Development and Validation of a Discrimination Model Between Primary PLA2R-Negative Membranous Nephropathy and Minimal Change Disease Confirmed by Renal Biopsy

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Research

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Abstract

Background

Membranous nephropathy (MN) and minimal change disease (MCD) are two common causes leading to nephrotic syndrome (NS). They have similar clinical feature but different treatment strategy and prognosis. M-type phospholipase A2 receptor (PLA2R) is considered as a specific marker of membranous nephropathy. However, its sensitivity is only about 70%. Therefore, there is a lack of effective and noninvasive tools to distinguish PLA2R-negative MN and MCD patients without renal biopsy. We aim to develop a discrimination model using noninvasive parameters for distinguishing the two diseases and support immediate treatment before result of renal biopsy.

Methods

A total 949 patients who were pathologically diagnosed as idiopathic MN or primary MCD in the First Affiliated Hospital of Zhengzhou University from January 2017 to August 2019 were enrolled in this study, including 805 idiopathic MN (605 PLAR2-positive MN and 200 PLA2R-negative MN) and 144 primary MCD. Based on the basic information and laboratory examination of 200 PLA2R-negative MN and 144 MCD, we used univariate and multivariate logistic regression analysis to select the relevant variables and develop the discrimination model. ROC curves and calibration curves were used to evaluate the diagnostic ability and calibration ability of the model. The decision curve was used to show the net benefit. We also tested the effectiveness of our model in all 949 patients.

Results

A novel model that included age, albumin, urea, high density lipoprotein, urea and red blood cell count was established for PLA2R-negative MN and MCD. The discrimination model has good differential capability (an AUC of 0.889 in training group and an AUC of 0.920 in test group) and calibration capability. When testing in all 949 patients, our model also showed good discrimination ability for all idiopathic MN and MCD.

Conclusion

We constructed a discrimination model with high diagnostic effectiveness for PLA2R-negative MN and MCD. The model could also be used for all idiopathic MN and MCD patients.

Introduction

Membranous nephropathy (MN) is one of the most common glomerular diseases causing primary nephrotic syndrome (NS) in China. The prevalence of MN has increased greatly in central China during the past two decades[1, 2]. Minimal change disease (MCD) is another primary glomerular disease with high prevalence characterized by rapid onset and development. Current studies suggest these two diseases account for about 60% NS patients in China[3, 4].

Renal biopsy is still the gold standard for diagnosing and distinguishing these two diseases. However, considering the poor physical condition and contraindications, many patients are unsuitable to undergo renal biopsy in daily clinical practice [5]. In addition, it does take a period of time to wait for the result. The onset of MCD is usually utterly urgent which may cause damage to renal function if the treatment is not timely and correct.

M-type phospholipase A2 receptor (PLA2R) is considered as a specific marker of idiopathic membranous nephropathy. Present evidences reveal PLA2R test has strong specificity, while its sensitivity is insufficient, 0.7 approximately[6-9]. Hence, quite amount of PLA2R-negative MN patients cannot be screened out. More importantly, the accuracy of PLA2R detection may be influenced by other diseases such as tumor-related diseases[7, 10].

Thus, it is urgent to develop a new convenient and noninvasive method to distinguish MN from MCD. The aim of our study is to develop and validate a model to distinguish MN, especially PlA2R-negative MN from MCD. The model can be used to the patients who are unsuitable or unwilling to undergo renal biopsy. We believe our model will help clinicians treat these patients in a timely manner and improve their prognosis.

Methods

Study population and Ethical approval

In this population-based retrospective analysis, we screened all the patients with NS who were hospitalized in the First Affiliated Hospital of Zhengzhou University from January 2017 to August 2019. The inclusion criteria were as follows: (1) aged 18-80 years (2) diagnosed as idiopathic MN or primary MCD by renal biopsy (3) experienced a PLA2R test. The exclusion criteria included application history of corticosteroid or immunosuppressant prior to renal biopsy. A total of 949 patients including 805 idiopathic MN (605 PLA2R-positive MN and 200 PLA2R-negative MN) and 144 primary MCD were enrolled potentially relevant cases.in this study. Among them, patients with PLA2R-negative MN and MCD were used to develop and validate the discrimination model. In the end, we also tested the differential ability of the model in all idiopathic MN and MCD. The enrollment flowchart of the participants in this study was shown in Figure1.

The First Affiliated Hospital of Zhengzhou University Ethics Review Committee granted ethical approval for the study and the ethics review approval ID was “2019-KY-361”.

Data collection

We collected the basic information and laboratory examination from all patients recruited at the time of renal biopsy, which might be involved in distinguishing the two diseases. The basic information included age, gender, onset time, systolic blood pressure (SBP) and diastolic blood pressure (DBP). The laboratory indices included red blood cell (RBC) count, white blood cell (WBC) count, platelet (PLT) count, hemoglobin (Hb) levels, mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), total protein (TP) levels, albumin (ALB) levels; total cholesterol (TCHO) levels,

triglyceride (TG) levels, low density lipoprotein (LDL) levels, high density lipoprotein (HDL) levels, estimated glomerular filtration rate (eGFR); serum creatinine (SCr) levels; uric acid (UA) levels, 24h uric total protein(24h TP) levels and 24h urine volume.

The urine and venous blood samples of all participants were collected after 12 hours of overnight fasting (except 24-hour urine samples) and sent to laboratory for testing straight away.

Statistical Analysis

The PLA2R-negetive MN and all MCD patients (n = 344) were randomly divided into the training group (n = 241) and the test group (n = 103).

There were a few missing data of several variables. For example, the HLD and LDL levels had 3.5% missing values, Scr and Urea levels had 0.9% missing values. To deal with these missing data, multivariate multiple imputation with chained equations was used to impute missing values so that we could maximize the statistical power and diminish bias[11]. Descriptive statistics of all variables including means, medians, and proportions are used to describe the characteristics of two groups. The categorical data expressed as the percentages, and means ± standard deviation (SD) or medians (quartile 1, quartile 3) were described as continuous variables satisfying or not satisfying the normal distribution, respectively. We used the univariate logistic regression to calculate the OR values of the variables, selected potential variables to perform multiple logistic regression subsequently, and calculated the collinearity of the variables to remove the colinear factors. The candidate variables with a p value <0.05 in the univariate analysis were enrolled to develop the multivariable model.

Based on the clinical features and variables with statistical sense, we attempted to develop the discrimination model for distinguishing PlA2R-negative MN from primary MCD patients.

Next, we drew a receiver operating characteristic curve (ROC) and used the area under the receiver operating characteristic curve (AUC) to evaluate the verification efficiency of the model. The calibration was assessed by constructing the calibration curve. The fitting degree of the model was assessed by the Akaike information criterion (AIC). After comprehensively evaluating the performance of each model, we obtained the best model and constructed a nomogram to make it convenient for the clinical application.

At last, we constructed the decision curve analysis to determine the clinical utility of the discrimination model by quantifying the net benefits at different threshold probabilities[12].

All the statistical analysis processes involved were completed by R software, version 4.0.2. P value <0.05 was considered statistically significant. Model validation and evaluation processes were independently performed on the training group and test group, respectively. In order to assess the diagnostic ability of the model distinguishing all idiopathic MN and MCD, we also performed validation processes on 949 potentially relevant cases.

Results

Baseline Characteristics

The baseline characteristics of PLA2R-negative MN (n=200) and MCD (n=144) were shown in Table 1. The results suggested that PLA2R-negative MN patients tended to be older and have higher TP, ALB levels and greater urine volume. While, the RBC and PLT count, median Hb, TCHO, TG, LDL level, HLD, Scr, Urea, and 24hTP levels of PLA2R-negative MN were lower than those of MCD. The baseline characteristics of all idiopathic MN and MCD were shown in Table S1.

All the participants were randomly divided into training group (n=241) and test group (n=103). The levels of these variables were similar and had no statistical difference, which represented similar clinical profiles between the two groups (Table 2).

Five potential predictors were selected to develop the discrimination model

By means of univariate logistic regression, 14 potential predictors from 22 candidates were considered to have statistical significance ($p<0.05$) in training group. After implementing the multivariable logistic regression analysis and removing the collinear candidates, five potential predictors including age, ALB levels, HDL levels, Urea levels and RBC counts were used to establish the discrimination model to distinguish PLA2R-negative MN from MCD (Table 3). The results indicated that patients with elder ages, higher ALB levels, lower HDL levels, lower serum Urea levels and lower RBC count were more likely suffering PLA2R-negative MN.

Good discrimination and calibration capability of the model

We drew the ROC to evaluate the diagnostic effectiveness of the model (Figure 2). The area under the ROC(AUC), which is referred to as the C-statistic, is considered to be an indicator for evaluating the effectiveness of the model. Surprisingly, we found that the discrimination model had a high efficiency with an AUC of 88.9% (cut-off value: 0.457, sensitivity: 0.802, specificity: 0.914; Figure 2A) in training group. We subsequently verified the effectiveness in the test group and the result showed an even higher efficiency with an AUC of 92.0% (cut-off value: 0.464, sensitivity: 0.791, specificity: 0.917; Figure 2B). The high value of AUC showed that the model had an good ability for discrimination the two disease.

The calibration curve was plotted to evaluate the calibration of the model and it demonstrated a good agreement between prediction and observation both in training group (mean absolute error=0.02) and test group (mean absolute error=0.022, Figure3 A, B). The calibration curve indicated that the model had a great calibration capability.

Construction and usage of the nomogram

In order to make the model convenient to use, we constructed the nomogram of our discrimination model based on 5 obtained predictive variables including age, HDL levels, ALB levels, Urea levels and RBC count

(Figure 4). The value of each variable represented as a score by drawing a straight line upward from the corresponding value to the “Points” line. Sum the total points and mark it at “Total points” line. Draw down a straight line to the corresponding “MN probability” axis and obtain the possibility of MN.

Decision curve showed it would add more net benefits for clinical decision

The result of decision curve analysis for the nomogram is shown in Figure 5. The y-axis represents net benefit and the thick red line represents the model. The thin gray line represents the assumption that all patients suffer from MN, while the thin black line represents that all patients are assumed to have MCD. Both in training group or test group, the decision curve showed that if the threshold probability of a patient is > 1%, using the nomogram in the present study to predict MN adds more benefits than performing renal biopsy on none or all patients.

Diagnosis efficiency testing in potentially relevant cases

In order to determine whether we can expand the application scope of our model, we also performed diagnosis efficiency testing in potentially relevant cases (including 605 PLA2R-positive MN, 200 PLA2R-negative MN and 144 MCD, Figure 2). The ROC showed the good diagnostic performance with an AUC of 0.856(cut-off value: 0.836, sensitivity: 0.771, specificity: 0.826), suggesting good discrimination ability for all idiopathic MN and MCD patients. The performances of calibration curve and decision were also good (Figure S1, S2).

Discussion

In this retrospective case-control study, we attempted to develop a discrimination model used to distinguish patients with PLA2R-negative MN and MCD. we collected and analyzed the basic information and laboratory examination of 949 patients with idiopathic MN or MCD. Based on 200 PLA2R-negative patients and 144 MCD patients, we developed a discrimination model to differentiate the two diseases. The results showed great diagnostic effectiveness with an AUC of 0.889 in training group and an AUC of 0.920 in test group as well as high calibration capability. To the best of our knowledge, it is the first study aiming to develop a discrimination model based on the basic information and the laboratory examination of participants to distinguish primary PLA2R-negative MN and MCD. In addition, our model also showed good diagnostic effectiveness with an AUC of 0.856 in all idiopathic MN patients (either PLA2R-negative or PLA2R-positive MN) and MCD patients. It is an attempt at translational medicine of our study, which can aid clinicians to treat patients with different methods in a timely manner and thus improve their prognosis.

Currently, it is difficult to distinguish MN and MCD patients by a noninvasive tool in clinical practice. A study tried to use soluble urokinase-type plasminogen activator receptor (suPAR) level to distinguish idiopathic focal segmental glomerulosclerosis (FSGS), MN and MCD, However, the study revealed that the three types of glomerulopathy cannot be distinguished using suPAR solely [13]. Therefore, there is no miraculous indicator or model to identify these two primary glomerular diseases currently.

Prediction and discrimination models based on clinical data have been developed increasingly in a wide variety of diseases recent years[14, 15]. In terms of kidney diseases, prediction and discrimination models are also rapidly growing owing to its scientific nature and accuracy[16-18]. The appearance of clinical models gave us great inspiration.

Present evidences suggested that MN had the largest proportion of morbidity in elderly patients, while MCD accounts for the highest proportion of primary nephrotic syndrome in young patients[3, 19, 20]. Our experiments reached similar results that the age at biopsy had a certain influence on the nomogram. In our study, patients with older age were more likely to be considered as PLA2R-negative MN, while younger age at onset was considered to be a higher risk of MCD.

ALB level was one of the predictive factors in this model. Some study showed MN patients always had higher ALB levels, which was consistent with our results[21, 22]. One of the most important clinical manifestations of nephrotic syndrome is increased urinary protein and decreased albumin level. Larger amounts of glomerular albumin filtration will also make serum albumin and serum total protein at a low level. MCD patients always presents as an acute onset, severe illness, and a greater amount of urinary protein, and rapid decline in renal function may result in increased Scr levels and decreased eGFR and 24h urine volume. Some patients will even progresses to AKI□which is relatively common actually□due to high-grade proteinuria[23]. The slit diaphragm between foot processes is regarded as a fine filter[24]. There is a common assumption that proteins leak from the slit pores due to reduced nephrin expression, leading to larger amounts of glomerular albumin filtration in MCD patients[25]. In our study, we chose ALB levels as one of the variables of the model by multivariable regression analysis to exclude the effect of collinearity.

HDL level is another variable that can be used to distinguish between two diseases according to our results. Nephrotic syndrome could cause upregulation of HDL endocytic receptor and downregulation of HDL docking receptor, causing dysregulation of lipid/lipoprotein metabolism[26]. MCD is also known as lipoid nephropathy because steatosis can be observed in epithelial cells of proximal convoluted tubules under light microscopy. In addition, increased hepatic lipoprotein synthesis and reduced lipoprotein degradation are also thought to be responsible for elevated blood lipid profiles. Takeshi Fujita compared lipid and fatty acid metabolism between 7 MCD and 11 MN patients. The results showed that the patients with MCD had higher level of blood lipids than MN[27]. Although the mean HDL level was much higher in patients with MCD, there was no statistical significance between the two groups, which was not exactly the same as our results. The reason might be that their sample size was not large enough, leading to the unobvious statistical significance.

Increased urea level was usually observed in a high protein decomposition status. After the renal filtration barrier disrupted, a large amount of proteins will leak into Bowman's space and renal tubules through glomerular barrier to form crude urine. When proximal tubules enhance the reabsorption of filtered proteins, the protein decomposition is also increased at the same time, resulting in elevated serum Urea level. Serum Urea level were also found different in MN patients and MCD patients in Jin Dong's

research[28], which was consistent with our results. The Urea level plays an important role in our discrimination model.

Red blood cell count and Hb levels had statistical significance by means of univariate regression in our study. They are usually recognized as the indices to evaluate anemia. Compared with younger patients, idiopathic membranous nephropathy patients over 65 years old were found to have lower Hb level than patients less than 65 years old in Choi JY's study[29]. However, the results in Yaeni Kim's study showed there was no difference of Hb levels between elderly patients and young patients[30]. The reason for this ambiguity might be different gender and illness state of included patients. In our study, univariate regression showed there was no statistical difference in gender. And the data we collected was from the time of renal biopsy, reducing influence of the illness state.

The decision curve showed the clinical utility of our model, indicating it may be beneficial for clinicians to distinguish the two diseases by using our model. And using the nomogram to distinguish the two diseases added more benefits than either all or no patients who underwent a renal biopsy if the threshold probability of a patient was > 1%. The results of decision curves suggest the good clinical application value of our model, reflecting the thinking mode of translational medicine.

The results of diagnosis efficiency test in potentially relevant cases suggested that our model is applicable to all idiopathic MN and MCD patients. Some hospitals are unable to perform PLA2R test, and our model might provide an alternative tool for these hospitals to distinguish MN and MCD.

Our study is an attempt in translational medicine and has a number of strengths. It had a large sample size with 949 idiopathic MN and MCD patients confirmed by renal biopsy. And the 5 items in the nomogram are routine clinical variables that can easily obtained by clinicians. We chose to collect the information and examination results at the time of renal biopsy, and excluded the influence of corticosteroid or immunosuppressive agents. What is more, our discrimination model has satisfying diagnostic effectiveness with an AUC of 0.889 in training group and an AUC of 0.920 in test group. The outstanding discrimination ability for all idiopathic MN and MCD patients even showed wider application prospects of our model. The operation of the model is simple and fast, which can help doctors diagnose patients timely. Unlike renal biopsy, our model doesn't have any contraindications so that it can be used more widely.

However, there are also several limitations in our study. First, we still need to expand the sample size for further reducing the heterogeneity. In addition, all the patients came from the First Affiliated Hospital of Zhengzhou University and we did not conduct multicenter external validation. Thus, we can't exclude the influence of diet, race and other related factors on the experimental results. Last, our model is only suitable for the identification of MN and MCD. There is still a lack of differential ability of our model for other types of nephrotic syndrome like focal segmental glomerulosclerosis (FSGS), IgA nephropathy (IgAN) and membranoproliferative glomerulonephritis (MPGN). Corrections to these shortcomings will be made in our subsequent research.

Conclusion

In this study, we developed and validated a discrimination model used for distinguishing PLA2R-negative MN and MCD patients. We further presented a nomogram including age, ALB levels, HDL levels, urea levels and RBC counts. The model showed good discrimination and calibration ability both in training group and test group. It also had a good diagnostic performance in all MN patients and MCD patients. Hopefully, it could provide a practical and convenient tool for clinicians to distinguish these two diseases.

Abbreviations

MN: Membranous nephropathy; MCD: Minimal change disease; NS: Nephrotic syndrome; PLA2R: M-type phospholipase A2 receptor; SBP, Systolic blood pressure; DBP, Diastolic blood pressure; RBC, Red blood cell; WBC, White blood cell; PLT, Platelet; Hb, Hemoglobin; MCH, Mean corpuscular hemoglobin, MCHC: Mean corpuscular hemoglobin concentration; TP: Total protein; ALB: Albumin; TCHO, Total cholesterol; TG: Triglyceride; LDL: Low density lipoprotein; HDL: High density lipoprotein; eGFR: Estimated glomerular filtration rate; Scr: Serum creatinine; UA: Uric acid; 24hTP: 24h uric total protein; ROC: Receiver operating characteristic curve; AUC: Area under the receiver operating characteristic curve; AIC: Akaike information criterion; suPAR: Soluble urokinase-type plasminogen activator receptor; FSGS: Focal segmental glomerulosclerosis, IgAN, IgA nephropathy; MPGN: membranoproliferative glomerulonephritis.

Declarations

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Authors' contributions

Zhanzheng Zhao, Xiao Jing and Jin Shang designed this study. Feng Wu and Yiding Zhang researched data and edited the manuscript. Wen Cui and Yijun Dong modified the manuscript. Yingyang Geng, Changhao Liu, Zemeng Li, Yandong Xie, and Xiaojing Cai collected the data. All authors have reviewed the manuscript. approved the final manuscript.

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Availability of data and materials

All the data and materials used in our study are available from the corresponding author on reasonable request.

Ethics approval and consent to participate

The First Affiliated Hospital of Zhengzhou University Ethics Review Committee granted ethical approval for the study and the ethics review approval ID was “2019-KY-361”. Informed consent was waived because of the retrospective analysis.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests

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Tables

Table 1. Baseline characteristics of PLA2R-negative MN and MCD. SBP, systolic blood pressure; DBP, diastolic blood pressure; RBC, red blood cell; WBC, white blood cell; PLT, platelet; Hb, hemoglobin; MCH, mean corpuscular hemoglobin, MCHC: mean corpuscular hemoglobin concentration; TP: total protein; ALB: albumin; TCHO, total cholesterol; TG: triglyceride; LDL: low density lipoprotein; HDL: high density lipoprotein; eGFR: estimated glomerular filtration rate; Scr: serum creatinine; UA: uric acid; 24hTP: 24h uric total protein.

Variables	MN (n=200)	MCD (n=144)	p Value
Age	50(34,58)	31(24,48)	<0.001
Gender, n (male, %)	114(57.0)	80(55.6)	0.790
SBP	130(120,140)	127(117,135)	0.114
DBP	80(76,87)	83(78,90)	0.117
RBC	4.33±0.58	4.64±0.64	<0.001
WBC	6.50(4.50,7.88)	6.2(5.1,7.6)	0.310
PLT	225(192,265)	252(203,313)	0.001
Hb	130.5±18.1	138.9±21.9	<0.001
MCH	30.3(29.3,31.4)	30.4(29.0,31.4)	0.878
MCHC	333(327,339)	333(326,338)	0.515
TP	51.0(44.7,58.1)	41.2(36.4,46.8)	<0.001
ALB	28.05(22.81,34.38)	19.78(16.83,23.18)	<0.001
TCHO	6.05(4.95,7.83)	10.06(7.86,11.93)	<0.001
TG	1.85(1.25,2.67)	2.18(1.51,3.20)	0.018
LDL	3.99(3.11,5.33)	7.82(5.34,9.66)	<0.001
HDL	1.25(1.05,1.57)	1.63(1.29,2.05)	<0.001
eGFR	104.03(90.24,114.49)	100.16(71.38,118.85)	0.383
Scr	68(56,80)	76(61,102)	<0.001
Urea	4.80(3.73,6.02)	5.50(3.99,8.38)	0.001
UA	310.0(259.3,375.3)	317.5(261.5,385.3)	0.650
24hTP	4.04(1.92,7.20)	6.43(4.28,9.04)	<0.001
Urine volume	1.6(1.2,2.2)	1.2(0.7,1.9)	<0.001

Table 2. Baseline characteristics showed there was no statistical difference in the training group and test group. SBP, systolic blood pressure; DBP, diastolic blood pressure; RBC, red blood cell; WBC, white blood cell; PLT, platelet; Hb, hemoglobin; MCH, mean corpuscular hemoglobin, MCHC: mean corpuscular hemoglobin concentration; TP: total protein; ALB: albumin; TCHO, total cholesterol; TG: triglyceride; LDL: low density lipoprotein; HDL: high density lipoprotein; eGFR: estimated glomerular filtration rate; Scr: serum creatinine; UA: uric acid; 24hTP: 24h uric total protein.

Variables	Training group(n=241)	Validation group(n=103)	p Value
Age	40(29,54)	48(31,55)	0.112
Gender, n (male, %)	131(54.3)	63(61.1)	0.244
SBP	130(119,139)	128(120,139)	0.745
DBP	81(77,90)	80(76,86)	0.278
RBC	4.47±0.63	4.52±0.61	0.706
WBC	6.50(5.30,7.90)	6.1(5.2,7.6)	0.268
PLT	234(197,279)	236(186,286)	0.694
Hb	134.4±20.6	133.3±19.3	0.648
MCH	30.4(29.3,31.5)	30.2(29.0,31.4)	0.863
MCHC	333(327,340)	333(327,337)	0.459
TP	46.6(40.6,54.4)	45.8(39.7,53.0)	0.648
ALB	24.0(19.5,30.8)	22.9(18.5,31.0)	0.439
TCHO	7.79(5.70,10.29)	6.90(5.09,9.64)	0.095
TG	2.08(1.36,2.94)	1.85(1.42,2.50)	0.219
LDL	5.14(3.65,7.88)	4.89(3.31,8.19)	0.472
HDL	1.41(1.12,1.79)	1.32(1.09,1.78)	0.490
eGFR	105.36(88.83,117.63)	98.96(80.73,110.67)	0.057
Scr	69.0(58.5,84.5)	73.0(60.0,90.9)	0.141
Urea	4.90(3.80,6.75)	5.00(4.00,6.60)	0.696
UA	309(254,377)	324.0(273,395)	0.094
24hTP	5.05(2.38,8.58)	5.16(2.55,7.81)	0.971
Urine volume	1.5(1.0,2.1)	1.5(1.0,2.2)	0.787

Table 3. Potential risk factors identified by univariate and multivariate logistic regression analysis. SBP, systolic blood pressure; DBP, diastolic blood pressure; RBC, red blood cell; WBC, white blood cell; PLT, platelet; Hb, hemoglobin; MCH, mean corpuscular hemoglobin, MCHC: mean corpuscular hemoglobin concentration; TP: total protein; ALB: albumin; TCHO, total cholesterol; TG: triglyceride; LDL: low density lipoprotein; HDL: high density lipoprotein; eGFR: estimated glomerular filtration rate; Scr: serum creatinine; UA: uric acid; 24hTP: 24h uric total protein.

Variable	Univariable		Multivariable	
	OR (95%CI)	P	OR (95%CI)	P
Age	1.053(1.033-1.073)	<0.001	1.057(1.024-1.090)	<0.001
Gender	0.927(0.555-1.550)	0.773		
SBP	1.016(1.000-1.032)	0.045		
DBP	0.987(0.963-1.011)	0.288		
RBC	0.385(0.243-0.611)	<0.001	0.313(0.102-0.959)	0.042
WBC	1.062(0.932-1.210)	0.365		
PLT	0.993(0.989-0.997)	0.001		
Hb	0.978(0.965-0.991)	0.001		
MCH	1.036(0.914-1.174)	0.582		
MCHC	1.012(0.988-1.038)	0.329		
TP	1.116(1.077-1.157)	<0.001		
ALB	1.174(1.118-1.234)	<0.001	1.190(1.044-1.357)	0.009
TCHO	0.656(0.582-0.740)	<0.001		
TG	1.010(0.893-1.142)	0.876		
LDL	0.647(0.570-0.734)	<0.001		
HDL	0.251(0.142-0.445)	<0.001	0.258(0.121-0.549)	<0.001
eGFR	1.007(0.998-1.017)	0.122		
Scr	0.992(0.986-0.999)	0.016		
Urea	0.851(0.775-0.934)	0.001	0.822(0.684-0.988)	0.037
UA	0.999(0.996-1.022)	0.500		
24hTP	0.865(0.803-0.930)	<0.001		
Urine volume	1.945(1.354-2.794)	<0.001		

Figures

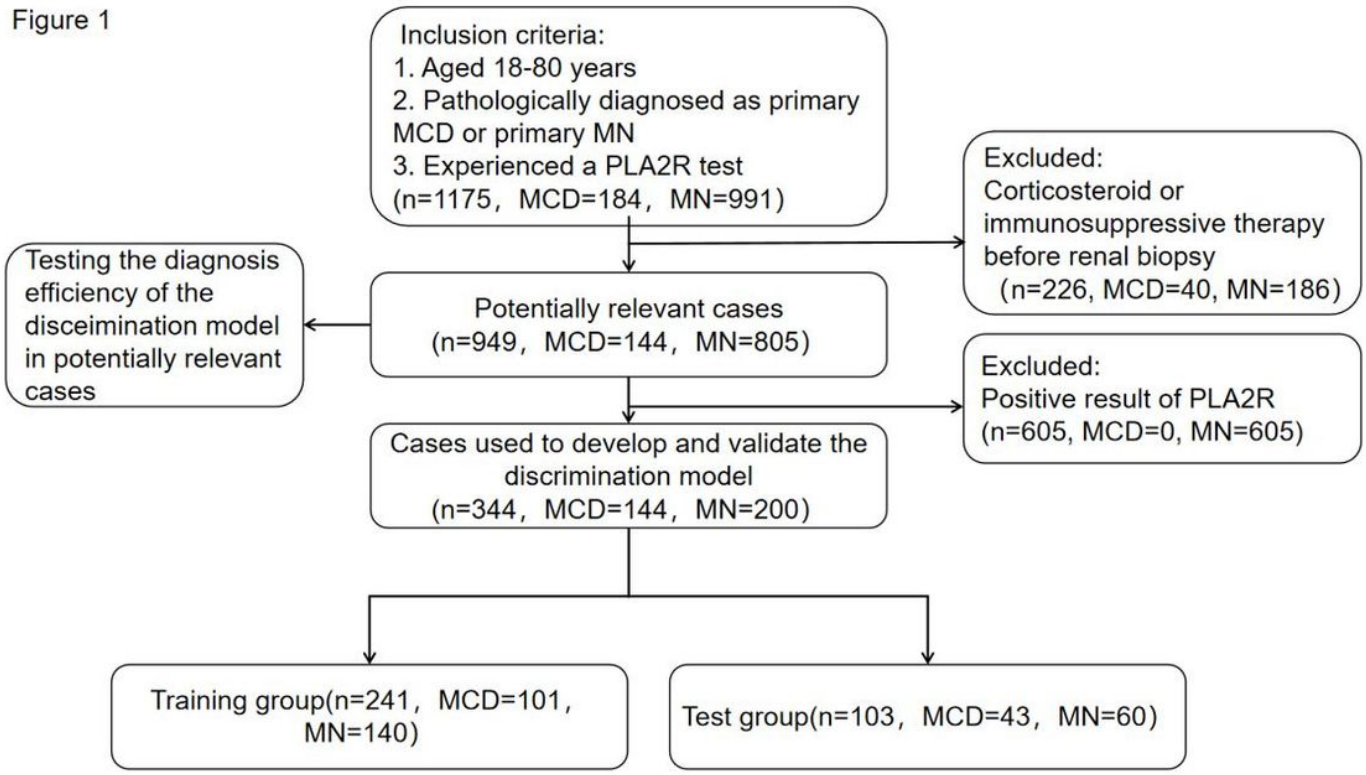

Figure 1

Enrollment flowchart of participants used for model development and validation. MCD, Minimal change disease; MN, Membranous nephropathy.

Figure 1
Inclusion criteria:
1. Aged 18-80 years
2. Pathologically diagnosed as primary MCD or primary MN
3. Experienced a PLA2R test
(n=1175, MCD=184, MN=991)
Excluded:
Corticosteroid or immunosuppressive therapy before renal biopsy
(n=226, MCD=40, MN=186)
Testing the diagnosis efficiency of the disceimination model in potentially relevant cases
Potentially relevant cases
(n=949, MCD=144, MN=805)
Excluded:
Positive result of PLA2R
(n=605, MCD=0, MN=605)
Cases used to develop and validate the discrimination model
(n=344, MCD=144, MN=200)
Training group(n=241, MCD=101, MN=140)
Test group(n=103, MCD=43, MN=60)

Figure 1

Enrollment flowchart of participants used for model development and validation. MCD, Minimal change disease; MN, Membranous nephropathy.

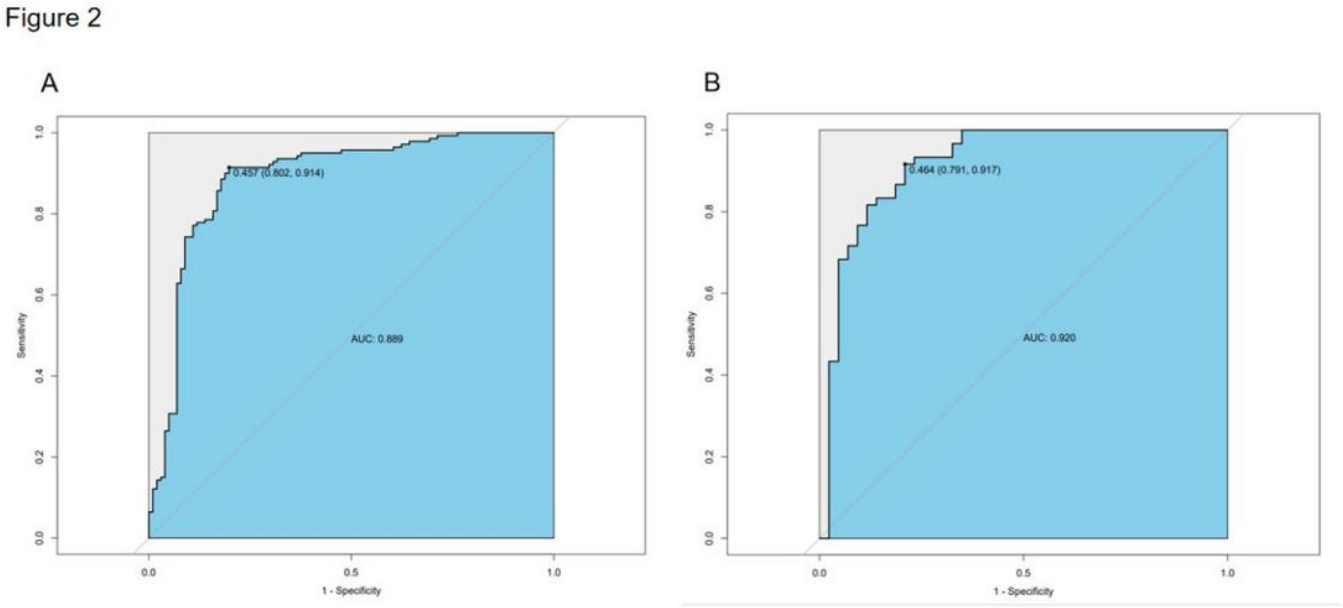

Figure 2

Differential capability of the nomogram. (A)ROC curve based on obtained potential risk factors identified by multivariate logistic regression analysis showing discrimination rate for PLA2R negative MN and MCD in the training group. (B)ROC curve based on obtained potential risk factors identified by multivariate logistic regression analysis showing discrimination rate for PLA2R negative MN and MCD in the test group. MCD, Minimal change disease; MN, Membranous nephropathy.

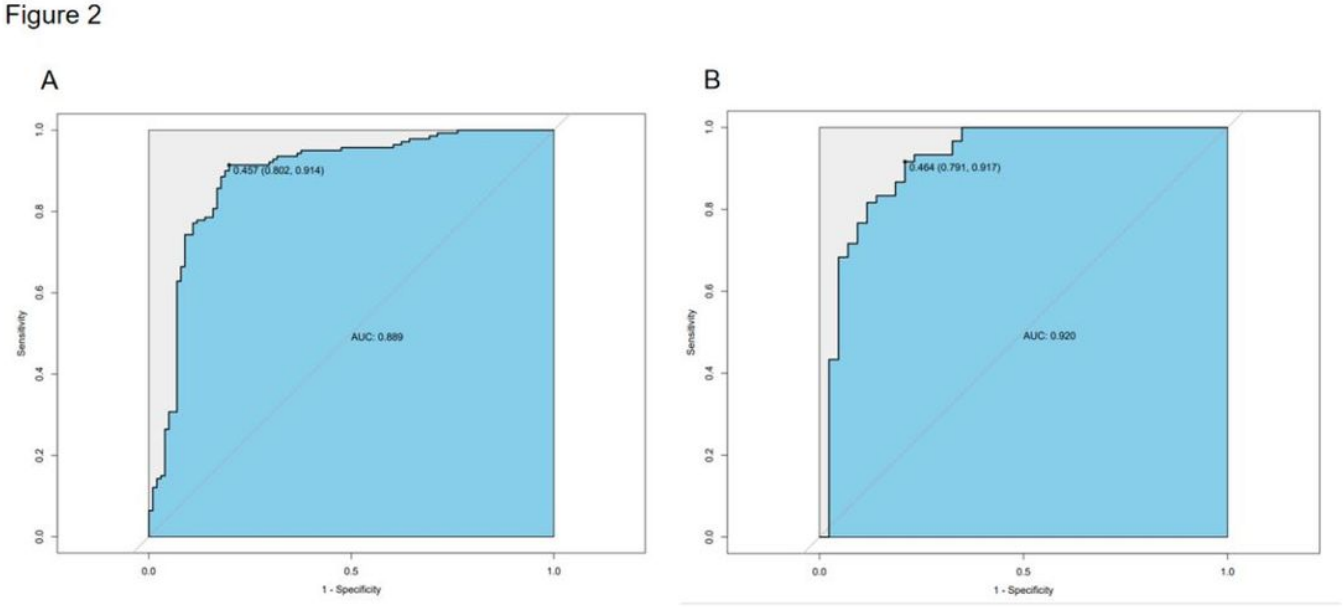

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Figure 3

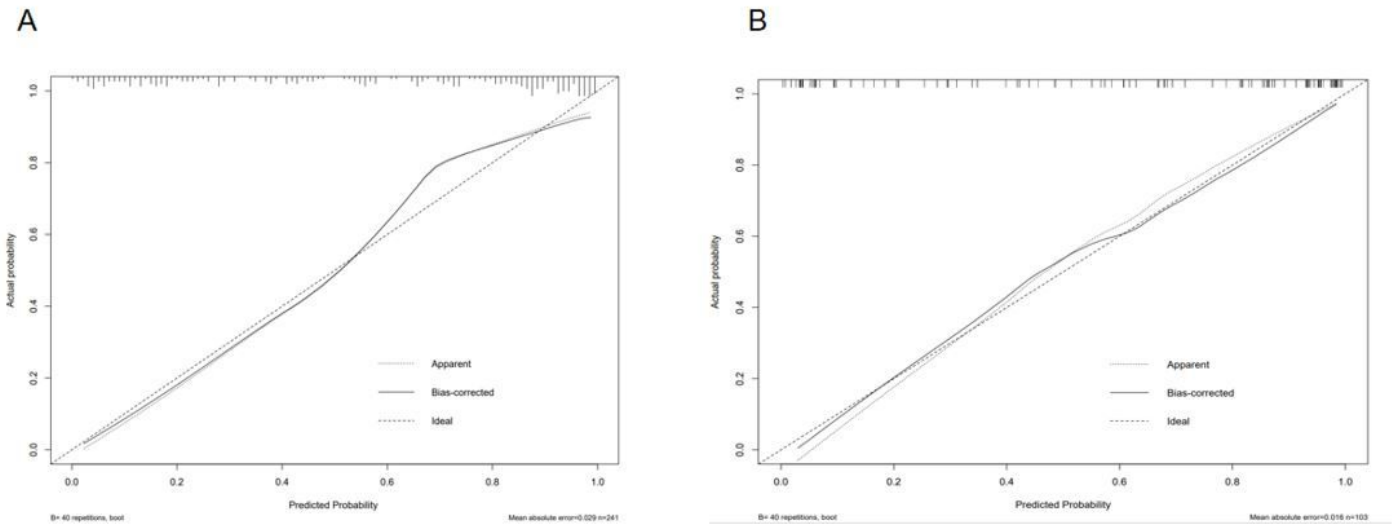

Figure 3

Calibration curve of the discrimination nomogram in the (A)training group or (B)test group. The x-axis represents the predicted probability of MN. The y-axis represents the actual pathologically diagnosed MN. The diagonal dotted line represents a perfect prediction by an ideal model. The solid line represents the performance of the nomogram, of which a closer fit to the diagonal dotted line represents a better prediction. MCD, Minimal change disease; MN, Membranous nephropathy.

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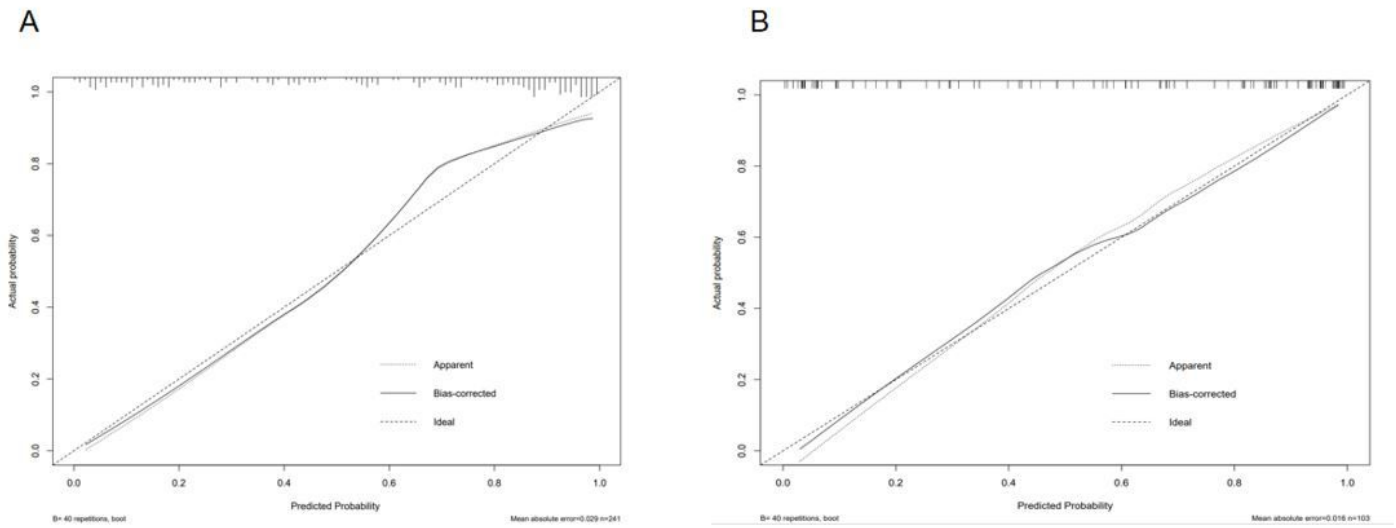

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Figure 4

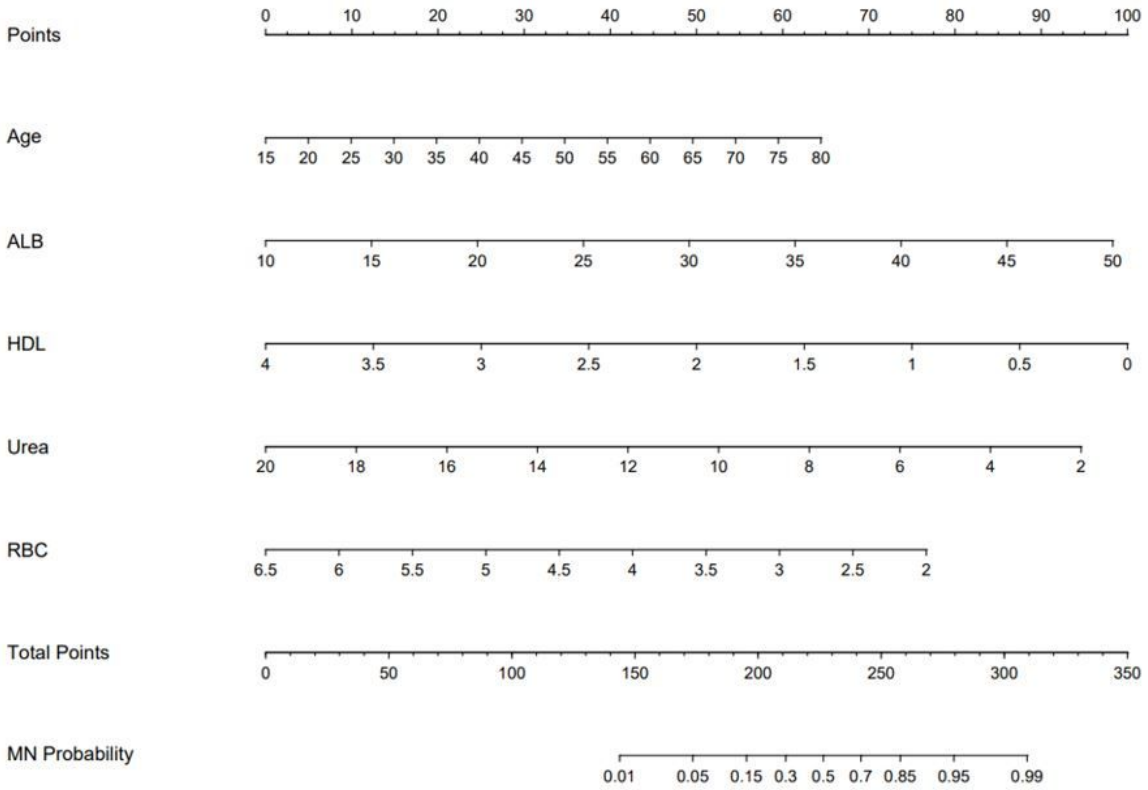

Figure 4

Nomogram based on the laboratory model; ALB, Albumin; HDL, High density lipoprotein; RBC, Red blood cell. MN, Membranous nephropathy.

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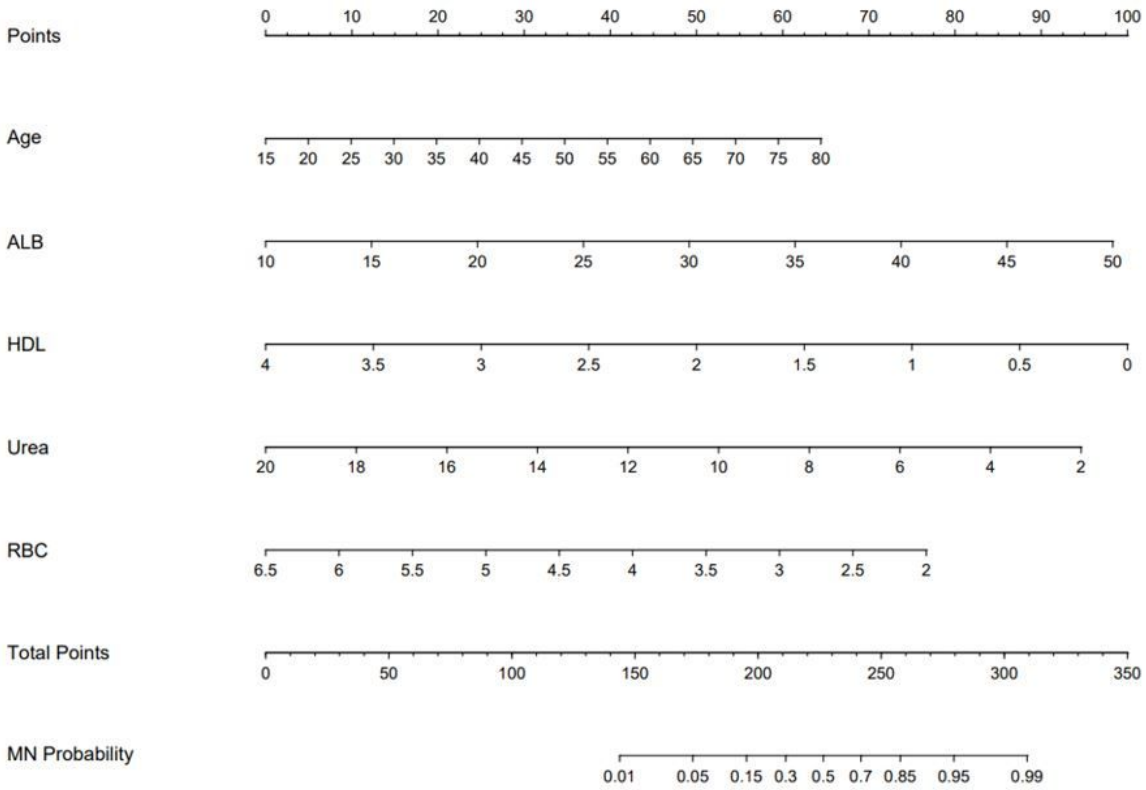

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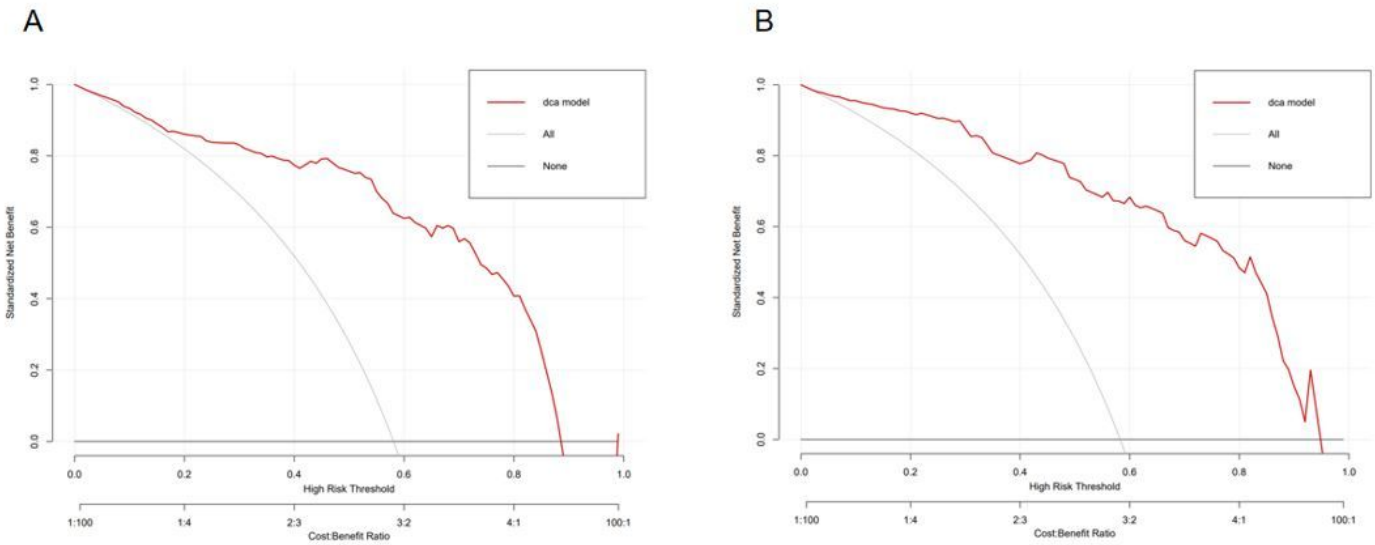

Figure 5

Decision curve for the nomogram predicting MN in (A)training group or (B)test group. Net benefit is shown on the y-axis. The thick red line represents the model; the thin gray line represents the assumption that all patients have MN; the thin black line represents the assumption that all patients have MCD. Both in training group or test group, the decision curve showed that if the threshold probability of a patient is > 1%, using the nomogram in the present study to predict MN adds more benefit than performing biopsy on all or no patients. MCD, Minimal change disease; MN, Membranous nephropathy.

Figure 5

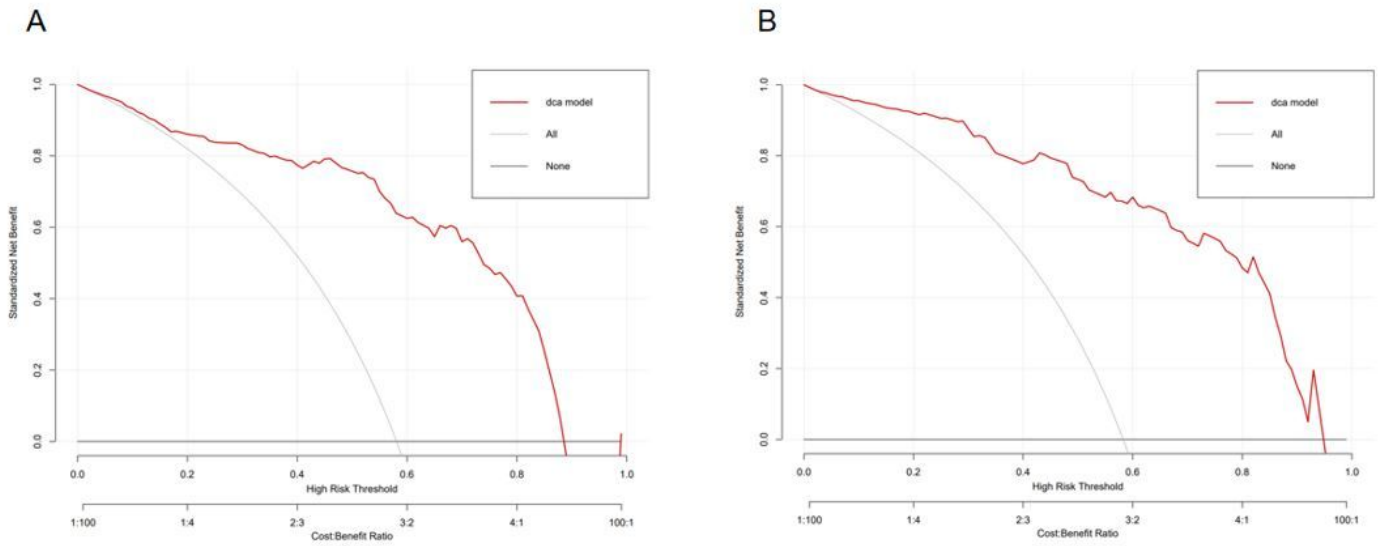

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Figure 6

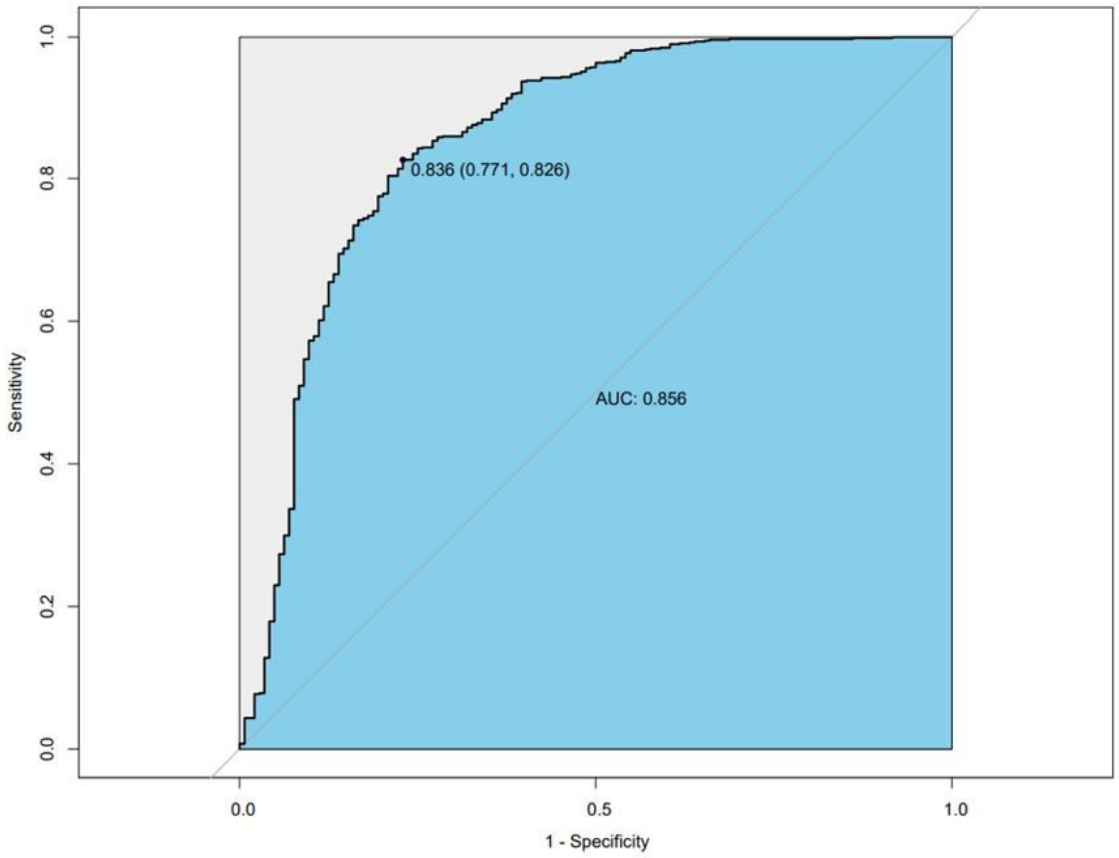
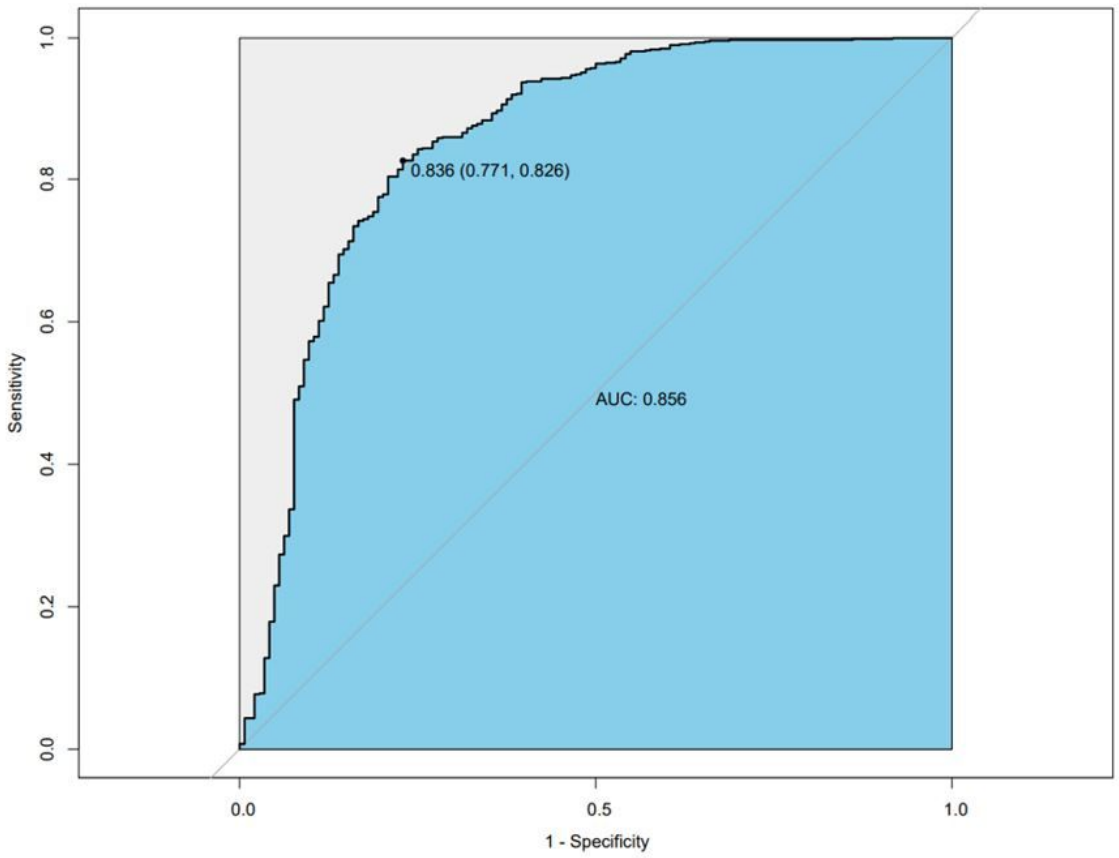


Figure 6

ROC curve showing discrimination effective of nomogram used in all MN and MCD. MCD, Minimal change disease; MN, Membranous nephropathy.

Figure 6



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ROC curve showing discrimination effective of nomogram used in all MN and MCD. MCD, Minimal change disease; MN, Membranous nephropathy.

Supplementary Files

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- FigureS2.pdf
- FigureS2.pdf
- TableS1.docx
- TableS1.docx