SERUM AND URINARY CONCENTRATIONS OF TYPE IV COLLAGEN IN THE
PATIENTS WITH RENAL DISEASES

Yoshikazu Hayashi, Hiroyoshi Makino, Kenichi Shikata, Zensuke Ota
Third Department of Internal Medicine, Okayama University Medical
School, Okayama, Japan

Abstract; We measured serum and urinary concentrations of type
IV collagen by enzyme-linked immunosorbent assay (ELISA) in
patients with renal diseases and compared them with normal
levels. In diabetics with macroproteinuria or with renal
insufficiency, serum and urinary concentrations of type IV
collagen were higher than in diabetics without nephropathy or
with early renal damage as determined by the presence of
microproteinuria. Serum concentrations of type IV collagen in
patients with minimal change nephrotic syndrome (MCNS) at the
remission phase, membranous nephropathy (MN),
mesangio proliferative glomerulonephritis (MPGN) and chronic
renal failure without diabetes mellitus (CRF without DM) showed
a significant increase compared with normal controls. Urinary
concentrations of type IV collagen in patients with MN, IgA
nephropathy and CRF without DM showed a significant increase
compared with normal controls. These increases may indicate that
a change in basement membrane metabolism has occurred in these
renal diseases.

Key words; basement membrane, type IV collagen, glomerulonephritis
urine, enzyme-linked immunosorbent analysis (ELISA)

Accepted January 28, 1991
Reprint requests to: Dr. Yoshikazu Hayashi
Third Department of Internal Medicine, Okayama University Medical
School, 2-5-1 Shikata-cho, Okayama 700, Japan
Hayashi Y et al: Collagen IV in renal diseases

Recently biochemical and immunobiochemical approaches have been used to characterize the changes in basement membrane components, especially those of the glomerular basement membrane, which include the collagenous component type IV collagen and noncollagenous component laminine, heparansulfate proteoglycan and fibronectine. Radioimmunochemical methods have become available for the study of basement membrane metabolism through the measurement of serum and urinary concentrations of basement membrane proteins such as type IV collagen and laminine (1,2,3,4). Earlier studies have demonstrated increased serum levels of 7S-collagen in rats with streptozotocin-induced diabetes which can be normalized by insulin treatment (5). Serum levels of 7S-collagen and laminine P1 in diabetes were also found to be increased compared to those in controls (6). To the best of our knowledge, there has been no report of measurement of the concentrations of type IV collagen in human urine. In the present study, we measured serum and urinary concentrations of type IV collagen by means of ELISA in patients with various renal diseases. The purpose of the present study was to apply these methods to the monitoring of possible change in basement membrane metabolism in the patients with renal diseases.

PATIENTS AND METHODS

Type IV collagen was measured with a sandwich EIA kit with two monoclonal antibodies to the 7S domain and some of the non-7S and non-NCl domains (IV-EIA :Fuji Chemical Industries, LTD.,Japan ). Eighty-six patients with renal diseases, 10 diabetics without
Hayashi Y et al: Collagen IV in renal diseases

nephropathy and 10 normal controls were admitted to the study. The degree of diabetic nephropathy was rated as 1 to 4 for each patient: 1: no nephropathy; 2: urinary protein (+); 3: urinary protein (++) or (+++); 4: serum creatinine levels >2mg/dl.

Patients with additional diseases associated with connective tissue changes, such as rheumatic disorders or chronic liver disease, were excluded from participation in the study.

RESULTS

In diabetic patients a significantly higher serum concentration of type IV collagen was found in groups 2, 3 and 4 than in normal controls (Fig.1). Furthermore a significantly higher urinary concentration of type IV collagen was noted in groups 3 and 4 than in normal controls (Fig.2). Serum concentrations of IV-EIA in patients with MCNS in the remission phase, MN, MPGN and chronic renal failure without diabetes mellitus (CRF without DM) showed a significant increase compared with normal controls (Fig.3). MN, IgA nephropathy and CRF without DM had significantly high levels of urinary concentrations of IV-EIA (Fig.4). There was a positive correlation between serum and urinary IV-EIA concentrations (p<0.01).

![Graph showing serum concentrations of IV-EIA in diabetes](image-url)
Hayashi Y et al: Collagen IV in renal diseases

**Fig. 2.** Urinary concentrations of IV-EIA in diabetics. (mean ± S.E.M.)
Significantly different at *p < 0.005 compared to normal control.
**p < 0.005, ***p < 0.05

**Fig. 3.** Serum concentrations of IV-EIA (mean ± S.E.M.)
Significantly different at *p < 0.025 and **p < 0.01 compared to normal control.

**Fig. 4.** Urinary concentrations of IV-EIA (mean ± S.E.M.)
Significantly different at *p < 0.025 and **p < 0.05 compared to normal control.
Diabetic nephropathy is characterized by thickening of capillary basement membranes. The current evidence suggests that an increase in basement membrane synthesis coupled with a normal or decreased rate of degradation leads to a buildup of basement membrane and thickening (7,8,9,10). Furthermore, serum levels of 7S collagen and laminine P1 in diabetes were found to be increased compared to those in controls (6). Earlier studies reported that a radioimmunoassay for specific 7S domains of type IV collagen detected a high molecular weight antigen which was probably intact type IV collagen and small oligomeric variants (10). In contrast, assay with IV-EIA detected a single peak of high molecular weight antigen which was probably intact type IV collagen (4). These results may indicate that measurement of type IV collagen with a radioimmunoassay for specific 7S domains reflects both the synthesis and degradation of glomerular basement membrane, and that the measurement of type IV collagen with IV-EIA primarily reflects the synthesis of glomerular basement membrane. In our study, serum and urinary concentrations of type IV collagen were measured in patients with renal diseases. In diabetic patients in groups 3 and 4, serum and urinary concentrations of type IV collagen were significantly increased. These results suggest that serum and urinary concentrations of type IV collagen are increased in patients with advanced diabetic nephropathy and suggest that a change in basement membrane metabolism occurs in diabetics. Urinary
concentrations of type IV collagen may reflect both the basement membrane metabolic level and a disorder of the glomerular filtration barrier. Thus the measurement of type IV collagen in serum and urine, which can be accompanied using non-invasive methods, will be useful in the detection of changes in the glomerular basement membrane.

REFERENCES