SZABO SCANDIC

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Produktinformation

- Forschungsprodukte & Biochemikalien
- Zellkultur & Verbrauchsmaterial
- Diagnostik & molekulare Diagnostik
- Laborgeräte & Service

Weitere Information auf den folgenden Seiten!
See the following pages for more information!

Lieferung & Zahlungsart

siehe unsere Liefer- und Versandbedingungen

Zuschläge

- Mindermengenzuschlag
- Trockeneiszuschlag
- Gefahrgutzuschlag
- Expressversand

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Product No: **K-4200**

Dextran Sulfate 5000 0.003-1.0 µg/mL
Dextran Sulfate 500000 0.003- 0.3 µg/mL
ELISA Buffer/Urine

Dextran Sulfate ELISA Kit for Buffer/Urine Samples

INTENDED USE: THIS PRODUCT IS FOR RESEARCH USE ONLY. NOT INTENDED FOR CLINICAL OR DIAGNOSTIC USE.

Kit includes:

Coated 96-well plate
Detector -Enzyme Conjugate vial
Conjugate Diluent
TMB Solution
Stop Solution
Wash Concentrate 10X, (dilute 1 part plus 9 parts water)

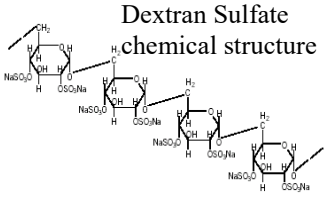
Researcher must provide:

Pipettes (8 channel multipipettor recommended)
Absorbance microplate reader
Dextran sulfate standards
Tris Buffered Saline (TBS) pH 7.5 (10mM Tris 150mM NaCl)
Plate Cover

Storage and Stability

Kit can be stored unopened at 4°C for up to six months. Reconstituted detector enzyme conjugate is unstable and should be used immediately. If you wish to run less than a full plate, it should be stored as frozen aliquots at -80°C. Aliquots must be thawed **immediately** before use. After one thaw, any unused detector enzyme conjugate must be discarded. TMB solution should be protected from light.

Background


Dextran Sulfate chemical structure

Dextran Sulfate is in the family glycosaminoglycan. It is a polyanionic dextran derivative which may be synthesized from various high purity and well-characterized dextran fractions. In clinical research, anticoagulant dextran sulfate properties have been tested as a possible substitute for heparin in anticoagulant therapy. Another source of interest relates to the effect of dextran sulfate on enzyme inhibition in certain biological systems. Dextran sulfate is used to precipitate LDL and VLDL in plasma fractionation procedures. Dextran sulfate must then be removed from the product. The K-4200 assay allows measurement of extremely low levels of dextran sulfate and gives manufacturers quantitative data that they have removed dextran sulfate from their product.

The dextran sulfate ELISA product number K-4200 is a quantitative enzyme-linked assay designed for the *in vitro* measurement of dextran sulfate levels in low protein content fluids such as buffer or urine. This assay measures dextran sulfate directly using a dextran sulfate binding protein which has been conjugated to HRP.

The dextran sulfate-ELISA is a competitive assay in which the colorimetric signal is inversely proportional to the amount of dextran sulfate present in the sample. Samples to be assayed are first mixed with the detector-enzyme conjugate in wells of the coated plate. Dextran sulfate in the sample competes with dextran sulfate bound to the plate for binding of the detector-enzyme conjugate. The concentration of dextran sulfate in the sample is determined using a standard curve of known amounts of dextran sulfate.

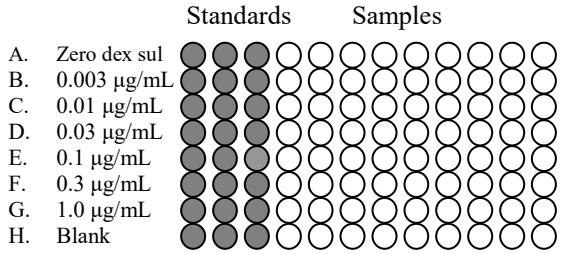
Reagent Preparation

Dextran Sulfate Standards: Make dilutions of your dextran sulfate using Tris Buffered Saline (TBS) pH 7.5 (10mM Tris 150mM NaCl) to obtain standards of 0.003, 0.01, 0.03, 0.1, 0.3, and 1.0 µg/mL. **Standardization should be performed using dextran sulfate that is the same dextran sulfate type contained in your unknowns.**

Working Detector-Enzyme Conjugate: Measure exactly **5.2 mL** of conjugate diluent and add to a clean tube. Perform a 'clean transfer' of the lyophilized Detector-Enzyme Conjugate into the 5.2 mL of conjugate diluent. This can be done by adding 500 microliters of the measured Diluent to reconstitute the Detector-Enzyme. Wait a minute to allow the lyophilized material to dissolve and then add to the liquid back to the tube. Repeat this step two more times to be sure all the Detector-Enzyme Conjugate has been transferred from the vial to the tube. If you wish to perform less than a full plate, the Reconstituted Detector-Enzyme Conjugate is not

stable and must be stored as aliquots at -80ºC and thawed immediately before re-use. After one thaw, any unused detector enzyme conjugate must be discarded.

1X Wash Buffer: Make a 1:10 dilution of 10X Wash Buffer in distilled or deionized water.



Dextran Sulfate ELISA

Assay Procedure

1. Set up the dextran sulfate ELISA plate as illustrated above. We suggest the dextran sulfate standard dilution series be run in triplicate for best results. Add **50 µL** of Standards and samples into corresponding wells. Add **50 µL** of Working Detector - Enzyme Conjugate to all wells except the Blank wells. Mix well. Cover plate and incubate for 30 minutes at room temperature. A rotator is highly recommended if available, as constant mixing significantly improves precision.

2. Discard the solution and wash the wells four times with 300 µL per well of 1X Wash Buffer. An automated plate washer is recommended if available. After washing, immediately proceed to the next step. Do not delay in removing wash buffer from the wells. Do not allow plate to dry.

3. Add 100 µL TMB Solution to each well. Incubate the plate in the dark at room temperature for 4-60 minutes waiting for the zero dextran sulfate wells to develop to a medium to dark blue color. Watch for color development and **DO NOT** overdevelop.

4. Add 50 µL Stop Solution which will change the color from blue to yellow.

5. Immediately measure the absorbance of each well at 450 nm.

6. Calculate the binding percentage for each sample using the formula:

$$[A_{450}(\text{Sample}) - A_{450}(\text{Blank})]/ [A_{450}(\text{Zero dextran sulfate}) - A_{450}(\text{Blank})] \times 100 = \% \text{ Binding}$$

Using linear or nonlinear regression, plot a standard curve of percent binding versus concentration of dextran sulfate standards. Determine dextran sulfate levels of unknowns by comparing their percentage of binding relative to the standard curve. Dextran sulfate can be estimated by comparing the values from the wells containing unknowns to the values in the standard curve.