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Original Article

Observation period for changes in proteinuria and risk prediction of end-stage renal disease in general population

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KEY WORDS:
chronic kidney disease, end-stage renal disease, general population, proteinuria, risk prediction.

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SUMMARY AT A GLANCE

Changes in dipstick proteinuria were an independent predictor of ESRD in the general population. This study on a large Japanese population of CKD with proteinuria shows that increase dipstick in proteinuria over 2 years may be appropriate for risk prediction of ESRD.

ABSTRACT:

Aim: **Proteinuria is known to be an independent risk factor of end-stage renal disease (ESRD). But the associations between changes in dipstick proteinuria and the risk of ESRD in the general population and its appropriate observation period to predict incident ESRD are unknown.**

Methods: **We assessed the changes in dipstick proteinuria in 69 021 participants aged ≥20 years who participated in health check-ups from 1993 and more than once until 1996 in Okinawa, Japan. Development of ESRD until 2011 was identified using dialysis registry. Cox proportional hazards model and receiver operating characteristic (ROC) curve were used.**

Results: **At baseline, proteinuria (±) and ≥(1+) were observed in 2.4% and 1.2% of total subjects. 1.5% of subjects had decreased and 9.4% of subjects had increased their proteinuria level after 2 years. After adjustment for confounding factors, hazard ratios (95% confidence interval) of ESRD for subjects with proteinuria change ≤ − 1, +1, +2, +3, and +4 level during 2 years compared to subjects with no change were 0.89 (0.43–1.87), 3.18 (2.21–4.60), 8.01 (5.55–11.55), 11.17 (6.59–19.95), and 16.59 (5.95–46.25), respectively. Heterogeneity existed between changes in proteinuria level during 1 or 3 years and the risk of ESRD among baseline proteinuria. Area under the ROC curve (95%CI) to predict ESRD by increase in proteinuria level during 1, 2, and 3 years were 0.650 (0.623–0.679), 0.779 (0.751–0.808), and 0.778 (0.748–0.808), respectively.**

Conclusions: **The changes in dipstick proteinuria were an independent predictor of ESRD in the general population. Changes in proteinuria over 2 years may be appropriate for the risk prediction of ESRD.**

Chronic kidney disease (CKD) is recognized as a worldwide problem, and the number of patients with end-stage renal disease (ESRD) is increasing.[1] CKD is a risk factor not only for ESRD, but also for cardiovascular disease (CVD) morbidity and mortality.[2,3] Proteinuria is a predictor of ESRD and CVD independent of renal function and other risk factors.[4,5] In addition to one time measurement, the changes in proteinuria were also associated with the risk of renal and cardiovascular outcome in observational and interventional studies of diabetic and hypertensive kidney disease, IgA nephropathy, and subjects with cardiovascular disease.[6–13] There are few studies that accessed the relationship between the changes in proteinuria and the risk of ESRD in the general population.[14] Also little is known whether any difference exists among different durations to observe the changes in proteinuria for risk prediction. Here we present the study that evaluated the changes in dipstick proteinuria and the risk of ESRD in the general population using the data of health check-up in Okinawa, Japan. We also examined the ability to predict ESRD by different durations between two measurements to observe changes in proteinuria.

MATERIALS AND METHODS

Study population

The study population comprised a total 69 727 subjects aged ≥20 years who participated in the 1993 and at least once

during 1994 through 1996 health check-ups by the Okinawa General Health Maintenance Association (currently Okinawa Health Promotion Foundation) in Okinawa, Japan.[15,16] Four-hundred and eight participants without baseline body mass index and serum total cholesterol, 22 subjects without urine test, systolic blood pressure (SBP), diastolic blood pressure (DBP), and serum at baseline and at least once during a 1–3-year observation period, and 276 subjects with outlier values were excluded. The remaining 69 021 were the final study subjects. The protocol of this study was approved by the ethics committee of the Japanese Society of Nephrology (No. 35).

Risk factors

Dipstick urinalysis was performed in fresh urine. Proteinuria was categorized to (−), (±), (1+), (2+), (3+), and (4+), and the changes in proteinuria was defined by the difference in levels between two measurements. Blood pressure was measured by standard mercury sphygmomanometer in the sitting position. Creatinine was measured by the Jaffe method and converted to enzymatic method.[17] Estimated glomerular filtration rate (eGFR) was calculated by the Japanese Nephrology Society equation.[18] 47 767 subjects had baseline data of fasting blood sugar (FBS).

Outcome

Subjects who developed ESRD were identified by using the Okinawa Dialysis Study (OKIDS) Registry. The ESRD cases were subjects who started chronic dialysis between the last health check-up and the end of 2011. The start date of chronic dialysis was used as the date of onset of ESRD.

Statistical analysis

The linear trends of risk factors were calculated using linear regression for continuous variables and logistic regression for categorical variables. We drew Kaplan–Meier failure curve and used Log-rank test to compare the cumulative incidence of ESRD by proteinuria level at baseline and changes during the observation period by groups of subject as follows: for baseline proteinuria level (−), no change, 1 level increase, 2 level increase, and ≥3 level increase; for baseline proteinuria (±), 1 level decrease, no change, 1 level increase, and ≥2 level increase; for baseline proteinuria ≥(1+), ≥1 level decrease, no change, 1 level increase, and ≥2 level increase. The multivariable-adjusted hazard ratios (HRs) with their 95% confidence intervals (CIs) according to proteinuria level at baseline and second measurements were estimated using Cox proportional hazards model, adjusted for age, sex, SBP, DBP, body mass index, serum total cholesterol, eGFR, changes in SBP, DBP, and eGFR during observation period. We estimated the HRs in three groups for each baseline proteinuria according to their changes in proteinuria level as follows: for baseline proteinuria (−), no change, 1 level increase, and ≥2 level increase; for baseline proteinuria ≥(±), decrease, no change, and increase. The age- and sex-adjusted incidence rate of ESRD according to changes in proteinuria level over 2 years was calculated by a person-year method using direct method with 10-year age and sex group of the overall study population. HRs and 95% CIs for changes in proteinuria level over 2 years were estimated using stratified Cox proportional hazards model by baseline proteinuria, adjusted for age, sex, SBP, DBP, body mass index, serum total cholesterol, eGFR, changes in SBP, DBP, and eGFR over 2 years. Subgroup analysis of multivariable-adjusted HRs of ESRD for every 1-level increment in proteinuria over 2 years was performed by dividing the subjects by age, sex, and using the definition of hypertension, diabetes mellitus, and CKD.[19–21] Heterogeneity between changes in proteinuria level over 1, 2, or 3 years and HRs of ESRD among baseline proteinuria, and of HRs of ESRD for every 1-level increment in proteinuria over 2 years among subgroups was tested by adding a multiplicative interaction term in the model. We calculated the sensitivity and specificity of ≥1 level increase in proteinuria during observation period for ESRD prediction. We drew receiver operating characteristic (ROC) curves to compare the ability to predict ESRD using continuous variables of increase in proteinuria level during the observation period, categorizing the subjects with decrease or no change as 0.[22] We performed sensitivity analysis using the subjects with baseline FBS. To examine the HRs for decreasing changes in proteinuria level, we estimated the multivariable-adjusted HRs for changes in proteinuria level during a 2-year period using participants with baseline proteinuria ≥(±). All the analyses performed using STATA 12.1 (StataCorp, TX, USA). Two-tailed $P < 0.05$ were considered statistically significant.

RESULTS

The number of subjects with proteinuria level measured at 1, 2, and 3 years from the baseline were 54 959, 52 758, and 49 685, and the mean follow-up period for incident ESRD after second measurements were 17.3, 16.3, and 15.3 years, respectively (Fig. S1).

The mean age of the total subjects was 56 years old and the prevalence of males was 43.0% (Table 1). The number of subjects with baseline proteinuria (±) was 1666 (2.4%), (1+) was 727 (1.1%), and ≥(2+) was 106 (0.2%). Age, SBP, DBP, body mass index, FBS, serum total cholesterol, and percentage of male were higher, and eGFR were lower for subjects with higher proteinuria level (all P for trend <0.001). Changes in DBP during observation period of 1, 2, and 3 years were significantly larger with higher baseline proteinuria ($P = 0.017$, $P < 0.001$, and $P < 0.001$, respectively). Changes in eGFR during 2 and 3 years of observation were larger with higher baseline proteinuria ($P = 0.020$ and $P < 0.001$, respectively).

Table 1 Baseline characteristics according to baseline proteinuria level (*n* = 69021)

		Baseline proteinuria					
	All	(–)	(±)	(1+)	(2+)	(3+)	*P* for trend
	(*n* = 69021)	(*n* = 66522)	(*n* = 1666)	(*n* = 727)	(*n* = 104)	(*n* = 2)	
Age, years	56 (15)	56 (15)	58 (14)	60 (14)	56 (13)	38 (16)	<0.001
Male, %	43.0	42.7	51.1	49.6	58.7	100.0	<0.001
Systolic blood pressure, mmHg	127.8 (17.4)	127.4 (17.2)	136.7 (18.0)	140.1 (20.0)	139.9 (21.3)	124.0 (31.1)	<0.001
Diastolic blood pressure, mmHg	76.7 (10.4)	76.5 (10.3)	81.7 (11.3)	82.9 (11.6)	84.8 (14.6)	76.0 (22.6)	<0.001
Body mass index, kg/m^2	24.1 (3.3)	24.0 (3.3)	25.3 (3.8)	25.5 (3.9)	25.4 (4.5)	30.9 (6.9)	<0.001
Serum total cholesterol, mg/dL	204 (36)	204 (35)	213 (40)	221 (43)	231 (49)	261 (2)	<0.001
Fasting blood sugar, mg/dL (*n* = 47767)	96 (19)	96 (18)	108 (34)	110 (36)	107 (30)	96 (8)	<0.001
eGFR, mL/min per 1.73m^2	80.0 (20.9)	80.3 (20.8)	73.7 (21.7)	69.7 (24.6)	70.3 (30.1)	91.0 (20.0)	<0.001
Change in systolic blood pressure, mmHg							
1 year	0.6 (12.3)	0.6 (12.2)	1.7 (14.5)	1.1 (14.6)	−2.4 (18.2)	10.0 (5.7)	0.11
2 years	1.8 (13.4)	1.8 (13.3)	2.5 (14.9)	1.7 (17.3)	−3.2 (15.6)	8.0	0.99
3 years	1.9 (13.8)	2.0 (13.7)	1.6 (15.9)	1.3 (16.8)	1.1 (17.8)	8.0	0.14
Change in diastolic blood pressure, mmHg							
1 year	0.3 (8.6)	0.3 (8.6)	0.3 (9.7)	−0.3 (9.0)	−2.4 (11.5)	5.0 (1.4)	0.017
2 years	0.7 (9.2)	0.7 (9.1)	0.1 (10.3)	−0.6 (11.4)	−2.3 (11.1)	10.0	<0.001
3 years	0.4 (9.5)	0.4 (9.4)	−0.9 (10.7)	−1.6 (11.3)	−1.9 (11.0)	4.0	<0.001
Change in eGFR, ml/min/1.73m^2							
1 year	−1.6 (16.1)	−1.6 (16.2)	−1.0 (14.7)	−1.4 (15.4)	−5.0 (10.4)	−15.4 (17.3)	0.97
2 years	−1.6 (16.5)	−1.6 (16.5)	−1.1 (16.8)	−2.9 (15.3)	−9.4 (14.6)	−11.7	0.020
3 years	−2.1 (17.2)	−2.1 (17.2)	−2.2 (16.8)	−4.4 (16.8)	−11.9 (16.2)	−3.9	<0.001

Age, systolic and diastolic blood pressures, body mass index, serum total cholesterol, fasting blood sugar, eGFR, changes in systolic and diastolic blood pressures, and eGFR are means (SD). eGFR, estimated glomerular filtration rate; SD, standard deviation.

Table 2 Number of subjects (%) and incident end-stage renal disease cases according to proteinuria level at baseline and 1, 2, and 3 years (*n* = 69021)

	Baseline proteinuria					
	(–)	(±)	(1+)	(2+)	(3+)	All
	Number of incident end-stage renal disease cases / Number of subjects (%)					
1-year proteinuria						
(–)	103 / 49089 (89.3)	7 / 421 (0.8)	3 / 81 (0.1)	0 / 11 (0.0)	0 / 0 (0.0)	113 / 49602 (90.3)
(±)	28 / 2946 (5.4)	25 / 478 (0.9)	40 / 280 (0.5)	3 / 27 (0.0)	0 / 1 (0.0)	96 / 3732 (6.8)
(1+)	14 /877 (1.6)	22 / 395 (0.7)	13 / 109 (0.2)	1 / 6 (0.0)	0 / 0 (0.0)	50 / 1387 (2.5)
(2+)	1 / 21 (0.0)	4 / 24 (0.0)	8 / 34 (0.1)	2 / 4 (0.0)	0 / 0 (0.0)	15 / 83 (0.2)
(3+)	3 / 35 (0.1)	4 / 21 (0.0)	19 / 61 (0.1)	13 / 32 (0.1)	1 / 1 (0.0)	40 / 150 (0.3)
(4+)	0 / 0 (0.0)	0 / 0 (0.0)	1 / 1 (0.0)	1 / 4 (0.0)	0 / 0 (0.0)	2 / 5 (0.0)
Total	149 / 52968 (96.4)	62 / 1339 (2.4)	84 / 566 (1.0)	20 / 84 (0.2)	1 / 2 (0.0)	316 / 54959 (100.0)
2-year proteinuria						
(–)	79 / 47442 (89.9)	4 / 450 (0.9)	3 / 108 (0.2)	0 / 9 (0.0)	0 / 1 (0.0)	86 / 48010 (91.0)
(±)	22 / 2275 (4.5)	8 / 252 (0.4)	2 / 39 (0.1)	0 / 4 (0.0)	0 / 0 (0.0)	32 / 2570 (4.9)
(1+)	25 / 893 (2.2)	15 / 292 (0.5)	8 / 100 (0.2)	0 / 2 (0.0)	0 / 0 (0.0)	48 / 1287 (2.4)
(2+)	10 / 280 (0.7)	27 / 204 (0.4)	38 / 218 (0.4)	4 / 20 (0.0)	0 / 0 (0.0)	79 / 722 (1.4)
(3+)	3 / 41 (0.1)	6 / 33 (0.1)	21 / 62 (0.1)	8 / 25 (0.0)	0 / 0 (0.0)	38 / 161 (0.3)
(4+)	0 / 0 (0.0)	1 / 2 (0.0)	2 / 2 (0.0)	4 / 4 (0.0)	0 / 0 (0.0)	7 / 8 (0.00)
Total	139 / 50931 (96.5)	61 / 1233 (2.3)	74 / 529 (1.0)	16 / 64 (0.1)	0 / 1 (0.0)	290 / 52758 (100.0)
3-year proteinuria						
(–)	73 / 44195 (89.0)	3 / 457 (0.9)	3 / 99 (0.2)	0 / 8 (0.0)	0 / 1 (0.0)	9 / 44760 (90.1)
(±)	14 / 2229 (4.9)	7 / 194 (0.4)	2 / 36 (0.1)	1 / 6 (0.0)	0 / 0 (0.0)	24 / 2465 (5.0)
(1+)	25 / 1114 (2.2)	10 / 270 (0.5)	5 / 102 (0.2)	0 / 5 (0.0)	0 / 0 (0.0)	40 / 1491 (3.0)
(2+)	18 / 355 (0.7)	26 / 204 (0.4)	33 / 189 (0.4)	5 / 20 (0.0)	0 / 0 (0.0)	82 / 768 (1.5)
(3+)	5 / 55 (0.1)	7 / 38 (0.1)	22 / 74 (0.1)	4 / 17 (0.0)	0 / 0 (0.0)	38 / 184 (0.4)
(4+)	0 / 5 (0.0)	0 / 1 (0.0)	1 / 3 (0.0)	4 / 8 (0.0)	0 / 0 (0.0)	5 / 17 (0.0)
Total	135 / 47953 (96.5)	53 / 1164 (2.3)	66 / 503 (1.0)	14 / 64 (0.1)	0 / 1 (0.0)	268 / 49685 (100.0)

The total number of subjects who had changes in proteinuria level during the observation period of 1, 2, and 3 years were 5278 (9.6%), 4944 (9.4%), and 5174 (10.4%), and the number of subjects who developed ESRD during the follow-up period were 316 (0.6%), 290 (0.6%), and 268 (0.6%), respectively (Table 2).

Compared to subjects with baseline proteinuria (−) and no change over 2 years, those with 1 level increase [(±)], 2 level increase [(1+)], and ≥3 level increase [(2+)(3+)(4+)] had significantly higher cumulative incidence of ESRD (all Log-rank test $P < 0.001$) (Fig. 1). For subjects with baseline proteinuria (±), those decreased to (−) had significantly lower, and those with ≥2 level increase [(2+)(3+)(4+)] had significantly higher cumulative incidence of ESRD compared to subjects with no change (Log-rank test $P = 0.025$ and $P < 0.001$, respectively). Subjects with baseline proteinuria ≥(1+) and ≥1 level decrease [(1+) → (−)(±), (2+) → (−)(±)(1+), (3+) → (−)(±)(1+)(2+)] had significantly lower, and those with 1 level increase [(1+) → (2+), (2+) → (3+), (3+) → (4+)] and ≥2 level increase [(1+) → (3+)(4+), (2+) → (4+)] had significantly higher cumulative incidence of ESRD compared to no change (Log-rank test: ≥1 level decrease, $P = 0.016$; 1 level increase, $P = 0.026$; ≥2 level increase, $P < 0.001$). The Kaplan–Meier curves of observation periods of 1 or 3 years were similar to 2 years (Fig. 2 and 3). However, for baseline proteinuria ≥(1+), there was no significant difference for subjects with ≥1 level decrease [(1+) → (−)(±), (2+) → (−)(±)(1+), (3+) → (−)(±)(1+)(2+)] during 1 year (Log-rank test $P = 0.47$) or 3 years of observation (Log-rank test $P = 0.12$) compared to subjects with no change.

Baseline SBP, DBP, and eGFR were significantly higher with increasing change in proteinuria level regardless of baseline proteinuria for observation period of 2 years (Table S2). Changes in SBP and DBP over 2 years were larger with increasing change in proteinuria level for subjects with baseline proteinuria (−) and (±) (all P for trend <0.001), but not for subjects with baseline proteinuria ≥(1+). Changes in eGFR over 2 years were larger with increasing change in proteinuria level

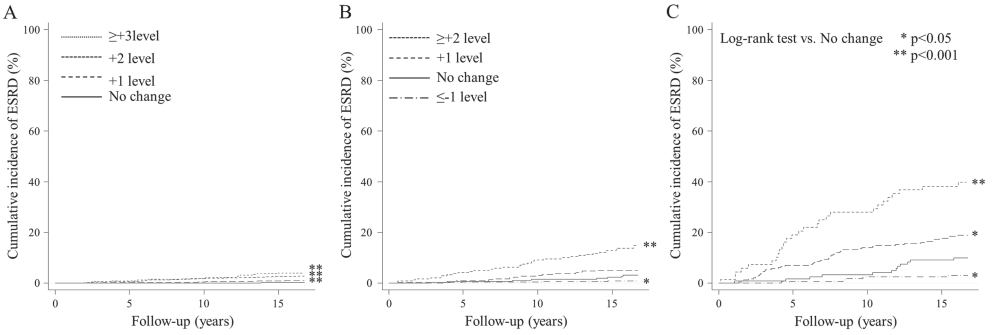

Fig. 1 Kaplan–Meier curves for the cumulative incidence of end-stage renal disease according to proteinuria level at baseline and change during 2-year ($n = 52758$).(A) Subjects with baseline proteinuria (−), ($n = 50931$). Log-rank test vs. no change (——): 1 level increase (------), $P < 0.001$; 2 level increase (·····), $P < 0.001$; ≥3 level increase (·····), $P < 0.001$. (B) Subjects with baseline proteinuria (±), ($n = 1233$). Log-rank test vs. no change (——): 1 level decrease (– · – ·), $P = 0.025$; 1 level increase (–·–·–), $P = 0.25$; ≥2 level increase (------), $P < 0.001$. (C) Subjects with baseline proteinuria ≥(1+), ($n = 594$). Log-rank test vs. no change (——): ≥1 level decrease (– · – ·), $P = 0.016$; 1 level increase (–·–·–), $P = 0.026$; ≥2 level increase (------), $P < 0.001$. ESRD, end-stage renal disease.

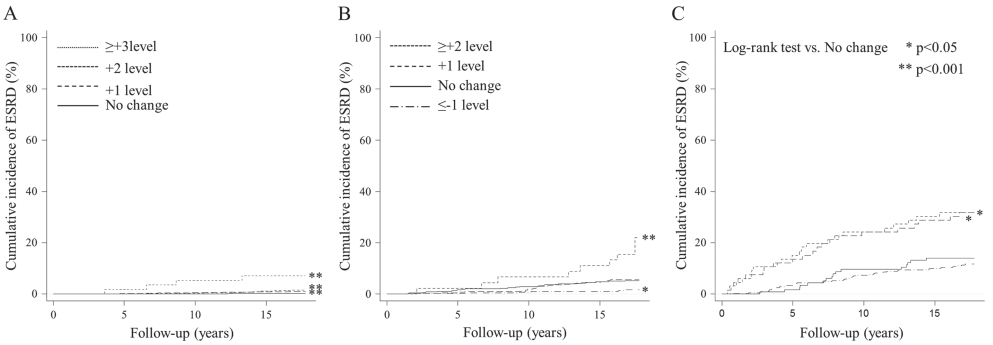

Fig. 2 Kaplan–Meier curves for the cumulative incidence of end-stage renal disease according to proteinuria level at baseline and change during 1-year ($n = 54959$).(A) Subjects with baseline proteinuria (−), ($n = 52968$). Log-rank test versus no change (——): 1 level increase (------), $P < 0.001$; 2 level increase (·····), $P < 0.001$; ≥3 level increase (·····), $P < 0.001$. (B) Subjects with baseline proteinuria (±), ($n = 1339$). Log-rank test vs. no change (——): 1 level decrease (– · – ·), $P = 0.004$; 1 level increase (------), $P = 0.84$; ≥2 level increase (·····), $P < 0.001$. (C) Subjects with baseline proteinuria ≥(1+), ($n = 652$). Log-rank test vs. no change (——): ≥1 level decrease (– · – ·), $P = 0.47$; 1 level increase (------), $P = 0.003$; ≥2 level increase (·····), $P = 0.003$. ESRD, end-stage renal disease.

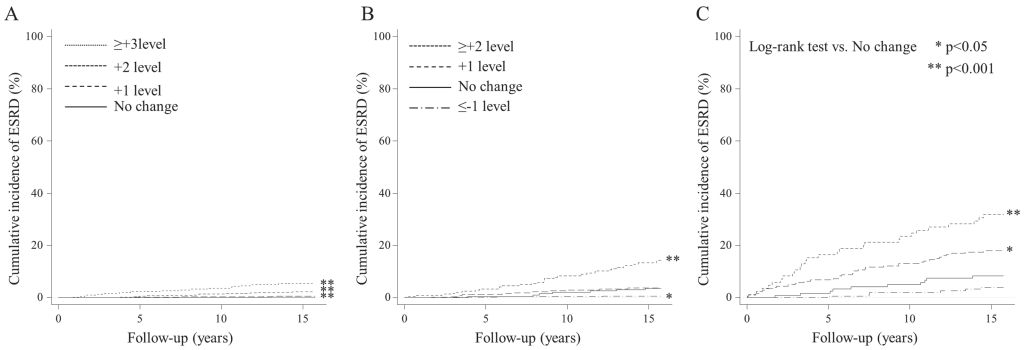

Fig. 3 Kaplan–Meier curves for the cumulative incidence of end-stage renal disease according to proteinuria level at baseline and change during 3-year (n = 49658).(A) Subjects with baseline proteinuria (−), (n = 47953). Log-rank test vs. no change (——): 1 level increase (------), $P < 0.001$; 2 level increase (·····), $P < 0.001$; ≥3 level increase (·····), $P < 0.001$. (B) Subjects with baseline proteinuria (±), (n = 1164). Log-rank test vs. no change (——): 1 level decrease (– · – ·), $P = 0.005$; 1 level increase (–·–·–), $P = 0.94$; ≥2 level increase (------), $P < 0.001$. (C) Subjects with baseline proteinuria ≥(1+), (n = 568). Log-rank test vs. no change (——): ≥1 level decrease (– · – ·), $P = 0.12$; 1 level increase (–·–·–), $P = 0.015$; ≥2 level increase (------), $P < 0.001$. ESRD, end-stage renal disease.

in all baseline proteinuria. A similar association was observed for 1- and 3-year observations (Tables S1 and S3).

Table 3 is the HRs of ESRD according to proteinuria level at baseline and second measurement compared to subjects with baseline proteinuria (−) with no change, adjusted for age, sex, SBP, DBP, body mass index, serum total cholesterol, eGFR, changes in SBP, DBP, and eGFR during observation period. The HRs were comparable among observation periods of 1, 2, and 3 years, and HRs became higher with higher proteinuria level at second measurement in each baseline proteinuria. The interaction between baseline and changes in proteinuria level for HRs of ESRD were significant in observation period of 1 and 3 years (P for interaction <0.001 and =0.002), but not for 2 years (P for interaction =0.25).

We estimated the multivariable-adjusted HRs according to change in proteinuria level over 2 years (Table 4). Multivariable-adjusted HRs (95% CI) of subjects became higher with increase in change in proteinuria level.

Table 3 Multivariable-adjusted hazard ratios (95% confidence interval) of end-stage renal disease according to proteinuria level at baseline and 1, 2, and 3 years (n = 69 021)

	Baseline proteinuria					Baseline-change
	(−)	(±)	(1+)	(2+)	(3+)	P for interaction
1-year proteinuria[1]						
(−)	**1.00 (reference)**	6.35 (2.95, 13.68)	26.76 (18.15, 39.46)			<0.001
(±)	3.71 (2.44, 5.65)	**14.14 (9.01, 22.19)**		21.83 (7.92, 60.15)	–	
(1+)			**30.74 (16.99, 55.60)**			
(2+)	6.29 (3.77, 10.48)	17.35 (11.38, 26.44)		**77.01 (18.08, 327.93)**		
(3+)			67.99 (42.66, 108.35)	64.80 (34.89, 120.37)	**858.58 (110.18, 6690.36)**	
(4+)					–	
2-year proteinuria[2]						
(−)	**1.00 (reference)**	4.27 (1.56, 11.68)	12.75 (5.12, 31.71)			0.25
(±)	4.47 (2.78, 7.20)	**12.37 (5.93, 25.80)**		–	–	
(1+)			**21.48 (10.22, 45.18)**			
(2+)	12.05 (8.07, 18.00)	25.25 (17.17, 37.10)		**32.40 (11.12, 94.38)**		
(3+)			50.73 (34.71, 74.15)	84.18 (43.08, 164.48)		
(4+)					–	
3-year proteinuria[3]						
(−)	**1.00 (reference)**	2.88 (0.90, 9.20)	10.95 (4.78, 20.90)			0.002
(±)	2.88 (1.62, 5.12)	**13.99 (6.41, 30.52)**		22.60 (3.10, 164.59)	–	
(1+)			**13.86 (5.52, 34.83)**			
(2+)	10.65 (7.27, 15.62)	22.28 (14.87, 33.38)		**26.40 (10.14, 68.72)**		
(3+)			37.64 (25.05, 56.55)	41.97 (18.40, 95.70)	–	
(4+)					–	

[1]Adjusted for age, sex, systolic and diastolic blood pressures, body mass index, serum total cholesterol, eGFR, changes in systolic and diastolic blood pressures, and eGFR over 1 year. [2]Adjusted for age, sex, systolic and diastolic blood pressures, body mass index, serum total cholesterol, eGFR, changes in systolic and diastolic blood pressures, and eGFR over 2 years. [3]Adjusted for age, sex, systolic and diastolic blood pressures, body mass index, serum total cholesterol, eGFR, changes in systolic and diastolic blood pressures, and eGFR over 3 years. Abbreviation: eGFR, estimated glomerular filtration rate.

Table 4 Age- and sex-adjusted incidence (per 1000 person-years) and multivariable-adjusted hazard ratios of end-stage renal disease according to change in proteinuria level during 2-year (n = 52758)

	Change in proteinuria level during 2-year						
	≤ − 1	0	+1	+2	+3	+4	*P* for trend
	(n = 613)	(n = 47814)	(n = 2810)	(n = 1163)	(n = 315)	(n = 43)	
Cumulative prevalence, %	100	98.8	8.2	2.9	0.7	0.1	–
Age- and sex-adjusted incidence	0.5	0.1	0.8	2.1	3.6	6.2	–
Multivariable-adjusted HR (95% CI)	0.88 (0.42, 1.85)	1.00 (reference)	3.26 (2.26, 4.69)	8.11 (5.63, 11.66)	11.79 (6.96, 19.96)	19.23 (7.00, 52.88)	<0.001

Adjusted for age, sex, systolic and diastolic blood pressures, body mass index, serum total cholesterol, eGFR, changes in systolic and diastolic blood pressures, and eGFR during 2-year. CI, confidence interval; eGFR, estimated glomerular filtration rate; HR, hazard ratio. Cumulative prevalence indicates change of this level or greater.

The overall HRs of ESRD for every 1-level increment in proteinuria over 2 years was 2.22 (95% CI: 1.98, 2.50), and there were no significant difference between sex, DBP <90 mmHg and ≥90 mmHg, eGFR <60 mL/min per 1.73m^2 and ≥60 mL/min per 1.73m^2, or among proteinuria (−), (±), and ≥(1+) (Fig. 4). Heterogeneity of HRs for every 1-level increment in proteinuria level existed between subjects with age < 65 years and ≥65 years (p for interaction =0.001), SBP <140 mmHg and ≥140 mmHg (*P* for interaction <0.001), and FBS <126 mg/dL and ≥126 mg/dL (*P* for interaction =0.010), but the HRs of each subgroups were all significant.

We calculated the sensitivity and specificity for prediction of ESRD for ≥1 level increase in proteinuria by each observation period. The sensitivity became higher from 1 year of observation (37.3%) to 2 years of observation (62.8%), and remained at the same level for 3 years of observation (63.1%), but the specificity was similar among three observation periods (92.1%, 92.1%, and 91.1%, respectively). The area under the ROC curve (AUC) (95%CI) for prediction of ESRD by increase in proteinuria level during observation period of 1 year became higher for 2 years (*P* = 0.001), and did not change for 3 years (*P* = 0.81).

Sensitivity analysis by 47 767 subjects with FBS was performed. Baseline FBS became higher with increase in proteinuria level in subjects with baseline proteinuria (−) for observation period of 1, 2, and 3 years (all *P* for trend <0.001), but there was no significant trend for subjects with baseline proteinuria (±) and ≥(1+) (Table S4). Adjusted for the above-stated confounding factors and FBS, the multivariable-adjusted HRs according to proteinuria level at baseline and 1, 2, or 3 year remained substantially unchanged (Table S5). We estimated the multivariable-adjusted HRs for change in proteinuria level during 2 years compared to no change using subjects with baseline proteinuria ≥(±) to evaluate the risk for decrease in proteinuria (Table S6). Multivariable-adjusted HRs with or without further adjustment for FBS became significantly higher from subjects with ≥2 level decrease to ≥2 level increase in proteinuria (both *P* for trend <0.001). We also performed sensitivity analysis with unified follow-up period of 15 years for each observation period, but the result did not change substantially (data not shown).

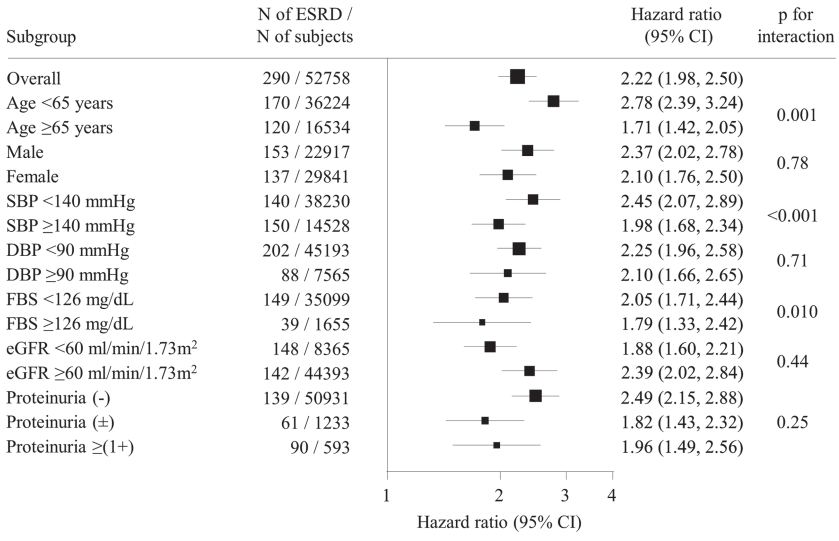

Fig. 4 Multivariable-adjusted hazard ratios of end-stage renal disease for every 1 level increment in proteinuria during 2-year by subgroup. Adjusted for sex, age, systolic and diastolic blood pressures, body mass index, serum total cholesterol, eGFR, changes in systolic and diastolic blood pressures, and eGFR during 2-year. CI, confidence interval; DBP, diastolic blood pressure; eGFR, estimated glomerular filtration rate; ESRD, end-stage renal disease; FBS, fasting blood sugar; SBP, systolic blood pressure.

DISCUSSION

Using data from health check ups of the general population, we have shown that an increase in dipstick proteinuria during the 1- to 3-year observation periods was associated with higher risk of ESRD independent of confounding factors at baseline and changes during the observation period. The predictive ability of ESRD was higher in the observation period for change in proteinuria of 2 years compared to 1 year, but there was no significant difference between 2 and 3 years. Also, heterogeneity existed between changes in proteinuria level and the risk of ESRD among baseline proteinuria for the observation periods of 1 and 3 years, but not for 2 years. To the best of our knowledge, this is the first study to evaluate changes in dipstick proteinuria and the risk of ESRD in the general population. Also, this is the first study that examined the duration of an observation period for changes in proteinuria and the ability to predict the outcome.

Changes in proteinuria have been suggested as a surrogate outcome for kidney disease progression.[23] There have been studies that examined the relationship between the magnitude of change and the outcome, but few studies examined the association between duration of change in proteinuria and the risk of outcome.[14] In our study, significant interaction between baseline and changes in proteinuria for HRs of ESRD existed for observation periods of 1 and 3 years, which means that the HRs for increase in proteinuria level are different among baseline proteinuria. Since the interaction was not significant for 2 years, we can estimate that the HRs of ESRD according to changes in proteinuria level regardless of baseline proteinuria. Considering the predictive ability of change in proteinuria and heterogeneity between changes in proteinuria level and the risk of ESRD among baseline proteinuria, changes in proteinuria during an observation period of 2 years may be appropriate for risk prediction of ESRD.

There were few observational studies with a large number of subjects that reported the association between the changes in proteinuria or albuminuria and renal outcomes. A total of 983 Pima Indians with type 2 diabetes had two albumin/creatinine ratio (ACR) measurements by means of 2.4 years and were followed for a median of 8.4 years after the second measurement.[6] Each doubling in the second ACR was associated with a 1.71-fold increase of incident ESRD after adjustment for confounding factors. A health care utilization cohort from Stockholm, Sweden, Stockholm CREAtinine Measurements (SCREAM) project followed 31 732 subjects for a median of 3 years.[14] Compared to stable ACR, a fourfold increase in ACR was associated with a 3.08-times higher, and a fourfold decrease in ACR was associated with a 0.34-times lower risk of ESRD.

Many clinical trials for type 2 diabetes,[7–10] vascular disease or high-risk diabetes[11] have shown that the reduction in albuminuria was associated with reduction in renal risk. A total of 810 African Americans with hypertensive renal disease were followed up for a median of 3.8 years, and the relationship between the changes in proteinuria and the risk of ESRD were examined in The African American Study of Kidney Disease (AASK), and the changes in proteinuria from baseline to 6 month were associated with the risk of ESRD.[12] Meta-analysis using 830 individual patient data of IgA nephropathy from 11 randomized trials examined the association between proteinuria reduction in median of 9 months and composite outcome of doubling of serum creatinine level, ESRD, or death.[13] Reduction in proteinuria was associated with lower risk, and was consistent across studies. In our study, HRs for increase in proteinuria level was significantly lower for subjects with high SBP and high FBS, but all the HRs were significant. Changes in proteinuria were a significant risk factor of ESRD regardless of age, sex, and comorbidities.

Albumin is the predominant protein filtered through glomerular membrane, and it is retrieved by proximal tubule.[24] Either structural or functional alteration of glomerular filtration barrier or dysfunction of proximal tubule may cause proteinuria. The mechanism of kidney injury induced by proteinuria, which may lead to ESRD, was examined by clinical and experimental studies. Kidney nephrectomized from children with congenital nephrotic syndrome of the Finnish type, which is characterized by constant heavy proteinuria from birth, had interstitial inflammation and fibrosis.[25] Proteinuria induced apoptosis in proximal tubular cells and dysfunctional autophagy in the proximal tubule, which resulted in the accumulation of intracellular proteins.[26,27] Albuminuria impaired renal tubular tight junctions and induced a proinflammatory and profibrotic response in cortical collecting duct.[28,29] In our study, the ability to predict ESRD for increase in proteinuria during an observation period of 1 year became higher for 2 years, with no further improvement by one more year prolongation to 3 years. This may be because effects to kidney tissues caused by proteinuria may need a certain amount of time to develop renal injury. Heterogeneity existed between 1- and 3-year changes in proteinuria level and the risk of ESRD among baseline proteinuria, but not between 2 years. One year may not be enough to cause kidney tissue change by increase in proteinuria for subjects without damage, while subjects with impaired kidney may easily progress in a shorter time. An observation period of 3 years may have increased competing risks such as death, which may have a stronger effect on older or advanced kidney disease subjects. Further basic and clinical studies are warranted to elucidate the pathogenic mechanisms of the contribution of proteinuria for CKD progression and ESRD.

The strengths of our study are large numbers of study subjects and long duration of follow-up. The limitations are a lack of information about comorbidities or medication and data for competing risks such as death.

In conclusion, the changes in dipstick proteinuria were an independent predictor of ESRD in the general population. The risk of ESRD according to the proteinuria level at baseline and

second measurement was comparable among different observation periods, but there was heterogeneity between changes in proteinuria level and the risk of ESRD among baseline proteinuria for the 1- and 3-year observation periods, but not for 2 years. Compared to changes in proteinuria over 1 year, the ability to predict ESRD was higher for changes over 2 years and no difference was made by prolongation of observation for one more year to 3 years. For risk prediction of ESRD, changes in proteinuria during a 2-year observation period may be appropriate.

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SUPPORTING INFORMATION

Additional Supporting Information may be found online in the supporting information tab for this article.

Table S1. Baseline and changes in blood pressures and estimated glomerular filtration rate according to proteinuria level at baseline and change over 1 year (n = 54 959).
Table S2. Baseline and changes in blood pressures and estimated glomerular filtration rate according to proteinuria level at baseline and change over 2 year (n = 52 758).
Table S3. Baseline and changes in blood pressures and estimated glomerular filtration rate according to proteinuria level at baseline and change over 3 year (n = 49 685).
Table S4. Baseline fasting blood sugar according to proteinuria level at baseline and change over 1, 2, and 3 years (n = 47767).
Table S5. Multivariable-adjusted hazard ratios of end-stage renal disease according to proteinuria level at baseline level and 1, 2, and 3 years by subjects with baseline fasting blood sugar (n = 47 767).
Table S6. Multivariable-adjusted hazard ratios according to change in proteinuria over 2 years by subjects with baseline proteinuria (±) or higher (n = 1827).
Figure S1. Number of study subjects and follow-up period.
Figure S2. Receiver operating characteristic curves for prediction of end-stage renal disease by increase in proteinuria level.