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**Product Information: ATTO 647N**

**ATTO 647N** belongs to a new generation of fluorescent labels for the red spectral region. Characteristic features of the label are strong absorption, excellent fluorescence quantum yield, high photostability, excellent ozone resistance, good solubility, and very little triplet formation. The dye is highly suitable for single-molecule detection applications and high-resolution microscopy.

**ATTO 647N** is a cationic dye. After coupling to a substrate the dye carries a net electrical charge of +1. In common with most **ATTO**-labels, absorption and fluorescence are independent of pH in the range of 2 to 11, used in typical applications.

As supplied **ATTO 647N** consists of a mixture of two isomers with practically identical absorption and fluorescence properties. For details of coupling see our recommended labeling procedure at [www.atto-tec.com](http://www.atto-tec.com) - Support - Downloads - [General Procedures](#).

**Optical data of the carboxy derivative (in PBS, pH 7.4):**

$$\lambda_{\text{abs}} = 646 \text{ nm}$$

$$\epsilon_{\text{max}} = 1.5 \times 10^5 \text{ M}^{-1} \text{ cm}^{-1}$$

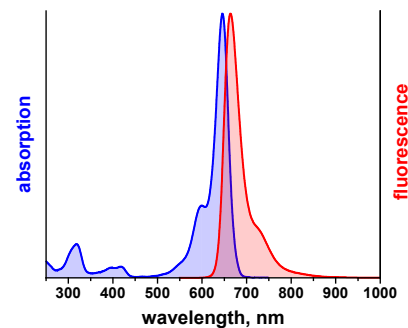
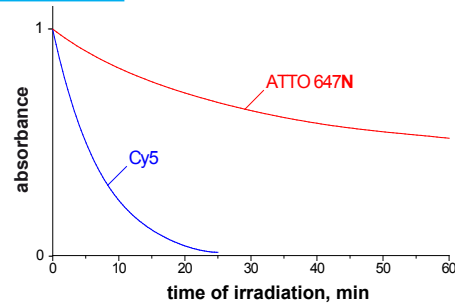
$$\lambda_{\text{fl}} = 664 \text{ nm}$$

$$\eta_{\text{fl}} = 65 \%$$

$$\tau_{\text{fl}} = 3.5 \text{ ns}$$

$$\text{CF}_{260} = 0.04$$

$$\text{CF}_{280} = 0.03$$



Spectra available in digitized form (excel file) on <http://www.atto-tec.com>

Modification	MW, g/mol	M <sup>+</sup> , g/mol	Order Code	
			Unit (1 mg)	Unit (5 mg)
carboxy	746	646	AD 647N-21	AD 647N-25
NHS-ester	843	743	AD 647N-31	AD 647N-35
maleimide	868	768	AD 647N-41	AD 647N-45
biotin	1057	956	AD 647N-71	AD 647N-75
phalloidin	1530	1415	AD 647N-81*	AD 647N-82**
amine	889	688	AD 647N-91	AD 647N-95
azide	960	846	AD 647N-101	AD 647N-105
iodoacetamide	956	856	AD 647N-111	AD 647N-115
hydrazide <i>new</i>	861	660	AD 647N-121	AD 647N-125
alkyne	783	683	AD 647N-141	AD 647N-145
cadaverine	958	731	AD 647N-231	AD 647N-235
tetrazine	930	830	AD 647N-2502#	AD 647N-2505##

\*10 nmol    \*\*20 nmol    #0.2 mg    ##0.5 mg

## General Information

**Storage:** The product is shipped solvent-free at ambient temperature. Upon receipt store at -20 °C. To avoid moisture condensation onto the product, vial must be equilibrated to room temperature before opening. When stored properly, protected from moisture and light, ATTO-TEC products are stable for at least three years.

**Risk and safety:** A material safety data sheet (MSDS) of each derivative can be downloaded from our website at [www.atto-tec.com](http://www.atto-tec.com).

**Solutions:** The product is soluble in polar solvents, e.g. dimethylformamide (DMF), dimethylsulfoxide (DMSO), or acetonitrile. However, due to their inherent reactivity, NHS-esters and maleimides must be well protected from OH-containing solvents like ethanol and, in particular, water. Prepare labeling solutions of NHS-esters and maleimides immediately before use by dissolving the vial content in anhydrous and amine-free DMF or DMSO. Depending on the quality of the solvent used, such solutions may be of limited stability.

Dye with **free carboxy group (COOH)** may be used for any kind of spectroscopy. Due to the high extinction coefficient and its high quantum yield of fluorescence this product is suitable for high-sensitivity detection including single-molecule work. The dye can be activated at the carboxy group for coupling purposes.

The **NHS-ester** of the dye reacts easily with amino-groups of proteins and other bio-molecules. Since the amino-group must be non-protonated to be reactive, the pH of the reaction solution has to be adjusted sufficiently high. As with all NHS-esters unavoidable hydrolysis takes place at high pH and competes with the desired labeling reaction. Therefore the solution has to be buffered carefully. For details see the Labeling Protocol on [www.atto-tec.com](http://www.atto-tec.com).

The **maleimide** is suitable for labeling sulfhydryl (thiol) groups of proteins, in particular cystein residues. See Labeling Protocol on [www.atto-tec.com](http://www.atto-tec.com).

The **biotin** derivative can be used as reagent for binding to proteins like avidin and streptavidin.

**Phalloidin**, a bicyclic heptapeptide, is a very strong binding reagent to actin. Fluorescent labeled phalloidin has become a useful tool to investigate the distribution of F-actin within the cytoskeleton of cells by fluorescence microscopy. To prepare a stock solution of the phalloidin-conjugate it is recommended dissolving the sample in 1 ml of methanol.

The **amine** derivative may be used for reactions with activated carboxy-groups like NHS-esters, TFP-esters etc.

The **azide** or **alkyne** modification is used in the Huisgen reaction ("Click Chemistry").

The **iodoacetamide** derivative reacts, like the maleimide, with a sulfhydryl group forming a thioether bond. It is predominantly used for tagging cystein residues of proteins.

The **hydrazide** derivative is used to modify aldehydes and ketones.

The **cadaverine** derivative can be used as a fluorescent amine donor substrate for transglutaminases.

The **tetrazine** derivative readily reacts in a bioorthogonal way with strained alkenes or alkynes such as trans-cyclooctenes (TCO) or cyclooctynes like bicyclo[6.1.0]non-4-yne (BCN), respectively.

### **Further Notes:**

- ATTO-TEC products are high-quality reagents intended for research purposes only.
- The use of ATTO-TEC products must be supervised by technically qualified personnel experienced in handling potentially hazardous chemicals. For safety instructions please read the corresponding Material Safety Data Sheet.
- Most ATTO-TEC products and product applications are covered by European and foreign patents.
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