CKD273 Enables Efficient Prediction of Diabetic Nephropathy in Nonalbuminuric Patients

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Diabetic nephropathy (DN) is the most common cause of end-stage renal disease in developed nations. Significant albuminuria has long been regarded as the hallmark of DN. Despite this clearly defined indicator, a significant fraction of patients with diabetes develop renal impairment without significant albuminuria, with a variable percentage of patients having advanced kidney disease (1). This observation has resulted in the hypothesis of two distinct pathways (Alb+ and Alb−) of progression toward DN, as suggested by different sets of covariates associated with the two phenotypes. Based on data from the National Health and Nutrition Examination Surveys (NHANES), the number of adults with an estimated glomerular filtration rate (eGFR) $<$60 mL/min/1.73 m^2 and absence of albuminuria increased during the last 30 years (2). Furthermore, mortality rates increased in those with an eGFR $<$60 mL/min/1.73 m^2 without albuminuria from 35 to 51 deaths per 1,000 person-years during this time. Improved strategies are required for the management of patients developing DN without albuminuria. Biomarkers are urgently needed to identify the nonalbuminuric patients at early stages of DN.

CKD273 is a multidimensional urinary proteome classifier consisting of 273 protein fragments, predicting deterioration of renal function (3). The U.S. Food and Drug Administration recently encouraged further studies of CKD273 as a biomarker for diagnosis and risk prediction of DN (www.fda.gov/downloads/Drugs/DevelopmentApprovalProcess/UCM508790.pdf). Therefore, we investigated the value of CKD273 as a biomarker detecting DN when albuminuria is absent. Urine samples from 1,014 individuals with type 1 or type 2 diabetes, baseline eGFR $\geq$70 mL/min/1.73 m^2, and urinary albumin excretion of $<$20 μg/min were examined. Over a follow-up time of 6 years, 810 participants remained with eGFR $\geq$60 mL/min/1.73 m^2, while 204 progressed to eGFR $<$60 mL/min/1.73 m^2. Within the group of patients with type 2 diabetes (39%), 224 (56%) patients had hypertension. The urinary proteome data were classified using CKD273 and accessed with respect to the incidence of an eGFR $<$60 mL/min/1.73 m^2 during a follow-up time of 6 years. This eGFR level was chosen because the European Medicines Agency proposed that the prevention or delay of renal function decline defined as time to or the incidence rate of chronic kidney disease (CKD) stage 3 with or without albuminuria should be used as primary efficacy end point for trials (www.ema.europa.eu/docs/en_GB/document_library/Scientific_guideline/2016/10/WC500214980.pdf).

Cox proportional hazards regression with single biomarker models based on CKD273, baseline eGFR, age, or systolic or diastolic blood pressure showed that CKD273 resulted in a significant hazard ratio (HR) of 2.6 (95% CI 1.8–3.7) (Fig. 1). Next, CKD273 was adjusted in a model for age, baseline eGFR, and systolic and diastolic blood pressure. This resulted in an HR of 1.9 (95% CI 1.3–2.7), statistically significant and substantially higher than the other covariates. The positive predictive value of CKD273 is 34% for the total cohort and 47% in patients with type 2 diabetes. The results demonstrate that CKD273 enables the identification of patients with diabetes who will progress to eGFR $<$60 mL/min/1.73 m^2 in the absence of albuminuria. This observation supports that CKD273 is independent of albumin excretion and eGFR.

Previous investigations have shown that modifications of various collagen fragments in early CKD stages suggest changes in extracellular matrix (ECM) turnover, a hallmark of kidney disease (4). During fibrosis, the formation of scar tissue in the interstitial space is the result of excessive accumulation of ECM components. In a recent article, a significant association of CKD273 with renal fibrosis was demonstrated (5). CKD273 consists

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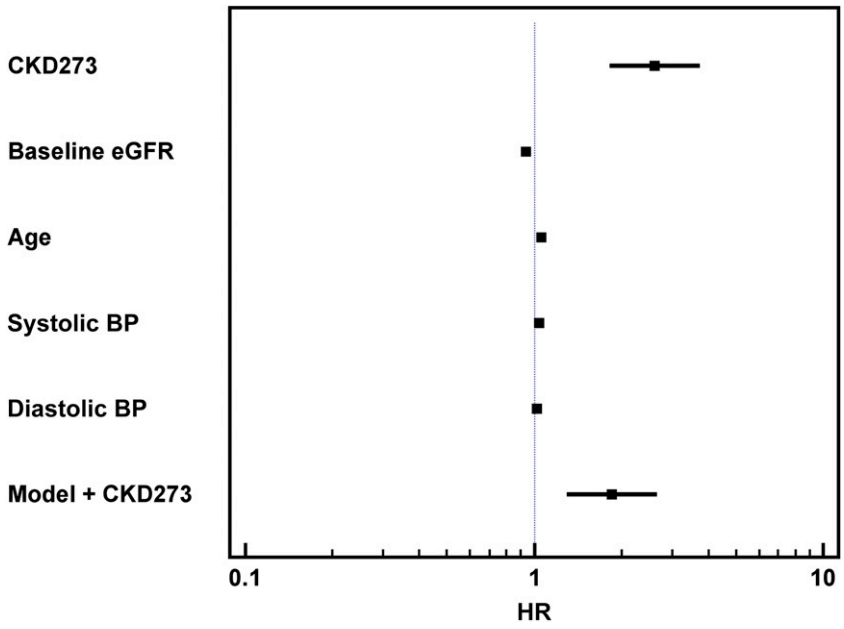

Figure 1—Forest plot. For the different Cox proportional hazards models (single and adjusted biomarker models), the HRs including the 95% CI are depicted. BP, blood pressure.

mostly of collagen peptides and may be an indicator of collagen degradation. Based on the multiple observations that urinary collagen fragments are associated with renal function, the hypothesis was generated that in CKD, especially in DN, physiological collagen degradation is attenuated, which leads to an increase of collagen in the ECM and to fibrosis. This attenuation of collagen degradation is displayed in CKD273, enabling the detection of patients with diabetes at risk for progression to DN independent of urinary albumin concentration. Based on the results presented, CKD273 currently appears to give optimal guidance on managing patients with diabetes without albuminuria.

One limitation of this work is the fact that the results are not based on a prospective study but based on data generated in multiple previous studies. However, the large number of samples included from multiple centers counteracts a possible introduced bias.

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