TRITC-lysine-dextran

CAS nr: N/A

Structure:

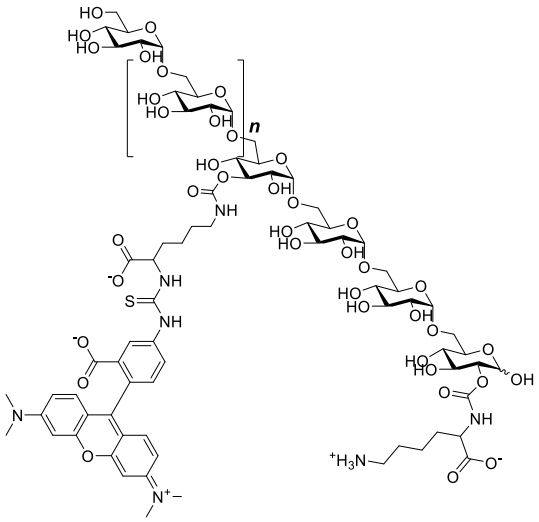

Fig. 1. Structural representation of TRITC-lysine-dextran. Whether Lysine is mainly conjugated with dextran via the ε- or the α-amino group of Lysine has not been investigated.

Synthesis and structure

Lysine-dextrans (or Dextran Lysine Fixable) are synthesised from well-characterized dextran fractions derived from Leuconostoc mesenteroides labelled with Lysine. Lysine-dextran is then functionalized with TRITC under mild conditions to form TRITC Lysine-dextran. After purification, the products are controlled for Mw, appearance, solubility, DS, pH, free Lysine and free TRITC. The products are designated by the approximate molecular weights of the dextran fractions used. Thus, for example, the product TRITC-lysine-dextran 4 has a molecular weight of approx. 4000 Da. The actual molecular weight is determined by GPC. This value is supplied with the Certificate of Analysis. The dextran used is from Leuconostoc mesenteroides B-512F which is essentially a linear α-(1-6)-linked glucose chain with however a low percentage (2-5%) of α-(1-3) branches distributed along the chain. The dextran fractions used are from Mw of 4000 to 500000 and are carefully controlled by GPC, optical rotation, absorbance and other control parameters

Physical properties

TRITC Lysine Dextrans are pink to purple powders that are readily soluble in water or electrolyte solutions. TRITC Lysine Dextrans are insoluble in most organic solvents, such as ethanol, methanol, acetone, chloroform, ethyl acetate etc. The degree of substitution of Lysine (DS) is between 0.005-0.03 (mol Lysine/mol Glucose) and the degree of substitution of TRITC (DS) is 0.001-0.01 (mol TRITC/mol Glucose). TRITC has an excitation maximum at 550 nm and an emission maximum at 574 nm (Fig.2).

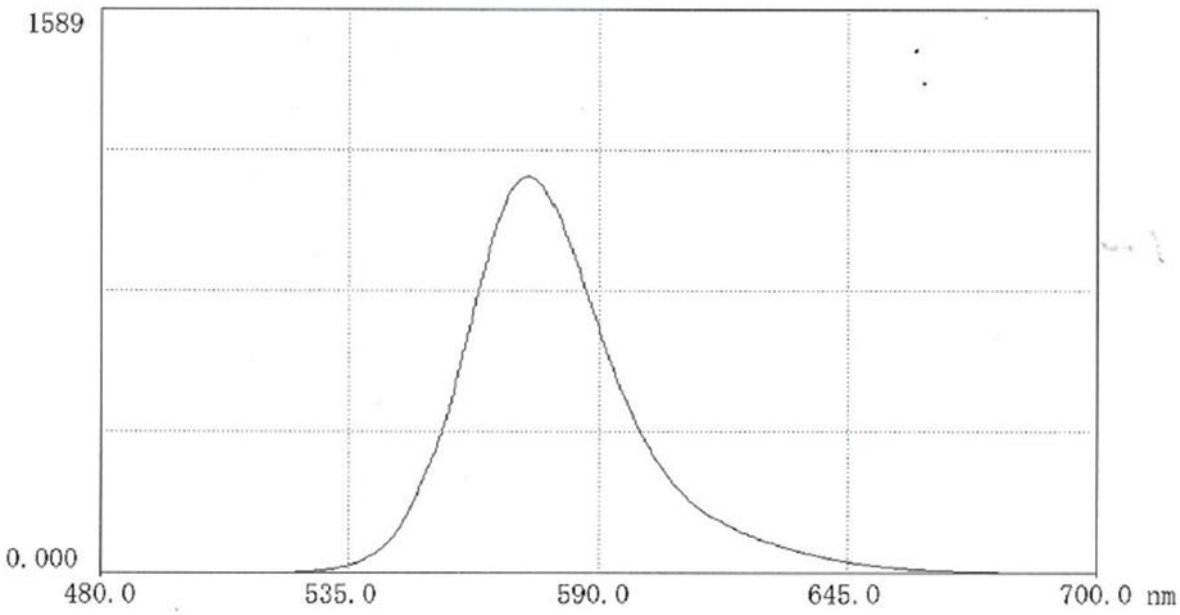

Fig.2 Fluorescence scan of TRITC Lysine Dextran 10 in 0.025 M Borate buffer pH 9 (10 mg in 50 mL). Ex/Em 550/574 nm.

Stability

No prospective stability studies on TRITC Lysine Dextrans have been performed yet. However, the structural properties of the dextran and of the carbamide linkage of the Lysine to the dextran chain would suggest high stability of the product. The thiourea coupling between TRITC and Lysine is stable. It is recommended that the products are stored in air-tight containers. TRITC Lysine Dextrans may be stored at ambient temperatures.

Applications

Dextrans carrying amino groups are valuable to the scientific community as versatile tools for bioconjugation and fixation in living systems1,2. For conjugation, the free amino group of the Lysine structure will be able to covalently bind to activated entities such as NHS-esters or isothiocyanates or be coupled to carboxylic acids using common peptide coupling reagents such as DCC, HOBt and or HATU. Cell and tissue fixation is performed in order to preserve components in a "life-like state" and to make cells permeable to allow antibodies to access cellular structures, for example for microscopy studies or immunostaining. The amino groups on Lysine Dextran reacts with the fixing agent (here: formaldehyde or glutaraldehyde) to form a covalent crosslinking with biomolecules such as proteins and lipids, immobilizing the whole system and the dextran. This is of particular importance when evaluating biological events qualitatively or quantitatively in molecular imaging. Without fixation, these structures within a living system would fall apart and diffuse rapidly.
Tetramethylrhodamine/TRITC is a fluorescent dye that has an excitation maximum at 550 nm and an emission maximum at 574 nm.

References

1. (a) Henley JR, Krueger EW, Oswald BJ, McNiven MA, J Cell Biol (1998) 141:85-99; (b) Fritzsch B, Christensen MA, Nichols DH; J Neurobiol. 1993 Nov;24(11):1481-99; (c) Fritzsch B., J Neurosci Methods (1993) 50:95-103.
2. Schmued, L., Kyriakidis, K., Heimer, L., Brain Res., 526, (1990) 127-134.