Renal impairment markers in type 2 diabetes patients with different types of hyperuricemia

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ABSTRACT
Aims/Introduction: Hyperuricemia (HUA) occurs because of decreased excretion of uric acid, increased synthesis of uric acid or a combination of both mechanisms. The proportions of these three types of HUA in type 2 diabetes patients are not known. In the mean time, we assume that different types of HUA might manifest with different renal damage, even in patients with normal renal filtration function.

Materials and Methods: We included 435 inpatients with type 2 diabetes at the Metabolic Disease Hospital of Tianjin Medical University from 2015 to 2016. Based on the clearance of uric acid, 90 patients with HUA were divided into three types: synthesis-increased HUA, excretion-decreased HUA and mixed type of HUA.

Results: Patients with the mixed type of HUA had the severest kidney injury manifested by a high level of 24 h urinary microalbumin, urinary immunoglobulin G, transferrin, α-galactosidase and β2-microglobulin compared with the normal uric acid group. Urinary immunoglobulin G, transferrin and α-galactosidase were also increased in patients with synthesis-increased HUA compared with the normal uric acid group. Patients with excretion-decreased HUA did not have an increased level of renal impairment markers; however, these patients had an increased body mass index, which might cause dysfunction of kidney excretion.

Conclusions: Excretion-decreased HUA is a more common type of HUA in type 2 diabetes patients that might be caused by dysfunction of tubular excretion instead of structural damage. The mixed type of HUA patients had the severest kidney glomerular and tubular damage compared with the normal uric acid group. Clinically, different types of hyperuricemia should be given individualized treatment according to their own characteristics.

INTRODUCTION
Hyperuricemia (HUA) is a metabolic disease of purine and its incidence has increased rapidly in recent years. It is closely related to metabolic syndrome, type 2 diabetes mellitus, hypertension, cardiovascular disease, chronic kidney disease and gout. The prevalence of HUA in the general population is 16.20%, and in type 2 diabetes mellitus patients is 17.24%. It can be divided into synthesis-increased HUA, excretion-decreased HUA and mixed type of HUA based on urinary uric acid excretion (UEUA) and fraction excretion of uric acid (FEUA). However, the distribution of the three types of HUA is not known in type 2 diabetes mellitus patients. In the mean time, renal damage occurs early in type 2 diabetes mellitus patients, even before microalbuminuria. We assume that different types of HUA can manifest with different renal damage, even in patients with normal renal filtration function. Therefore, the present study investigated for the first time the distribution, kidney damage, clinical characteristics and risk factors of the three types of HUA in type 2 diabetes mellitus patients with normal renal function.

METHODS
Study population
A total of 435 type 2 diabetes mellitus inpatients with normal renal function in the Metabolic Disease Hospital of Tianjin
### Table 1 | General characteristics and biochemical data of the participants

|                          | NUA (n = 345) | Synthesis-increased HUA (n = 25) | Mixed type of HUA (n = 21) | Excretion-decreased HUA (n = 44) |
|--------------------------|---------------|----------------------------------|---------------------------|---------------------------------|
| **Age (years)**          | 54.21 ± 10.21 | 51.76 ± 10.84                    | 49.67 ± 8.70              | 53.14 ± 10.74                   |
| **Male/female**          | 197/148       | 12/13                            | 14/7                      | 20/24                           |
| **Duration of T2DM**     | 9.37 ± 6.31   | 7.94 ± 6.02                      | 7.85 ± 4.00               | 7.44 ± 5.54                     |
| **BMI (kg/m²)**          | 26.12 ± 4.17  | 26.24 ± 2.97                     | 27.21 ± 4.09              | 29.68 ± 4.95**                  |
| **SBP (mmHg)**           | 129.09 ± 15.93| 130.60 ± 16.09                   | 129.00 ± 8.67             | 129.89 ± 11.93**                |
| **DBP (mmHg)**           | 78.84 ± 8.60  | 81.20 ± 9.16                     | 77.50 ± 7.86              | 79.55 ± 8.19                    |
| **FPG (mmol/L)**         | 10.40 ± 4.10  | 8.96 ± 3.27                      | 9.42 ± 2.33               | 10.84 ± 3.45                    |
| **HbA1c (%)**            | 8.77 ± 1.97   | 7.43 ± 1.62**                    | 7.97 ± 1.65               | 8.57 ± 2.12                     |
| **TG (mmol/L)**          | 2.19 ± 2.00   | 3.54 ± 2.49*                     | 4.13 ± 3.78*              | 2.38 ± 2.15                     |
| **TC (mmol/L)**          | 5.35 ± 1.31   | 5.40 ± 1.33                      | 5.57 ± 1.65               | 5.14 ± 1.05                     |
| **HDL-C (mmol/L)**       | 1.40 ± 0.35   | 1.23 ± 0.26*                     | 1.36 ± 0.24               | 1.27 ± 0.23*                    |
| **ALT (U/L)**            | 25.42 ± 17.72 | 21.47 ± 13.23                    | 23.85 ± 12.81             | 25.32 ± 17.09                   |
| **AST (U/L)**            | 20.48 ± 10.35 | 19.47 ± 6.51                     | 22.24 ± 8.51              | 21.60 ± 10.21                   |
| **SCr (µmol/L)**         | 59.11 ± 11.23 | 66.48 ± 13.15*                   | 58.04 ± 9.84              | 59.98 ± 12.28                   |
| **BUN (mmol/L)**         | 5.50 ± 3.08   | 5.48 ± 1.35                      | 4.99 ± 0.98               | 5.38 ± 1.20                     |
| **SUA (µmol/L)**         | 278.36 ± 65.64| 439.57 ± 83.06**                 | 460.13 ± 45.81**          | 445.31 ± 83.02**                |
| **eGFR (mL/min·1.73 m²)**| 120.34 ± 20.19| 102.71 ± 13.12**                 | 127.43 ± 18.61            | 114.80 ± 18.12                  |
| **24hUCr (mg/24 h)**     | 11.04 ± 3.78  | 12.56 ± 2.95*                    | 15.35 ± 4.24*             | 10.25 ± 3.45                    |
| **24hUAU (mg/24 h)**     | 649.37 ± 317.55| 924.90 ± 211.80**                | 881.23 ± 172.36**         | 474.99 ± 174.36**               |
| **UEUA (mg/day [1.73 m²]⁻¹)** | 578.08 ± 277.52 | 839.42 ± 166.03**             | 765.84 ± 140.04**         | 408.28 ± 139.69**               |
| **FEUA (%)**             | 7.94 ± 3.99   | 6.67 ± 1.10                      | 4.37 ± 0.53*              | 3.77 ± 0.85*                    |

Data are expressed as mean ± standard deviation. Compared with the normal plasma uric acid (NUA) group *P < 0.05, **P < 0.01. 24HUCr, 24-h urinary creatinine; 24HUAU, 24-h urinary uric acid; ALT, alanine aminotransferase; AST, aspartate transaminase; BMI, body mass index; BUN, blood urea nitrogen; DBIL, direct bilirubin; DBP, diastolic blood pressure; eGFR, estimated glomerular filtration rate; FEUA, fraction excretion of uric acid; FPG, fasting plasma glucose; HbA1c, hemoglobin A1c; HDL-C, high density lipoprotein cholesterol; HUA, hyperuricemia; SBP, systolic blood pressure; SCr, serum creatinine; SUA, serum uric acid; T2DM, type 2 diabetes mellitus; TBIL, total bilirubin; TC, total cholesterol; TG, triglyceride; UEUA, urinary uric acid excretion.
Medical University, Tianjin, China, from 2015 to 2016 were enrolled. Type 2 diabetes mellitus was diagnosed according to World Health Organization 2009 criteria. To exclude the effects of kidney impairment on uric acid excretion, we included the patients with estimated glomerular filtration rate (eGFR) >90 mL/min·1.73 m⁻². We also excluded patients with acute complications of diabetes, primary kidney disease, abnormal liver function, anemia, malignant tumors, severe cardiovascular and cerebrovascular diseases, alcohol abuse, previous history of gout, hypothyroidism, hyperparathyroidism, and patients who were pregnant, as well as patients taking medications for lowering uric acid, diuretics, glucocorticoids, antituberculosis drugs and cytotoxic drugs that might affect the uric acid level.

Our study obtained the approval of the ethical committee. Informed consent was obtained from all participants.

Measurements
The following data were collected. General information: age, sex, body mass index (BMI), blood pressure and diabetes course. Biochemical data: hemoglobinA1c (HbA1c) was measured by high performance liquid chromatography, fasting plasma glucose was measured by the glucose oxidase method, and blood urea nitrogen, serum creatinine, serum uric acid (SUA), triglyceride (TG), total cholesterol, high-density lipoprotein cholesterol, total bilirubin (TBIL), direct bilirubin, alanine aminotransferase and aspartate transaminase were tested by automatic biochemical analyzer. In addition, 24-h urine specimens were collected after blood glucose was controlled, and patients had no strenuous exercise, fever, pscyentonia and high-protein diet; 24-h urinary creatinine (24hUCr) and uric acid (24hUUA) were measured by the enzyme colorimetric method; 24-h urinary microalbumin was measured by the immunoturbidimetric method. The first morning midstream urine was collected in all patients. Urinary immunoglobulin G (IgG) and transferrin (TF) were measured by transmission immunosassay, and the immunoturbidimetric method was used to measure retinol-binding protein (RBP), N-acetyl-β-glucosaminidase (NAG), β2-microglobulin (β2-MG) and α-galactosidase.

The Modification of Diet in Renal Disease formula was used to estimate the glomerular filtration rate (eGFR). The diagnostic criteria for hyperuricemia was >420 μmol/L for men and >360 μmol/L for women. The level of uric acid excretion was expressed as the UEUA rate and the fraction of uric acid excretion (FEUA). UEUA was calculated by the equation of 24hUUA/body surface area. FEUA was calculated by the following equation: urinary uric acid × serum creatinine/(urinary creatinine × body ureic acid). The definitions for various types of HUA are listed as follows: synthesis-increased HUA if FEUA ≥5.5% and UEUA >600 mg/day (1.73 m²)⁻¹, mixed type of HUA if FEUA <5.5% and UEUA >600 mg/day (1.73 m²)⁻¹ and excretion-reduced HUA if FEUA <5.5% and UEUA ≤600 mg/day (1.73 m²)⁻¹.

Statistical analysis
SPSS 19.0 statistical software (SPSS, Chicago, IL, USA) was used for statistical analysis. Data with normative distribution or approximate normal distribution were expressed as mean ± standard deviation or mean ± standard error of the mean. The one-factor analysis of variance (ANOVA) test was used to analyze the difference of data with normal distribution, whereas a non-parametric test was used to analyze the difference of data with non-normal distribution. The χ²-test was used to evaluate the differences of proportions. The independent risk factors of various types of HUA were analyzed by logistic regression.

RESULTS
Distribution of different types of HUA in Type 2 diabetes mellitus patients
Among the 435 patients with type 2 diabetes mellitus, 20.69% (90 cases) had HUA. Based on FEUA and UEUA, these patients were grouped into three types of HUA: synthesis-increased HUA (28%, 25 cases), excretion-decreased HUA (49%, 44 cases) and mixed type of HUA (23%, 21 cases).

Figure 1 | Level of (a) urinary microalbumin (UMA), (b) transferrin (TF) and (c) immunoglobulin G (IgG) in patients with normal plasma uric acid (NUA), synthesis-increased hyperuricemia (HUA), mixed type of HUA and excretion-decreased HUA. Damage of kidney glomeruli in different types of hyperuricemia (HUA) compared with the normal plasma uric acid (NUA) group. Data are expressed as mean ± standard error of the mean. *P < 0.05, **P < 0.01. IgG, urinary immunoglobulin G; TF, urinary transferrin; UMA, 24-h urinary microalbumin.
General characteristics of different types of HUA

Compared with patients with normal plasma uric acid (NUA), SUA, TG, serum creatinine, SUA, 24hUCr, 24hUUA and UEUA were higher, and eGFR, high-density lipoprotein, HbA1c, TBIL and direct bilirubin were lower for patients with synthesis-increased HUA (all \( P < 0.05 \); Table 1).

Compared with patients with NUA, TG, SUA, 24hUCr, 24hUUA and UEUA were higher, in addition, FEUA was lower in the mixed type of HUA (all \( P < 0.05 \)).

Compared with patients with NUA, the BMI and SUA were higher, but the high-density lipoprotein, 24hUUA, UEUA and FEUA were lower in patients with excretion-decreased HUA (all \( P < 0.05 \)).

Markers reflecting glomeruli damage

As shown in Figure 1, urinary IgG and TF were higher for patients with synthesis-increased HUA compared with NUA. The level of 24-h urinary microalbumin, urinary IgG and TF were significantly higher than patients with the mixed type of HUA. There were no differences in urinary protein excretion and markers reflecting glomeruli damage between the excretion-reduced HUA and NUA group.

Markers reflecting tubuli damage

As shown in Figure 2, urinary \( \alpha \)-galactosidase was higher for patients with synthesis-increased HUA compared with the NUA group. However, there was no statistically significant difference in \( \beta_2 \)-MG, RBP and NAG between the two groups. The level of urinary \( \alpha \)-galactosidase and \( \beta_2 \)-MG were significantly higher than the NUA group in the mixed type of HUA. However, RBP and NAG were not statistically different. Patients with excretion-reduced HUA and NUA were not statistically different in \( \beta_2 \)-MG, \( \alpha \)-galactosidase, RBP and NAG.

Risk factors of various types of HUA

The independent risk factors of synthesis-increased HUA were eGFR (odds ratio [OR] 0.939, 95% confidence interval [CI] 0.891–0.990, \( P < 0.05 \)), TBIL (OR 0.755, 95% CI 0.594–0.960, \( P < 0.05 \)) and TG (OR 1.215, 95% CI 1.001–1.473, \( P < 0.05 \); Table 2). The independent risk factor was TG (OR 1.230, 95% CI 1.083–1.396, \( P < 0.01 \)) in the mixed-type of HUA (Table 3).

The independent risk factor of excretion-reduced HUA was BMI (OR 1.163, 95% CI 1.086–1.247, \( P < 0.001 \); Table 4).

DISCUSSION

In our previous study, we found that a great proportion of type 2 diabetes mellitus patients manifested with kidney glomerular and tubular damage even before the occurrence of microalbuminuria. As HUA is closely associated with renal damage, we assume that different types of HUA might have different manifestations of renal damage. NAG and \( \alpha \)-galactosidase are two enzymes of renal tubular epithelial cells, and their increase in urine reflects a structural injury of the renal tubule. Urinary

![Figures 2](http://wileyonlinelibrary.com/journal/jdi)
Table 2 | Risk factors of synthesis-increased hyperuricemia in type 2 diabetes mellitus patients

| Risk Factor | B (OR) | Wald | P | OR 95% CI |
|-------------|--------|------|---|----------|
| HbA1c (%)   | -0.445 | 3.09 | 0.079 | 0.641, 0.390–1.052 |
| eGFR (mL/min·1.73 m²) | -0.063 | 5.428 | 0.020 | 0.939, 0.891–0.990 |
| TBIL (μmol/L) | -0.281 | 5.237 | 0.022 | 0.755, 0.594–0.960 |
| TG (mmol/L) | 0.194  | 3.899 | 0.048 | 1.215, 1.001–1.473 |
| HDL-C (mmol/L) | 0.061  | 6.043 | 0.961 | 1.063, 0.991–1.2410 |

95% CI, 95% confidence interval; eGFR, estimated glomerular filtration rate; HbA1c, hemoglobin A1c; HDL-C, high-density lipoprotein cholesterol; OR, odds ratio; TBIL, total bilirubin; TG, triglyceride.

Table 3 | Risk factors of mixed type of hyperuricemia in type 2 diabetes

| Risk Factor | B (OR) | Wald | P | OR 95% CI |
|-------------|--------|------|---|----------|
| TG (mmol/L) | 0.207  | 10.134 | 0.001 | 1.230, 1.083–1.396 |

95% CI, 95% confidence interval; OR, odds ratio; TG, triglyceride.

Table 4 | Risk factors of excretion-decreased hyperuricemia in type 2 diabetes mellitus patients

| Risk Factor | B (OR) | Wald | P | OR 95% CI |
|-------------|--------|------|---|----------|
| BMI (kg/m²) | 0.151  | 18.432 | 0.000 | 1.163, 1.086–1.247 |
| HDL-C (mmol/L) | -0.0922 | 2.136 | 0.144 | 0.938, 0.865–0.937 |

95% CI, 95% confidence interval; BMI, body mass index; HDL-C, high-density lipoprotein cholesterol; OR, odds ratio.

RBP8 and β2-MG9 are classic indicators of tubular reabsorption ability, and their increase in urine infers a functional injury of the renal tubule. Urinary IgG and TF10 are macromolecules, and their increase in urine reflects damage of glomeruli.

Excretion-decreased HUA is a more common type of HUA in patients with type 2 diabetes mellitus. It is generally assumed that excretion-reduced HUA in patients might be caused by the decreased glomerular filtration rate and increased reabsorption of the renal tubule; however, no statistical difference in eGFR, urinary microalbumin, and biomarkers of glomerular and renal tubular injury were found in excretion-decreased HUA in the present study. Therefore, it might be caused by dysfunction of tubular excretion instead of structural damage. Furthermore, we found that BMI was an independent risk factor of excretion-reduced HUA. Obesity could result in elevated UA levels by a reduction in urinary uric acid excretion11. The possible reason is that obese patients are always accompanied by insulin resistance, which can activate the sympathetic nervous system and the renin–angiotensin system and then produce lactic acid, thus leading to competitive inhibition of UA secretion and ultimately causing higher SUA. Previous studies have shown that weight loss was an effective non-pharmacological treatment for lowering SUA12. Therefore, the BMI should also be controlled properly.

Compared with the NUA group, patients with the mixed type of HUA manifested the severest damage to the kidney tubule, as well as glomeruli. The 24hUUA was significantly higher than in NUA patients at the same time, so in terms of clinical medicine, we should try to avoid using drugs that can aggravate kidney damage. In addition, drugs promoting the excretion of uric acid can lead to further increases of 24hUUA, thereby increasing the kidney damage, so it is advised to use drugs that inhibit synthesis instead of promoting excretion of UA to protect kidneys from the damage of high UUA. TG was the independent risk factor in the type of HUA. On the one hand, TG can make small arteries of kidney stenosed or even occlusive by long-term lipid metabolism disorder, which eventually results in reduced excretion of UA in the kidney. On the other hand, it is reported that the activity of glyceraldehyde 3-phosphate dehydrogenase is reduced by hyperglycemia and hyperlipidemia. Therefore it can accelerate UA synthesis. Whereas lipid-lowering treatment has been proved to reduce the level of SUA in patients with type 2 diabetes mellitus.

We found that compared with NUA patients, synthesis-increased HUA patients had increased urinary levels of TF, IgG and α-galactosidase, which reflect damage both in the glomeruli and tubule. Therefore, it might be helpful to pay early attention to kidney damage in this type of HUA. In the present study, we also studied the risk factors in patients with synthesis-increased HUA. We found that TBIL was a protective factor. It has been proved that serum bilirubin can inhibit oxidative stress13 and inflammation14, which might be helpful in lowering plasma levels of uric acid. The present results also showed that TG was a risk factor. Therefore, to control synthesis-increased HUA, we need to lower the TG level.

A limitation of the present study was that we could rule out other causes of hyperuricemia, such as Down syndrome, vitamin B12 deficiency and genetic disorders that might also affect the level of plasma uric acid. Considering the low prevalence of these diseases, we believe that excluding or including these patients might not have made a great difference to the final result.

In summary, excretion-reduced HUA is a more common type of HUA in patients with type 2 diabetes mellitus, which might be caused by dysfunction of tubular excretion instead of structural damage. Patients with the mixed type of HUA have the severest kidney glomerular and tubular damage compared with NUA patients. Clinically, different types of hyperuricemia should receive individualized treatment according to their own characteristics.

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DISCLOSURE
The authors declare no conflict of interest.

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