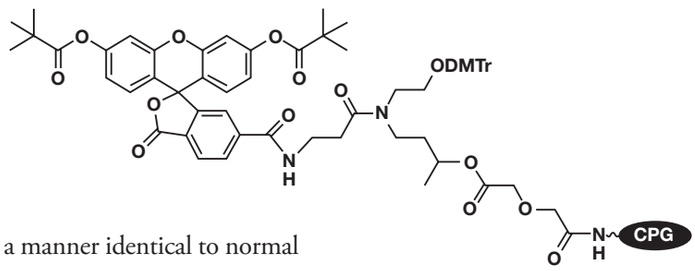
Store labeled oligo in the dark, either dry or in a neutral aqueous media at -20°C.
Do not store crude fluorescently labeled oligonucleotides in ammonia.

METKINEN
chemistry

FAM Phosphoramidities

3'-(6-Fluorescein)-CPG

1-Dimethoxytrityloxy-3-aza-3-[1-oxo-3-{(di-O-pivaloyl-fluorescein)-5-carboxamido}propyl]heptyl-6-O-diglycoyl-long chain alkylamino-CPG



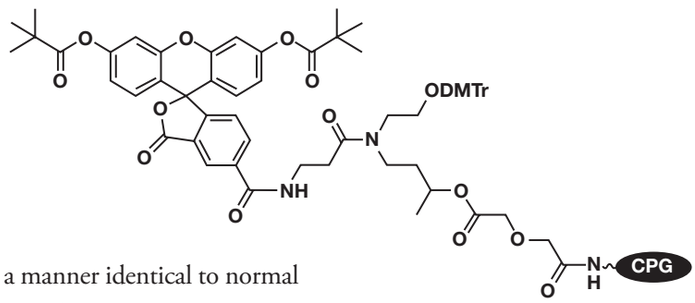
Catalogue Number: 303-07
Diluent: Not Applicable
Coupling: This support should be used in a manner identical to normal protected nucleoside support since it contains the DMT group.
Deprotection: No changes needed from standard method recommended by synthesizer manufacturer.
Storage: Freezer storage, -10 to -30°C, dry.
Stability in Solution: Not Applicable

Price:	1 g	590 euro
	10 g	2 900 euro

Store labeled oligo in the dark, either dry or in a neutral aqueous media at -20°C.
Do not store crude fluorescently labeled oligonucleotides in ammonia.

3'-(5-Fluorescein)-CPG

1-Dimethoxytrityloxy-3-aza-3-[1-oxo-3-{(di-O-pivaloyl-fluorescein)-5-carboxamido}propyl]heptyl-6-O-diglycoyl-long chain alkylamino-CPG



Catalogue Number: 303-08
Diluent: Not Applicable
Coupling: This support should be used in a manner identical to normal protected nucleoside support since it contains the DMT group.
Deprotection: No changes needed from standard method recommended by synthesizer manufacturer.
Storage: Freezer storage, -10 to -30°C, dry.
Stability in Solution: Not Applicable

Price:	1 g	590 euro
	10 g	2 900 euro

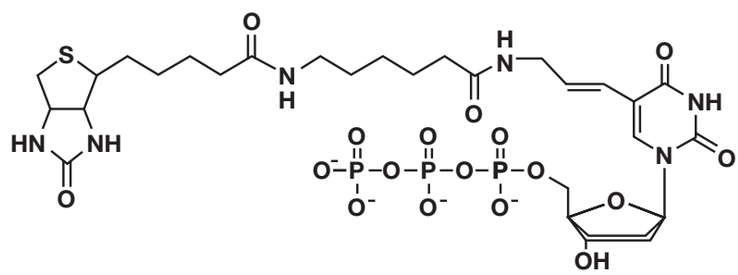
Store labeled oligo in the dark, either dry or in a neutral aqueous media at -20°C.
Do not store crude fluorescently labeled oligonucleotides in ammonia.

METKINEN
chemistry

Labeled 2'-Deoxyuridine 5'-Triphosphates

Biotin-11-dUTP

5-[3-(6-(Biotinylamido)hexanoylamido)propenyl]-2'-deoxyuridine-5'-triphosphate, tetra(triethylammonium) or tetralithium (optional) salt



Catalogue Number: 303-09

Diluent: dd-H_2O

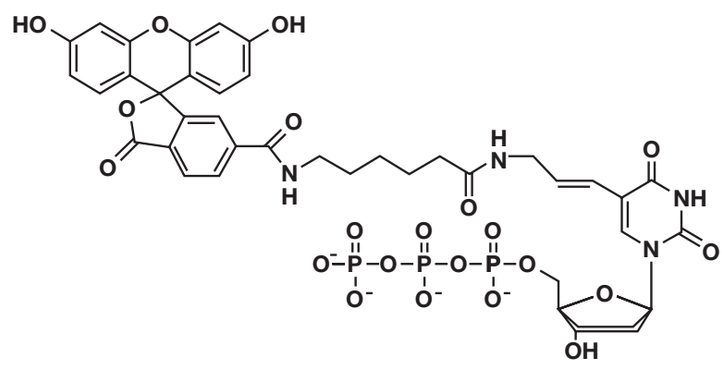
Storage: Freezer storage, -10 to -30°C, dry.

Stability in Solution: 1 month as 1 mM solution in water or neutral buffers at +4°C, 6 months as 1 mM solution in water or neutral buffers at -20°C.

Price:	1 mg	750 euro
	10 mg	3 750 euro
	100 mg	15 000 euro

6-FAM-11-dUTP

5-[3-(6-(fluoresceinyl-6-carboxamido)hexanoylamido)-propenyl]-2'-deoxyuridine-5'-triphosphate, tetra(triethylammonium) or tetralithium (optional) salt



Catalogue Number: 303-12

Diluent: dd-H_2O

Storage: Freezer storage in dark, -10 to -30°C, dry.

Stability in Solution: 1 month as 1 mM solution in water or neutral buffers at +4°C in dark, 6 months as 1 mM solution in water or neutral buffers at -20°C in dark.

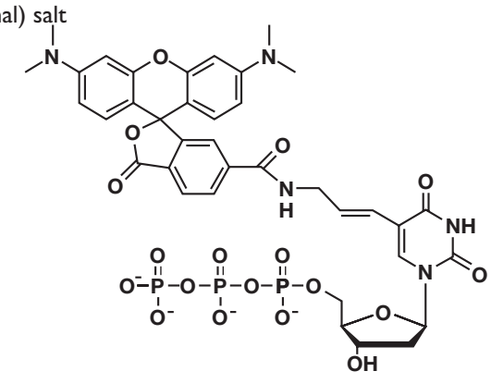
Price:	1 mg	750 euro
	10 mg	3 750 euro
	100 mg	15 000 euro

METKINEN
chemistry

Labeled 2'-Deoxyuridine 5'-Triphosphates

6-TAMRA-5-dUTP

5-[3-(tetramethylrhodamine-6-carboxamido)propenyl]-2'-deoxyuridine-5'-triphosphate, tetra(triethylammonium) or tetralithium (optional) salt



Catalogue Number: 303-10 **Diluent:** dd-H_2O

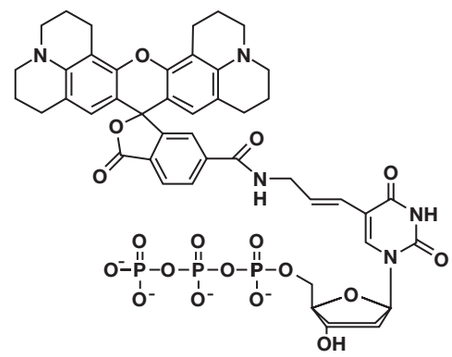
Storage: Freezer storage in dark, -10 to -30°C, dry.

Stability in Solution: 1 month as 1 mM solution in water or neutral buffers at +4°C in dark, 6 months as 1 mM solution in water or neutral buffers at -20°C in dark.

Price:	1 mg	1 200 euro
	10 mg	6 000 euro
	100 mg	24 000 euro

6-ROX-5-dUTP

5-[3-(X-Rhodamine-6-carboxamido)propenyl]-2'-deoxyuridine-5'-triphosphate, tetra(triethylammonium) or tetralithium (optional) salt



Catalogue Number: 303-11 **Diluent:** dd-H_2O

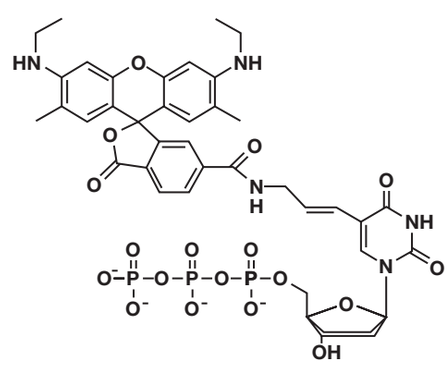
Storage: Freezer storage in dark, -10 to -30°C, dry.

Stability in Solution: 1 month as 1 mM solution in water or neutral buffers at +4°C in dark, 6 months as 1 mM solution in water or neutral buffers at -20°C in dark.

Price:	1 mg	1 200 euro
	10 mg	6 000 euro
	100 mg	24 000 euro

R6G-5-dUTP

5-[3-(rhodamine6G-6-carboxamido)propenyl]-2'-deoxyuridine-5'-triphosphate, tetra(triethylammonium) or tetralithium (optional) salt



Catalogue Number: 303-13 **Diluent:** dd-H_2O

Storage: Freezer storage in dark, -10 to -30°C, dry.

Stability in Solution: 1 month as 1 mM solution in water or neutral buffers at +4°C in dark, 6 months as 1 mM solution in water or neutral buffers at -20°C in dark.

Price:	1 mg	1 200 euro
	10 mg	6 000 euro
	100 mg	24 000 euro

ORDERING INFORMATION

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info@metkinenchemistry.com
www.metkinenchemistry.com

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