Glomerular Basement Membrane (GBM; dissociated)

Antigen Specification

Product Number: 16800

Description:
Human α3 chain of collagen IV; identical with the antigen called "glomerular basement membrane antigen" (GBM).
Recombinant antigen for in vitro research and manufacturing use only.

Immunological function:
Binds IgG-type human auto-antibodies.

Origin:
Recombinant. Expressed by recombinant baculovirus (*Autographa californica* multiple nuclear polyhedrosis virus; AcMNPV) infection of *Spodoptera frugiperda* Sf9 insect cells.

Expression construct:
cDNA coding for a minicollagen version of the human collagen IV α3 chain fused to a hexa-histidine purification tag. The term minicollagen designates the removal of most of the epitope-less triplehelical collagenous region (situated between the N-terminal 7S domain and the C-terminal noncollagenous NC1 domain), which is a requirement for recombinant production of this antigen.

Biochemical tests:
SDS-PAGE (purity > 80%); Western blot with i: anti-GBM autoantibody-positive sample; ii: monoclonal anti-His-tag antibody.

Calculated molecular weight:
44 kDa

Calculated isoelectric point:
pH 8.9

Immunological tests/Functionality:
Standard ELISA test (checkerboard analysis of positive/negative samples); immunodot analyses with positive/negative samples.

Recommended buffer/storage and handling conditions:
Recommendations for storage buffer: neutral to slightly alkaline pH and 4 M urea as dissociating agent. Storage conditions: -70°C or below.
Repeated freeze/thaw cycles should be avoided.

Coating concentration:
0.12-0.5 µg/mL (depending on the type of ELISA plate and coating buffer). Suitable for labeling of functional groups.

CAUTION: It has been reported that the immunodominant epitope of GBM is a cryptic epitope that is not easily accessible to the corresponding autoantibodies. It is necessary to treat the protein under non-reducing conditions with a denaturant such as urea to unmask the epitopes *(see Hellmark et al. in Autoantibodies, Peter, J.B. and Shoenfeld, Y., eds., Elsevier B.V., 1996, pp 291-298).*

This GBM antigen product is produced in dissociated form and does not require additional unmasking of the epitope before coating.