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Diabetes mellitus and renal disease: when to perform a renal biopsy?

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ABSTRACT

Background: Several studies suggest that non-diabetic renal disease (NDRD) is common in patients with diabetes mellitus. The aim of this analysis of renal biopsies in diabetic patients was (a) to assess the prevalence and type of NDRD and (b) to identify its clinical and laboratory predictors.

Methods: This retrospective study analysed clinical and laboratory data and biopsy findings in diabetic patients observed by a single pathologist over the past 25 years. Based on biopsy findings, patients were categorised as (i) isolated diabetic nephropathy, (ii) isolated NDRD and (iii) NDRD superimposed on diabetic nephropathy.

Results: Of the 236 patients studied, 60% were male and the mean age was 56.3 (±14.2) years. Of these, 91% had known diabetes mellitus at the time of biopsy (13% type 1 and 87% type 2). Isolated diabetic nephropathy was found in 125 (53%), isolated NDRD in 89 (38%) and NDRD superimposed on diabetic nephropathy in 22 (9%) patients.

The main indication for biopsy in the three groups was nephrotic proteinuria. Patients with isolated NDRD and NDRD superimposed on diabetic nephropathy presented acute deterioration of renal function more frequently ($p<0.001$) and had more microhaematuria ($p<0.001$) as indications for renal biopsy. Focal segmental glomerulosclerosis and membranous nephropathy were the most frequent diagnoses in patients with NDRD.

Patients with isolated diabetic nephropathy were younger ($p=0.02$), presented a longer duration of diabetes mellitus ($p<0.001$) and had more frequent retinopathy ($p<0.001$). The prevalence of microhaematuria was higher in patients with isolated or superimposed NDRD ($p=0.01$).

Conclusion: The prevalence of NDRD (either isolated or superimposed on diabetes mellitus) is remarkably frequent in diabetic patients in whom nephrologists consider renal biopsy an appropriate measure. Predictors of NDRD were older age, shorter duration of diabetes mellitus, absence of retinopathy and presence of microhaematuria.

Key-Words:
Diabetes mellitus; diabetic nephropathy; non-diabetic renal disease; renal biopsy.

INTRODUCTION

Diabetic nephropathy (DN) is a major cause of end-stage renal disease[1,2]. The diagnosis of DN is almost always based on clinical and laboratory data and few studies have analysed the prevalence of non-diabetic renal disease (NDRD) in diabetic patients.

Persistent and slow progressive proteinuria is the most characteristic feature of DN and diabetic renal failure[3,4]. When a patient with type 1 diabetes mellitus (DM) for more than 10 years with either diabetic retinopathy or neuropathy develops renal impairment, the probability of having histologically proven DN is more than 95%[5]. Studies based on renal biopsies from type 2 DM have shown that the risk of NDRD is much higher. Other glomerular diseases, superimposed on DN or occurring as isolated NDRD, comprise up to 27-79% in type 2 DM patients and have important implications for both therapy and prognosis[6-11].

Different predicting factors have been identified in diabetic patients found to have NDRD, including late onset of DM, absence of neuropathy, absence of retinopathy and presence of other systemic diseases[12]. However, these factors were found to have variable predictive values in different series with only the absence of retinopathy and autonomic neuropathy found to be useful clinical markers[13]. Overall, it remains unclear whether biopsy should be offered to all such patients, or reserved for patients with atypical features associated with proteinuria, i.e. sudden onset, haematuria, acute renal insufficiency, extra-renal manifestations, absence of retinopathy and short duration of diabetes.

The aim of this analysis of renal biopsies in DM patients was (a) to assess the prevalence and nature of NDRD and (b) to identify its clinical and laboratory predictors.

PATIENTS AND METHODS

Study design

This was a retrospective study which analysed the clinical and laboratory data of diabetic patients and their renal biopsies observed in our renal morphology unit over the last 25 years.

Population

The study included 236 patients. Mean age was 56.3 (±14.2) years and 60% of the patients were male. Two hundred and fourteen (91%) had known DM at the time of renal biopsy. Of those with known DM, twenty-eight (13%) had type 1 DM and one hundred and eighty-four (87%) type 2 DM. Mean DM time when renal biopsy was performed was 8.1±7.1 years (range 0 to 39 years).

Methods

Renal biopsies were all observed by a single nephropathologist. Tissue samples were routinely processed by light microscopy and immunofluorescence. Electron microscopy was performed only when needed to reach diagnosis. For light microscopy, tissue sections were stained with hematoxylin-eosin, Schiff's periodic acid, Masson trichrome, and methenamine silver. Immunofluorescence examinations with antisera against immunoglobulin G (IgG), IgM, IgA, complement factor 3 (C3), C4, C1q, fibrinogen and albumin were performed. In particular cases, antisera against serum amyloid AA, light chain κ and λ and P component were used.

DN was diagnosed based on the presence of the following diabetic lesions: glomerulosclerosis either of the nodular (Kimmelstiel-Wilson) (Fig. 1) or diffuse (mesangial expansion) type (Fig. 2), hyalinisation of the renal arterioles, exsudative lesions such as

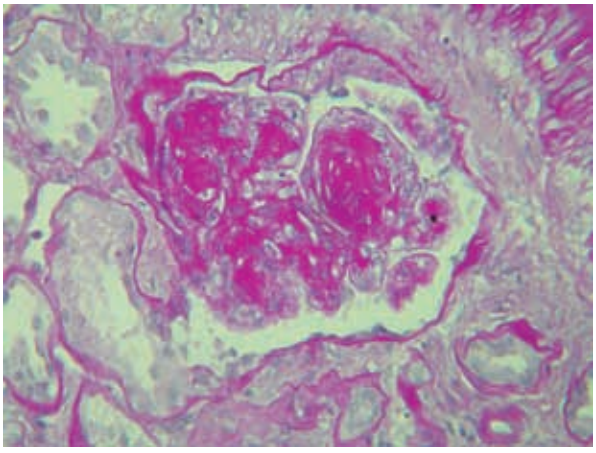

Figure 1

Diabetic nephropathy - nodular type (PAS × 400).

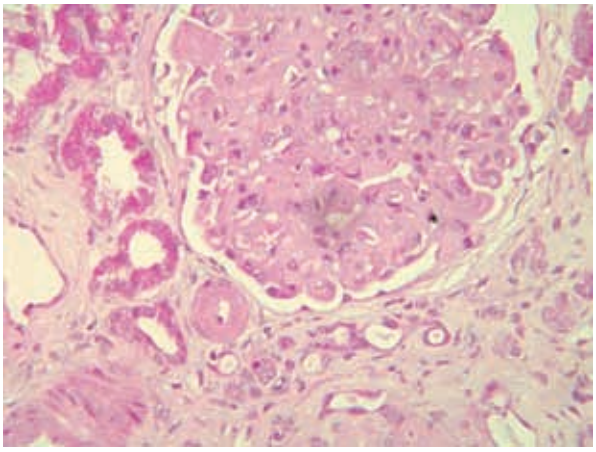

Figure 2

Diabetic nephropathy - diffuse type (HE × 200).

"fibrin cap", "capsular drop" or "hialine trombus", uniform glomerular capillary basement membrane thickening, and linear deposits of IgG and albumin in glomerular and tubular basement membrane on immunofluorescence studies[14]. NDRD was categorised following orthodox pathological criteria[15].

The patients were divided into the following three groups according to histological diagnosis: (i) the DN group (125 patients) which had isolated diabetic nephropathic lesions, (ii) the non-DN group (89 patients) which had only NDRD (with absence of diabetic nephropathic lesions) and (iii) the complicated group (22 patients) which had renal histopathological changes of NDRD superimposed on those of DN.

Clinical data included age, gender, duration of diabetes, presence of diabetic retinopathy and hypertension. Indications for renal biopsy were also recorded. Laboratory data included serum creatinine, urinary protein excretion in 24-hours and microscopic haematuria in urinary sediment.

Statistical analysis

Data were expressed as mean±SD. Differences between groups were assessed using the $\chi 2$ test for categorical variables and one-way analysis of variance (ANOVA) for continuous variables. Statistical analysis was performed with SPSS system 14.0 (SPSS Inc., Chicago, IL). For all comparisons, a $p<0.05$ was considered statistically significant.

Results

Demographic characteristics of the total 236 patients included in the study are listed in Table I. Two hundred and fourteen (91%) had known DM at the time of renal biopsy. Over the course of this review, 22 (9%) patients did not have a clinical diagnosis of DM and were categorised as diabetics because their renal biopsies showed characteristic features of DN. Of those with previously known DM, twenty-eight (13%) had type 1 DM and one hundred and eighty-four (87%) type 2. Comparing these three groups, we found that patients with type 1 DM were significantly younger ($p<0.001$) and had both less hypertension ($p=0.01$) and less microscopic haematuria ($p=0.01$). Patients with type 1 DM also had a longer duration of DM ($p<0.001$) and a higher incidence of diabetic retinopathy ($p<0.001$). Twenty-five (89%) patients with type 1 DM and 93 (52%) of those with type 2 had morphological features of DN (Table II).

The DN group consisted of 125 (53%) patients, the isolated NDRD group of 89 (38%) patients and the NDRD superimposed on DN group of 22 (9%) patients.

The main indication for renal biopsy in the three groups was nephrotic proteinuria, although the percentage was significantly higher in DN patients than in the other two groups ($p=0.03$). Patients with isolated NDRD or NDRD superimposed on DN presented more acute renal failure (ARF) or rapidly progressive renal failure (RPRF) ($p<0.001$) and more microscopic haematuria ($p<0.001$) than DN patients, as a cause for renal biopsy. Non-nephrotic proteinuria as an indication for renal biopsy was similar in all groups (Table III).

Table I

Demographic characteristics of the total group

	Total group (n=236)
Age at biopsy (years)	56.3±14.2 (14-89)
Gender (M/F)	151/85
Duration of diabetes (years)	8.1±7.1 (0-39)
Diabetic retinopathy (%)	28%
Hypertension (TA< 140/90 mmHg) (%)	62%
Serum creatinine (mg/dL)	3.1±2.3 (0.6-13)
Proteinuria (g/24 h)	6.4±5.8 (0-32)
Microhaematuria (%)	47%

Table II

Clinical and laboratory data of type 1 DM, type 2 DM and "not known to be diabetic" patients

	Type 1 DM (n=28)	Type 2 DM (n=178)	Not known to be diabetic (n=22)	P*
Age at biopsy (years)	34.5±13.3 (14-69)	59.6±10.9 (36-84)	57.5±14.6 (33-89)	‹0.001
Gender (M/F)	13/15	107/71	18/4	NS
Duration of diabetes (years)	14.2±8.3 (1-39)	7.0±6.3 (0-30)	NA	‹0.001
Diabetic retinopathy (%)	72%	24%	NA	‹0.001
Hypertension (TA› 140/90 mmHg) (%)	36%	68%	57%	0.01
Serum creatinine (mg/dL)	2.9±1.3 (1.2-7)	3.1±2.5 (0.6-13)	3.3±2.7 (0.7-11)	NS
Proteinuria (g/24 h)	5.9±5.5 (0-20)	6.4±5.8 (0-32)	7.6±6.6 (1-25)	NS
Microhaematuria (%)	28%	51%	61%	0.02
Diabetic nephropathy on renal biopsy (%)	89%	52%	100%	‹0.001

In type 1 diabetics, 4 (11%) patients presented either isolated NDRD (1 had minimal change disease and 2 had hypertensive nephroangiosclerosis) or NDRD superimposed on DN (1 patient with acute interstitial nephritis and DN).

In type 2 diabetics, 107 (48%) patients presented either isolated NDRD or NDRD superimposed on DN. Focal segmental glomerulosclerosis (FSGS) and membranous nephropathy were the most frequent diagnoses in both groups, accounting for more than 30 and 50% of the renal biopsy diagnosis in isolated NDRD or NDRD superimposed on DN respectively. Renal disease entities identified in type 2 DM patients are listed in Table IV.

In DN patients, 71 (59%) had DN of nodular type and the remaining (41%) had diffuse lesions. Serum creatinine levels were higher in patients with nodular type DN (3.9±1.8 *vs.* 2.8±1.6; p‹0.001). Clinical and other laboratory parameters were similar in both groups.

Patients with DN were younger than patients without DN (either with isolated NDRD or NDRD superimposed on DN) (p=0.02); older age seems to be a risk factor for having either an NDRD or a combined disease. Males outnumbered females in all groups. Duration of DM as well as the presence of diabetic retinopathy was significantly lower in patients with isolated NDRD than in the others (p‹0.001). Incidence of hypertension and level of proteinuria were similar in all three groups, while serum level of creatinine was higher in patients with DN (p=0.03). Incidence of microscopic haematuria was significantly higher in patients with either isolated NDRD or NDRD superimposed on DN (p=0.01) (Table V).

DISCUSSION

DN is one of the most frequent and clinically important complications of diabetes, affecting appro-

Table III

Indications for renal biopsy

	Group I (n=125) (%)	Group II (n=89) (%)	Group III (n=22) (%)	P*
Nephrotic proteinuria	95 (76%)	44 (49%)	12 (55%)	0.03
Non-nephrotic proteinuria	17 (14%)	17 (19%)	4 (18%)	NS
ARF / RPRF	11 (9%)	24 (27%)	5 (23%)	‹0.001
Haematuria	2 (1%)	4 (5%)	1 (4%)	‹0.001

ARF (acute renal failure); RPRF (rapidly progressive renal failure)

Group I, diabetic nephropathy; group II, isolated non-diabetic renal disease; group III, non-diabetic renal disease superimposed on diabetic nephropathy

Table IV

Histological diagnoses in type 2 diabetic patients

Histology	Group II (n=86) (%)	Group III (n=21) (%)
Glomerular diseases		
Minimal change	5 (6%)	0
FSGS	14 (16%)	7 (33%)
Membranous	13 (15%)	5 (23%)
IgA nephropathy	3 (4%)	3 (14%)
PIGN	1 (1%)	0
Crescentic GN	6 (7%)	1 (5%)
Mesangioproliferative GN	6 (7%)	0
Endocapilar proliferative	2 (2%)	1 (5%)
Membranoproliferative GN	2 (2%)	1 (5%)
Lupus nephritis	1 (1%)	0
Goodpasture's disease	1 (1%)	0
Vascular diseases		
Chronic ischaemia including HT nephroangiosclerosis	10 (13%)	0
Tubulointerstitial disease		
Acute tubular necrosis	2 (2%)	2 (10%)
Acute interstitial nephritis	1 (1%)	0
Chronic interstitial nephritis	6 (7%)	0
Amyloidosis	9 (11%) (6-AA; 3-AL)	1 (5%) (1-AL)
Light-chain disease	1 (1%)	0
Hereditary diseases		
Fabry's disease	1 (1%)	0
Histologically normal kidney	2 (2%)	0

GN, glomerulonephritis; HT, hypertensive; PIGN, postinfectious glomerulonephritis.
Group II, isolated non-diabetic renal disease.
Group III, non-diabetic renal disease superimposed on diabetic nephropathy.

ximately 40% of patients who have had DM for more than 20 years and contributing to a substantial number of patients beginning dialysis[1,2].

It is generally held that NDRD complicating type 1 DM is comparatively rare, probably around 4% in unselected cases with proteinuria and duration of DM of more than 10 years[16]. Usual criteria for suspecting and carrying out renal biopsy in type 1 DM are microhaematuria, absence of diabetic retinopathy, uncharacteristic change in renal function and presence of other systemic disease or immunological abnormalities[17]. In this study, the incidence of NDRD in this group of patients was higher (11%), probably because these patients were younger and only 72% had diabetic retinopathy. Almost one-third of these patients also presented microscopic haematuria, an uncharacteristic feature of DN.

Among patients with type 2 diabetes with renal biopsy performed, the prevalence of NDRD in the published research varies widely, from 27 to 79%, depending on the selection criteria and the populations being studied[6-11]. In our study, 48% of the type 2 diabetic patients had NDRD (either isolated or superimposed). Patients with NDRD or combined disease were older than patients with DN. Duration of diabetes was smaller in the isolated NDRD group and higher in the superimposed group. Thus, a shorter duration of diabetes was a risk factor for isolated NDRD and a higher age for superimposed or isolated NDRD in our study. A study by Lee *et al.*[18] also concluded that a short duration of diabetes was significantly associated with NDRD, while a study by Bertani *et al.*[19] found no significant difference in the duration of diabetes in different histological classes.

One of the important predictors of NDRD is said to be the absence of retinopathy. This association is stronger in people with type 1 diabetes than in those with type 2 diabetes. This study confirms the

Table V

Clinical and laboratory parameters in the different groups

	Group I (n=125)	Group II (n=89)	Group III (n=22)	*P**
Age at biopsy (years)	53.2±14.6 (15-83)	59.9±12.0 (14-84)	58.4±16.4 (26-89)	0.02
Gender (M/F)	72/53	55/34	14/8	NS
Duration of diabetes (years)	12.7±10.2 (1-30)	5.3±6.2 (0-39)	10.0±6.6 (1-30)	⇐0.001
Diabetic retinopathy (%)	57%	28%	49%	⇐0.001
Hypertension (TA⇒ 140/90 mmHg) (%)	56%	44%	58%	NS
Serum creatinine (mg/dL)	3.6±2.1 (0.7-11)	3.2±2.8 (0.6-13)	2.7±2.4 (0.7-8)	0.03
Proteinuria (g/24 h)	6.6±5.5 (0-32)	6.0±5.8 (0-26)	7.3±7.2 (1-25)	NS
Microhaematuria (%)	36%	61%	54%	0.01

Group I, diabetic nephropathy; Group II, isolated non-diabetic renal disease; Group III, non-diabetic renal disease superimposed on diabetic nephropathy.

accepted view that the absence of retinopathy should raise the possibility of a non-diabetic lesion and hence, the need for a renal biopsy. Retinopathy absence predicted isolated NDRD in 72% of the cases in our study. It also predicted NDRD in 87% of the patients in a study by Jacob *et al.*[20]. Although retinopathy has been strongly correlated with the presence of DN, discordance in the occurrence of the two complications is not uncommon. We found that 43% of patients with DN did not have retinopathy and 28% of diabetic patients with diabetic retinopathy had isolated NDRD. It has been suggested that the two complications show dissimilar genetic predisposition[21,22].

Either the incidence of hypertension or the level of proteinuria was similar in all three groups in our study, while levels of serum creatinine were higher in patients with DN. In a study by Martin *et al.*[23] coexisting renal disease was found to be associated with a significantly higher creatinine, independent of the severity of diabetic glomerulopathy. Studies into the impact of superimposed disease on the natural history of DN are contradictory. Martin *et al.*[23] reported that these lesions and their underlying causes influence the renal function and natural history of renal disease in individuals with diabetes, while Wong *et al.*[24] found the contrary.

Our study showed that the presence of microscopic haematuria predicted the existence of NDRD (either isolated or superimposed). A study by Wong *et al.*[24] also showed that the association of haematuria with the absence of retinopathy constitutes the strongest indication for a non-diabetic lesion. Thus, a combination of indications seems to constitute a more sensitive predictor of NDRD than any one indication alone.

Histologically, NDRD comprised a heterogeneous group of diseases. FSGS and membranous nephropathy were the commonest lesions in type 2 DM patients with isolated NDRD or NDRD superimposed on DN, probably as nephrotic proteinuria was the main reason for biopsy in both groups. In patients with DN, nodular type lesion was the more frequent lesion in our population and, as described in other studies[9-11], those patients presented a higher serum creatinine.

The mechanisms implicated in the development of NDRD in diabetic patients, with or without DN, remain the subject of speculation. The predisposition of DN to a superimposed nephritis is still unknown, but it has been attributed to an enhanced exposure of antigenic cellular components, which would favour the triggering of immune responses. Pre-existing glomerular alterations might favour an immune reaction in the subepithelial space[10,24]. For example, injury and loss of podocytes could occur, which would favour the occurrence of FSGS.

Another finding in our study was that the histological changes of DN may precede other clinical manifestations of DM in a minority of patients, in whom the first diagnosis of diabetes was made based on characteristic histological features of DN present in the renal biopsy. This has been also reported in some small series[6,25].

This study advocates a higher degree of suspicion of NDRD in diabetic patients, particularly in type II

DM, which favour performing a renal biopsy. The prevalence of NDRD (either isolated or superimposed on DN) is high in appropriate clinical settings. Older age, shorter duration of diabetes, absence of retinopathy and the presence of microscopic haematuria strongly predict NDRD.

Conflict of interest statement. None declared.

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