

# Product Specifications

Custom Oligo Synthesis, antisense oligos, RNA oligos, chimeric oligos, Fluorescent dyes, Affinity Ligands, Spacers & Linkers, Duplex Stabilizers, Minor bases, labeled oligos, Molecular Beacons, siRNA, phosphonates Locked Nucleic Acids (LNA); 2'-5' linked Oligos

## Oligo Modifications

For research use only. Not for use in diagnostic procedures for clinical purposes.

## **ICG NHS**

Category Fluorescent Dyes

Modification Code ICG-N

Reference Catalog Number 26-6522

5 Prime Y

3 Prime Y

Internal Y

Molecular Weight(mw) 734.05

5'-Amino C6 example
Other amino mods available

HO. p.
0.
0.
wwoOligo -3' or 5'

ICG Indocyanine Green NHS [26-6522-XX]

#### Click here for a list of fluorophores.

This modification is a post synthesis conjugation to a primary amino group thus an additional modification with an amino group is required. A C3, C6 or C12 amino group can be placed at the 5' or for the 3' end a C3 or C7 amino and for internal positions an amino modified base is used, e.g Amino dT C6.

Yield of Post Synthesis NHS, Maleimide & Click Ligand Conjugation\* Oligo Scale of Synthesis Yield, nmols 50 nmol 2 nmol 200 nmol 5 nmol 1 umol 16 nmol 2 umol 30 nmol 5 umol 75 nmol 10 umol 150 nmol 15 umol 225 nmol \* The yield will be lower for oligos longer than 50mer. Click here for yield table of long oligos. \* Click here for RNA Oligos scale of synthesis and yield. **NHS Ligand conjugation** requires a primary amino group. Gene Link offers a wide selection of amino modifications for 5', 3' and internal sites. Click here for a list of conjugation chemistry modifications. **Maleimide Ligand conjugation** requires a thiol group. Gene Link offers a wide selection of thiol modifications for 5', 3' and internal sites.



Click here for a list of conjugation chemistry modifications. **Click Chemistry Ligand conjugation** requires a corresponding Click modification; examples Alkyne:Azide, Azide:DBCO, BCN:Azide, BCN: TCO:Tetrazine. Gene Link offers a wide selection of click modifications for 5', 3' and internal sites. Click here for a list of click chemistry modifications.

#### ICG (Indocyanine Green) NHS

ICG NHS ester is an advanced, amine-reactive form of the near-infrared (NIR) fluorescent dye Indocyanine Green (ICG), specifically designed for stable and efficient biomolecule labeling in bioimaging applications. This modified version of ICG combines the exceptional optical properties of the parent compound with enhanced reactivity, making it an invaluable tool for researchers in various fields of life sciences and medical imaging. The dye can be effectively excited using 750-800 nm laser lines or LED sources, positioning it in the optimal NIR region for deep tissue imaging. This spectral profile allows for visualization of structures several millimeters below the skin surface, significantly reducing background autofluorescence and enhancing signal-to-noise ratios in biological samples. The NHS ester reactive group readily forms stable amide bonds with primary amines found in proteins, antibodies, and amino-modified oligonucleotides. This reactivity ensures efficient and durable labeling of various biomolecules, including the ε-amino groups of lysine residues and the amine termini of nucleotides.

#### Cyanine 7.5 Sulfo is a suitable substitute for Indocyanine Green NHS.

## Cyanine 7.5 Sulfo

Cyanine7.5 Sulfo is a near-infrared water-soluble and hydrophilic dye for the NIR imaging applications. The structure and spectra of the dye resemble indocyanine green (ICG) which has been approved for use in humans for years. However, unlike ICG, sulfo-Cyanine7.5 contains a trimethylene bridge that increases its quantum yield compared to ICG and has a linker arm for its attachment to proteins, peptides, and other molecules. This derivative is an NHS ester for the modification of amine groups.

#### Cyanine7.5

Cyanine 7.5 (Cy7.5) NHS ester is a fluorescent dye that belongs to the Cyanine family of synthetic polymethine dyes. Cy7.5 is reactive, water-soluble, and has an absorbance maximum of 788 nm and an emission maximum of 808 nm, which is in the near IR. It is available as an NHS ester, and is used to fluorescently label oligonucleotides at either the 5' or 3' end, or internally. As a near IR dye, Cy7.5 has very little background fluorescence associated with it (1). It is thus an excellent choice for labeling oligo probes slated for in vivo applications, because the minimal scattering and absorption of near-IR photons by cellular tissue ensures higher S/N ratio, and better sensitivity. For example, Fluorescent Resonance Energy Transfer (FRET) oligonucleotide duplexes using Cy5.5 as the donor on one strand and Cy7.5 as the acceptor on the complementary strand have been used to detect and characterize transcription factor NF-kappaB p50 protein binding to DNA (2)

Caution: Cy7.5 is intensely colored and very reactive. Care should be exercised when handling the vial containing the C7.5-labeled oligo to avoid staining clothing, skin, and other items. Also, because Cy7.5 is in the form of an NHS ester, the oligo first must be synthesized with an Amino C6 Linker (for the ends) or the Amino C6 version of the base phosphoramidite (for internal labeling). The Cy7.5-NHS ester is then manually attached to the oligo through the amino group in a separate reaction post-synthesis.

Near Infrared Fluorophore Spectral Data & Quencher Selection Guide

Fluorophore Name
Absorbance Max, nm +/-10
Emission Max, nm +/-10
Extinction Coefficient*
Color**
Quencher
Cy5 650 665 250,000
IRDye 650 NHS 650 665 230,000
AZ647 NHS 655 680 191,800
AZ680 NHS 678 701 185,000
Cy5.5 684 710 198,000

IRDye 700 NHS 684 710 288,000

AZdye700 NHS 696 719 192,000
Atto 700 NHS 700 716 120,000
Atto 725 NHS 728 751 120,000
Atto 740 NHS 743 763 120,000
Cy7 NHS 740 773 199,000
IRDye 750 NHS 756 776 260,000
cy7.5 NHS 788 808 223,000
IRDye 800 NHS 795 819 240,000

\* Extinction coefficient at  $\lambda$  (max) in cm-1M-1. \*\* Typical emission color seen through the eyepiece of a conventional fluorescence microscope with appropriate filters. Near-IR region. Human vision is insensitive to light beyond ~650 nm; it is not possible to view near-IR fluorescent dyes.

Click here for a list of fluorophores.

Click here for list of quenchers.

- References
- 1. Benson, R.C., Kues, H.A. Absorption and Fluorescence Properties of Cyanine Dyes. *J. Chem. Eng. Data* (1977), 22: 379-383.
- 2. Zhang, S., Metelev, V., Tabatadze, D., Zamecnik, P.C., Bogdanov, A. Fluorescence resonance energy transfer in near-infrared fluorescent oligonucleotide probes for detecting protein-DNA interactions. *Proc. Nat. Acad. Sci. USA.* (2008), 105: 4156-4161.

