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*Original Article*

# Efficacy of different regimens for treatment of idiopathic membranous nephropathy

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**Abstract:** treatment regimens for idiopathic membranous nephropathy (IMN) are heavily debated. To date, a consensus on first-line treatment has not been reached. This study aimed to explore the optimal treatment for IMN patients by comparing the efficacy and safety of five treatment methods. Records of 107 patients with IMN proved by renal biopsy in our hospital from January 2004 to January 2016 were retrospectively reviewed. Twenty-two patients received oral prednisone combined with oral cyclophosphamide (group A); 20 patients were treated with oral prednisone combined with intravenous cyclophosphamide (group B). The modified Ponticelli protocol was administered to 19 patients (group C); 21 patients received oral prednisone and intravenous cyclosporine A (group D); 25 patients received oral prednisone combined with oral tacrolimus (group E). Efficacy evaluated by the multilevel model analysis showed that the no remission (NR) rate of female patients with normal lipid values, who had received regimen E was remarkably lower than that in other regimens (compared to group E, A: OR=2.250, $\chi^2$=5.044, P=0.025; B: OR=2.063, $\chi^2$=4.001, P=0.045; C: OR=2.286, $\chi^2$=4.947, P=0.026; D: OR=1.160, $\chi^2$=0.159, P=0.048, respectively). We observed no significant differences in the incidence of adverse events and relapse rate among the five groups (P>0.05). Immunosuppressive treatment with tacrolimus and prednisone seems to be more effective and well tolerated by patients with IMN.

**Keywords:** Efficacy, idiopathic membranous nephropathy, nephrotic syndrome, safety, tacrolimus

## Introduction

Membranous nephropathy is a type of autoimmune glomerular disease mediated by autoantibodies, and is the leading cause of nephrotic syndrome. The mortality associated with idiopathic membranous nephropathy (IMN) is one per million population. Adults aged 40-50 years are at higher risk and the sex ratio (male: female) is approximately 2:1. The classic optical microscope feature of IMN is basement membrane diffuse thickening with spike formation. Immunofluorescence shows particle-like deposition of IgG and C3 mainly along the capillary wall. Electron microscope characteristics include basement membrane thickening and epithelial foot process fusion [1].

A variety of causes lead to the occurrence of membranous nephropathy, such as lupus nephritis, hepatitis B virus-associated nephritis, drugs, and tumor-associated nephropathy. These etiologies lead to secondary membranous nephropathy, which can be reversed by removing or correcting the underlying etiological factors. Conversely, another class of membranous nephropathy which is caused by an unidentifiable factor is termed idiopathic membranous nephropathy (IMN). Clinical studies have found that secondary membranous nephropathies are more common in children and the elderly (75%) than in adults (25%) [2]. Therefore, the diagnosis of IMN should only be made after ruling out secondary membranous nephropathy.

Although the majority of IMN patients experience a benign or indolent course of disease with up to one third undergoing spontaneous remission, 30-40% of IMN patients will progress to end-stage renal failure within 5-15 years [3]. Studies demonstrate that there is no significant effect in treatment of IMN with glucocorticoid alone, either in achieving remission or in reducing the risk of progression towards renal failure [4]. Emerging research suggests that immunosuppressive agents can reduce pro-

teinuria and remarkably improve long-term renal function, but most immunosuppressants have significant side effects which greatly limit their clinical application [5]. Accordingly, the combination of glucocorticoids with immunosuppressive therapy in IMN became an area of new research. A multicenter randomized controlled trial of methylprednisolone plus chlorambucil versus methylprednisolone plus cyclophosphamide in the treatment of IMN suggested that the total remission rate with methylprednisolone and chlorambucil was 82% and with methylprednisolone and cyclophosphamide was 93%, and the recurrence rates were 30.5% and 25%, respectively. It was concluded that both regimens were proved to be effective on IMN, but the cyclophosphamide group had slighter adverse effects compared with the chlorambucil group [6]. In 2010, Goumenos and colleagues evaluated the efficacy of cyclosporin A alone or combined with corticosteroids in patients with IMN. That study suggested that low-dose cyclosporin A combined with or without corticosteroids can lead to induced remission and well-preserved renal function [7].

In recent years, tacrolimus, which was widely used to prevent graft rejection in post-renal transplantation, has been applied to the treatment of IMN [8, 9]. Tacrolimus exerts immunosuppressive effects mainly by interfering with the calcium ion signaling pathway to inhibit the dephosphorylation and proliferation of T lymphocytes. A randomized controlled study from China displayed significantly greater improvements in remission and a better treatment tolerance in the tacrolimus group compared with the cyclophosphamide group [10]. Another prospective analysis of tacrolimus suggested that it has the potential to be a novel therapeutic option for patients with IMN [11].

An increasing amount of clinical research has indicated that a satisfactory curative effect was observed in IMN patients treated by glucocorticoids combined with immunosuppressive agents. Nevertheless, the standard or unified first-line therapy approach for IMN still remains controversial. Besides this, the severe adverse effects of immunosuppressants cannot be ignored. Given the current uncertainties in the management of IMN, it is crucial for us to explore the optimal treatment of IMN. Therefore, we aimed to explore the optimal treatment for IMN patients by comparing the efficacy and safety of five treatment methods.

## Material and methods

### *Subjects*

In this retrospective study, 107 patients were recruited from January 2004 to January 2016 in our hospital. Inclusion criteria included: (1) patients histopathologically diagnosed with IMN; (2) all were followed up for at least one year with complete clinical data; (3) those patients had persistent proteinuria did meet the diagnostic criteria for nephrotic syndrome (>3.5 g/d). Exclusion criteria were: (1) secondary membranous nephropathy caused by systemic lupus erythematosus, hepatitis B and hepatitis C virus, cancer, drugs, or poisons (2) medication history of corticosteroids and immunosuppressants; (3) patients who terminated or changed the treatment arbitrarily.

### *Grouping*

All patients were divided into the following five groups according to different treatment plans. Group A: oral cyclophosphamide (CTX) was administered at a dose of 50 mg twice daily and stopped when the cumulative dose reached 8-9 g. Group B: i.v. CTX pulses (0.8-1.2 g/month) were administered every 2 or 4 weeks, and stopped when the cumulative dose reached 8-9 g. Group C: for the first month, i.v. methylprednisolone pulses were implemented at 1 g/d for 3 days, followed by oral methylprednisolone 0.5 mg/kg/day for 27 days; then for the second month, oral CTX 2.0 mg/kg/day for 1 month. The above treatments were repeated 3 times for a total of 6 months. Group D: oral cyclosporin A (CSA) was given with a dose of 3-5 mg/kg/day for 2 weeks to reach the target CSA blood levels of 100-200 ng/ml, then the dose of CSA was adjusted according to blood concentrations. Group E: oral tacrolimus (TAC) was introduced at 0.05-1 mg/kg/d twice per day initially. The dose was adjusted according to the blood level of TAC as monitored every 2 weeks until achieving a blood trough concentration of 5-10 ng/ml. Oral corticosteroid was administered simultaneously in groups A, B, D and E with 0.6-1 mg/kg/d (the maximum dose cannot exceed 60 mg/d) for 2 to 3 months, tapered gradually to the maintenance dose of 5-10 mg/d. Each group of patients was routine-

**Table 1.** Baseline characteristics of patients ($\bar{x} \pm s$)

| Characteristics | Groups | | | | | P |
|---|---|---|---|---|---|---|
| | A (n=22) | B (n=20) | C (n=19) | D (n=21) | E (n=25) | |
| Gender (%) | | | | | | |
| Male | 64.3 | 77.5 | 56.4 | 57.1 | 71.1 | 0.657 |
| Female | 35.7 | 22.5 | 43.6 | 42.9 | 28.9 | |
| Ages (%, $\bar{x} \pm s$) | 46.21±12.77 | 47.87±11.73 | 46.92±12.03 | 43.46±12.74 | 37.89±13.63 | 0.002* |
| ≤38 y | 19.0 | 12.5 | 23.1 | 20.0 | 48.9 | 0.039* |
| 39~44 y | 21.4 | 27.5 | 23.1 | 31.4 | 20.0 | |
| 45~53 y | 31.0 | 30.0 | 20.5 | 28.6 | 17.8 | |
| ≥54 y | 28.6 | 30.0 | 33.3 | 20.0 | 13.3 | |
| Combinations (%) | | | | | | |
| Hypertension | 19.0 | 25.0 | 33.3 | 11.4 | 11.1 | 0.065 |
| Diabetes | 19.0 | 20.0 | 23.1 | 14.3 | 2.2 | 0.065 |
| Renal failure | 0.00 | 0.00 | 2.60 | 0.00 | 0.00 | 0.383 |
| ACEI/ARB | 64.3 | 67.5 | 71.8 | 77.1 | 68.9 | 0.793 |
| Blood biochemistry ($\bar{x} \pm s$) | | | | | | |
| CHOL | 8.28±2.24 | 8.77±2.72 | 8.33±2.66 | 8.27±2.67 | 8.00±2.72 | 0.761 |
| TG | 2.68±1.98 | 3.01±1.73 | 2.91±2.48 | 2.58±1.24 | 2.44±1.48 | 0.618 |
| GLU | 5.17±1.10 | 5.45±1.44 | 5.49±1.34 | 5.21±1.34 | 5.32±0.85 | 0.230 |
| HB | 136.64±22.04 | 139.83±19.91 | 134.36±19.64 | 140.57±18.95 | 143.47±17.68 | 0.255 |
| ALB | 24.47±5.03 | 24.07±5.50 | 24.00±4.91 | 23.33±5.31 | 22.82±6.32 | 0.650 |
| 24 h UP | 7.51±3.00 | 8.42±3.37 | 7.78±2.67 | 8.25±2.62 | 8.49±4.50 | 0.842 |
| Cr | 73.59±23.73 | 69.90±16.63 | 63.79±20.69 | 70.85±15.89 | 71.74±17.92 | 0.209 |
| ALT | 21.71±8.79 | 28.78±22.79 | 21.72±11.72 | 22.83±8.43 | 24.04±10.52 | 0.116 |

Note: CHOL: cholesterol; TC: triglyceride; GLU: glucose; HB: hemoglobin; ALB: serum albumin; 24 h UP: 24 hours urinary protein; Cr: creatinine; ALT: alanine aminotransferase; *P<0.05 means the statistically significant difference.

ly given supportive therapy consisting of anticoagulant, calcium agents, hypotensive drugs and other symptomatic treatment.

*Observation indicators*

Clinical parameters were obtained by review of subjects' records including gender, age and comorbidities. laboratory measurements including serum creatinine, serum albumin, blood glucose, serum lipid, liver function and renal function were performed at 3, 6 and 12 months. The outcomes which were measured by CR, PR, NR, and adverse effects were evaluated at 3, 6 and 12 months.

*Criteria of clinical efficacy*

Complete remission (CR) was defined as proteinuria level of <0.3 g/24 h with normal renal function (serum creatinine ≤140 μmol/l), and serum albumin ≥35 g/l. Partial remission (PR) was defined as a daily proteinuria of 0.3-3.5 g, or at least 50% initial proteinuria level with unchanged or improved renal function. No response (NR) did not meet the above criteria for remission. Relapse was defined as an increase in proteinuria to ≥3.5 g/day for at least 2 weeks after a period of CR or PR, and failure to recover even after pathogenic causes (e.g. exertion, infection) were removed.

*Multi-level model*

Given that our study involves follow-up and repeated measurement, data information cannot be fully elucidated by simply merging data from different time points. As our study does not belong to a randomized controlled trial, multi-level model analysis is preferable to traditional variance analysis in processing non-treatment factors among groups. In addition, this model can effectively control these influencing factors by introducing gender, age, dis-

**Table 2.** Main parameters of patients in 5 groups during follow-up period ($\bar{x} \pm s$)

| Treatment course | Baseline | 3 months later | 6 months later | 12 months later |
|---|---|---|---|---|
| A | | | | |
| CHOL | 8.28±2.24 | 7.23±2.12* | 5.81±1.70* | 5.49±1.75* |
| TG | 2.68±1.98 | 2.61±2.38 | 2.16±1.72 | 1.75±0.78* |
| GLU | 5.17±1.10 | 5.09±1.11 | 5.31±1.27 | 5.35±0.81 |
| ALB | 24.47±5.03 | 33.44±5.17* | 36.23±5.28* | 40.20±5.29* |
| 24 h UP | 7.51±3.00 | 3.83±3.46* | 2.44±2.26* | 1.34±2.02* |
| Cr | 73.59±23.73 | 74.08±21.96 | 75.57±20.85 | 76.23±20.44 |
| ALT | 21.71±8.79 | 25.93±11.90 | 25.45±13.71 | 23.57±9.02 |
| B | | | | |
| CHOL | 8.77±2.72 | 7.67±2.49 | 6.73±2.91* | 5.64±1.80* |
| TG | 3.01±1.73 | 2.50±1.29 | 2.28±1.25 | 1.87±0.79 * |
| GLU | 5.45±1.44 | 5.42±1.64 | 5.33±1.19 | 5.74±1.24 |
| ALB | 24.07±5.50 | 31.13±5.46* | 36.01±5.72* | 38.94±5.05* |
| 24 h UP | 8.42±3.37 | 4.31±2.69* | 2.39±2.91* | 1.44±2.38* |
| Cr | 69.90±16.63 | 75.39±20.76 | 76.17±24.60 | 86.70±28.02* |
| ALT | 28.78±22.79 | 25.83±13.47 | 27.43±16.10 | 23.58±11.54 |
| C | | | | |
| CHOL | 8.33±2.66 | 7.29±2.22 | 6.17±1.65* | 5.84±1.90* |
| TG | 2.91±2.48 | 2.66±1.44 | 2.41±1.13 | 1.88±0.91* |
| GLU | 5.49±1.34 | 5.86±1.47 | 5.77±1.58 | 5.84±1.34 |
| ALB | 24.00±4.91 | 31.91±6.50* | 36.24±6.92* | 39.72±6.51* |
| 24 h UP | 7.78±2.67 | 3.99±3.40* | 2.30±2.58* | 1.55±2.22* |
| Cr | 63.79±20.69 | 66.53±17.88 | 67.64±16.68 | 71.04±23.07 |
| ALT | 21.72±11.72 | 25.15±17.88 | 21.33±12.45 | 20.46±9.27 |
| D | | | | |
| CHOL | 8.27±2.67 | 7.77±3.96 | 6.28±1.91* | 5.13±1.60* |
| TG | 2.58±1.24 | 2.36±1.27 | 1.96±0.98 | 1.59±0.69* |
| GLU | 5.21±1.34 | 5.01±0.91 | 4.97±0.87 | 5.19±0.65 |
| ALB | 23.33±5.31 | 34.00±5.26* | 37.25±5.46* | 40.74±5.30* |
| 24 h UP | 8.25±2.62 | 3.15±2.74* | 1.65±2.37* | 1.51±3.31* |
| Cr | 70.85±15.89 | 77.43±31.31 | 72.25±14.87 | 75.15±22.20 |
| ALT | 22.83±8.43 | 28.34±12.17 | 29.14±18.13* | 20.54±6.75 |
| E | | | | |
| CHOL | 8.00±2.72 | 7.43±2.35 | 6.33±1.97* | 5.69±1.67* |
| TG | 2.44±1.48 | 2.48±1.64 | 2.44±1.33 | 1.62±1.10* |
| GLU | 5.32±0.85 | 5.31±1.11 | 5.49±1.48 | 5.46±1.20 |
| ALB | 22.82±6.32 | 34.51±6.71* | 37.92±6.88* | 39.93±6.23* |
| 24 h UP | 8.49±4.50 | 3.67±3.67* | 2.15±3.21* | 1.46±2.50* |
| Cr | 71.74±17.92 | 74.44±16.41 | 78.23±18.84 | 77.99±18.61 |
| ALT | 24.04±10.52 | 28.82±15.73 | 24.04±13.71 | 30.16±16.09 |

Note: *P<0.05 compared with baseline.

ease history and biochemical indicators as covariates. We set the curative effects at different follow-up time points as level 1 (i) and the study subjects as level 2 (j). The model formula is as follows:

$$Resp_{ijk} = \beta_{0\ jk}\ con_{ijk} + \beta_1\ time.01_{jk} + \beta_2\ group.01_{jk} + \beta_3\ x.01_{jk}$$

Note: $Resp_{ijk}$: therapeutic effect; *Con*: intercept; *x*: covariates.

*Statistical analysis*

Statistical analysis and baseline description were conducted in SPSS 21.0 software. Measurement data were described as the mean ± standard deviation (SD) and categorical variables were presented as numbers and percentages. Intergroup difference was analyzed by paired t-test; the Kruskal-Wallis H-test was used when the data had non-normal distribution, and the Chi-square test was used for comparison of remission rates between groups. For the comparison of the incidence of remission between 5 groups, multi-level linear model of ranked data was established by MLwiN2.25 software to take into account the availability of different data points from the same study [12]. P<0.05 is considered as statistically significant.

## Results

*Baseline information of patients*

There were no significant differences regarding sex, comorbidities including diabetes mellitus, hypertension, renal failure, proportion of ACEI/ARB, level of serum lipid and blood glucose, hepatic function, and renal function among the baseline measurements of the 5 groups (P>0.05). However, significant differences were observed in age among the 5 groups: the age of patients in group E (TAC) was lower than that in other groups (P<0.05) (**Table 1**).

**Table 3.** Remission rates of 5 groups at different stages of treatment (%)

| Groups | 3 months later | | | 6 months later | | | 12 months later | | |
|---|---|---|---|---|---|---|---|---|---|
| | CR | PR | NR | CR | PR | NR | CR | PR | NR |
| A (n=42) | 2.38 | 66.67 | 30.95 | 14.29 | 69.05 | 16.67 | 52.38 | 38.10 | 9.52 |
| B (n=40) | 0.00 | 62.50 | 37.50 | 17.50 | 45.00 | 37.50 | 45.00 | 30.00 | 25.00 |
| C (n=39) | 5.13 | 58.97 | 35.90 | 33.33 | 46.15 | 20.51 | 46.15 | 41.03 | 12.82 |
| D (n=35) | 8.57 | 68.57 | 22.86 | 48.57 | 42.86 | 8.57 | 57.14 | 34.28 | 8.57 |
| E (n=45) | 11.11 | 68.89 | 20.00 | 37.78 | 53.33 | 8.89 | 60.00 | 35.33 | 4.67 |

*Changes of laboratory parameters*

The statistical results showed that the proteinuria and cholesterol of group A was reduced at 3, 6 and 12 months compared with baseline, and the level of serum albumin had the opposite trend (P<0.05); whereas the changes in serum creatinine, glucose and alanine aminotransferase were not significant from baseline to 12 months (P>0.05). In group B, the levels of 24 h urinary protein and serum lipids were lower at 12 months than at baseline (P<0.05), and the level of serum albumin appeared to have an increasing trend during the follow-up period (P<0.05); in addition, an escalating trend can be observed in serum creatinine from baseline to 12 months (P<0.05). There were no significant differences in glucose and alanine aminotransferase levels at different time points (P>0.05). In group C, both 24 h urinary protein and serum lipid levels decreased from baseline to 12 months (P<0.05), serum albumin levels were higher than those before treatment (p<0.05); and there were no significant differences in renal function, blood glucose level, and liver function at 3, 6, 12 months compared with the baseline (P>0.05). Similarly, in group D, urinary protein and serum lipid levels showed a declining trend, and serum albumin level showed a rising trend (P<0.05). But compared with the baseline, the level of alanine aminotransferase significantly improved at 6 months (P<0.05). In group E, 24 h urinary protein excretion and serum lipid level significantly decreased during the treatment period (P<0.05), and serum albumin level increased after treatment (P<0.05). The aforementioned data suggest that all of these five regimens were effective for IMN patients (**Table 2**).

*Comparison of treatment efficacy*

After one year of follow-up, five treatment regimens obtained different proportions of complete remission and partial remission, as shown in **Table 3**. In particular, the total remission (CR and PR) rates of group E were 80.0%, 91.11% and 95.33% at 3, 6 and 12 months, respectively. Multi-level model analysis was performed to compare the efficacy of five treatment regimens in consideration of the influence of baseline parameters including gender, age, hypertension, diabetes mellitus, renal function and history of ACEI/ARB (**Table 4**). When regarding group E (prednisone plus tacrolimus) as the control group, the odds ratios (OR) of non-remission in group A, B, C, D were 2.250, 2.036, 2.286 and 1.160 times, respectively, and these differences were statistically significant (P<0.05). Furthermore, remission rates were significantly associated with gender and serum lipids level: the risk of non-remission in females was 42.4% that of males (P=0.001), and hyperlipidemia tended to increase the risk of non-remission (P=0.003). However, remission rates were not related to age, hypertension, diabetes mellitus, renal function and history of ACEI/ARB. Comprehensive analysis indicated that prednisone plus tacrolimus is the best treatment program for IMN. In addition, male gender and hyperlipidemia were independent risk factors for poor prognosis. See **Table 5** for details.

*Adverse event*

Adverse events during the treatment period are described in **Table 6**. A total of 15 patients suffered from hepatic damage: 5 in group A, 4 in group B, 3 in group C, 2 in group D and 1 in group E. Pulmonary infection was contracted by 3 patients in group A, 3 in group B, 2 in group C, 4 in group D and 5 in group E. One case of leukopenia can be observed in each of groups A and E. Two viral infection events were recorded in group D and E. In the majority of patients, these complications could be controlled adjusting the dose of immunosuppressant and symptomatic treatment, with the exception of 1 patient in group A whose treatment regimen was prematurely discontinued due to severe liver damage. As shown in **Table 7**, there were no significant differences in the rates of complications among the 5 groups (P>0.05).

**Table 4.** Variable assignment of multiple level model

| Variate | Definition and assignment | | | | |
|---|---|---|---|---|---|
| Dependent variable | | | | | |
| Efficacy | 0 No response | 1 Partial response | 2 Complete response | | |
| Schemes | 1 CTX po | 2 CTX ivgtt | 3 Ponticelli | 4 Ciclosporin | 5 Tacrolimus |
| Demographic characteristic | | | | | |
| Gender | 1 Male | 2 Female | | | |
| Ages (years) | 1 ≤38 | 2 39~44 | 3 45~53 | 4 ≥54 | |
| Combinations | | | | | |
| Hypertension | 0 No | 1 yes | | | |
| Diabetes | 0 No | 1 yes | | | |
| Renal failure | 0 No | 1 yes | | | |
| ACEI/ARB | 0 No | 1 yes | | | |

Note: po means oral, ivgtt means intravenous.

**Table 5.** Multivariate Analysis of 5 treatment regimens

| Parameters | Estimate value | SE | OR | $\chi^2$ | *p* |
|---|---|---|---|---|---|
| Fixed part | | | | | |
| Intercept (CR+PR)* | | | | | |
| ≤NR | 7.361 | 0.763 | 1573.409 | 92.978 | 0.000 |
| Time | -2.182 | 0.127 | 0.113 | 296.868 | 0.000 |
| E (tacrolimus) | | | | | |
| A | 0.811 | 0.361 | 2.250 | 5.044 | 0.025 |
| B | 0.724 | 0.362 | 2.063 | 4.001 | 0.045 |
| C | 0.827 | 0.372 | 2.286 | 5.947 | 0.016 |
| D | 0.148 | 0.372 | 1.960 | 3.159 | 0.048 |
| Age | 0.012 | 0.010 | 1.012 | 1.460 | 0.227 |
| Gender (male) | -0.859 | 0.247 | 0.424 | 12.109 | 0.001 |
| Combinations | | | | | |
| Hypertension | -0.256 | 0.333 | 0.774 | 0.590 | 0.442 |
| Diabetes | -0.644 | 0.370 | 0.525 | 3.022 | 0.082 |
| Renal failure | 2.998 | 1.996 | 20.045 | 2.255 | 0.133 |
| ACEI/ARB | -0.065 | 0.252 | 0.937 | 0.067 | 0.796 |
| Blood biochemistry ($\bar{x} \pm s$) | | | | | |
| Blood lipid | 0.237 | 0.080 | 1.267 | 8.737 | 0.003 |
| GLU | 0.004 | 0.092 | 1.004 | 0.002 | 0.964 |
| ALT | -0.015 | 0.007 | 0.985 | 4.744 | 0.629 |
| Random part | | | | | |
| $\sigma_v^2$ | 1.209 | 0.253 | 3.350 | 22.887 | 0.000 |

Note: *CR was complete response; NR was no response; PR was partial response; P<0.05 means the statistically significant difference.

*Relapse rate*

We observed the relapse rates of the five groups for 1 year by telephone and outpatient follow-up after the termination of the intervention. The number of patients in follow-up was decreased compared with the original enrolled number on account of withdrawal, incomplete information, and other sources of attrition. The relapse rates in groups A, B, C, D and E were 22.22%, 26.67%, 28.27%, 29.41% and 28.57%, respectively. There were no significant differences in relapse rates among the 5 groups (P>0.05) as shown in **Table 8**.

## Discussion

IMN is one of the most common pathological types of nephrotic syndrome, accounting for one fifth of nephrotic syndrome in adults, which is associated with a large economic burden [13-15]. The natural course of IMN is complex and diverse, so it is essential to know how to choose the timing of treatment, which patients should receive immunosuppressive therapy, and which treatment regimen is best for IMN patients.

Some studies have pointed out that patients with mild proteinuria who had higher spontaneous remission rates should only be administered ACEIs, ARBs, anti-hypertensive agents, lipid-lowering drugs, anticoagulants and other supportive therapy [16]. It is worth mentioning that urinary protein levels and serum creatinine should be periodically monitored. In 2011, the

**Table 6.** The occurrence of adverse events

| Groups | Hepatic damage | Pulmonary infection | Leukocytopenia | Virus infection |
|---|---|---|---|---|
| A | 5 | 3 | 1 | 0 |
| B | 4 | 3 | 0 | 0 |
| C | 3 | 2 | 0 | 0 |
| D | 2 | 4 | 0 | 2 |
| E | 1 | 5 | 1 | 2 |

clinical practice guideline for glomerulonephritis developed by the Kidney Disease: improving global outcomes (KDIGO) nonprofit organization recommended that glucocorticoids and immunosuppressants should be administered only after meeting the following requirements: ① proteinuria persistently >4 g/d or above 50% of the baseline level without downward trend after at least 6 months of antiproteinuric treatment; ② Patients present with severe nephrotic syndrome-related clinical symptoms which may lead to disability or life threatening symptoms; ③ serum creatinine level has risen by ≥30% within 6-12 months, but GFR ≥25-30 ml/min, and the above changes were not caused by superimposed complications [1, 17].

CTX and CSA, as classical immunosuppressive agents, play a pivotal role in therapeutic treatment of IMN. In order to clarify whether the administration method of CTX can affect its curative effect, we compared the remission rate, recurrence rate and side effects of oral CTX, intravenous CTX and modified Ponticelli protocol. Our study found that there were no significant differences among these groups A, B and C. In addition, Dede et al. also demonstrated that the administration route of CTX was not the key factor determining its therapeutic effect [18]. CSA is usually used in IMN patients who are not sensitive to supportive treatment, hormone-dependent, or who have repeated recurrence [19]. However, its long-term efficacy still needs to be further investigated. A ten-year study from India reported that the remission rate of CTX at the end of 10 years was only 58.6%, which was far lower than previously reported [20]. In recent years, a number of novel immunosuppressive agents such as TAC [21], rituximab [22], immunoglobulins [23] and mycophenolate mofetil [24] have been applied to the therapy of IMN. The calcineurin inhibitor TAC has shown promise in IMN treatment and may become the alternative agent for IMN patients. The mechanism of TAC in acting on IMN was demonstrated in a previous study. TAC can markedly reduce proteinuria and promote podocyte repair by decreasing angiopoietin-like-4 [25]. Our results showed that TAC was more effective than CTX at 3 months and 12 months in IMN (CR and PR). We also found that TAC had superior potency compared to CSA. One recent study also demonstrated that TAC possessed more advantages on remission rates than CTX in treating IMN patients, especially in the first 3 months (76.7% vs 59.0%, $P<0.05$) [26].

We compared the baseline features of the five treatment groups and found significant differences in participants' age among the five groups ($P<0.05$). We carried out a subgroup analysis based on age to exclude the confounding factor. Our data suggested that age was not relevant to remission rate, which was not consistent with the traditional viewpoint that advanced age is a poor prognostic factor for IMN patients. Notably, another study recruited 171 IMN patients who were divided into different age subgroups. 90 cases (52.6%) patients <65 years, 40 cases (23.4%) patients 65-70 years old and 41cases (24%) patients >70 years were analyzed and followed up for an average of 37 months. A total of 103 patients obtained complete remission of proteinuria, but there were no significant differences among different age groups ($P=0.831$) [27]. Therefore, large sample multicenter randomized controlled trails are required to further confirm this correlation.

In the present study, we also found that female gender and hypolipidemia were protective factors for IMN patients, who were then prone to a better prognosis. This is consistent with previous findings that the male gender, increasing age, massive proteinuria, impaired renal function and certain histologic features including glomerulosclerosis, interstitial fibrosis and vascular disease are high risk factors for progressing to renal failure in IMN [28]. Patients with the above risk factors should be given active treatment. Whereas, the significant differences in age, comorbidities and blood biochemistry were not observed in our data.

The multiple adverse effects caused by immunosuppressant treatment cannot be ignored, as many of them are irreversible and even life-threatening. Common complications include hepatic damage, pulmonary infection, low leucocyte amount and virus infection. In addition,

**Table 7.** Statistical analysis of the adverse events

| Groups | Number | Adverse event (n) | Incidence of adverse event (%) | Chi-square | P |
|---|---|---|---|---|---|
| A | 22 | 9 | 40.91 | 0.119 | 0.730 |
| B | 20 | 7 | 35.00 | 0.005 | 0.945 |
| C | 19 | 5 | 26.32 | 0.467 | 0.495 |
| D | 21 | 8 | 38.10 | 0.022 | 0.883 |
| E | 25 | 9 | 36.00 | - | - |

Note: Compared with the E group.

**Table 8.** Relapse rate in 5 groups

| Groups | Number | Recurrence (n) | Recurrence rate (%) | P |
|---|---|---|---|---|
| A | 9 | 2 | 22.22 | 0.612 |
| B | 15 | 4 | 26.67 | 0.553 |
| C | 7 | 2 | 28.27 | 0.460 |
| D | 17 | 5 | 29.41 | 0.394 |
| E | 14 | 4 | 28.57 | - |

Note: Compared with the E group.

rare complications have been documented. It has been reported that one case of an IMN patient in Poland receiving TAC developed into Kaposi's sarcoma [29]. In our study, although the total occurrence of side effects among the 5 groups had no significant difference, we observed that the profile of adverse effects was distinctly different. We found that liver damage and lung infection happened in all 5 groups of patients. Hepatic function of these patients was recovered through adjustment of the doses of immunosuppressants and administration of hepatinica, and lung infection was controlled with antibiotics. Leukopenia tended to occur in the CTX group and the TAC group. Patients could continue the original program of treatment after symptomatic treatment. IMN patients receiving CSA and TAC had a higher occurrence of viral infection than other groups. In particular, patients taking the calcineurin inhibitors (TAC, CSA) had a higher prevalence of herpes zoster virus infection than CTX, likely due to the powerful immunosuppressive action of calcineurin inhibitors. These symptoms disappeared after adjusting the dose of drugs and antiviral therapy. No life-threatening side effects occurred in our observation, which may be related to our short follow-up period and small sample size. Therefore, patients receiving immunosuppressive agents should be closely monitored for the occurrence of side effects. It is essential that early intervention is provided once related complications arise [30].

Finally, we estimated the risk of 1-year post-treatment relapse among the 5 treatment regimens. In the present study, the 1-year relapse rates in the 5 groups were no more than 30%, and there were no significant differences in relapse rates among the 5 groups. However, they were limited by high attrition rate of follow-up and inadequate follow-up time. Similarly, a recurrence rate of 25% between 6 and 30 months was seen with CTX in a randomized controlled trial [6]. Obviously, long-term follow up is needed to evaluate the efficacy and safety of TAC. In addition, re-treatment should be considered when an appropriate risk-benefit ratio exists.

In conclusion, the data from this retrospective study show that glucocorticoids combined with immunosuppressive agents are effective in the treatment of IMN. Treatment with the combination of TAC and prednisolone shows higher efficacy and comparable tolerability, and is superior to CTX and CSA. Our study provides powerful evidence for selecting an optimal treatment strategy for patients with IMN. However, complications arising during the period of TAC administration should be closely monitored and should receive timely treatment.

## Disclosure of conflict of interest

None.

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## References

[1] Beck L, Bomback AS, Choi MJ, Holzman LB, Langford C, Mariani LH, Somers MJ, Trachtman H and Waldman M. KDOQI US commentary on the 2012 KDIGO clinical practice guideline for glomerulonephritis. Am J Kidney Dis 2013; 62: 403-41.

[2] Ponticelli C and Glassock RJ. Glomerular diseases: membranous nephropathy--a modern view. Clin J Am Soc Nephrol 2014; 9: 609-16.

[3] Chen Y, Schieppati A, Chen X, Cai G, Zamora J, Giuliano GA, Braun N and Perna A. Immunosuppressive treatment for idiopathic membranous nephropathy in adults with nephrotic syndrome. Cochrane Database Syst Rev 2014; CD004293.
[4] Cattran DC, Delmore T, Roscoe J, Cole E, Cardella C, Charron R and Ritchie S. A randomized controlled trial of prednisone in patients with idiopathic membranous nephropathy. N Engl J Med 1989; 320: 210-5.
[5] Couser WG. Primary membranous nephropathy. Clin J Am Soc Nephrol 2017; 12: 983-97.
[6] Ponticelli C, Altieri P, Scolari F, Passerini P, Roccatello D, Cesana B, Melis P, Valzorio B, Sasdelli M, Pasquali S, Pozzi C, Piccoli G, Lupo A, Segagni S, Antonucci F, Dugo M, Minari M, Scalia A, Pedrini L, Pisano G, Grassi C, Farina M and Bellazzi R. A randomized study comparing methylprednisolone plus chlorambucil versus methylprednisolone plus cyclophosphamide in idiopathic membranous nephropathy. J Am Soc Nephrol 1998; 9: 444-50.
[7] Kalliakmani P, Koutroulia E, Sotsiou F, Vlachojannis JG and Goumenos DS. Benefit and cost from the long-term use of cyclosporine-A in idiopathic membranous nephropathy. Nephrology (Carlton) 2010; 15: 762-7.
[8] Ramachandran R, Hn HK, Kumar V, Nada R, Yadav AK, Goyal A, Kumar V, Rathi M, Jha V, Gupta KL, Sakhuja V and Kohli HS. Tacrolimus combined with corticosteroids versus modified ponticelli regimen in treatment of idiopathic membranous nephropathy: randomized control trial. Nephrology (Carlton) 2016; 21: 139-46.
[9] Rojas-Rivera J, Fernandez-Juarez G, Ortiz A, Hofstra J, Gesualdo L, Tesar V, Wetzels J, Segarra A, Egido J and Praga M. A European multicentre and open-label controlled randomized trial to evaluate the efficacy of Sequential treatment with TAcrolimus-Rituximab versus steroids plus cyclophosphamide in patients with primary membranous nephropathy: the STARMEN study. Clin Kidney J 2015; 8: 503-10.
[10] Xu J, Zhang W, Xu Y, Shen P, Ren H, Wang W, Li X, Pan X and Chen N. Tacrolimus combined with corticosteroids in idiopathic membranous nephropathy: a randomized, prospective, controlled trial. Contrib Nephrol 2013; 181: 152-62.
[11] Chen M, Li H, Li XY, Lu FM, Ni ZH, Xu FF, Li XW, Chen JH, Wang HY; Chinese Nephropathy Membranous Study Group. Tacrolimus combined with corticosteroids in treatment of nephrotic idiopathic membranous nephropathy: a multicenter randomized controlled trial. Am J Med Sci 2010; 339: 233-8.
[12] Biener L, Hamilton WL, Siegel M and Sullivan EM. Individual, social-normative, and policy predictors of smoking cessation: a multilevel longitudinal analysis. Am J Public Health 2010; 100: 547-54.
[13] Yoon HE, Shin MJ, Kim YS, Choi BS, Kim BS, Choi YJ, Kim YO, Yoon SA, Kim YS and Yang CW. Clinical impact of renal biopsy on outcomes in elderly patients with nephrotic syndrome. Nephron Clin Pract 2011; 117: c20-27.
[14] Brown CM, Scheven L, O'Kelly P, Dorman AM and Walshe JJ. Renal histology in the elderly: indications and outcomes. J Nephrol 2012; 25: 240-4.
[15] Verde E, Quiroga B, Rivera F and Lopez-Gomez JM. Renal biopsy in very elderly patients: data from the spanish registry of glomerulonephritis. Am J Nephrol 2012; 35: 230-7.
[16] Lonnbro-Widgren J, Molne J, Haraldsson B and Nystrom J. Treatment pattern in patients with idiopathic membranous nephropathy-practices in Sweden at the start of the millennium. Clin Kidney J 2016; 9: 227-33.
[17] Radhakrishnan J and Cattran DC. The KDIGO practice guideline on glomerulonephritis: reading between the (guide) lines--application to the individual patient. Kidney Int 2012; 82: 840-56.
[18] Dede F, Ayili D and Sahiner S. Effective treatment administration of cyclophosphamide in membranous nephropathy. J Nephrol 2008; 21: 560-5.
[19] Ponticelli C. Membranous nephropathy. J Nephrol 2007; 20: 268-87.
[20] Ram R, Guditi S and Kaligotla Venkata D. A 10-year follow-up of idiopathic membranous nephropathy patients on steroids and cyclophosphamide: a case series. Ren Fail 2015; 37: 452-5.
[21] Horvatic I and Galesic K. Membranous glomerulonephritis--recent advances in pathogenesis and treatment. Lijec Vjesn 2012; 134: 328-39.
[22] Wang X, Cui Z, Zhang YM, Qu Z, Wang F, Meng LQ, Cheng XY, Liu G, Zhou FD and Zhao MH. Rituximab for non-responsive idiopathic membranous nephropathy in a Chinese cohort. Nephrol Dial Transplant 2017; [Epub ahead of print].
[23] Muller-Deile J, Schiffer L, Hiss M, Haller H and Schiffer M. A new rescue regimen with plasma exchange and rituximab in high-risk membranous glomerulonephritis. Eur J Clin Invest 2015; 45: 1260-1269.
[24] Choi JY, Kim DK, Kim YW, Yoo TH, Lee JP, Chung HC, Cho KH, An WS, Lee DH, Jung HY, Cho JH, Kim CD, Kim YL and Park SH. The effect of mycophenolate mofetil versus cyclosporine as combination therapy with low dose corticoste-

roids in high-risk patients with idiopathic membranous nephropathy: a multicenter randomized trial. J Korean Med Sci 2018; 33: e74.
[25] Peng L, Ma J, Cui R, Chen X, Wei SY, Wei QJ and Li B. The calcineurin inhibitor tacrolimus reduces proteinuria in membranous nephropathy accompanied by a decrease in angiopoietin-like-4. PLoS One 2014; 9: e106164.
[26] Cui W, Lu X, Min X, Liu M, Guan S, Wang Y, Luo M, Li W, Li Q, Dong W, Miao L and Luo P. Therapy of tacrolimus combined with corticosteroids in idiopathic membranous nephropathy. Braz J Med Biol Res 2017; 50: e5976.
[27] Yamaguchi M, Ando M, Yamamoto R, Akiyama S, Kato S, Katsuno T, Kosugi T, Sato W, Tsuboi N, Yasuda Y, Mizuno M, Ito Y, Matsuo S and Maruyama S. Patient age and the prognosis of idiopathic membranous nephropathy. PLoS One 2014; 9: e110376.
[28] Cattran D. Management of membranous nephropathy: when and what for treatment. J Am Soc Nephrol 2005; 16: 1188-94.
[29] Rukasz D, Krajewska M, Augustyniak-Bartosik H, Letachowicz K, Halon A, Ekiert M, Jakuszko K, Madziarska K, Weyde W and Klinger M. Effective treatment of Kaposi sarcoma with everolimus in a patient with membranous glomerulonephritis. Intern Med J 2015; 45: 230-1.
[30] Goumenos DS, Ahuja M, Davlouros P, El Nahas AM and Brown CB. Prednisolone and azathioprine in membranous nephropathy: a 10-year follow-up study. Clin Nephrol 2006; 65: 317-23.