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**Product Information: ATTO 465**

**ATTO 465** is a novel fluorescent label derived from the well-known dye Acriflavine. The dye is intended for application in the area of life science, e.g. labeling of DNA, RNA or proteins. Characteristic features of the label are strong absorption and good fluorescence, large Stokes-shift, good water solubility and high triplet quantum yield. The dye shows intense and long-lived phosphorescence in solid matrix. 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):**

$$\lambda_{\text{abs}} = 453 \text{ nm}$$

$$\epsilon_{\text{max}} = 7.5 \times 10^4 \text{ M}^{-1} \text{ cm}^{-1}$$

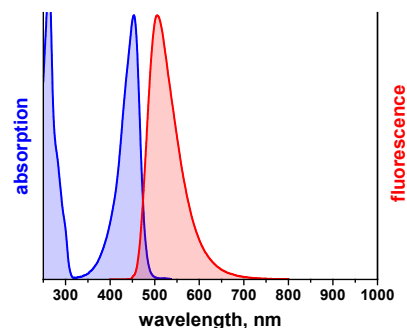
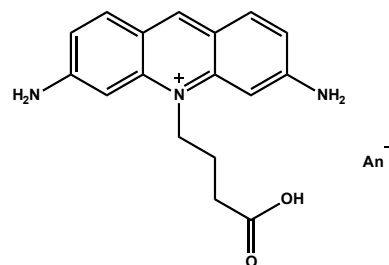
$$\lambda_{\text{fl}} = 506 \text{ nm}$$

$$\eta_{\text{fl}} = 75 \%$$

$$\tau_{\text{fl}} = 5.0 \text{ ns}$$

$$\text{CF}_{260} = 1.09$$

$$\text{CF}_{280} = 0.48$$



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	396	296	AD 465-21	AD 465-25
NHS-ester	493	393	AD 465-31	AD 465-35
maleimide	518	418	AD 465-41	AD 465-45
biotin	706	606	AD 465-71	AD 465-75
phalloidin	1179	1065	AD 465-81*	AD 465-82**
azide <i>new</i>	610	496	AD 465-101	AD 465-105
tetrazine (MeTet) <i>new</i>	593	479	AD 465-2502 <sup>#</sup>	AD 465-2505 <sup>##</sup>

\* 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).

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

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

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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