

Longitudinal follow-up study of the retrobulbar and intrarenal hemodynamics in patients with T2DM

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Abstract

Objective: The primary aim of this study is to examine the hemodynamics of retrobulbar and intrarenal in the changes of early stage of type 2 diabetes mellitus (T2DM) patients from 2000 to 2015 and to assess incidence associated with diabetic kidney disease (DKD) and diabetic retinopathy (DR).

Method: Our study contained 60 subjects newly diagnosed of T2DM were divided into 2 groups base on the mean resistive index (RI) (≤ 0.7 and > 0.7) of hemodynamic and to compare between-group differences of the early changes in hemodynamics of retrobulbar and intrarenal and also to conclude the incidences of diabetic kidney disease (DKD) and diabetic retinopathy (DR) subsequently with a long follow-up duration(2000–2015). First, to compare the mean RI of central retinal artery (CRA) between 2 groups. Second, to compare the mean RI of intrarenal hemodynamics in the bilateral interlobular renal arteries, renal function parameters (blood urea nitrogen (BUN), creatinine (Cr), blood glucose parameters (glycosylated hemoglobinA1c (HbA1c), fasting plasma glucose (FBG), and 2-hour postprandial blood glucose (2hPBG)), glomerular filtration rate (GFR), albumin excretion rate (AER), and urine albumin-to-creatinine ratio (UACR) between 2 groups.

Results: First part of our follow-up studies was to compare hemodynamic RI index of retrobulbar in years of 2000 and 2015, both renal function and blood glucose parameters were fund significantly enhanced in subject group RIs ≤ 0.7 . Incidence of DKD and DR was notably lower in group RIs ≤ 0.7 than group RIs > 0.7 , difference was statistically significant ($P < .05$). Incidence of HbA1c $\leq 7\%$ was higher in group RIs ≤ 0.7 than group RIs > 0.7 , but difference was not statistically significant ($P > .05$). Incidence of proliferative diabetic retinopathy (PDR) was notably lower in group RIs ≤ 0.7 than group RIs > 0.7 , but the difference was not statistically significant ($P > .05$). Second part of our follow-up studies was to compare hemodynamic RI index of interlobular renal in years of 2000 and 2015, both renal function and blood glucose parameters were fund significantly enhanced in subject group RIs ≤ 0.7 . Compared data of various incidences from first part of study were coherent with second part. (Incidence of DKD and DR was notably lower in group RIs ≤ 0.7 than group RIs > 0.7 , difference was statistically significant ($P < .05$). Incidence of HbA1c $\leq 7\%$ was higher in group RIs ≤ 0.7 than group RIs > 0.7 , but difference was not statistically significant ($P > .05$). Incidence of PDR was notably lower in group RIs ≤ 0.7 than group RIs > 0.7 , but the difference was not statistically significant ($P > .05$).

Conclusions: RIs of retrobulbar and interlobular renal which would serve as a good predictors for the hemodynamics changes in retrobulbar and intrarenal would assess incidence of DKD and DR during the preclinical stage in long-term range excluding renal function and HbA1c in T2DM patients.

Abbreviations: 2hPBG = 2-hour postprandial blood glucose, AER = albumin excrete rate, AGEs = advanced glycated end-products, BP = blood pressure, BUN = blood urea nitrogen, CDI = color Doppler imaging, CMECs = cardiac microvascular endothelial cells, Cr = creatinine, CRA = central retinal artery, DBP = diastolic blood pressure, DKD = diabetic kidney disease, DN =

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diabetic nephropathy, DR = diabetic retinopathy, EDV = end-diastolic velocity, ESRD = end-stage renal disease, FBG = fasting plasma glucose, GFR = glomerular filtration rate, HbA1c = glycosylated hemoglobin A1c, HDL-C = high-density lipoprotein cholesterol, K/DOQI = kidney disease outcome quality initiatives, LDL-C = low-density lipoprotein cholesterol, LS = least squares, OA = arteria ophthalmica, OAPP = advanced oxidation protein products, OGTT = oral glucose tolerance test, PCA = posterior ciliary artery, PSV = peak systolic velocity, RAS = renin-angiotensin system, RBS = random blood sugar, RI = resistivity index, SBP = Systolic blood pressure, SD = standard deviation, TC = Total cholesterol, TG = triglycerides, T2DM = type 2 diabetes mellitus, UACR = urinary albumin/creatinine ratio.

Keywords: diabetic kidney disease, diabetic retinopathy, intrarenal hemodynamics, resistivity index, retrobulbar hemodynamics, type 2 diabetes mellitus

1. Introduction

The incidence and prevalence of diabetes mellitus have increased remarkably worldwide, primarily due to the increasing incidence of type 2 diabetes mellitus (T2DM). Patients with T2DM develop abnormal glucose and lipid metabolism, which are associated with multiple organ dysfunction syndromes. Diabetes mellitus can cause by metabolic disorders in different organs, including intraocular tissue structures. Diabetic retinopathy (DR), a type of microangiopathy that primarily occurs several years after the development of diabetes (an average of 5–10 years), is the most common ocular complication of diabetes. It is responsible for the development of blindness in patients between 20 and 74 years of age in developed countries.^[1] New terminology to describe kidney disease attributable to diabetes has been introduced in recent guidelines (National Kidney Foundation, 2007). Thus, the term diabetic nephropathy (DN) has been replaced with diabetic kidney disease (DKD), a prevalent microvascular disease. Specifically, as a long-term complication, DKD is defined as structural, functional, and clinical abnormalities of the kidneys due to diabetes. This complication has become the single most frequent cause of end-stage renal disease (ESRD).^[2] Indeed, DKD and DR are leading causes of ESRD and acquired blindness, respectively.^[3]

Color Doppler imaging (CDI) is a safe, noninvasive approach applied in ophthalmology that has been used to evaluate hemodynamic changes in the orbital vessels for 2 decades. In this technique, a simultaneous B-mode sonogram is combined with colors representing movements based on Doppler frequency shifts to assess blood flow velocities, including peak systolic velocity (PSV) and end-diastolic velocity (EDV), in retrobulbar vessels. In addition, the resistivity index (RI) of every retrobulbar vessel is calculated to measure peripheral vascular resistance.^[4] The Doppler RI, $(PSV-EDV)/PSV$, reflects the intrarenal vascular resistance and has been widely used to quantify the changes in renal blood flow caused by renal disease. Alterations in intrarenal arterial waveforms have been shown to be associated with urinary obstruction, renal vascular disease and several types of intrinsic renal disorders.^[5]

Novelty is promised when to apply the retrobulbar and intrarenal RI to assess incidence of DKD and DR in long-term. We presume that by exploring hemodynamic correlation between the 2 organs is significant as this indicator is very valuable for predicting vascular complications during the preclinical stage.

2. Patients and methods

In our longitudinal follow-up studies, 154 subjects initially diagnosed with T2DM (82 males and 72 females) in year 2000 were divided into 2 groups based on the mean central retinal artery (CRA) and mean interlobular renal arteries hemodynamics (resistive index, RI) (≤ 0.7 and > 0.7). However, only 60 subjects

(32 males and 28 females) could be allocated in 2015. The retrobulbar and intrarenal blood glucose (FBG, 2hPBG, and HbA1c), RI, UACR, AER, BUN, and Cr were last measured in 2015 and compared to the values obtained in 2000. The diabetes duration (range: 4 to 12 years), glycosylated hemoglobin A1c (HbA1c) levels [range: 7.0% (53 mmol/mol) to 10% (86 mmol/mol)], retrobulbar hemodynamic parameters (RI), renal function parameters [blood urea nitrogen (BUN) and creatinine (Cr) levels], albumin excretion rate (AER), urine albumin-to-creatinine ratio (UACR)], and blood pressure (BP) were recorded (Table 1 and Table 2). T2DM was diagnosed in accordance with the guidelines of the American Diabetes Association^[6]:

1. symptoms of diabetes (thirst, polydipsia, diuresis, and weight loss) that cannot be interpreted as any other pathology;
2. random plasma sugar (RPS) level ≥ 11.1 mol/L, fasting plasma glucose (FPG) level ≥ 7.0 mol/L, the outcome of the oral glucose tolerance test (OGTT) was a 2-hour postprandial plasma glucose (2hPG) level ≥ 11.1 mol/L; or
3. an absence of symptoms of diabetes but the RPS level was ≥ 11.1 mol/L or the FPG level was ≥ 7.0 mol/L.

DKD was confirmed according to US Kidney Disease Outcome Quality Initiatives (K/DOQI) guidelines (<https://www.kidney.org/professionals/KDOQI/guidelines> commentaries). Based on the Fukuda classification, we evaluated the DR stage as follows: no signs of diabetic retinopathy-A0, nonproliferative diabetic retinopathy (mild)-A1, nonproliferative diabetic retinopathy (moderate)-A2, nonproliferative severe/preproliferative diabetic retinopathy-B1.^[7] The DKD stage was evaluated categorically according to the AER levels in at least 2 of 3 timed urine collections: normal AER (< 30 mg/24 hour or < 20 μ g/minutes), microalbuminuria (30–300 mg/24 hours or 20–200 μ g/minutes), and macroalbuminuria (> 300 mg/24 hours or > 200 μ g/minutes). The presence of ESRD was determined based on whether patients had undergone a kidney transplant or were receiving dialysis (patients with ESRD were excluded at baseline). The GFR was estimated with a formula based on cystatin C.^[8] We used the NGSP converter to convert HbA1c levels from Diabetes Control and Complications Trial (DCCT)-derived units (%) to the units recommended by the International Federation of Clinical Chemistry and Laboratory Medicine (IFCC) (mmol/mol) (<http://www.ngsp.org/convert1.asp>).

The selection criteria were

1. patients diagnosed with T2DM by the guidelines of the American Diabetes Association^[6];
2. had no history of smoking, pulmonary disease, cold, or pulmonary infection within a 2-week period;
3. were not diagnosed with hepatopathy, nephropathy, hyperuricemia, or gastrointestinal disease;
4. were likely to have good compliance and were able to visit our hospital for periodic assessments.

Table 1
The change of RI in the CRA from baseline to year 15.

Content	Group (mean RI in CRA)		t/ χ^2 value	P value	
	≤ 0.7	> 0.7			
NO. (n)	25	35	-	-	
Sex, n (%)					
	Male	19 (54.29)	0.031	.861	
	Female	16 (45.71)			
Age (years) in 2000	52.44 ± 5.77	52.49 ± 5.64	0.031	.976	
BMI (kg/m ²) in 2000	27.44 ± 1.36	27.31 ± 1.64	0.314	.755	
Diabetes duration (years) in 2000	7.68 ± 2.12	7.23 ± 1.80	0.890	.377	
HbA1c in 2000					
	%	7.18 ± 0.35	7.79 ± 0.45	5.124	.000*
	mmol/mol	29.30 ± 6.31	29.57 ± 7.43	5.124	.000*
HbA1c in 2015					
	%	7.46 ± 0.40	8.05 ± 0.49	4.932	.000*
	mmol/mol	54.21 ± 4.35	60.64 ± 5.38	4.932	.000*
Baseline HbA1c, n (%) in 2000					
	≤ 7%/ (53 mmol/mol)	6 (24.00)	3 (8.57)	2.723	.099
	> 7%/ (53 mmol/mol)	19 (76.00)	32 (91.43)		
Baseline HbA1c, n (%) in 2015					
	≤ 7%/ (53 mmol/mol)	2 (8.00)	2 (5.71)	0.122	.726
	> 7%/ (53 mmol/mol)	23 (92.00)	33 (94.29)		
FBG (mmol/l)					
	baseline	7.64 ± 0.38	8.09 ± 0.32	5.083	.000*
	year 15	7.89 ± 0.42	8.26 ± 0.27	4.159	.000*
2 hPBG (mmol/l)					
	baseline	11.76 ± 0.70	12.32 ± 0.57	3.438	.001*
	year 15	12.18 ± 0.74	12.68 ± 0.61	2.853	.006*
DKD, n (%) in the 2015					
	DKD	6 (24.00)	23 (65.71)	10.162	.001*
	NDKD	19 (76.00)	12 (34.29)		
DR, n (%) in the 2015					
	DR	10 (40.00)	24 (68.57)	4.848	.012*
	NDR	15 (60.00)	11 (31.43)		
PDR, n (%) in the 2015					
	PDR	2 (20.00)	9 (37.50)	0.988	.320
	NPDR	8 (80.00)	15 (62.50)		
Mean RI in interlobular renal arteries					
	baseline	0.67 ± 0.03	0.75 ± 0.02	12.741	.000*
	year 15	0.71 ± 0.03	0.80 ± 0.03	11.644	.000*
Mean RI in CRA					
	baseline	0.67 ± 0.03	0.75 ± 0.02	13.345	.000*
	year 15	0.72 ± 0.03	0.79 ± 0.03	11.460	.000*
AER					
	baseline	16.88 ± 1.48	19.97 ± 1.20	8.919	.000*
	year 15	24.84 ± 7.06	35.00 ± 6.85	5.591	.000*
UACR					
	baseline	16.04 ± 1.54	18.74 ± 1.24	7.507	.000*
	year 15	27.16 ± 5.68	34.46 ± 6.02	4.741	.000*
Cr					
	baseline	74.16 ± 5.47	75.34 ± 4.19	0.949	.347
	year 15	87.72 ± 7.46	92.26 ± 5.20	2.777	.007*
BUN					
	baseline	5.49 ± 0.59	5.90 ± 0.35	3.313	.002*
	year 15	6.43 ± 0.49	6.78 ± 0.44	2.911	.005*

* P < .05, the difference between the 2 groups had statistical significance.

AER = albumin excrete rate, BMI = body mass index, BUN = blood urea nitrogen, Cr = creatinine, DKD = diabetes kidney disease, DR = diabetic retinopathy, NDKD = no diabetes kidney disease, NDR = no diabetic retinopathy, NPDR = nonproliferative diabetic retinopathy, PDR = proliferative diabetic retinopathy, UACR = urinary albumin/creatinine ratio.

The exclusion criteria were

1. type 1 diabetes mellitus (T1DM), pregnancy, and lactation;
2. renal inadequacy, a serum Cr level >132 μmol/L for males or >123 μmol/L for females, AER >30 mg/24 hours or >20 mg/minute;
3. hyperhepatic, in which the liver enzyme levels are 2-fold higher than normal;
4. patients in intensive care receiving insulin treatment;
5. patients with both DR and hypertension (antihypertensive drugs were used);
6. patients with other eye conditions that may interfere with blood flow, such as maculopathy of any origin, glaucoma, high degree myopia, or patients with a history of laser treatment or intraocular surgery;
7. patients with both DKD and hypertension (antihypertensive drugs were used);
8. patients with other renal conditions that may interfere with blood flow, such as urolithiasis, urinary infections, and renal cysts (diameter >3 cm);
9. New York Heart Association class III or IV heart failure, a history of coronary angioplasty, coronary bypass surgery, coronary stent placement, or myocardial infarction within 6 months;
10. patients who use cholesterol-lowering drugs with inadequate control of the blood lipid levels (TC >250 mg/dl, HDL-C <30 mg/dl, LDL-C >170 mg/dl, and TG >200 mg/dl); and
11. patients who used systemically injected glucocorticoids within 3 months prior to our study.

This trial was conducted in accordance with the guidelines of the Declaration of Helsinki. The clinical research was approved by the Medical Ethics Committee (Number ICE20160204) at Shenyang the Fourth Hospital of People. The study protocol was reviewed by an independent ethics committee or institutional review board at every research site. Each patient and their family members provided written informed consent. Our research has been reviewed and registered on ClinicalTrials.gov (NCT02805543).

Table 2
The change of RI in the retrobulbar blood vessels from baseline to year 15.

Content	Group (mean RI in interlobular renal arteries)		<i>t/χ²</i> value	<i>P</i> value	
	≤ 0.7	> 0.7			
NO. (<i>n</i>)	23	37	-	-	
Sex, <i>n</i> (%)					
	Male	12 (52.17)	20 (54.05)	0.031	.861
	Female	11 (47.83)	17 (45.95)		
Age (years) in 2000	52.13 ± 5.93	52.68 ± 5.54	0.361	.719	
BMI (kg/m ²) in 2000	27.43 ± 1.41	27.32 ± 1.60	0.272	.787	
Diabetes duration (years) in 2000	7.65 ± 2.21	7.27 ± 1.76	0.741	.462	
HbA1c in 2000					
	%	7.22 ± 0.34	7.76 ± 0.46	4.852	.000*
	mmol/mol	51.60 ± 3.68	57.51 ± 5.06	4.852	.000*
HbA1c in 2015					
	%	7.45 ± 0.37	8.02 ± 0.51	4.698	.000*
	mmol/mol	54.07 ± 4.06	60.38 ± 5.57	4.698	.000*
Baseline HbA1c, <i>n</i> (%) in 2000	≤ 7%/ (53 mmol/mol)	5 (21.74)	4 (10.81)	1.329	.249
	> 7%/ (53 mmol/mol)	18 (78.26)	33 (89.19)		
Baseline HbA1c, <i>n</i> (%) in 2015	≤ 7%/ (53 mmol/mol)	2 (8.70)	2 (5.41)	0.247	.619
	> 7%/ (53 mmol/mol)	21 (91.30)	35 (94.59)		
FBG (mmol/l)					
	baseline	7.62 ± 0.36	8.08 ± 0.34	4.957	.000*
	year 15	7.86 ± 0.40	8.26 ± 0.28	4.539	.000*
2 hPBG (mmol/l)					
	baseline	11.73 ± 0.72	12.30 ± 0.56	3.462	.001*
	year 15	12.15 ± 0.76	12.67 ± 0.60	2.971	.004*
DKD, <i>n</i> (%) in the 2015	DKD	6 (26.09)	23 (62.16)	7.392	.007*
	NDKD	17 (73.91)	14 (37.84)		
DR, <i>n</i> (%) in the 2015	DR	9 (39.13)	25 (67.57)	4.671	.031*
	NDR	14 (60.87)	12 (32.43)		
PDR, <i>n</i> (%) in the 2015	PDR	1 (12.50)	9 (36.00)	1.585	.208
	NPDR	7 (87.50)	16 (64.00)		
Mean RI in interlobular renal arteries					
	baseline	0.67 ± 0.02	0.75 ± 0.02	14.405	.000*
	year 15	0.71 ± 0.03	0.79 ± 0.03	10.820	.000*
Mean RI in CRA					
	baseline	0.67 ± 0.03	0.74 ± 0.02	11.650	.000*
	year 15	0.71 ± 0.03	0.79 ± 0.03	9.862	.000*
AER					
	baseline	16.96 ± 1.49	19.76 ± 1.50	7.050	.000*
	year 15	25.17 ± 7.27	34.24 ± 7.40	4.648	.000*
UACR					
	baseline	16.09 ± 1.59	18.57 ± 1.42	6.266	.000*
	year 15	27.26 ± 5.92	34.00 ± 6.16	4.182	.000*
Cr					
	baseline	74.43 ± 5.59	75.11 ± 4.22	0.639	.598
	year 15	87.70 ± 7.58	92.03 ± 5.34	2.595	.012*
BUN					
	baseline	5.53 ± 0.60	5.85 ± 0.40	2.498	.015*
	year 15	6.48 ± 0.48	6.73 ± 0.48	1.998	.048*

* *P* < .05, the difference between the 2 groups had statistical significance.

AER = albumin excrete rate, BMI = body mass index, BUN = blood urea nitrogen, Cr = creatinine, DKD = diabetes kidney disease, DR = diabetic retinopathy, NDKD = no diabetes kidney disease, NDR = no diabetic retinopathy, NPDR = nonproliferative diabetic retinopathy, PDR = proliferative diabetic retinopathy, UACR = urinary albumin/creatinine ratio.

2.1. Major equipment

A Logiq 400 MD Pro Series scanner (GE Medical Systems, Milwaukee, WI) with a 3.5-4-MHz vector array transducer, and a Powervision SSA-380A (Toshiba, Tokyo, Japan) with a 7-MHz transducer were used for the CDI recordings.

2.2. Study assessments and endpoints

Blood specimen collection and laboratory tests: Venous blood was extracted between 6–8 AM following a fast of ≥8 hours and was used to measure the levels of FPG, HbA1c; the glucose oxidase method was used to determine plasma glucose levels. For the OGTT, subjects were orally administered 75 g of glucose (a 50% anhydrous glucose solution in 150 ml of warm water in 7.5 bottles). Venous blood was collected to measure the 2hPBG; 5 ml of venous blood were collected in a glass tube and incubated for at least 10 minutes, centrifuged (3000 r/minute) for 10 minutes, and then the serum was separated, which was stored in a -70°C cryogenic refrigerator. BUN, and Cr levels were measured

according to the manufacturer’s instructions. All specimens were measured within 1 week of collection.

Urine sample collection and laboratory tests: Urine parameters were quantified using a single 24-hour urine sample. We measured the urinary albumin concentration using a double antibody radioimmunoassay with a sensitivity of 0.5 mg/L, an intra-assay coefficient of variation of 4.5%, and an inter-assay coefficient of variation of 5.3% in the range of 10 to 50 mg/L. None of the patients with diabetes exhibited persistent micro-albuminuria (defined as an AER greater than 20 µg/minute in 2 out of 3 overnight urine collections within 6 months).

Measuring BP: Systolic blood pressure (SBP) and diastolic blood pressure (DBP) were measured using an electronic sphygmomanometer. Each subject’s BP was measured in a sitting position by placing an automatic device at the brachial artery after a 5-minute resting period.

Hemodynamic parameters of retrobulbar and intrarenal (PSV, EDV, and RI) in bilateral retrobulbar blood vessels (central retinal artery (CRA)) and interlobular renal arteries: The parameters were detected using CDI, bilateral CRA and

interlobular renal arteries were examined in the present study. Patients were measured in a sitting position, as described in a previous study.^[9,10] The measurements were conducted in patients who had fasted for at least 8 hours after documenting the BP and pulse rate. The examinations were initiated 15 minutes after the patients rested in the supine position, as described previously. The PSV and peak ESV were recorded in centimeters per second. Subsequently, we calculated the RI [(PSV-EDV)/PSV] for each vessel measured using a computer. These procedures were performed by the same ophthalmologist. For each recording, the RI was exclusively measured when at least 3 consecutive waveforms with a similar appearance were recorded. The mean value of 3 measurements was obtained for each patient. All examinations were performed twice by the same operator who had no knowledge of all the patients.

2.3. Statistical analysis

Data are presented as the means \pm standard deviations (SD). The statistical analysis was conducted using the SPSS statistical package (Version 17.0, SPSS Inc. Chicago, IL, USA). Differences in categorical variables between the 2 groups were evaluated using the Chi-Squared test; differences in continuous variables between the 2 groups were evaluated using the independent-samples *t* test; before and after treatment within-group differences in continuous variables were assessed using the paired-sample *t* test. $P < .05$ was considered statistically significant.

3. Results

First part of longitudinal follow-up studies of the retrobulbar hemodynamics of the 2 subject groups categorized by their RIs, significantly better renal function parameters (BUN and Cr), AER, UACR, and blood glucose (FBG, 2hPBG, and HbA1c) were observed in subjects with RIs ≤ 0.7 in 2000 and 2015. The incidence of HbA1c $\leq 7\%$ in the group with RIs ≤ 0.7 (6/25, 24.00%) was higher than that in the group with RIs > 0.7 (3/35, 8.57%), but the difference was not statistically significant ($P > .05$) in 2000; the incidence of HbA1c $\leq 7\%$ in the group with RIs ≤ 0.7 (2/25, 8.00%) was higher than that in the group with RIs > 0.7 (2/35, 5.71%), but the difference was not statistically significant ($P > .05$) in 2015. The incidence of DKD in the group with RIs ≤ 0.7 (6/25, 24.00%) was significantly lower than that in the group with RIs > 0.7 (23/35, 65.71%) ($P < .05$). The incidence of DR was significantly lower in the group with RIs ≤ 0.7 (10/25, 40.00%) than in the group with RIs > 0.7 (24/35, 68.57%) ($P < .05$). The incidence of proliferative diabetic retinopathy (PDR) was significantly lower in the group with RIs ≤ 0.7 (2/20, 20.00%) than in the group with RIs > 0.7 (9/24, 37.50%), but the difference was not statistically significant ($P > .05$) (Table 1).

In the second longitudinal follow-up study of the interlobular renal hemodynamics of the 2 subject groups categorized by their RIs, significantly better renal function parameters (BUN and Cr), AER, UACR, and blood glucose (FBG, 2hPBG, and HbA1c) were observed in subjects with RIs ≤ 0.7 in 2000 and 2015. The incidence of HbA1c $\leq 7\%$ in the group with RIs ≤ 0.7 (5/23, 21.74%) was higher than that in the group with RIs > 0.7 (4/37, 10.81%), but the difference was not statistically significant ($P > .05$) in 2000; the incidence of HbA1c $\leq 7\%$ in the group with RIs ≤ 0.7 (2/25, 8.70%) was higher than that in the group with RIs > 0.7 (2/37, 5.41%), but the difference was not statistically

significant ($P > .05$) in 2015. The incidence of DKD in the group with RIs ≤ 0.7 (6/23, 26.09%) was significantly lower than that in the group with RIs > 0.7 (23/37, 62.16%) ($P < .05$). The incidence of DR was significantly lower in the group with RIs ≤ 0.7 (9/23, 39.13%) than in the group with RIs > 0.7 (25/37, 67.57%) ($P < .05$). The incidence of proliferative diabetic retinopathy (PDR) was significantly lower in the group with RIs ≤ 0.7 (1/8, 12.50%) than in the group with RIs > 0.7 (9/25, 36.00%), but the difference was not statistically significant ($P > .05$) (Table 2).

4. Discussion

Based on the results of the present study, RIs of retrobulbar and intrarenal may serve as a good hemodynamic predictors of the changes in retrobulbar and intrarenal in addition to GFR and it is considered a better predictor than UACR and AER in T2DM adult patients during the preclinical stage of DKD and DR. Strategies is to regulate glycemia which not only improve intrarenal hemodynamics but also improve retrobulbar hemodynamics.

HbA1c is an indicator of diabetes control. The higher the HbA1c level, the poorer the diabetic control and the higher the circulating glucose concentration. If the circulating glucose concentration is consistently elevated for 3 months (as measured by HbA1c levels), an increase in the nonenzymatic glycosylation of tissue proteins may occur.^[11] Our results are consistent with the results of other studies, although the current study enrolled patients with HbA1c levels as low as 7.0% (53 mmol/mol), whereas patients with baseline HbA1c levels $< 7.5\%$ (58 mmol/mol) were excluded in other studies.^[11] The difference is significant because of the seemingly greater efficacy of antidiabetic agents in patients with higher baseline HbA1c levels.^[11] A major factor contributing to poor compliance in maintaining HbA1c levels $< 7\%$ (53 mmol/mol) is a lack of patient awareness.^[12] As shown in the study by Prabhu et al,^[12] only 23 patients (11.5%) were aware of their HbA1c levels, 10 patients (5%) misinterpreted the HbA1c levels, and approximately 164 patients (82%) were not aware of the significance of or the terminology for HbA1c. The proportion of patients with DR who achieved the target level of $< 7\%$ (53 mmol/mol) remained low. In our study, the proportion of patients (11/90; 12.22%) reaching this standard HbA1c level [$< 7.0\%$ (53 mmol/mol)] was also low; most patients did not adequately control their glycemia. Thus, the mean FPG (8.00 mmol/L) and 2hPG (11.58 mmol/L) levels were higher than the target levels (7 mmol/L and 10 mmol/L, respectively) in the diabetes group.

Renal Doppler RI is widely applied to evaluate blood flow in patients with renal parenchymal diseases. RI is generally accepted as a measure to quantify the changes in renal blood flow caused by renal disease. Thus, intrarenal vascular resistance is reflected and remarkably linked to vascular compliance (i.e., the rate of change in the volume of a vessel as a function of pressure).^[5] Additionally, the application of the captopril test to renal Doppler sonography in screening studies is a noninvasive and inexpensive tool for diagnosing renovascular hypertension.^[13] Therefore, we evaluated intrarenal hemodynamics by examining the RI using Doppler sonography.

DKD is typically responsible for end-stage renal disease (ESRD) in developed nations, which is thought to be attributable to interactions between metabolic and hemodynamic factors. Specific, metabolically driven, glucose-dependent pathways are

activated within the renal tissues of patients with diabetes. These pathways trigger oxidative stress, hexosamine flux, polyol pathway flux, and the accumulation of advanced glycation endproducts (AGEs). Hemodynamic factors are also involved in the pathogenesis of DKD; the pathophysiological mechanism of DKD poses a substantial challenge to medical researchers.^[13]

Despite the unclear nature of the exact pathogenesis of the disease, several theories have been proposed regarding the processes affecting hemodynamics in DKD. Researchers have focused on the hemodynamic changes in the diabetic kidney. Activation of the renin-angiotensin system (RAS) has been reported to induce abnormalities in intrarenal hemodynamics in patients with diabetes.^[14] According to Taniwaki and colleagues, the intrarenal RAS might be activated in patients with diabetes; however, the activation might be impacted by poor glycemic control. In addition, blockade of RAS activation with captopril might reduce intrarenal vascular resistance in patients with diabetes.^[15]

Elevated RI has been reported to be associated with vascular-interstitial disease, including DKD (with the exception of glomerulopathies), which might be caused by decreased tissue and vascular compliance, as well as by increased vascular resistance.^[5] However, early stages of DN are related to an increased GFR and variable increases in renal plasma flow and filtration fraction in both clinical and experimental settings. Diabetic hyperperfusion and hyperfiltration at the single-nephron level are characterized by a disproportionate decrease in afferent arteriolar resistance, leading to an elevation of the glomerular capillary pressure. These features may also be reflected by an increase in RI.^[16] Duplex Doppler sonography is a noninvasive and inexpensive tool used to measure the RI and identify hemodynamic abnormalities in patients with DKD during the preclinical stage.^[17] Based on the results of biopsy studies using children, basement membrane thickening, and mesangial expansion in the kidney develop prior to the onset of microalbuminuria.^[18] Evidently, Doppler sonography does not replace renal biopsy, but it provides a readily applicable and noninvasive tool to investigate renal hemodynamics. In addition, Doppler sonography is also a credible approach to provide both morphological and physiological data when studying renal blood flow in children.^[16] To our knowledge, our study is the first to examine the early changes in intrarenal hemodynamics in adults with T2DM without any evidence of renal dysfunction. Although the renal function was normal in patients with diabetes; therefore, RI can be used to predict changes in renal function in the preclinical stage of DKD. Our results are similar to the results reported by Pelliccia P and colleagues, who selected children as study subjects.^[16]

Nevertheless, a general agreement regarding the significance and the predictive value of renal RI in patients with DKD has not been achieved. Several published studies have examined the application of Doppler sonography in evaluating abnormalities in intrarenal hemodynamics in adults with DKD,^[5,19] but studies of adults during the preclinical stage of DKD (with normal renal function) are still lacking. In our study, we aimed to explore whether Doppler sonography could be applied to detect alterations in renal RI in diabetic adults who had normal renal function in laboratory tests. Although all RI values were less than or equal to 0.70 for both kidneys, the widely recognized boundary value is applied to healthy adults and children older than 6 years (rather than younger children).^[20]

The pathophysiological mechanism of DR poses a substantial challenge in medical science. Although the precise pathogenesis of the disease has not been described, several theories have been posed to describe the processes that impact ocular blood flow in DR. Several researchers have focused more on the hemodynamic changes in the diabetic retina and choroid. Ocular blood flow is measured as blood flow velocity multiplied by the cross-sectional area, as measured in other vascular beds. In terms of abnormalities in ocular blood flow at various stages of DR, some notable deviations were documented in different reports. However, the precise nature of the deviations remains somewhat controversial, presumably due to various techniques in measuring retinal blood flow.^[21,22] Few research studies have assessed hemodynamic changes in patients without DR; thus, we studied the hemodynamic changes in T2DM patients without DR. Inhibiting progression to DR is the goal of the treatment period. DR is one of the main factors contributing to preventable blindness in developed nations and is becoming the primary factor responsible for blindness in middle-income nations.^[23] Up to 39% of patients who receive a first diagnosis of T2DM are estimated to present with signs of DR.^[24] Population-based diabetic eye screening programs have been initiated in several western European countries. The incidence and prevalence of blindness is reduced in patients with diabetes who have access to screening programs compared with populations in which organized, population-based screening has not been implemented.^[25] However, population-based diabetic eye screening programs cannot be widely implemented in China because of the limited healthcare conditions compared to those in western European countries. The frequency of DR, a well-known microvascular complication of DM, increases with age and disease duration. An overwhelming majority of T1DM patients and over 60% of T2DM patients will develop DR within 20 years.^[26] Because of the exclusion of DR and DKD, we chose patients with a diabetes duration ranging from 3 to 12 years (<15 years). CDI is used to evaluate circulatory parameters of retrolubar blood vessels. Scanning laser Doppler flowmetry (SLDF), a useful method for evaluating blood flow in retinal tissues, has been widely applied in ophthalmic pathology for many years.^[27,28]

Therefore, in our longitudinal follow-up study, the quantum was 15 years (from 2000 to 2015). As shown in the study by Dimitrova G, the results of a retinal tissue evaluation using SLDF were positively correlated with the retrolubar circulatory parameters of CDI in diabetes patients without DR.^[29] Dimitrova G also reported significantly increased PSV, EDV, and RI in CRV, due to the progression of retinopathy during a 21-month period, but no significant alterations in the circulatory parameters in the CRA or PCA were recorded after the progression of DR.^[30] In our first part of follow-up study, the retrolubar hemodynamics of the 2 subject groups categorized by their RIs indicated that subjects with RIs ≤ 0.7 showed significantly improved renal function parameters (BUN and Cr levels), AER, UACR, and blood glucose (FBG, 2hPBG, and HbA1c) in 2000 and 2015. The incidence of DKD was significantly lower in subject group RIs ≤ 0.7 (6/25, 24.00%) than group RIs > 0.7 (23/45, 65.71%) ($P < .05$). The incidence of DR was significantly lower in group RIs ≤ 0.7 (10/25, 40.00%) than group RIs > 0.7 (24/35, 68.57%) ($P < .05$). The incidence of PDR was significantly lower in group RIs ≤ 0.7 (2/20, 20.00%) than group RIs > 0.7 (9/24, 37.50%), but the difference was not significant ($P > .05$) (Table 1). In our second longitudinal follow-up study of the interlobular renal

hemodynamics of the 2 subject groups categorized by their RIs, significantly better renal function parameters (BUN and Cr), AER, UACR, and blood glucose (FBG, 2hPBG, and HbA1c) were observed in subjects with RIs ≤ 0.7 in 2000 and 2015. The incidence of HbA1c $\leq 7\%$ in subject group RIs ≤ 0.7 (5/23, 21.74%) was higher than group RIs > 0.7 (4/37, 10.81%), but the difference was not statistically significant ($P > .05$) in 2000; the incidence of HbA1c $\leq 7\%$ in group RIs ≤ 0.7 (2/25, 8.70%) was higher than group RIs > 0.7 (2/37, 5.41%), but the difference was not statistically significant ($P > .05$) in 2015. The incidence of DKD in the group with RIs ≤ 0.7 (6/23, 26.09%) was significantly lower than that in the group with RIs > 0.7 (23/37, 62.16%) ($P < .05$). The incidence of DR was significantly lower in the group with RIs ≤ 0.7 (9/23, 39.13%) than in the group with RIs > 0.7 (25/37, 67.57%) ($P < .05$). The incidence of proliferative diabetic retinopathy (PDR) was significantly lower in the group with RIs ≤ 0.7 (1/8, 12.50%) than in the group with RIs > 0.7 (9/25, 36.00%), but the difference was not statistically significant ($P > .05$) (Table 2). The RIs increased gradually and significantly over period of 15 years, similar to the results reported in the 10-year follow-up study by Neudorfer.^[31]

As shown in the study by Hirose A et al,^[32] HbA1c levels may better predict retinopathy if metabolic memory-free data are included. In addition, some studies have reported a correlation between the severity of retinopathy and the extent of decreased blood flow. Hyperglycemia enhances blood flow and interferes with retinal autoregulation. In addition, retinal blood flow is regulated by changes in ocular vascular resistance and the RI was notably higher in the diabetes than in the healthy people;^[33,34] therefore, RI plays an important role in assessing hemodynamic changes in DR patients.

In summary, the RI can be used to evaluate the hemodynamic changes of retrobulbar and intrarenal in DKD/DR and its progression and it can also predict hemodynamics changes in retrobulbar and kidney during the preclinical stage of DR and DKD, similar to the HbA1c levels and diabetes duration.

In our future study, several concerns shall be addressed as follow. First, we could not observe the morphological changes in retinal and renal tissues and did not identify the protein that induced damage to these tissues. Because not all patients agree to undergo a biopsy, we will adopt an animal model to study the pathology. Second, we did not assess the long-term changes, and we only examined the index in the early period (without DKD and DR). Third, we studied patients with T2DM during the preclinical stage and did not study them at different stages of DKD and DR. Moreover, retrobulbar, and intrarenal IR may be used as the as good predictors of ophthalmic diseases and nephropathy, these studies will be carried out in the future.

In conclusion, this study is anticipated as the first to use the RI to predict the progression of DKD and DR. For patient's lack of proper control of glycemia, RI of retrobulbar and intrarenal can predict the degree of damages to hemodynamic during the treatment which include patients not presented with DKD and DR. Therefore, retinal and kidney function should be monitored by both doctors and patients during the preclinical stage of diabetes.

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