

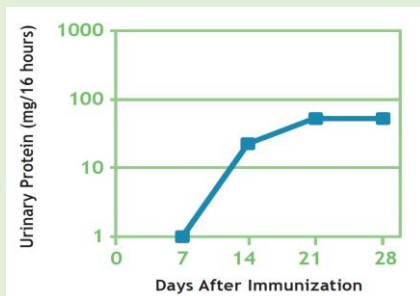


Rat Nephritis Reagents



Goodpasture Syndrome, an autoimmune disorder mediated by autoantibodies against the glomerular basement membrane (GBM), can lead to severe renal disease that is often associated with pulmonary hemorrhage. These autoantibodies target the NC1 domain of type IV collagen, which is a major constituent of the basement membrane in kidney and lung tissues. Chondrex, Inc. provides the NC1 fraction of type IV collagen and nephritogenic monoclonal antibodies to induce nephritis in rats (1-2). For more information about these products, please contact Chondrex, Inc. at support@chondrex.com.

NC1 Fraction of Bovine Type IV Collagen (Catalog # 1102)



Protocol:

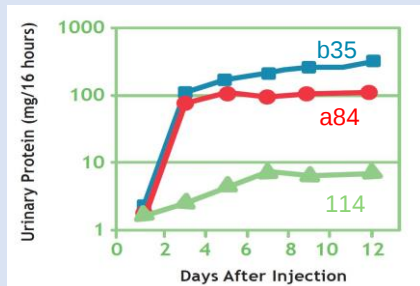
WKY rats (Female, 7 - 8 weeks old)

Immunize 50 µg of the NC1 fraction emulsified with CFA (Catalog # 7001) by subcutaneous injection at the base of the tail.

Measure protein levels in 16-hour urine samples taken every 7 days using the Rat Urinary Protein Assay Kit (Catalog # 9040)

Nephritogenic Monoclonal Antibodies

Clone	Quantity (Catalog #)	
b35	1 mg (70201)	5 mg (70205)
a84	1 mg (70211)	5 mg (70215)
114	1 mg (70221)	5 mg (70225)



Protocol:

WKY rats (Female, 7 - 8 weeks old)

Inject 100 µg of nephritogenic monoclonal antibodies by intraperitoneal injection.

Measure protein levels in a 16-hour urine sample every other day using the Rat Urinary Protein Assay Kit (Catalog # 9040)

NOTE: Urine volumes vary among individual rats, therefore total amounts of protein excreted in a 16-hour urine sample should be determined instead of protein concentration in order to accurately evaluate nephritis severity.

Comparing Two Models

	NC1 of Type IV collagen	Monoclonal Antibodies
Study period	2 - 3 weeks	7 - 10 days
Injection method	SC at the base of the tail	IV or IP
Severity	Mild - severe	Dependent on clones



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Rat Urinary Protein Assay Kit (Catalog # 9040)

Urinary protein levels in human specimens can be determined with a turbidity assay to assess renal damage. Chondrex, Inc.'s rat urinary protein assay kit (Catalog # 9040) uses the turbidity method with rat serum standards in 96-well microtiter plates and works for assaying a large number of rat urine samples due to the wide assay range. Moreover, the turbidity assay has good correlation with the Bradford assay.

Bromophenol Blue Protein Assay Kit (Catalog # 6026)

Urinary protein levels, a useful marker of renal disease in rodents, are commonly determined by a simple dipstick method which is affected by urine volume and color, leading to inaccurate results. Chondrex, Inc.'s bromophenol blue (BPB) protein assay kit (Catalog # 6026) is available as a simple, precise, and accurate alternative to determine proteinuria. Because BPB has a higher affinity to albumin than many types of globulin, this assay may better reflect glomerular albuminuria.

Rat Albumin ELISA Kit (Catalog # 3020)

Excretion of albumin into urine (albuminuria) is significantly increased by renal damage as seen in rat models of sepsis, glomerulonephritis, and diabetic nephropathy. Plasma albumin levels decrease with severe liver disease as well as other critical illnesses, such as kidney disease, inflammation, and sepsis due to increased capillary leakage. Chondrex, Inc.'s rat albumin ELISA kit (Catalog # 3020) is designed to measure albumin levels in both serum and urine.

Comparing The Three Assay Methods

Product	Specificity	Sensitivity	Sample Volume	Assay Time
Urinary Protein Assay	Total Protein	0.4 - 4 mg/ml	100 µl	15 minutes
BPB Protein Assay	Primarily Albumin	0.03 - 2 mg/ml	100 µl	10 minutes
Rat Albumin ELISA	Albumin Only	1.6 - 100 ng/ml	Less than 10 µl	4 hours

References

1. T. Kohda *et al.*, High nephritogenicity of monoclonal antibodies belonging to IgG2a and IgG2b subclasses in rat anti-GBM nephritis. *Kidney International*. **66**, 177–186 (2004).
2. T. Kado *et al.*, Immunohistochemical characterization of glomerular inflammatory cells and expression of adhesion molecules in anti-glomerular basement membrane (anti-GBM) glomerulonephritis induced in WKY rats with monoclonal anti-GBM antibodies of different subclasses. *Pathol Int*. **56**, 55–61 (2006).