Natural history and risk factors for diabetic kidney disease in patients with T2D: lessons from the AMD-Annals

Francesca Viazzi, MD[1], Giuseppina Russo, MD, PhD[2], Antonio Ceriello MD[3,4], Paola Fioretto MD[5], Carlo Giorda MD[6], Salvatore De Cosmo, MD[7], and Roberto Pontremoli, MD, PhD[1*]

[1]Università degli Studi and IRCCS Policlinico San Martino-IST, Genova, Italy, [2]Department of Clinical and Experimental Medicine, University of Messina, Messina, Italy, [3]Institut d'Investigacions Biomèdiques August Pii Sunyer (IDIBAPS) and Centro de Investigación Biomédicaen Red de Diabetes y Enfermedades Metabólicas Asociadas (CIBERDEM), Barcelona, Spain; [4]U.O. Diabetologia e Malattie Metaboliche, Multimedica IRCCS, Sesto San Giovanni, Milano, Italy; [5]Department of Medicine, University of Padua, Italy and Department of Medical Sciences, [6]Diabetes and Metabolism Unit ASL Turin 5, Chieri, Italy; [7]Scientific Institute "Casa Sollievo della Sofferenza", San Giovanni Rotondo (FG), Italy.

***Corresponding author:** Roberto Pontremoli

roberto.pontremoli@unige.it

Running title: Diabetic Kidney Disease: the AMD Annals initiative

Word count for the abstract: 235

Word count for the text: 4054

Compliance with Ethical Standards

Conflict of interest: The authors declare that they have no conflict of interest.

Informed consent: For this type of study formal consent is not required

Welfare of animals: This article does not contain any studies with animals performed by any of the authors

Acknowledgements: The authors acknowledge the Associazione Medici Diabetologi that has provided financial support for this research.

Abstract

The AMD (Associazione Medici Diabetologi)Annal initiative is an ongoing observational survey promoted by AMD. It is based on a public network of about 700 italian diabetes clinics, run by specialists who provide diagnostic confirmation and prevention and treatment of diabetes and its complications. Over the last several years, the analysis of the AMD Annals dataset contributed several important information on clinical features and their prognostic and therapeutic implications of type 2 diabetes kidney disease. Among these: (a) non-albuminuric renal impairment is the predominant clinical phenotype, although associated to a lower risk of progression as compared to overt albuminuria, it contributes significantly to the burden of end stage renal disease morbidity; (b) optimal blood pressure control provides significant but incomplete renal protection, it reduces albuminuria but there may be a J curve phenomen with eGFR at very low blood pressure values; (c) Hyperuricemia and diabetic hyperlipidemia, namely hypertriglycerides and low HDL cholesterol are strong independent predictor of chronic kidney disease onset in diabetes, although the pathogenetic mechanisms underlying these associations remain uncertain; (d) long term intra individual variability in HbA1c, lipid parameters, uric acid and blood pressure plays a greater role in the appearance and progression of chronic kidney disease than the absolute values of each one of these variables. These data help clarify the natural history of chronic kidney disease in patients with type 2 diabetes and provide important clues to devise future interventional studies.

Key words: T2 diabetes, glomerual filtration rate, albuminuria, chronic kidney disease, hypertension

Introduction

Diabetes mellitus is the leading cause of chronic kidney disease (CKD). In fact, approximately 40% of patients with diabetes develop diabetic kidney disease (DKD) resulting in albuminuria, reduction of glomerular filtration rate (GFR), or both. The typical natural history of diabetic nephropathy has been derived mainly from longstanding epidemiologic studies in patients with type 1 diabetes mellitus (T1D). In these patients, microalbuminuria is the first sign of renal impairment and may progress to macroalbuminuria, which predicts subsequent decline of GFR and, eventually, end stage renal disease (ESRD). The natural history of DKD in type 2 diabetes mellitus (T2D) appears to be more heterogeneous. In fact, several studies have recently shown that a majority of patients with DKD may progress to ESRD despite having modest or no albuminuria [1-3].

Since its early stages, CKD is also associated with an increased risk of death, particularly from cardiovascular disease (CVD). The presence of a mild or moderate renal impairment entails a risk for cardiovascular (CV) complications and death that has been shown to be even greater than progression to ESRD in some [4], although not all studies [5]. Both features of DKD, albuminuria and reduced GFR values have independent prognostic values in terms of CV and renal outcome [6].

It is therefore of paramount importance to detail natural history of different renal phenotypes and their prognostic implications in patients with T2D.

Here, we report a summary of recent studies from the AMD (Associazione Medici Diabetologi) Annals initiative that have provided important information on presenting clinical features and their prognostic and therapeutic implications in a very large cohort of Italian patients with T2D.

The AMD annals initiative

In Italy, diabetes care is mainly provided by a public network of about 700 diabetes clinics, run by specialists who provide diagnostic confirmation and prevention and treatment of diabetes and its complications through close patient follow-up and regular check-ups [7]. The Italian AMD Annals initiative has been devised to analyze the AMD database and identify a set of indicators that can be used in the context of continuous quality improvement. Participating centers adopted the same software system for the everyday management of outpatients, while a specially developed software package allowed for extraction of specific information.

Data from all participating centers were collected and centrally analyzed anonymously. T2D was diagnosed at participating diabetes centers according to the American Diabetes Association 2003 criteria. The biochemical measurements were performed at clinical laboratories at each participating center. This initiative includes measuring and monitoring renal function (creatinine, uric acid and albuminuria), hemoglobin A1c (HbA1c), blood pressure (BP) and lipid profile (LDL cholesterol or total and HDL cholesterol and triglycerides). The use of specific classes of drugs (insulin, oral antidiabetic, statins, antihypertensive agents) was also evaluated. Because normal ranges for HbA1c varied among centers, the percentage change with respect to the upper normal value (measured value/upper normal limit) was estimated and multiplied by 6.0 to allow comparisons among the centers. Kidney function was assessed by serum creatinine and urinary albumin excretion measurements. GFR was estimated (eGFR) for each patient using a standardized serum creatinine assay and the CKD-Epidemiology Collaboration equation [8]. Increased urinary albumin excretion was diagnosed and defined as albuminuria if urinary albumin concentration was >30 mg/L, urinary albumin excretion rate was >20 mg/min, or urinary albumin-to-creatinine ratio was >2.5 mg/mmol in men and >3.5 mg/mmol in women. CKD was defined as albuminuria or low eGFR (<60 ml/min/1.73m^2 per 1.73 m^2) or both. There are both weaknesses and strengths that should be kept in mind when interpreting study results. In fact, the AMD Annals initiative, does not make use of a central laboratory and this may have led to some degree of variability in laboratory results. Although creatinine determination cannot always be referred to IDMS procedures, most laboratories around the country currently use the Jaffè method, which has been shown to have good reproducibility. In addition, information on albuminuria were available only as a categorical trait. On the other hand, the study has several strengths since the large size of the database and the homogeneous geographical distribution of participating centres provide a realistic snapshot of real life clinical situation representative of the Italian health care system.

Natural history of diabetic renal disease

DKD phenotypes and related risk factors were investigated in a large cohort of T2D patients (n= 120.903) with a mean age of 66.6 ± 11.0 years, 58.1% male and a mean duration of diabetes of 11.1 ± 9.4 years, the proportion of patients with albuminuria was 36%, while low GFR was found in 24% and both albuminuria and low GFR in 12% of patients [7]. It is noteworthy that out of 28.806 patients with low eGFR (i.e. <60

ml/min/1.73m^2), 13.660 (48%) had an isolated reduction of GFR (i.e. normoalbuminuric renal impairment). In our series, diabetic patients with isolated reduction of GFR were more often females, showed similar age, shorter duration of diabetes, higher GFR and HDL cholesterol, and lower HbA1c, systolic BP and triglycerides as compared to patients with both low GFR and albuminuria. Although observational data such as these cannot provide conclusive information and should be considered as hypothesis-generating, our study suggests that reduced eGFR and albuminuria, showing at least in part, distinct sets of clinical correlates, might therefore entail different pathogenetic mechanisms.

Moreover, the AMD Annals Initiative provided an opportunity to describe that CKD components, mainly high albuminuria, are associated with failure to reach therapeutic targets, especially for HbA1c and BP, despite a greater use of drugs in patients with T2D [9]. In contrast with T1D, where albuminuria usually precedes and predicts renal function deterioration, in patients with T2D increased albuminuria carries a less specific predictive power with respect to renal function worsening [10]. It has recently been reported that a significant portion of T2D diabetic patients may eventually progress toward ESRD even in the absence of increased albuminuria [11]. In the Nefron study, more than half of patients with Type 2 diabetes had urinary albumin excretion rate persistently in the normal range, although renal impairment in the absence of albuminuria was less common in those with diabetes than in the general population [1]. Based on the rising trend in the prevalence of diabetes, it has been suggested that more studies are needed to clarify the pathogenesis, prevention, and treatment of nonalbuminuric renal impairment, which is, in reality, the most common cause of renal impairment in the western world.

To address this issue 17,160 T2D patients with baseline eGFR value >60 ml/min/1.73m^2. were prospectively studied over a 4-year follow-up. Baseline albuminuria was equally distributed among patients with eGFR values above and below 90 mL/min/1.73m^2 (22.3% and 22.7%, respectively). When compared to patients with eGFR >90 mL/min71,73m^2 and normoalbuminuria [12], the OR of developing the composite renal endpoint over the 4-year study period was 1.67 for patients with albuminuria and baseline eGFR values above 90 mL/min/1.73m^2 and rose progressively to 2.9 in patients with normoalbuminuria and GFR values between 90 and 60 mL/min/1.73m^2 and then further up to 5.2 when albuminuria was concomitant with mild GFR reduction (i.e. GFR between 90 and 60 mL/min/1.73m^2). At any given level of GFR, the concomitant presence of albuminuria entails an elevated renal risk, although even modest reductions in GFR (e.g. 5

mL/min/1.73m^2) seem to confer a significantly greater renal risk in the long term. This translates into a greater absolute number of non-albuminuric as compared to albuminuric patients progressing to stage 3 CKD over a four-year follow-up. In fact, two third of those patients reaching eGFR below 60 ml/min/1.73m^2 showed no albuminuria at baseline.

Hypertension and renal risk

Antihypertensive treatment and BP reduction are known to retard the progression of CKD in T2D but long-term real-life clinical data on the incidence of DKD are lacking. The AMD annals database provides a unique opportunity to perform an observational, prospective cohort study, investigating the association between achievement and maintenance of recommended BP values and the incidence of DKD and its components. To this purpose, clinical records from a total of about 13,000 patients with normal renal function and normal urine albumin excretion at baseline and with regular visits during a 4-year follow-up were retrieved and analyzed [13]. The relationship between recommended, time-updated BP control (BPC) (i.e. ≥75% of visits with SBP and DBP <140/85 mmHg) and the incidence of renal outcomes was evaluated. At baseline, 28% of patients (n=3612) had recommended BP values. Over the follow-up period, 37% developed DKD, 16% low GFR and 27% albuminuria. Patients who failed to achieve and maintain BPC over the study period showed an increased risk of developing DKD [odds ratio (OR) 1.38, P<0.001], low GFR (OR 1.18, P.0.03) and albuminuria (OR 1.47, P<0.001) as compared with those with persistent BPC. These results were confirmed after adjustment for confounders and in different subgroups.

This large, real-life cohort study shows that achieving and maintaining BP values below 140/85 mmHg is associated with a significant reduction in the incidence of CKD and its components in patients with hypertension and T2D over a 4-year follow-up. However, despite optimal BP reduction, residual renal risk remains high in patients with T2D and hypertension (Figure 1).

In a different study, the predictive role of changes in albuminuria on the loss of renal function under antihypertensive treatment was investigated on a total of 12 611 hypertensive T2D patients with normal eGFR at baseline over a 4-year period [14]. The association between changes in albuminuria status during a 1-year baseline period and time updated BP and eGFR loss overtime was assessed. Mean age at baseline was 65±9 years, known duration of diabetes11±8 years, eGFR 85±13 ml/min/1.73m^2 and BP 142±17/81±9

mmHg. Patients with persistent albuminuria showed the highest 4-year risk of eGFR loss more than 30% from baseline or onset of stage 3 CKD (eGFR<60 ml/min/1.73m^2) as compared with those with persistent normal albuminuria (odds ratio 2.00, confidence interval 1.71–2.34; P<0.001). Female gender, disease duration, age, BMI, lipid profile, low baseline eGFR, the number of antihypertensive drugs and variations in albuminuria status were relatated with renal risk in the whole study population. Furthermore, lower time updated BP values and the use of RAAS- inhibitors were asssociated to the development of renal endpoints only in patients without albuminuria. The study shows that under real-life clinical conditions, changes in albuminuria parallel changes of renal risk. Thus, albuminuria status could be a guide to optimize therapeutic strategy. Reduction of albuminuria or its prevention should be sought to maximize renal protection in T2D with hypertension (Figure 2). Furthermore, in the absence of albuminuria less strict BP target could be advisable and RAS-i may not necessarily represent the preferred antihypertensive drug. An individually tailored strategy both in terms of BP reduction and type of antihypertensive agents could be desirable depending on the presenting renal phenotype. This issue deserves to be further investigated by adequately powered RCTs conducted in patients with nonalbuminuric renal impairment.

Another important clinical issue in T2D patients at high renal risk is the presence and prognostic impact of apparent treatment resistant hypertension (aTRH). Resistant hypertension has been defined as failure to achieve recommended blood pressure levels despite concurrent use of three antihypertensive agents of different classes including a diuretic. TRH is becoming an increasingly frequent challenge in clinical practice, in light of current guidelines recommendations favoring more ambitious therapeutic targets in high risk patients. To investigate this issue, clinical records from a total of 29 923 patients with T2D and hypertension, with normal baseline eGFR and regular visits during a 4-year follow-up, were retrieved and analyzed [15]. The association between time-updated BP control (ie, 75% of visits with BP <140/90 mm Hg) and the occurrence of eGFR <60 ml/min/1.73m^2 and/or a reduction ≥30% from baseline was assessed. At baseline, 17% of patients had aTRH. Over the 4-year follow-up, 19% developed low eGFR and 12% an eGFR reduction ≥30% from baseline. Patients with aTRH showed an increased risk of developing both renal outcomes (adjusted odds ratio, 1.31 and 1.43; P<0.001 respectively), as compared with those with non-aTRH. No association was found between BP control and renal outcomes in non-aTRH, whereas in aTRH, BP control was associated with a 30% (P=0.036) greater risk of developing the renal end points. Thus, it

appears that under real-life conditions in patients with T2D and hypertension the presence of aTRH entails a significantly greater risk of developing CKD and/or a clinically relevant reduction in eGFR. The achievement and maintenance of recommended BP values (ie, 75% of visits with BP <140/ 90 mm Hg) are associated with a worse renal outcome in aTRH patients. The relationship between achieved BP and renal function seems to be J-shaped, at least at very low levels, with optimal SBP values between 120 and 140 mm Hg (Figure 3). There is a need for early identification and management of patients to prevent the development of aTRH and associated increase in renal morbidity. Reduction of antihypertensive treatment should be considered in a small, but relevant, proportion of patients in order to improve renal outcome. A similar study was also conducted in a cohort of 2,778 diabetic patients with hypertension and stage 3 CKD at baseline to assess the role of aTRH and time-updated BP control (BPC) on the progression of CKD [16]. Apparently TRH was present in 33% of patients at baseline. Over the 4-year follow-up, 20% of study patients developed an eGFR reduction >30%, with the aTRH subgroup showing an increased risk of reaching this endpoint (OR 1.31; P<0.007). Furthermore, in patients with aTRH, BPC was associated with a 79% (P=0.029) greater risk of eGFR reduction despite a 58% (P=0.001) lower risk of albuminuria status worsening. In non-aTRH, no association was found between renal outcome and BPC. In patients with aTRH, the loss of eGFR over a four-year follow-up period was faster and achievement and maintenance of recommended BP values is associated with a worse renal prognosis despite greater albuminuria reduction. Thus, aTRH seems to be associated with a burden of risk modifiable only in part by BP reduction. Further studies are clearly needed to investigate the pathophysiological mechanism underlying the effect of BP reduction per se as well as different pharmacologic strategies on renal outcome in high risk hypertensive patients such as those with diabetes and CKD.

Uric acid and the metabolic syndrome

A hotly debated and currently unresolved issue is the role of serum uric acid (SUA) in the development of DKD. Several cross-sectional observational studies have documented a strong association between increased SUA levels and reduction of GFR or albuminuria in patients with T2D. Indeed, some studies have yielded conflicting results, mainly after adjustment for confounders. We prospectively explored the association of

SUA and incident CKD in a large cohort of patients with T2D with preserved renal function at baseline. We found a strict correlation at 4-year follow up between SUA and DKD (i.e. eGFR < 60 ml/min/1.73m^2) with a progressive and constant increased of patients who develop low eGFR from second to fifth quintile of baseline SUA values, also after adjusting for several variables, such as BP, BMI, glycometabolic control, and lipid profile [17]. The association between SUA with the incidence of albuminuria appeared to be weaker, with albuminuria increased only in the fifth quintile of baseline SUA levels (Figure 4). In addition, the duration of diabetes appeared to be a modulator of the link between SUA and incident of DKD being it weaker in patients with short disease duration. In conclusion, results of our study strongly suggest a causative role of UA in DKD in patients with T2D.

It is well known as hyperuricemia (HU) is strongly associated with metabolic syndrome (Mets) and its individual components, even after adjusting for obesity and Insulin Resistance (IR). Being both UA and Mets predictors of DKD, in another study we sought to dissect whether they contribute to the onset of DKD independently of each other. We found that, after adjustment for confounders such as high BP, high triglycerides low HDL, HU remained independently associated to the development of DKD [18]. In addition, the incidence of low eGFR was higher in patients with HU also when the coexistence or absence of Mets was taken into account, while albuminuria developed more frequently in those with HU and Mets as compared to the group of reference.

Taken all together these results highlight the harmful role of both SUA and Mets and their potential interaction on kidney and may be relevant to help focusing on better prevention and therapeutic strategies of kidney damage in patients with T2D. Randomized controlled trials will further clarify the effect of SUA lowering treatment on DKD risk. A multicenter randomized, placebo-controlled study, Prevention of Early Renal function Decline (PERL), is currently investigating the effect of SUA lowering by allopurinol in patients with T1D [19]. No information is available about such a trial in patients with T2D.

Diabetic dyslipidemia

In spite of the adequate control of traditional DKD risk factors including LDL cholesterol as recommended by international guidelines, the occurrence of DKD remains high in T2D patients. Several studies indicate that diabetic dyslipidemia, i.e. high triglycerides (TG) and/or low high density lipoprotein cholesterol (HDL-

C) levels, has been consistently shown to be an independent residual risk for DKD in patients with at-target values of blood glucose and BP. Although the pathophysiological bases linking dyslipidemia to DKD are still largely undefined, it has been suggested that either deficient and/or dysfunctional HDL particles, hampering reverse cholesterol transport from renal cells, may contribute to intra-renal lipid accumulation leading to glomerulosclerosis and tubule-interstitial damage [20]. On the other hand, high TG/low HDL-C is the typical lipid pattern of insulin-resistant states and it is plausible that this metabolic derangement in T2D facilitates the toxic effects of accumulated lipids on the kidney microvascular bed [21-24]. In the large cohort of T2D subjects at high cardiovascular risk participating to the ADVANCE (The Action in Diabetes and Vascular Disease: Preterax and Diamicron-MR Controlled Evaluation Study) Study, after 5 years of follow-up, HDL-C levels in the lowest tertile were associated with a 19% higher risk of DKD risk than those in the highest tertile (OR 1.19 ; 95% CI 1.08-1.32, P = 0.0005) [25]. In another prospective study including ~ 900 T2D subjects, TG/HDL-C ratio was positively associated with the composite end-point of retinopathy and nephropathy, independently from LDL-C, HbA1c, smoking, hypertension and other variables [26]. The results of the RIACE (Renal Insufficiency And Cardiovascular Events) study demonstrating an independent association of high TG levels with DKD [27], and the slow-down of GFR decline and albuminuria with fenofibrate [3] are in line with these results.

To further investigate this issue, data collected from a large cohort of >15,000 DKD-free T2D subjects, with controlled LDL-C values at baseline, participating in the AMD Annals Initiative were analyzed longitudinally over a 4-year follow-up period [28]. Overall, 32% of study subjects developed DKD during study period, 23.2% developed albuminuria, 19% low eGFR values or eGFR decline >30%, and 4% of them developed both renal abnormalities. Notably, after controlling for LDL-C levels and a large set of potential confounding risk factors, the occurrence and progression of these renal abnormalities were significantly associated with out-of target baseline TG and/or HDL-C levels: TG levels ≥150 mg/dl increased the risk of albuminuria by 19%, of low GFR by 26%, of eGFR decline by 29%, and of developing low eGFR or albuminuria by 35% ; the risk of developing albuminuria associated with low HDL-levels was 24% and that of developing low eGFR and/or albuminuria was 44%, with linear trend of the risk with increasing TG or decreasing HDL-C values (Figure 5). Several other smaller epidemiological studies have also pointed to the role of diabetic dyslipidemia as risk factor for incidence and progression of DKD,

whereas other Authors did not confirm its independent association with microangiopathy [29-31]. These controversial results may depend upon several factors, including the modulating effect of gender and/or genetic background on these associations. Clearly, further interventional studies are warranted to explore the possibility of improving renal outcome by targeting lipid metabolism and especially triglycerides levels in T2D patients.

Risk factors variability

Targeting HbA_{1c} reduces the risks of micro- and macrovascular complications. However, glycaemic variability, more deleterious on endothelial cells than stable high glucose levels, has been associated with diabetes vascular complications both in epidemiological as well as experimental studies [32-33] even in subjects with at -target HbA1c values [34]. Long-term glycemic variability, which refers to variations of HbA1c levels over several weeks or months, has been associated with the risk of developing long term complications, including DKD. Notably, a recent meta-analysis reported that HbA_{1c}variability is a risk factor for renal disease in both T1D and T2D [35], being associated, in T2D individuals, with a higher risk of renal disease (1.34 [1.15–1.57]), as well as of macrovascular events, ulceration/gangrene, cardiovascular disease and mortality.

Data on the role of glycaemic variability on DKD have been reported both in the ADVANCE [36] in the RIACE study [37], and in the DCCT study [38] with not completely overlapping results. Also BP variability has been associated with vascular complications [39], and increasing evidences suggest that also BP and HDL-C variability may be related to the development of DKD in T2D. On the other hand, risk factors variability may also be the expression of the variability in the quality of care/efficacy of treatments that may be associated with poorer outcomes. While, it is logical to hypothesize that long-term variability in major risk factors for DKD, i.e. glucose, BP and lipid control may have an impact on its occurrence and progression, data on the possible interaction between the variability of all these risk factors on both the development of albuminuria and GFR decline in T2D are still sparse.

These issues were explored in the AMD database by looking at the correlation between intra-individual variability in lipid parameters, serum UA, HbA1c, BP, and the incidence of albuminuria or CKD [40]. All

patients with at least five measurements of the following parameters (HbA1c, SBP and diastolic BP (DBP), serum UA, total cholesterol, HDL, LDL and triglycerides) recorded over a period of 3 consecutive years were identified and those patients with normal albuminuria or a GFR ≥60 mL/min/1.73 m^2 were followed-up for up to 5 years (longitudinal phase). It was found that lipid, UA, BP and HbA1c variability might have different impact on the development of CKD components in T2D.

Variability in HbA1c was associated with a significantly higher risk of developing albuminuria [upper quartile hazard ratio (HR) = 1.3; 95% confidence interval (CI) 1.1-1.6]. Variability in UA, LDL-C, HDL-C, systolic and diastolic BP,predicted the decline in eGFR, the association with UA variability being the strongest (upper quartile HR = 1.8; 95% CI 1.3-2.4). Moreover, the application of RECPAM techniques allowed for the identification of distinct subgroups showing a different likelihood of developing DKD features. Five subgroups, characterized by an increased risk of developing albuminuria and GFR decline, were identified by RECPAM. The concomitance of an increased variability in HbA1c and HDL-C conferred the highest risk of developing albuminuria (HR = 1.47; 95% CI 1.17-1.84), while variability in UA (HR = 1.54; 95% CI 1.19-1.99) or DBP (HR = 1.47; 95% CI 1.11-1.94) conferred the highest risk of decline in eGFR (Figure 6). Furthermore, data showed that HbA1c variability exerts a greater impact on the appearance of albuminuria than on the decline of GFR, which is consistent with a previous report., The most intriguing data from RECPAM analysis has to do with renal function, and indicates that UA variability is the strongest risk factor for GFR decline with the significant contribution of concomitant BP variability.

Conclusions

CKD remains a feared complication of T2D and its prevention and treatment is a therapeutic challenge, despite achievement of recommended therapeutic targets. The AMD database provides unique opportunities to characterize disease natural history and therapeutic issues under real life clinical conditions in Italy. Recent studies, from the AMD Annals group emphasize the high prevalence/incidence of DKD, the growing figure of non albuminuric renal presentation, the role of traditional (BP, AlbU) and emerging risk predictor (TG, SUA, long term variability). While there is growing expectations on the CV and renal effects of recently developed glucose lowering drugs, glucose and BP control, together with RAAS-I remain the cornerstones of treatment. Over the next few years, further, well performed real-life clinical studies will be

helpful to integrate results of RCTs in the assessment of therapeutic advances for people with diabetes at renal risk.

References

1. Thomas MC, Macisaac RJ, Jerums G, Weekes A, Moran J, Shaw JE, Atkins RC (2009). Nonalbuminuric renal impairment in type 2 diabetic patients and in the general population (national evaluation of the frequency of renal impairment cO-existing with NIDDM [NEFRON] 11). Diabetes Care 32: 1497e502.

2. Ninomiya T, Perkovic V, de Galan BE, Zoungas S, Pillai A, Jardine M, Patel A, Cass A, Neal B, Poulter N, Mogensen CE, Cooper M, Marre M, Williams B, Hamet P, Mancia G, Woodward M, Macmahon S, Chalmers J; ADVANCE Collaborative Group (2009) Albuminuria and kidney function independently predict cardiovascular and renal outcomes in diabetes. J Am Soc Nephrol 20: 1813e21.

3. Drury PL, Ting R, Zannino D, Ehnholm C, Flack J, Whiting M, Fassett R, Ansquer JC, Dixon P, Davis TM, Pardy C, Colman P, Keech A (2011) Estimated glomerular filtration rate and albuminuria are independent predictors of cardiovascular events and death in type 2 diabetes mellitus: the Fenofibrate Intervention and Event Lowering in Diabetes (FIELD) study. Diabetologia 54: 32e43

4. Adler AI, Stevens RJ, Manley SE, Bilous RW, Cull CA, Holman RR, UKPDS Group (2003) Development and progression of nephropathy in type 2 diabetes: the United Kingdom Prospective Diabetes Study (UKPDS 64). Kidney Int 63: 225e32.

5. Packham DK, Alves TP, Dwyer JP, Atkins R, de Zeeuw D, Cooper M, Shahinfar S, Lewis JB, Lambers Heerspink HJ (2011) Relative incidence of ESRD versus cardiovascular mortality in proteinuric type 2 diabetes and nephropathy: results from the DIAMETRIC (diabetes mellitus treatment for renal insufficiency consortium) database. Am J Kidney Dis 59: 75e83.

6. Matsushita K, Coresh J, Sang Y, Chalmers J, Fox C, Guallar E, Jafar T, Jassal SK, Landman GW, Muntner P, Roderick P, Sairenchi T, Schöttker B, Shankar A, Shlipak M, Tonelli M, Townend J, van Zuilen A, Yamagishi K, Yamashita K, Gansevoort R, Sarnak M, Warnock DG, Woodward M, Ärnlöv J; CKD Prognosis Consortium (2015) Estimated glomerular filtration rate and albuminuria for prediction of

cardiovascular outcomes: a collaborative meta-analysis of individual participant data. Lancet Diabetes Endocrinol. 3: 514-525. doi: 10.1016/S2213-8587(15)00040-6.

7. De Cosmo S, RossiMC, Pellegrini F, Lucisano G, Bacci S, Gentile S, Ceriello A, Russo G, Nicolucci A, Giorda C, Viazzi F, Pontremoli R; AMD-Annals Study Group (2014) Kidney dysfunction and related cardiovascular risk factors among patients with type 2 diabetes. Nephrol Dial Transplant 29: 657-662.

8 . Levey AS, Stevens LA, Schmid CH, Zhang YL, Castro AF 3rd, Feldman HI, Kusek JW, Eggers P, Van Lente F, Greene T, Coresh J; CKD-EPI (Chronic Kidney Disease Epidemiology Collaboration) (2009) A new equation to estimate glomerular filtration rate. Ann Intern Med 150: 604-612

9 . De Cosmo S, Viazzi F, Pacilli A, Giorda C, Ceriello A, Gentile S, Russo G, Rossi MC, Nicolucci A, Guida P, Di Bartolo P, Pontremoli R; AMD-Annals Study Group (2015) Achievement of therapeutic targets in patients with diabetes and chronic kidney disease: insights from the Associazione Medici Diabetologi Annals initiative. Nephrol Dial Transplant. 30: 1526-1533. doi: 10.1093/ndt/gfv101.

10 . Penno G, Solini A, Bonora E, Fondelli C, Orsi E, Zerbini G, Trevisan R, Vedovato M, Gruden G, Cavalot F, Cignarelli M, Laviola L, Morano S, Nicolucci A, Pugliese G; Renal Insufficiency And Cardiovascular Events (RIACE) Study Group (2011) Clinical significance of nonalbuminuric renal impairment in type 2 diabetes. J Hypertens. 29: 1802-1809.

11 . Retnakaran R, Cull CA, Thorne KI, Adler AI, Holman RR; UKPDS Study Group (2006) Risk factors for renal dysfunction in type 2 diabetes: U.K. Prospective Diabetes Study 74. Diabetes 55: 1832–1839.

12 . Viazzi F, Piscitelli P, Giorda C, Ceriello A, Genovese S, Russo GT, Fioretto P, Guida P, De Cosmo S, Pontremoli R; AMD-Annals Study Group (2017) Association of kidney disease measures with risk of renal function worsening in patients with hypertension and type 2 diabetes. J Diabetes Complications 31: 419-426. doi:10.1016/j.jdiacomp.2016.10.030.

13 . De Cosmo S, Viazzi F, Piscitelli P, Giorda C, Ceriello A, Genovese S, Russo G, Guida P, Fioretto P, Pontremoli R; AMD-Annals Study Group (2016) Blood pressure status and the incidence of diabetic kidney disease in patients with hypertension and type 2 diabetes. J Hypertens. 34: 2090-2098. doi: 10.1097/HJH.0000000000001045.

14 . Viazzi F, Ceriello A, Fioretto P, Giorda C, Guida P, Russo G, Greco E, De Cosmo S, Pontremoli R; AMD-Annals Study Group (2018) Changes in albuminuria and renal outcome in patients with type 2 diabetes and hypertension: a real-life observational study J Hypertens. 36: 1719-1728. doi: 10.1097/HJH.0000000000001749.

15 . Viazzi F, Piscitelli P, Ceriello A, Fioretto P, Giorda C, Guida P, Russo G, De Cosmo S, Pontremoli R; AMD-Annals Study Group (2017) Resistant Hypertension, Time-Updated Blood Pressure Values and Renal Outcome in Type 2 Diabetes Mellitus. J Am Heart Assoc. 22; 6(9). pii: e006745. doi: 10.1161/JAHA.117.006745

16 . Viazzi F, Greco E, Ceriello A, Fioretto P, Giorda C, Guida P, Russo G, De Cosmo S, Pontremoli R; AMD-Annals Study Group (2018) Apparent Treatment Resistant Hypertension, Blood Pressure Control and the Progression of Chronic Kidney Disease in Patients with Type 2 Diabetes. Kidney Blood Press Res. 43: 422-438. doi: 10.1159/000488255.

17 . De Cosmo S, Viazzi F, Pacilli A, Giorda C, Ceriello A, Gentile S, Russo G, Rossi MC, Nicolucci A, Guida P, Feig D, Johnson RJ, Pontremoli R; AMD-Annals Study Group (2015) Serum Uric Acid and Risk of CKD in Type 2 Diabetes. Clin J Am Soc Nephrol. 10: 1921-1929. doi: 10.2215/CJN.03140315.

18 . Viazzi F, Piscitelli P, Giorda C, Ceriello A, Genovese S, Russo G, Guida P, Fioretto P, De Cosmo S, Pontremoli R; AMD-Annals Study Group (2017) Metabolic syndrome, serum uric acid and renal risk in patients with T2D. PLoS One. 12:e0176058. doi: 10.1371/journal.pone.0176058. eCollection 2017.

19 . Maahs DM, Caramori L, Cherney DZ, Galecki AT, Gao C, Jalal D, Perkins BA, Pop-Busui R, Rossing P, Mauer M, Doria A; PERL Consortium (2013) Uric acid lowering to prevent kidney function loss in diabetes: the preventing early renal function loss (PERL) allopurinol study. Curr Diab Rep. 13: 550-559.

20 . Vaziri ND (2010) Lipotoxicity and impaired high density lipoprotein-mediated reverse cholesterol transport in chronic kidney disease. J Ren Nutr 20: S35-43.

21 . Russo GT, Baggio G, Rossi MC, Kautzky-Willer A (2015) Type 2 diabetes and cardiovascular risk in women. Int J Endocrinol. 2015:832484. doi: 10.1155/2015/832484.

22 . Russo GT, Horvath KV, Di Benedetto A, Giandalia A, Cucinotta D, Asztalos B (2010) Influence of menopause and cholesteryl ester transfer protein (CETP) TaqIB polymorphism on lipid profile and HDL subpopulations distribution in women with and without type 2 diabetes. Atherosclerosis. 210: 294-301.

23 . Russo GT, Meigs JB, Cupples LA, Demissie S, Otvos JD, Wilson PW, Lahoz C, Cucinotta D, Couture P, Mallory T, Schaefer EJ, Ordovas JM (2001) Association of the Sst-I polymorphism at the APOC3 gene locus with variations in lipid levels, lipoprotein subclass profiles and coronary heart disease risk: the Framingham offspring study. Atherosclerosis. 158:173-1781.

24 . Russo GT, Giandalia A, Romeo EL, Muscianisi M, Ruffo MC, Alibrandi A, Bitto A, Forte F, Grillone A, Asztalos B, Cucinotta D (2017) HDL subclasses and the common CETP TaqIB variant predict the incidence of microangiopatic complications in type 2 diabetic women: A 9years follow-up study. Diabetes Res Clin Pract. 132: 108-117.

25 . Morton J, Zoungas S, Li Q, Patel AA, Chalmers J, Woodward M, Celermajer DS, Beulens JW, Stolk RP, Glasziou P, Ng MK; ADVANCE Collaborative Group (2012) Low HDL cholesterol and the risk of

diabetic nephropathy and retinopathy: results of the ADVANCE study. Diabetes Care 35: 2201-2206. doi: 10.2337/dc12-0306.

26 . Zoppini G, Negri C, Stoico V, Casati S, Pichiri I, Bonora E (2012) Triglyceride -high-density lipoprotein cholesterol is associated with microvascular complications in type 2 diabetes mellitus. Metabolism. 61: 22-29.

27 . Penno G, Solini A, Zoppini G, Fondelli C, Trevisan R, Vedovato M, Gruden G, Lamacchia O, Pontiroli AE, Arosio M, Orsi E, Pugliese G; Renal Insufficiency And Cardiovascular Events (RIACE) Study Group (2015) Hypertriglyceridemia is independently associated with renal, but not retinal complications in subjects with type 2 diabetes: a cross-sectional analysis of the Renal Insufficiency And Cardiovascular Events (RIACE) Italian Multicenter Study. PLoS One 10:e0125512. doi: 10.1371/journal.pone.0125512

28 . Russo GT, De Cosmo S, Viazzi F, Pacilli A, Ceriello A, Genovese S, Guida P, Giorda C, Cucinotta D, Pontremoli R, Fioretto P; AMD-Annals Study Group (2016) Plasma Triglycerides and HDL-C Levels Predict the Development of Diabetic Kidney Disease in Subjects With Type 2 Diabetes: The AMD Annals Initiative. Diabetes Care 39: 2278-2287.

29 . Hanai K, Babazono T, Yoshida N, Nyumura I, Toya K, Hayashi T, Bouchi R, Tanaka N, Ishii A, Iwamoto Y (2012) Gender differences in the association between HDL cholesterol and the progression of diabetic kidney disease in type 2 diabetic patients. Nephrol Dial Transplant 27: 1070-1075.

30 . Lin J, Hu FB, Mantzoros C, Curhan GC (2010) Lipid and Inflammatory Biomarkers and Kidney Function Decline in Type 2 Diabetes. Diabetologia 53: 263-267.

31 . Wong TY, Klein R, Islam FM, Cotch MF, Folsom A, Klein BE, Sharret AR, Shea S (2006) Diabetic retinopathy in a multi-ethnic cohort in the United States. Am J Ophtalmol 141: 446-455.

32 . Riso A, Mercuri F, Quagliaro L, Damante G, Ceriello A (2001) Intermittant high glucose enhances apoptosis in human umbilical vein endothelial cells in culture. Am J Physiol Endocrinol Metab 281: E924–E930.

33 . Quagliaro L, Piconi L, Assaloni R, Martinelli L, Motz E, Ceriello A. Intermittent high glucose enhances apoptosis related to oxidative stress in human umbilical vein endothelial cells: the role of protein kinase C and NAD(P)H-oxidase activation. Diabetes 2003;52:2795–2804.

34 . Hirsch IB (2015) Glycemic Variability and Diabetes Complications: Does It Matter? Of Course It Does! Diabetes Care 38: 1610-1614. doi: 10.2337/dc14-2898.

35 . Gorst C, Kwok CS, Aslam S, Buchan I, Kontopantelis E, Myint PK, Heatlie G, Loke Y, Rutter MK, Mamas MA (2015) Long-term glycemic variability and risk of adverse outcomes: a systematic review and meta-analysis. Diabetes Care 38: 2354-2369.

36 . Hirakawa Y, Arima H, Zoungas S, Ninomiya T, Cooper M, Hamet P, Mancia G, Poulter N, Harrap S,Woodward M, Chalmers J (2014) Impact of visit-to-visit glycemic variability on the risks of macrovascular and microvascular events and all-cause mortality in type 2 diabetes: the ADVANCE trial. Diabetes Care37: 2359-2365.

37 . Penno G, Solini A, Bonora E, Fondelli C, Orsi E, Zerbini G, Morano S, Cavalot F, Lamacchia O, Laviola L, Nicolucci A, Pugliese G; Renal Insufficiency And Cardiovascular Events Study Group (2013) HbA1c variability as an independent correlate of nephropathy, but not retinopathy, in patients with type 2 diabetes: the Renal Insufficiency And Cardiovascular Events (RIACE) Italian multicenter study. Diabetes Care 36: 2301-2310.

38 . Chang YH, Chang DM, Lin KC, Hsieh CH, Lee YJ (2013) High-density lipoprotein cholesterol and the risk of nephropathy in type 2 diabetic patients. Nutr Metab Cardiovasc Dis 23: 751-757.

39 . Rothwell PM (2010) Limitations of the usual blood-pressure hypothesis and importance of variability, instability, and episodic hypertension. Lancet 375: 938-948.

40 . Ceriello A, De Cosmo S, Rossi MC, Lucisano G, Genovese S, Pontremoli R, Fioretto P, Giorda C, Pacilli A, Viazzi F, Russo G, Nicolucci A; AMD-Annals Study Group (2017) Variability in HbA1c, blood pressure, lipid parameters and serum uric acid, and risk of development of chronic kidney disease in type 2 diabetes. Diabetes Obes Metab 19: 1570-1578.

Figure Legends

Fig. 1 Blood pressure status and the incidence of chronic kidney disease in patients with hypertension and T2D

Blood pressure control indicates patients with over 75% of BP values <140/85 mmHg over a 4 year follow-up period (Modifiied from ref 13)

Fig. 2 Changes in albuminuria and renal outcome in patients with hypertension and T2D

Four year cumulative incidence of the combination of estimated glomerular filtration rate reduction more than 30% from baseline and onset of eGFR less than 60 ml/min/1.73m^2 in study patients stratified on the basis of albuminuria status

eGFR, estimated glomerular filtration rate. NN, Sustained normoalbuminuria; N-AlbU, Progression to albuminuria; AlbU-N, Remission to normoalbuminuria; AlbU-AlbU, Persistent albuminuria (Modifiied from ref 14)

Fig. 3 The J curve relationship between blood pressure and renal function

Odds ratios with 95% confidence interval of reaching renal end point (eGFR <60 mL/min per 1.73 m^2) on the basis of time-update mean SBP in patients with T2D and treatment resistant hypertension (aTRH), taking 140 mm Hg in non aTRH patients as reference category. Patients were grouped into 10 mm Hg subsets. The subset of patients with No-aTRH and 140 mm Hg SBP was taken as the reference group.

ATRH indicates apparent treatment resistant hypertension; eGFR, estimated glomerular filtration rate

Fig. 4 Odds ratio for new onset renal damage by gender specific serum uric acid quintiles

* p< 0.001; vs I gender specific quintile (Modifiied from ref 15)

Fig. 5 Plasma triglycerides (>150 mg/dl) predict the development of diabetic kidney disease in subjects with type 2 diabetes: the AMD Annals initiative

(Modifiied from ref 17)

Fig. 6 Recursive partitioning techniques (RECPAM) analysis of developing albuminuria (A) and eGFR <60 (B)

The RECPAM tree-growing algorithm models the hazard of developing GFR<60 ml/min/1.73 m^2 or albuminuria during follow-up for up to 5 years based on a multivariable Cox regression analysis. At each partitioning step, the method selects the covariate with the binary split that maximizes the difference in risk. The algorithm stops when user-defined conditions (stopping rules) are met. The minimum set considered in our analysis comprised 50 cases of GFR declineor albuminuria incidence and 250 patients per node. Splitting baseline variables were quartiles of variability in HbA1c, systolic blood pressure (SBP) and diastolic blood pressure (DBP), serum uric acid, total, HDL, LDL cholesterol and triglycerides. The following baseline variables were included in the model as global variables: age, gender, duration of diabetes, smoking, hypertension, baseline HbA1c, blood pressure, presence of albuminuria, serum uric acid, lipid parameters and eGFR values. Values associated with patient assignment to each of the 2 subgroups (circles or rectangles, the latter representing final RECPAM classes) are shown on the branches leading to the subgroup. The data in the circles and rectangles represent the ratios of the number of patients who develop GFR<60 ml/min/1.73 m^2 or albuminuria, respectively, to the total number of patients in the subgroup (Modifiied from ref 40)