Chondrex, Inc.

TRITC-Dextran

Catalog # 4014

For Research Use Only - Not Human or Therapeutic Use

DESCRIPTION: Tetramethylrhodamine isothiocyanate (TRITC) labeled dextran

APPLICATIONS: Use to assess the permeability of semi-permeable membranes either *in vivo* or *in vitro* (1-4).

Note 1: It is possible to simultaneously use FITC-dextran and TRITC-dextran of different sizes as FITC and TRITC fluoresce at different wavelengths.

Note 2: Black 96-well plates are available for reading fluoresce (Catalog # 9045).

QUANTITY: 5 ml

FORM: 25 mg/ml solution in PBS (Red color).

MOLECULAR WEIGHT: 70 kDa

FLUORESCENT PROPERTIES: Excitation: 550 nm, Emission: 572 nm

IN VIVO PROTOCOL:

1. Fast mice 4 hours before the oral feeding, and for the duration of the experiment.
2. Feed 20 ml/kg by oral gavage.
3. Maintain fasting conditions and wait 3 hours (may vary depending on individual animals).
4. Collect blood by retro-orbital bleeding, then spin and collect plasma. Dilute plasma 1:2 (or more) with PBS.
5. Optional: Prepare a standard curve by making dilutions of the stock TRITC-dextran with diluted (1:2) normal mouse plasma with PBS (12.5 μg/ml is a recommended starting point). See next page.*

   Caution: Protein in the samples may intefere and reduce the fluorescence intensity (FI). Therefore, to ensure an accurate determination of TRITC-dextran permeability, the Plasma to PBS ratio must be consistent throughout the standard curve, as well as with the dilution factor of the sample. Use a diluent prepared by the same ratio of normal mouse plasma to PBS in sample and standard dilution.
6. Transfer 50 or 100 μl of diluted samples to a black 96-well plate and read on a fluorescent plate reader.

IN VITRO PROTOCOL: Please refer to protocol of choice.

STORAGE TEMPERATURE: 4°C in the dark

STABILITY: 1 year

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REFERENCES:

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3. D. B. Pink, W. Schulte, M. H. Parseghian, A. Zijlstra, J. D. Lewis, Real-time visualization and quantitation of vascular permeability in vivo: implications for drug delivery. PLoS One 7, e33760 (2012).
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*** Preparation of Standards**

If you use 1:2 diluiton for sample dilution, prepare 33% normal mouse plasma in PBS (300 μl plasma in 600 μl PBS) as a dilutent. The dilutent should be same dilution of samples with PBS.

1. 10 μl of 25 mg/ml TRITC-Dextran in 990 μl of PBS (250 μg/ml)
2. 15 μl of the diluted TRITC-Dextran in 2.85 ml of 33% normal mouse plasma in PBS (12.5 μg/ml)
3. Mix 125 μl of the 12.5 μg/ml solution with an equal volume of 33% mouse normal plasma (6.3 μg/ml)
4. Repeat 5 times for the 3.1, 1.6, 0.8, 0.4 and 0.2 μg/ml.
5. Use 50 μl or 100 μl for reading.

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