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Case Report

Mizoribine Pulse Therapy for a Pediatric Patient with Steroid-Resistant Nephrotic Syndrome

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TANAKA, H., TSUGAWA, K., NAKAHATA, T., KUDO, M., ONUMA, S., KIMURA, S. and ITO, E. *Mizoribine Pulse Therapy for a Pediatric Patient with Steroid-Resistant Nephrotic Syndrome*. Tohoku J. Exp. Med., 2005, **205** (1), 87-91 —— A 12-year-old Japanese boy was referred to our hospital with a 2-month history of persistent proteinuria. Despite urinary protein excretion in the nephrotic range, associated with hypoproteinemia, the patient did not complain of any disability. A percutaneous renal biopsy revealed minor glomerular abnormalities, without any evidence of immune complex deposition. Therapy with prednisolone (60 mg/day) was initiated, and while the proteinuria decreased after 4-week therapy, elevated urinary protein excretion persisted thereafter, at 1-2 g/day. Because of the steroid-resistant proteinuria, mizoribine (MZR), was started at 150 mg/day (3 mg/kg), administered as a single daily dose an immunosuppressive agent, in combination with prednisolone. Although there was some fluctuation in the urinary protein excretion, heavy proteinuria persisted for the next 4 weeks. The peak blood level of MZR was 0.9 μg/ml. Since we have previously reported the efficacy and safety of oral MZR pulse therapy, which is associated with higher peak serum MZR levels than conventional MZR therapy in selected patients with lupus nephritis, we adopted MZR pulse therapy for this patient, after obtaining informed consent. MZR was started at the daily dose of 300 mg (6 mg/kg), administered as a single dose before breakfast, twice a week (on Monday and Thursday). The peak blood level of MZR then increased to 1.29 μg/ml. Thereafter, despite a gradual reduction of the concomitantly administered prednisolone dose, the urinary protein excretion decreased rapidly to around 0.3 g/day and remained at this level thereafter. No adverse effects of MZR were observed. Based on these clinical observations, we suggest that oral MZR pulse therapy may be the treatment of choice in selected patients of steroid-resistant nephrotic syndrome, in addition to those of lupus nephritis. ———— anti-proteinuric effect; mizoribin; oral mizoribine pulse therapy; steroid-resistant nephrotic syndrome

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Mizoribine (MZR) is a novel immunosuppressive agent that was developed for the first time in Japan. It has been reported to cause selective inhibition of inosine monophosphate dehydrogenase, which results in inhibitory effects against T-cell and B-cell proliferation (Yokota 2002). The drug has been successfully used without serious adverse effects in cases undergoing renal transplantation (Sonda et al. 1996) and cases of nephrotic syndrome (NS) (Igarashi et al. 1994; Hamasaki et al. 1997; Yoshioka et al. 2000; Honda 2002; Shibasaki et al. 2004), IgA nephropathy (Kawasaki et al. 2004) and lupus nephritis (Tanaka et al. 2003a, b, 2004a, b; Yoshidome et al. 2004). However, perhaps in part due to its relatively low efficacy, the clinical use of MZR for the treatment of NS is not as widespread as that of cyclophosphamide (CPA) or cyclosporin A (CsA).

We have previously reported the clinical usefulness of oral MZR pulse therapy, which is associated with increased serum levels of MZR, may be associated with higher clinical efficacy without any significant increase clinical toxicity as compared to conventional MZR therapy at least in selected patients of lupus nephritis (Tanaka et al. 2003a, b, 2004a). We, therefore, adopted oral MZR pulse therapy for a boy with steroid-resistant NS, in whom the conventional MZR therapy in combination with prednisolone (PSL) proved to have inadequate anti-proteinuric effect.

Case Report

A previously healthy Japanese boy aged 12 years was found to have 3+ of proteinuria during a mass-screening examination at school. He was referred to a regional hospital, where mild dependent peripheral edema was recorded. Urinalysis revealed a proteinuria of 497 mg/100 ml and 10-19 red blood cells (RBC) per high-powered field (HPF), while the urine culture was sterile. The laboratory examination results were as follows: serum total protein, 5.8 g/100 ml; serum albumin, 3.9 g/100 ml; serum total cholesterol, 226 mg/100 ml; blood urea nitrogen (BUN), 16 mg/100 ml; serum creatinine, 0.5 mg/100 ml; serum sodium, 140 mmol/liter; serum potassium, 4.3 mmol/liter and serum chloride, 106 mmol/liter. The results of immunological tests were unremarkable. Abdominal ultrasonography revealed no abnormalities. Since the patient had no disability, he was observed conservatively. However, the urinary protein excretion fluctuated around 3 g/day, and he was referred to Hirosaki University Hospital for further examination 2 months after he was first seen by a doctor.

On admission, the patient appeared quite well. Physical examination revealed a well-developed boy, with a height of 145.4 cm (–0.3 S.D.) and body weight of 51.0 kg (+1.2 S.D.); his blood pressure was 94/56 mmHg, and he had no skin or mucosal lesions. Urinalysis revealed a urinary specific gravity of 1.024, 24-hour urinary protein excretion of 4.0 g and 2 RBCs per HPF. The urinary β2-microglobulin was 320 μg/liter (normal, < 300 μg/liter). Other laboratory studies data were as follows: normal hemogram (total leukocyte, 6120/μl; hemoglobin, 13.1 g/100 ml; hematocrit, 36.7%; platelet count, 292,000/μl); serum total protein, 5.5 g/100 ml; serum albumin, 3.1 g/100 ml; serum total cholesterol, 276 mg/100 ml; BUN, 12 mg/100 ml; serum creatinine, 0.6 mg/100 ml; serum sodium, 143 mmol/liter; serum potassium, 4.5 mmol/liter; serum chloride, 110 mmol/liter; serum C-reactive protein, < 0.1 mg/100 ml. Immunological studies revealed the following results: IgG, 796 mg/100 ml; IgA, 323 mg/100 ml; IgM, 156 mg/100 ml; IgE, 623 IU/ml (normal, < 295 IU/ml); C3, 118 mg/100 ml (normal range, 79-152 mg/100 ml); C4, 18 mg/100 ml (normal range, 16-38 mg/100 ml); hemolytic complement activity, 51.2 U/ml (normal range, 23-46 U/ml); anti-nuclear antibody, 1:160, and anti-dsDNA antibody, 4.0 IU/ml (normal, < 12.0 IU/ml). The serum was negative for hepatitis B antigens and hepatitis C antibody. The creatinine clearance rate was 174.3 ml/minute per 1.43 m^2. Intravenous pyelography disclosed no urinary tract abnormalities. A percutaneous renal biopsy on Hospital day 4 revealed minor glomerular abnormalities with no segmental or tubulointerstitial lesions. There was no evidence of immune complex deposition.

The patient was initiated on PSL, 60 mg/day, from Hospital day 9; while the urinary protein ex-

cretion decreased to around 1-2 g/day after 4 weeks, it remained persistent at this level thereafter. Considering the patient as a case of steroid-resistant proteinuria, the PSL dose was reduced to 40 mg/alternate day and a combination with MZR was initiated at 150 mg/day (3 mg/kg), administered as a single-dose, form Hospital day 37. The peak blood level of MZR, determined 2 hours post dosing, was 0.91 μg/ml (Tanaka et al. 2003a, b). Cyclophosphamide was not administered to this patient to avoid gonadal toxicity. Since the proteinuria decreased to the non-nephrotic range proteinuria at this time, methylprednisolone pulse therapy was not considered. Cyclosporin A was avoided as it is more expensive than MZR.

However, despite some fluctuation in the urinary protein excretion, the proteinuria persisted over 4 weeks, therefore, oral MZR pulse therapy was initiated after obtaining informed consent, i.e., 300 mg (6 mg/kg) of the drug was administered as a single daily dose before breakfast twice a week (on Monday and Thursday) (Tanaka et al. 2003a, b, 2004a). The peak blood level of MZR then increased to 1.29 μg/ml, and despite a gradual reduction of the concomitantly administered PSL dose, the urinary protein excretion decreased rapidly to around 0.3 g/day and remained at this level thereafter (Fig. 1). Total protein, albumin and total cholesterol values in the serum also returned to normal (6.2 g/100 ml, 3.9 g/100 ml and 160 mg/100 ml, respectively) within a month following oral MZR pulse therapy. No adverse effects of MZR, such as leukocytopenia, gastrointestinal symptoms, and liver dysfunction, were observed in the patient.

Discussion

Although MZR has been successfully applied without serious adverse effects in the treatment of NS (Igarashi et al. 1994; Hamasaki et al. 1997; Yoshioka et al. 2000; Honda 2002; Shibasaki et al. 2004), its clinical use is still not widespread, perhaps in part due to its relatively low efficacy. In regard to the blood levels of MZR, it has been reported that during regular MZR therapy, i.e., administration at 3 mg/kg per day in three divided daily doses, the peak levels of the drug are apparently, at least around 0.5 μg/ml (Hamasaki et al. 1997), lower than those required to inhibit experimental human mixed-lymphocyte reaction, which occurs in the range of 3.0-6.0 μg/ml (Sonda et al. 1996). In this context, Honda (2002) recently reported that large doses of MZR of more than 5 mg/kg might be effective

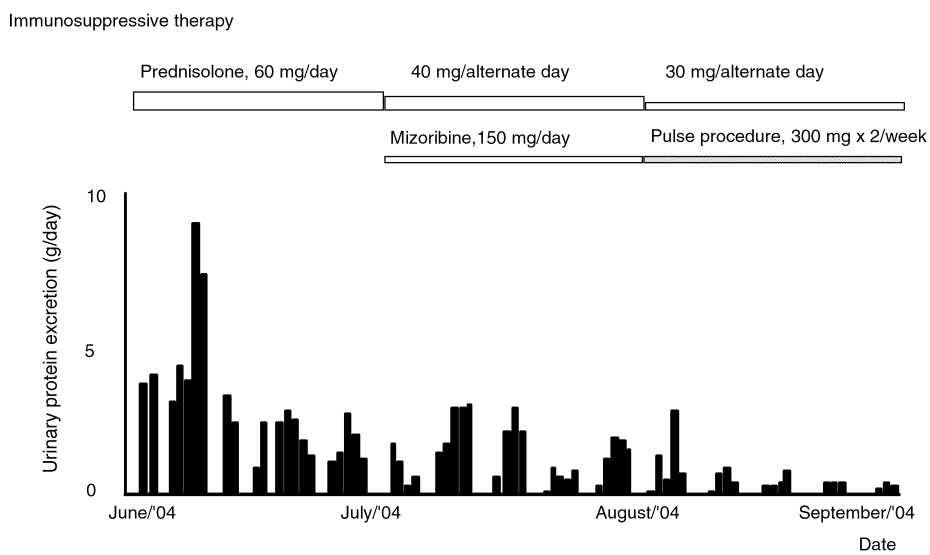

Fig 1. Clinical course of the patient. Mizoribine pulse therapy resulted in a rapid decrease of the urinary protein excretion of the patient despite a gradual reduction of the concomitantly administered prednisolone dose.

in patients with steroid-resistant NS. Since MZR is excreted rapidly into the urine (Yokota 2002), accumulation of the drug is, in general, not a problem, at least under the condition of normal renal function (personal communication from Dr. W. Yumura, Department of Medicine, Kidney Center, Tokyo Women's Medical University). Indeed, it has been reported that higher doses of MZR increase the area under the serum concentration-time curve in an adolescent lupus patient with normal renal function, which improved clinical efficacy without serious adverse effects (Abe et al. 2004). We therefore speculate that the efficacy of MZR may depend on the peak blood level of the drug, which, in turn, may be closely correlated with the area under the concentration curve of the drug. Since we have previously demonstrated the efficacy and safety of oral MZR pulse therapy, which is associated with significantly higher peak blood MZR levels than conventional MZR therapy, in selected patients of lupus nephritis (Tanaka et al. 2003a, b, 2004a), we adopted it for this patient after obtaining informed consent from his family.

Oral MZR pulse therapy increased the peak blood level of MZR to 1.29 μg/ml, which resulted in a rapid decrease of the urinary protein excretion despite a gradual reduction of the concomitantly administered PSL dose, nonetheless, the optimal peak blood level of the drug for the treatment of NS still remains to be elucidated. Although spontaneous remission or late response to prior prednisolone in combination with regular daily MZR therapy cannot be excluded, the patient's clinical course as described strongly suggested the efficacy of oral MZR pulse therapy. With the protocol used in this patient, the cumulative dose over 1 week (600 mg: 300 mg × 2 days) was less than that associated with regular daily administration (1050 mg: 150 mg × 7 days), making it also cost-effective for the patient (Tanaka et al. 1999).

Apart from causing selective inhibition of inosine monophosphate dehydrogenase in the *de novo* pathway, MZR has also been reported to reduce the urinary protein excretion levels in rat nephrosis models (Shibasaki et al. 1996). It has been reported that 14-3-3 proteins, that is, MZR-binding proteins, interact with glucocorticoid receptors after MZR administration, to enhance the transcriptional activity of these receptors (Takahashi et al. 2000). These laboratory observations suggest that this might be another favorable anti-proteinuric mechanism of MZR, at least in a proportion of patients with NS, although this still remains speculative.

Since persistent proteinuria has been reported to be harmful to the kidney by itself (Tanaka et al. 2000), oral MZR pulse therapy may be the treatment of choice in selected patients of NS. Further studies are needed on a larger number of patients to confirm the long-term efficacy and safety of this therapy.

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