Effectiveness of cryofiltration and mizoribine combination with oral steroid therapy in a patient with membranoproliferative glomerulonephritis due to essential cryoglobulinemia

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Abstract

A 65-year-old male patient with nephrotic syndrome was admitted to our hospital due to worsening systemic edema and purpura on the limbs. He had an impaired renal function, low serum complement level, and elevated rheumatoid factor level. He was positive for cryoglobulin (monoclonal IgM-κ and polyclonal mixed-type IgG), and the results of his kidney biopsy showed a tissue profile of membranoproliferative glomerulonephritis (MPGN). Due to the fact that the secondary cause was unclear, he was diagnosed with MPGN due to essential mixed cryoglobulinemia. On hospital day 20, he was initiated on 50 mg/day prednisolone (PSL). On hospital day 43, oral mizoribine (MZR) at a dose of 150 mg/day was prescribed. On hospital day 49, cryofiltration was performed because the disease was steroid resistant. The treatment promptly decreased urine protein levels. Serum albumin and serum complement levels increased, and complete remission was achieved approximately three months after the initiation of treatment. The PSL and MZR doses were gradually reduced to 2 mg/day and 100 mg/day, respectively, without any reemergence of the symptoms of cryoglobulinemia or relapse of the nephrotic syndrome for three years. Here, we report this case with essential mixed cryoglobulinemia in whom we could achieve complete remission of the disease by adding cryofiltration to the oral corticosteroid and immunosuppressant therapy with mizoribine and could maintain for a long time.

Keywords Cryoglobulin · Cryofiltration · Mizoribine · Membranoproliferative glomerulonephritis

Introduction

Approximately, 90% of cases with mixed cryoglobulinemia are related to hepatitis C infection. Cases of essential mixed cryoglobulinemia, with unknown underlying disease, are rare, accounting for approximately 5% of all cases [1]. Approximately, 55% and 20% of patients with essential mixed cryoglobulinemia have been reported to develop renal impairment and nephrotic syndromes, respectively.

Currently, this disease has no well-established therapy and its 10-year survival rate is 49% [2]. It has been shown in a report that this 10-year survival rate has significantly improved to 65% using corticosteroid drugs, immunosuppression, and plasma exchange [3]; however, these therapies have not yet been established for essential mixed cryoglobulinemia. We report here a case of a patient with membranoproliferative glomerulonephritis (MPGN) due to essential mixed cryoglobulinemia, who achieved long-term complete remission by immunosuppressant therapy with mizoribine (MZR) and cryofiltration in addition to a corticosteroid drug.

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Case report

A 65-year-old male patient had been well until several weeks before admission, when he found edema of his face and lower legs with 8 kg weight gain in two weeks. Chest

X-ray showed a dilated cardiothoracic ratio and pleural effusion, and he was admitted to a local hospital. The symptoms improved with diet therapy (NaCl 5 g/day) and administration of diuretics, and he was discharged. After two months, low-grade fever persisted, and purpura appeared on all of his limbs. He had elevated levels of inflammatory reaction and rheumatoid factor (RF), and he was positive for antinuclear antibodies. Skin biopsy was performed to investigate the purpura, but the findings were nonspecific. Several weeks later, the facial and bilateral lower leg edema aggravated, and urinalysis revealed urinary protein excretion of approximately 6.8 g/gCr and 2+ hematuria, and the sediment contained 30–49 red blood cells and 1–4 white blood cells per high-power field, as well as various types of casts. Renal function deteriorated with serum creatinine of 2.13 from 1.09 mg/dL. He was hospitalized at our university hospital.

History of past diseases

The patient had pulmonary tuberculosis when he was 13 years old and hypertension when he was 64 years old.

History of smoking and alcohol consumption

The patient has been smoking 10 cigarettes per day for 40 years and consumed approximately 180 mL of shōchū (a distilled spirit) per day.

Oral drugs per day at admission

The patient was given 40 mg of Olmesartan, 10 mg of amlodipine, 80 mg of furosemide, 25 mg of spironolactone, and 100 mg of ferrous citrate.

Physical condition at admission

The patient's height was 163.0 cm, weight 62.4 kg, temperature 36.6 °C, blood pressure 146/73 mmHg, and pulse 60 beats/min, with pale palpebral conjunctiva. He had purpura on the joints of the right hand and the lower legs, as well as prominent lower leg edema. No distinct neurological abnormality was noticed.

Laboratory findings at admission (Table 1)

Urinalysis results showed that the patient showed 3+ both for proteinuria and for urine occult blood and the presence of various types of casts. The 24 h urinary protein excretion was 3.63 g/day, and the serum albumin level was 2.99 g/dL. The selectivity index was low at 0.31. The levels of urea nitrogen and serum creatinine were 33.1 mg/dL and 2.00 mg/dL, respectively, indicating advanced impairment of renal function. Severe anemia at hemoglobin of 6.0 g/dL

was detected and the platelet count was low, at 9.4×10^4 μ L. Immunological tests showed elevated IgM levels at 1309 mg/dL and decreased complement levels of C3 and C4 at 46 mg/dL and 3 mg/dL, respectively. Serum and urine immunoelectrophoresis revealed monoclonal IgM- κ monoclonal protein and polyclonal IgG, but histological examination of the bone marrow showed normocellular bone marrow. The rheumatoid arthritis particle agglutination (RAPA) titer, RF level, and antinuclear antibody titer were 1280, 2441 U/mL, and 2560, respectively. The patient was negative for antibodies of various connective tissue diseases. He had hepatitis B surface antigen at 0.01 IU/mL and he tested negative for hepatitis C virus (HCV) antibody, which ruled out hepatitis B and C infections. The patient's serum was negative for *Mycoplasma*, cytomegalovirus, Epstein–Barr virus, and Group A β -hemolytic streptococci. Cryoglobulin was positive in the qualitative cryoglobulin test. A large amount of precipitate was formed at 4 °C, which redissolved at 37 °C (Fig. 1).

Image analysis findings at admission

Chest X-rays showed cardiomegaly and bilateral pleural effusion. Abdominal ultrasonography showed bilateral enlargement of the kidneys, with the right and left kidneys measuring 11.7×5.0 cm and 12.7×5.2 cm, respectively. However, whole-body computed tomography scans did not show any lymph node enlargement or splenomegaly.

Kidney biopsy findings (hospital day 8)

The renal biopsy specimen included only the renal cortex, with 32 glomeruli. The glomeruli showed lobulation and duplication of the basement membrane, an increase in the mesangial matrix, and proliferation of mesangial cells [Fig. 2a, b]. Narrowing of the lumens and protein clot thrombus were also seen [Fig. 2a]. Infiltration of inflammatory cells, including neutrophils, into the renal tubules and interstitium was also noted. Some of the arterioles showed signs suggestive of vasculitis with fibrinoid necrosis [Fig. 2c]. In the immunofluorescence analysis, staining was positive for C3 and IgM and negative for IgG, IgA, C4, C1q, and light chains [Fig. 2f]. Electron microscopy showed subendothelial electron dense deposits, with a fine tubulo-fibrous structure [Fig. 2d, e].

Clinical course

Figure 3 shows the clinical course of the patient after admission. The patient was diagnosed with MPGN according to the pathologic findings. The cause of the MPGN seemed to be type II essential mixed cryoglobulinemia. Treatment with prednisolone (PSL) 50 mg/day was started

Table 1 Laboratory data on admission

Urinalysis		Serological (Immunology)	
Proteinuria	(3+) 3.63 g/day	C-reactive protein	1.56 mg/dL
Occult blood	(3+)	IgA	270 mg/dL
Sugar	(–)	IgM	1309 mg/dL
Red blood cell	1–4 HPF	IgG	1087 mg/dL
White blood cell	10–19 HPF	C3	46 mg/dL
Deformed red blood cells	(+)	C4	3 mg/dL
Fatty cast	(+)	Others	
Oval fat body	(+)	ASK	320 fold
Chemistry		ASO	131 IU/mL
AST	14 IU/L	RAPA	1280 fold
ALT	5 IU/L	Rheumatoid factor	2441 U/mL
LDH	282 IU/L	Antinuclear antibody	2560 fold
ALP	204 IU/L	Anti ds-DNA antibody	1.4 IU/mL
Gamma-GTP	58 IU/L	Anti ss-DNA antibody	4.4 AU/mL
Total protein	6.11 g/dL	Anti SS-A antibody	≤ 1.0 U/mL
Albumin	2.99 g/dL	Anti SS-B antibody	≤ 1.0 U/mL
Blood urea nitrogen	33.1 mg/dL	MMP-3	112 ng/mL
Creatinine	2.00 mg/dL	Anti CCP antibody	< 0.5 U/mL
Na	139 mEq/L	PR3-ANCA	< 10 EU
K	3.4 mEq/L	MPO-ANCA	< 10 EU
Cl	105 mEq/L	HBs antigen	0.01 IU/mL
Corrected calcium	8.98 mg/dL	HBs antibody	Negative
Phosphate	3.95 mg/dL	HBc antibody	Negative
Free T4	1.17 ng/dL	HCV antibody	Negative
TSH	11.6 μIU/mL	Mycoplasma	< 40 fold
Serum iron	44 μg/dL	Cytomegalovirus	Negative
UIBC	139 μg/dL	Cytomegalovirus IgM	Negative
Ferritin	351 ng/mL	EBV anti VCA IgG	160 fold
ESR	156 mm/h	EBV anti VCA IgM	< 10 fold
Complete blood cell count		Streptococcus pyogenes	Negative
White blood cell	5500 μL	CEA	0.9 ng/mL
Red blood cell	203 × 10 ⁴ μL	CA19-9	5.0 U/mL
Hemoglobin	6.0 g/dl	IL-2R	1601 U/mL
Hematocrit	17.3%	Procarcitonine	≤ 0.1 mg/mL
Platelet count	9.4 × 10 ⁴ μL	Clyoglobulin	positive
		Selectivity index	0.31
		M protein	IgM-κ

HPF highper per field, *AST* aspartate transaminase, *ALT* alanine aminotransferase, *LDH* lactate dehydrogenase, *ALP* alkaline phosphatase, *γ-GTP*, gamma-glutamyl transpeptidase, *TSH* thyroid stimulating hormone, *UIBC* unsaturated iron binding capacity, *ESR* erythrocyte sedimentation rate, *ASK* anti-streptokinase antibody, *ASO* anti-streptolysin O antibody, *RAPA* rheumatoid arthritis particle agglutination, *MMP* metalloproteinase, *CCP* cyclic citrullinated peptides, *ANCA* antineutrophil cytoplasmic antibody, *HB* hepatitis B, *HCV* hepatitis C virus, *EBV* epstein-barr virus, *CEA* carcinoembryonic antigen

on hospital day 20. Although the urine protein levels decreased temporarily with this treatment, they increased again. Thus, we added MZR 150 mg/day from hospital day 43, and cryofiltration was started once a week on hospital day 49. A Plasauto iQ21 (Asahi Kasei Corp., Tokyo, Japan) was used for the cryofiltration. Plasmaflo OP-08 (Asahi Kasei Corp.) was fitted as the plasma filtration

membrane, Cascadeflo EC-50 W (Asahi Kasei Corp.) was fitted as the plasma component separation membrane, a dedicated circuit DFPP-21 (Asahi Kasei Corp.) was fitted as the blood circuit, and a coiled tube that was ice cooled to 0–4 °C was fitted to the plasma outlet. At the start of the filtration, 2000 units of heparin Na were injected intravenously as an anticoagulant, and 1000 units

Fig. 1 Cryoglobulinemia was confirmed by the detection of the protein that precipitates in the serum maintained at 4 °C and dissolves at 37 °C

were administered hourly as maintenance. The vascular access was through the femoral vein for removing blood from the patient and the median cubital vein for returning

the blood to the patient. Approximately, 2000–3000 mL of plasma was treated per session. Clogging during cryofiltration became progressively less as the sessions of cryofiltration progressed. The cryocrit was 16.5% before cryofiltration and decreased to 2.0% after four rounds of cryofiltration (Fig. 4). After cryofiltration, the IgG and IgM immunoglobulin levels decreased to 214 mg/dL and 296 mg/dL, respectively, which subsequently showed a gradual improvement. Because urine protein levels rapidly decreased to 0.72 g/day (incomplete remission) and the serum albumin and complement levels increased, cryofiltration was discontinued at the fifth session. The serum level of MZR was 3.78 µg/mL. Therefore, the patient was discharged on hospital day 88. One month after the discharge, the patient showed complete remission, with urinary protein excretion of 0.27 g/gCr/day. Currently, three years after the disease onset, the PSL and MZR doses have been gradually reduced to 2 mg/day and 100 mg/day, respectively. Currently, his serum creatinine and serum albumin levels are 1.49 mg/dL and 3.80 g/dL, respectively, and urinary protein excretion is 0.19 g/gCr/day. As for the complements, C3 has increased to 69 mg/dL, but C4 remains low at 5 mg/dL.

Fig. 2 Renal biopsy findings. **a** An increase in the mesangial substance and intraluminal thrombus is observed in the glomeruli (arrow) (periodic acid–Schiff staining, original magnification $\times 400$). **b** Glomerulus showing thickening of the glomerular basement membrane with a double-contoured appearance (periodic acid methenamine silver staining, original magnification $\times 400$). **c** Arterioles showing fibrinoid necrosis (arrow) (hematoxylin and eosin staining,

original magnification $\times 400$). **d** Electron deposits are present in the subendothelial space on electron microscopy (original magnification $\times 5000$). **e** Subendothelial deposits with organized tubular structures (original magnification $\times 30,000$). **f** Immunofluorescence microscopy image showing intense staining of C3 along the glomerular capillary walls (original magnification $\times 400$)

Fig. 3 Clinical course of the patient. PSL prednisolone, MZR mizoribine, Cr creatinine

Fig. 4 Cryocrit throughout the patient's treatment course. The patient's cryocrit was 16.5% at the time of hospitalization. His cryocrit continued to decrease down to 2.0% after cryofiltration and MZR treatment

Discussion

In this case, we could achieve long-term complete remission of MPGN caused by essential mixed cryoglobulinemia using MZR and cryofiltration combined with a corticosteroid drug. Cryoglobulinemia is classified into three types: monoclonal (type I), mixed (polyclonal and monoclonal, type II), and multipolyclonal (with two polyclones,

type III). Type I develops after lymphoproliferative diseases, such as multiple myeloma, primary macroglobulinemia, and malignant lymphoma. Approximately, 19% of cases with type II cryoglobulinemia occur after hepatitis C infection, and type III often develops when an underlying connective tissue disease or chronic inflammation is present. Essential mixed cryoglobulinemia with an unknown underlying disease is very rare, accounting for approximately 5% of the cases [1]. Cryoglobulinemic glomerulonephritis is caused by intracapillary, capillary wall (often subendothelial), and mesangial cryoglobulin deposits, giving an MPGN pattern of injury. Patients develop nephritic/nephrotic syndromes with various levels of kidney function [4]. MPGN is one of the most common causes of nephrotic syndromes in both children and adults [5, 6]. In the Japan Renal Biopsy Registry from 2007 to 2015, 261 out of 593 patients were registered as “secondary” MPGN, including 32 patients with cryoglobulinemia [7].

Age ≥ 50 years, purpura, splenomegaly, Cre > 1.5 mg/dL, IgG < 800 mg/dL, C3 < 54 mg/dL, and cryocrit $> 10\%$ have been listed as prognostic determining factors for essential mixed cryoglobulinemia [2]. In addition, a severity score based on peripheral neuropathy, cutaneous symptoms, vasculitis, peripheral ischemia, and joint symptoms has also been reported [8]. The present case seems to belong to the severe category, considering the age, purpura, elevated Cre level, decreased C3 level, and high cryocrit.

Essential cryoglobulinemia has no established treatment. Generally, a corticosteroid drug, immunosuppressant therapy, and plasmapheresis are used in combination [9, 10]. In plasmapheresis for type I cryoglobulinemia, simple plasma

exchange is basically performed because the immunoglobulins to be removed are relatively abundant. Alternatively, plasmapheresis using a secondary membrane and cryofiltration can also be employed. The amount of cryoglobulin is relatively less, and its molecular weight is large in types II and III. Therefore, they can be efficiently removed even by plasmapheresis using a secondary membrane with a relatively large pore size, which is generally used to remove IgM. Cryofiltration can also be used. The cryofiltration method used in the present case was developed by Nose and his group in 1980 [11]. It is a modification of double-filtration plasma exchange, where the plasma separated by the membrane is cooled down to precipitate the cryoglobulin for removal, which is warmed before being returned to the body of the patient. Unlike in double-filtration plasma exchange, in the modified method, cryogels, which are complexes of immunoglobulins, fibrinogen, and other components, form when the plasma is cooled down, and the cause of the disease can be efficiently removed [11]. Taniyama et al. [12] reported four cases of mixed cryoglobulinemia where cryofiltration was used successfully. Among these four, three cases were HCV-associated cryoglobulinemia and one was essential mixed cryoglobulinemia, which had the complications of a nephrotic syndrome and acute kidney injury; the concurrent use of cryofiltration and an adrenocortical drug was very effective. Wada et al. [13] also used cryofiltration on a patient with essential mixed cryoglobulinemia with the complication of acute renal failure and they succeeded in freeing the patient from dialysis. To date, evidence on the effectiveness of cryofiltration in treating essential mixed cryoglobulinemia is still insufficient, but it has been reported to provide immediate relief to patients with severe clinical symptoms, similar to the present case. We need to continue accumulating evidence.

With regard to immunosuppressant therapy for essential mixed cryoglobulinemia, the effectiveness of anti-B cell therapies (e.g., with rituximab), in addition to corticosteroids and cyclophosphamide, has also been reported recently [14, 15]. Cryoglobulin is produced as a result of genetic and environmental factors, additionally playing some role in the onset mechanism of cryoglobulinemia. For example, persistent infections, such as HCV, stimulate the proliferation of B-lymphocytes. Thus, anti-B-cell therapies are considered promising. MZR is a purine metabolism antagonist that was developed in Japan. In the body, it is metabolized to mizoribine-5'-phosphate, thus inhibiting inosine monophosphate dehydrogenase, a rate-limiting enzyme of the de novo purine biosynthesis pathway. This, in turn, inhibits the proliferation and activity of not only activated T-lymphocytes, but also B-lymphocytes [16]. Because of its anti-B-cell activity, MZR may potentially be effective for cryoglobulinemia. In fact, it has been shown in a report that the combination of Predonine with MZR yielded high effectiveness

against skin purpura and ulcers caused by cryoglobulinemia [17]. In addition, the effectiveness of MZR against steroid-resistant nephrotic syndromes [18], like the present case, has been reported. Moreover, MZR has been reported to have an enhancing action on the effect of corticosteroid drugs. It was shown to act on the conformation of 14–3–3 proteins, affecting their binding with glucocorticoid receptors and enhancing the transcriptional activity of these receptors in a dose-dependent manner [19]. Therefore, MZR would be effective in steroid-resistant conditions. The advantages of MZR include the fact that it has relatively mild adverse effects on elderly patients and it can be used for a long time easily. The combination use of MZR has been successful also as a maintenance therapy after remission. Therefore, MZR needs further investigation as one of the immunosuppressant choices against essential cryoglobulinemia.

The complement activation system plays an important role in triggering cryoglobulin-induced MPGN. The complement system is constructed from three activation pathways: classical, alternative, and lectin pathways. Particularly, the classical pathway is associated with the progression of immunoglobulin-positive MPGN, such as cryoglobulinemic glomerulonephritis [20].

Cryoglobulinemic glomerulonephritis includes low C4 levels in three quarters and low C3 levels in half [4]. With regard to hypocomplementemia, C3 is included as an item in the severity classification, and our patient responded to the treatment with an improvement in the C3 level, along with an improvement in proteinuria and other symptoms. The C3 level here probably reflects the status of the disease. However, the patient's C4 level remained low. The lowering of C4 is believed to be due to its excess consumption caused by the immunological response. This is often seen in patients with cryoglobulinemia [21, 22]. C4 is not always correlated with the severity of cryoglobulinemic vasculitis [23], and the low C4 level in our patient also did not seem to reflect the severity of the disease.

Conclusion

In the present case, we could achieve long-term complete remission of steroid-resistant MPGN caused by essential cryoglobulinemia by a combination therapy of MZR and cryofiltration in addition to a steroid. An immediate effect could be achieved in this severe case by cryofiltration in the early stage of treatment.

Compliance with ethical standards

Conflict of interest All authors declare that they have no conflict of interest.

Ethical approval This case report was approved by the Institutional Ethics Committee of Kurume University School of Medicine (2017-045).

Informed consent Informed consent was obtained from the patient.

References

- Charles ED, Dustin LB. Hepatitis C virus-induced cryoglobulinemia. *Kidney Int.* 2009;76(8):818–24. <https://doi.org/10.1038/ki.2009.247>.
- Tarantino A, Campise M, Banfi G, Confalonieri R, Bucci A, Montoli A, et al. Long-term predictors of survival in essential mixed cryoglobulinemic glomerulonephritis. *Kidney Int.* 1995;47(2):618–23.
- Terrier B, Carrat F, Krastinova E, Marie I, Launay D, Lacraz A, et al. Prognostic factors of survival in patients with non-infectious mixed cryoglobulinaemia vasculitis: data from 242 cases included in the CryoVas survey. *Ann Rheum Dis.* 2013;72(3):374–80. <https://doi.org/10.1136/annrheumdis-2012-201405>.
- Fogo AB, Lusco MA, Najafian B, Alpers CE. AJKD Atlas of renal pathology: cryoglobulinemic glomerulonephritis. *Am J Kidney Dis.* 2016;67(2):e5–7. <https://doi.org/10.1053/j.ajkd.2015.12.007>.
- Sugiyama H, Yokoyama H, Sato H, Saito T, Kohda Y, Nishi S, et al. Japan renal biopsy registry and Japan kidney disease registry: committee report for 2009 and 2010. *Clin Exp Nephrol.* 2013;17(2):155–73. <https://doi.org/10.1007/s10157-012-0746-8>.
- Yokoyama H, Sugiyama H, Sato H, Taguchi T, Nagata M, Matsuo S, et al. Renal disease in the elderly and the very elderly Japanese: analysis of the Japan Renal Biopsy Registry (J-RBR). *Clin Exp Nephrol.* 2012;16(6):903–20. <https://doi.org/10.1007/s10157-012-0673-8>.
- Nakagawa N, Hasebe N, Hattori M, Nagata M, Yokoyama H, Sato H, et al. Clinical features and pathogenesis of membranoproliferative glomerulonephritis: a nationwide analysis of the Japan renal biopsy registry from 2007 to 2015. *Clin Exp Nephrol.* 2018;22(4):797–807. <https://doi.org/10.1007/s10157-017-1513-7>.
- Stefanutti C, Vivenzio A, Di Giacomo S, Labbadia G, Mazza F, D'Alessandri G, et al. Immunoabsorption apheresis and immunosuppressive drug therapy in the treatment of complicated HCV-related cryoglobulinemia. *J Clin Apher.* 2009;24(6):241–6. <https://doi.org/10.1002/jca.20222>.
- Rockx MA, Clark WF. Plasma exchange for treating cryoglobulinemia: a descriptive analysis. *Transfus Apher Sci.* 2010;42(3):247–51. <https://doi.org/10.1016/j.transci.2010.03.001>.
- Ramos-Casals M, Stone JH, Cid MC, Bosch X. The cryoglobulinaemias. *Lancet.* 2012;379(9813):348–60. [https://doi.org/10.1016/S0140-6736\(11\)60242-0](https://doi.org/10.1016/S0140-6736(11)60242-0).
- Nosé Y, Horiuchi T, Malchesky PS, Smith JW, Matsubara S, Abe Y. Therapeutic cryogel removal in autoimmune disease: what is cryogel, 1982. *Ther Apher.* 2000;4(1):38–43.
- Taniyama Y, Nakatani Y, Matsuoka T, Takahashi M, Shimizu K, Yamamoto S, et al. Efficacy of cryofiltration for treatment of mixed cryoglobulinemia: a report of four cases. *Ther Apher Dial.* 2017;21(3):238–42. <https://doi.org/10.1111/1744-9987.12556>.
- Wada Y, Saeki T, Miyamura S, Osawa Y, Ueno M, Nishi S, et al. Successful treatment of acute renal failure in a patient with essential mixed cryoglobulinemia using prednisolone and cryofiltration. *Nihon Jinzo Gakkai Shi.* 1999;41(1):29–33.
- Terrier B, Launay D, Kaplanski G, Hot A, Larroche C, Cathébras P, et al. Safety and efficacy of rituximab in nonviral cryoglobulinemia vasculitis: data from the French Autoimmunity and Rituximab registry. *Arthritis Care Res (Hoboken).* 2010;62(12):1787–95. <https://doi.org/10.1002/acr.20318>.
- Terrier B, Krastinova E, Marie I, Launay D, Lacraz A, Belenotti P, et al. Management of noninfectious mixed cryoglobulinemia vasculitis: data from 242 cases included in the CryoVas survey. *Blood.* 2012;119(25):5996–6004. <https://doi.org/10.1182/blood-2011-12-396028>.
- Kamada H, Itoh H, Shibata H, Koshio T, Hayashi A, Nakagami K. Inhibitory mechanism of mizoribine on the antibody production of mouse B cells stimulated with lipopolysaccharide. *Jpn J Pharmacol.* 1997;74(4):323–30.
- Hamaguchi M, Kawahito Y, Tsubouchi Y, Kohno M, Yamamoto A, Ishino H, et al. Combination therapy of prednisolone and mizoribine improves cryoglobulinemic vasculitis with purpura and skin ulcers. *Clin Rheumatol.* 2007;26(7):1170–2. <https://doi.org/10.1007/s10067-006-0295-1>.
- Shibasaki T, Koyama A, Hishida A, Muso E, Osawa G, Yamabe H, et al. A randomized open-label comparative study of conventional therapy versus mizoribine onlay therapy in patients with steroid-resistant nephrotic syndrome (postmarketing survey). *Clin Exp Nephrol.* 2004;8(2):117–26. <https://doi.org/10.1007/s10157-004-0276-0>.
- Takahashi S, Wakui H, Gustafsson JA, Zilliacus J, Itoh H. Functional interaction of the immunosuppressant mizoribine with the 14-3-3 protein. *Biochem Biophys Res Commun.* 2000;274(1):87–92. <https://doi.org/10.1006/bbrc.2000.3104>.
- Mizuno M, Suzuki Y, Ito Y. Complement regulation and kidney diseases: recent knowledge of the double-edged roles of complement activation in nephrology. *Clin Exp Nephrol.* 2018;22(1):3–14. <https://doi.org/10.1007/s10157-017-1405-x>.
- Hiramatsu R, Hoshino J, Suwabe T, Sumida K, Hasegawa E, Yamanouchi M, et al. Membranoproliferative glomerulonephritis and circulating cryoglobulins. *Clin Exp Nephrol.* 2014;18(1):88–94. <https://doi.org/10.1007/s10157-013-0810-z>.
- Ferri C, Mascia MT. Cryoglobulinemic vasculitis. *Curr Opin Rheumatol.* 2006;18(1):54–63.
- Ferri C, Sebastiani M, Giuggioli D, Cazzato M, Longombardo G, Antonelli A, et al. Mixed cryoglobulinemia: demographic, clinical, and serologic features and survival in 231 patients. *Semin Arthritis Rheum.* 2004;33(6):355–74.

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