

# **CYOGLOBULINEMIA RELATED RENAL/GLOMERULAR CHANGES**

**FRANCO FERRARIO  
RENAL IMMUNOPATHOLOGY CENTER  
SAN CARLO BORROMEO HOSPITAL  
MILAN\_ITALY**

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## **Introduction**

Mixed cryoglobulins (MC) are plasma proteins that precipitate reversibly at low temperatures and are composed of immunoglobulins including one with rheumatoid activity. Two types of MCs can be defined on the basis of Brouet's classification (1). Type II cryoglobulins consist of one monoclonal immunoglobulin (usually IgMk) having rheumatoid activity against a polyclonal IgG while type III cryoglobulins are characterised by a combination of polyclonal immunoglobulins. Most MCs, defined as "secondary mixed cryoglobulins", have been detected in patients with connective tissue disorders, lymphoproliferative disorders, chronic infections, non-infectious hepatobiliary diseases or immunologically mediated glomerular diseases. In approximately 30% of cases, no underlying disease was present, and cryoglobulinaemia was defined as essential. The clinical syndrome of essential mixed cryoglobulinaemia was first described by Meltzer et al. in 1966 (2). It was characterised by purpura, weakness, arthralgia as well as by glomerular lesions in some patients. Several subsequent reports further defined this syndrome, indicating that its incidence varies in different geographical areas, with the majority of cases having been reported in the Mediterranean countries, namely Italy, France, Spain and Israel (3,4).

An important step forward in the knowledge of this disease was achieved when HCV infection was found in the majority of patients with "essential" mixed cryoglobulinaemia of either type (5,6,7,8,9,10,11), suggesting that these cases should no longer be considered "essential".

The clinical symptoms range from mild palpable purpura, arthralgias and fatigue, to severe vasculitis with skin necrosis, glomerulonephritis, involvement of peripheral nerves, central nervous system, gastrointestinal tract, lungs, myocardium (12,13,14,15,16).

In rheumatologic surveys, type III MC patients out-numbered those with type II MCs (2,12). On the contrary, surveys based on renal involvement indicated a large prevalence of type II MCs, with IgMk usually being the monoclonal IgM (17,18).

I would like to present a large multicentre , retrospective study, of the Italian Group of Renal Immunopathology that collected the clinical and serological data of 146 cases of biopsy-proven cryoglobulinaemic glomerulonephritis recorded in the Italian Registry of Renal Biopsies in 1995

### **Histological analysis**

Histological specimens were available for all 146 patients and were representative enough to characterise the patterns of glomerular involvement. The histological material was independently examined by 2 investigators. Immunofluorescence results were available for all patients. A mean of  $17 \pm 11.2$  glomeruli (average 5-70) were observed by means of light microscopy. Cases with advanced sclerosis were excluded from the study (90% showed less than 20% glomerular hyalinosis). Enough histological material was available for the quantitative and semi-quantitative evaluation of 130 out of 145 cases. This included the following features: 1) percentage of global glomerular sclerosis, segmental glomerular sclerosis, extracapillary proliferation 2) amount of mesangial sclerosis (grade 0 to 3), mesangial proliferation (0-3), endocapillary proliferation and/or exudation (0-3), diffuse thickening of the capillary wall (0-3), double contours (0-3), endoluminal hyaline thrombi and tubular atrophy (1:<30%; 2:>30<60%; 3:>60% of glomeruli or cortex), interstitial infiltrates (0-3 as per tubular atrophy), interstitial fibrosis (0-3 as per tubular atrophy) , and the presence or absence of: arteriolar hyalinosis, intimal fibrosis, miointimal hyperplasia, arteritis, thrombotic microangiopathy.

On the basis of the above described features, arbitrary chronic and activity histologic indexes were calculated for each case.

### **Morphologic analysis of kidney biopsy and the clinical-pathological correlations.**

Morphologic analysis identified the following histological patterns :

- 1) Membranous nephropathy (2 cases): characterised by the same morphology as the primary form. Prevalent sub-epithelial deposits of IgM, IgG and C3 , but with no proliferative lesions. Electron microscopy examination was available for one of the 2 cases , but showed no crystalloid structured deposits.
- 2) Mesangial proliferative glomerulonephritis (10 cases, group A): characterised by mesangial expansion and proliferation, without exudation or endocapillary proliferation. Isolated proteic endoluminal thrombi were only found in few cases. Immunofluorescence examination showed segmental and irregular deposits of IgM, IgG and C3 in mesangial and paramesangial localisations and within very few endocapillary thrombi.
- 3) Focal membranoproliferative glomerulonephritis (10 cases, group B): having a typical immunohistological pattern of cryoglobulinaemic glomerulonephritis, but involving < 50% of the glomeruli.
- 4) Diffuse membranoproliferative GN (108 cases, group C): having the same lesions as group 2, but involving more than 50% of the glomeruli. Four cases of

membranoproliferative GN showed in addition to the common IF pattern the presence of IgA deposits with prevalent mesangial but also parietal localization.

The main clinical features of the 3 histological groups (A, B and C) are shown in Table III

Table III: Clinical features at biopsy. Results are expressed as percentages unless indicated otherwise

histologic groups (# patients)	Mesangial proliferative "A" (10)	Focal membranoproliferative "B" (10)	Diffuse membranoproliferative "C" (108)
Age (mean $\pm$ s.d.)	51.5 $\pm$ 13.3	55.4 $\pm$ 9.7	57.9 $\pm$ 10.53
M/F	60/40	40/60	43/57
Time from onset of nephropathy (mean $\pm$ s.d.)	13 $\pm$ 21.2	28.4 $\pm$ 37.14	18 $\pm$ 24.6
Serum Creatinine (mg/dL) (mean $\pm$ s.d.)	1.3 $\pm$ 0.7	2.5 $\pm$ 2.2	2.6 $\pm$ 1.8
Serum creatinine >1.5 mg	40	30	33
Proteinuria (g/24h) (mean $\pm$ s.d.)	2.5 $\pm$ 3.0	2.9 $\pm$ 2.9	4.0 $\pm$ 9.7
Hematuria	100	100	100
Hypertension	60	40	66.7
$\downarrow$ C3	20	30	52
$\downarrow$ C4	60	60	93
$\uparrow$ ALT	30	30	22
Nephrotic Syndrome	30	30	41.5
Acute Nephritic Syndrome	-	-	7.5
Orinary Abnormalities	50	60	42.5
Chronic renal insufficiency	20	-	7.5
Acute renal failure	-	10	1

Serum creatinine levels were lower in group A than in the others groups, even though the percentage of patients with renal failure was substantially the same. Patients in group 3 showed higher levels of proteinuria, a higher incidence of nephrotic syndrome, and lower C4 levels.

Extrarenal symptoms and signs were more frequently observed in patients with a membranoproliferative pattern, while on the contrary, only 40% of patients with mesangioproliferative GN had extrarenal symptoms and signs.

The characterisation of cryoprecipitate was available in 117 cases. IgM-IgG was the component in 116 cases, while it was IgG-IgA in 1 case. Type III cryoglobulin was more frequent in group A (55%) than it was in groups B (33%) or C (20%). A monoclonal IgM $\kappa$  was present in 44%, 66%, and 77% of cases, respectively, in groups A, B, and C. An IgM $\lambda$  component was only found in 3 Group C cases.

The main lesions in Group A included global and diffuse, slight mesangial matrix expansion and mesangial cell proliferation. Focal endoluminal thrombi were found in very few cases. Proliferation, exudation, and thickening of the capillary wall in the Group B patients were mild and irregularly distributed within the same glomerulus and among glomeruli. Endoluminal thrombi were only found in 1 out of 10 cases. The proliferative and exudative abnormalities we observed in Group C were usually severe, with segmental distribution in 15% of cases. Centriolobular sclerosis was present in 50% of cases. The capillary wall abnormalities were not correlated to proliferative lesions. Double contour appearance of the capillary wall was present in 70% of cases, and endoluminal thrombi were found in more than 50%. Extracapillary proliferation was present in 16% of cases. Interstitial leukocyte infiltration, which was usually focal, was more often present in the membranoproliferative forms (60% in Groups B and C), and was correlated to the intensity of the proliferative glomerular lesions. Interstitial fibrosis, which was usually focal, was still present in the membranoproliferative forms: 90% in Group B and 70% in Group C, but only 50% in Group A. Arteriosclerotic lesions were present in 30% of cases, and no differences were observed among groups. Arteritis was rare (5.5%).

No differences were found with regards to the chronicity index among the 3 histological groups we took into consideration. It was, respectively, : Group A -  $4.3 \pm 2.8$ , Group B -  $4.1 \pm 1.9$ , and Group C -  $4.0 \pm 2.3$ . On the contrary, the activity index increased as the number of proliferative and inflammatory lesions grew and was, respectively, : group A -  $3.6 \pm 2.3$ , group B -  $4.8 \pm 2.5$ , and group C -  $8.4 \pm 9.0$ .

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## BULLET POINTS:

Mixed cryoglobulins are plasma proteins that precipitate reversibly at low temperatures.

Type II cryoglobulins consist of one monoclonal immunoglobulin (usually IgM kappa) having rheumatoid activity against a polyclonal IgG.

HCV infection is found in the majority of patients with “essential” mixed cryoglobulinemia suggesting that these cases should no longer be considered “essential”.

Cryoglobulinemic glomerulonephritis may present a variety of light microscopy appearances.

The most common morphological pattern is a membranoproliferative exudative glomerulonephritis with frequent presence of intraluminal thrombi.

By immunofluorescence the deposits are positive for IgM, IgG and C3 (cryoimmunoglobulins).