

Tetramethylrhodamine isothiocyanate (TRITC) mixed isomers

CAS Nr.: 95197-95-8

Empirical Formula: C₂₅H₂₁N₃O₃S

MW: 443.52 g/mol (excluding Chloride anions)

Structure:

Fig. 1 Structure of TRITC isomers

Brief description

Tetramethylrhodamine isothiocyanate widely known as TRITC is a prominent member of the family of rhodamine dyes (Fig. 1 for structure). It exhibits strong orange fluorescence (excitation max. at λ = 560 nm and fluorescence max. at λ =590 nm in DMSO; see Fig. 2). Its intense fluorescence is nearly insensitive to pH changes. Its high brightness and fluorescence quantum yield along with its capacity to covalently conjugate to proteins and polysaccharides efficiently under relatively mild conditions, render this rhodamine dye an indispensable fluorescent labeling agent. This fluorescent dye is currently available in the form of a mixture of two isomers (5- and 6- isothiocyanate isomers; see Fig.1).



Synthesis and Structure

TRITC (isomer mix) is synthesized via a well-developed TdB Labs synthetic method which results in a product of high purity. The purity of the final product is assessed using HPLC. Each batch produced is validated by reacting a sample to dextran and spectrally evaluating the absorbance, excitation, and fluorescence characteristics of the final product.

Properties

TRITC (isomer mix.) is delivered as a dark red powder (Fig. 3C). The powder is highly soluble in polar organic solvents such as DMSO and DMF and exhibits significant solubility in MeOH and EtOH as well. It is incompatible with strong oxidizing agents and should be stored in a dry environment. When desiccated, it can be safely stored at room temperature. However, it is generally recommended that it is stored at temperatures between 4-8°C.

Solutions of TRITC in organic solvents are colored vivid-red and exhibit significant orange emission when observed under white light (Fig. 3D). The excitation and emission spectra of TRITC (isomer mix.) are depicted in Fig. 2. An excitation max. at λ =560 nm and fluorescence max. at λ =590 nm are observed in DMSO solutions. The absorption band observed in DMSO solutions exhibits a shoulder between 530-550 nm which is attributed to the coexistence of the two isomers.

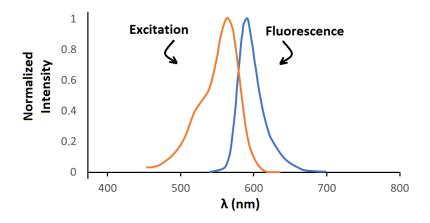


Fig. 2 Excitation and fluorescence spectra of TRITC (mixed isomers) recorded in DMSO

TRITC can efficiently conjugate to -NH₂ involving biomolecules such as proteins and antibodies or biorelevant macromolecules such as modified polysaccharides (e.g. lysine-dextran) at mild basic conditions. The rhodamine is thus conjugated to the macromolecules via a thiourea linker. TRITC can also label dextran via a well-developed



TdB Labs methodology allowing for TRITC-dextrans with high degree of substitution (See TdB Labs TRITC-Dextran products).

TRITC and TRITC-labelled products are highly important for a range of biorelevant applications e.g. in cell imaging, permeability and microcirculation studies (see for example Fig. 3A and 3B) as well as research on drug delivery as a molecular size marker and in various biosensor applications.

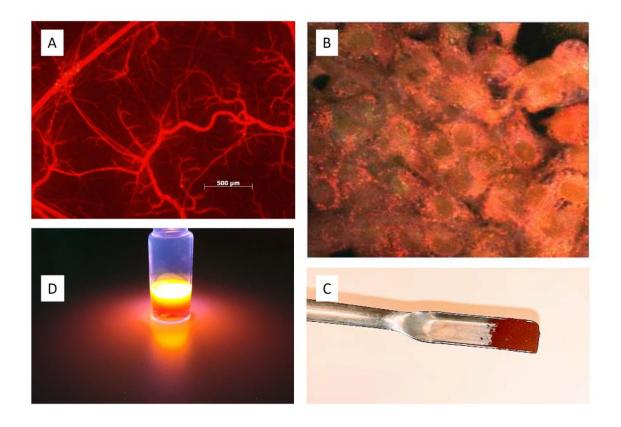


Fig. 3 A. Fluorescence microscopy image of a microvasculature of hamster cheek pouch obtained using TRITC-dextran 150 kDa **B.** TRITC-dextran 10 kDa fixation on NHPTK cells (a human proximal tubule cell line) **C.** Spatula currying a small amount of TRITC powder (mix. of isomers) **D.** The bright orange fluorescence of a TRITC DMSO solution (all images are courtesy of TdB Labs AB)