- Original Report -

Genome-wide association study of renal function traits: Results from the J-MICC Study

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Abstract

Background: Chronic kidney disease (CKD) is a rapidly growing worldwide public health problem. Recent advances in genome-wide-association studies (GWAS) revealed several genetic loci associated with renal function traits worldwide. ***Methods:*** We investigated the association of genetic factors with the levels of serum creatinine (SCr) and estimated glomerular filtration rate (eGFR) in Japanese population-based cohorts analyzing the GWAS imputed data with 11,221 subjects and 12,617,569 variants, and replicated the findings with the 148,829 hospital-based Japanese subjects. ***Results:*** In the discovery phase, 28 variants within four loci (chromosome [chr] 2 with eight variants including rs3770636 in the *LRP2* gene locus, on chr 5 with two variants including rs270184, chr 17 with 15 variants including rs3785837 in the *BCAS3* gene locus, and chr 18 with three variants including rs74183647 in the *NFATC1* gene locus) reached the suggestive level of $p < 1\times10^{-6}$ in association with eGFR and SCr, and 2 variants on chr 4 (including rs78351985 in the *MTTP* gene locus) fulfilled the suggestive level in association with the risk of CKD. In the replication phase, 25 variants within three loci (chr 2 with seven variants, chr17 with 15 variants and chr 18 with three variants) in association with eGFR and SCr, and two variants on chr 4 associated with the risk of CKD became nominally statistically significant after Bonferroni correction, among which 15 variants on chr 17 and three variants on chr 18 reached genome-wide significance of $p < 5\times10^{-8}$ in the combined study meta-analysis. The associations of the loci on chr 2 and 18 with eGFR and SCr as well as that on chr 4 with CKD risk have not been previously reported in the Japanese and East Asian populations. ***Conclusion:*** Although the present GWAS of renal function traits included the largest sample of Japanese participants to date, we did not identify novel

loci for renal traits. However, we identified the novel associations of the genetic loci on chr 2, 4, and 18 with renal function traits in the Japanese population, suggesting these are transethnic loci. Further investigations of these associations are expected to further validate our findings, for the potential establishment of personalized prevention of renal disease in the Japanese and East Asian populations.

Key Words

Chronic kidney disease, genome-wide association study, population-based cohort

Introduction

Chronic kidney disease (CKD) is a worldwide public health problem that is growing rapidly. Recent advances in genome-wide-association studies (GWASs) revealed several genetic loci associated with renal function traits in European populations [1, 2], Asian populations [3] and worldwide [4]. For example, recent GWASs investigating the genetic loci associated with kidney function identified the *glucokinase regulator* (*GCKR*) locus on chromosome (chr) 2 as highly significant loci [5, 6]. Additionally, genotype distributions and factors related to lifestyles are known to differ according to races, ethnicity and nations, which may lead to the existence of diverse population-specific gene-environmental interactions, making the nation-based reports of genetic associations still meaningful for the effective prevention of disease in each population [7]. Recently, a meta-analysis in an East Asian population revealed 17 loci that were newly associated with renal function traits. However, the participants of which were multi-ethnic and were mostly diseased with regard to the Japanese individuals [3, 8]. We conducted a nation-wide genome cohort study to find genetic factors for the possible establishment of personalized prevention of human chronic diseases in Japanese, named the Japan Multi-institutional Collaborative Cohort (J-MICC) Study. Regarding this study, 100,000 participants from 12 areas in Japan have been recruited, and the GWAS genotyping with 14,000 subjects for 1,000,000 single nucleotide polymorphisms (SNPs) have been completed [9, 10]. Herein, we systematically and comprehensively investigated the association of genetic factors with the levels of serum creatinine (SCr) and estimated glomerular filtration rate (eGFR) in Japanese population-based cohorts, using GWAS data from the J-MICC Study.

Subjects and Methods

Study subjects

We analyzed the data repository of J-MICC Study, launched in 2005 in 10 areas of Japan, in which about 100,000 volunteers aged 35-69 years old provided their blood and lifestyle data based on a questionnaire, after providing informed consent [9, 10].

The present study included 14,539 randomly selected J-MICC Study participants from 12 areas (Chiba, Sakuragaoka, Shizuoka-Daiko, Okazaki, Aichi, Takashima, Kyoto, Tokushima, Fukuoka, Kagoshima and Kyushu-KOPS [Kyushu Okinawa Population Study]) where the J-MICC Study took place. As a sample quality check (sample QC), participants with a proportion of identity by descent (IBD) over 0.1875, and outliers in the principal component analysis (PCA) were excluded, resulting in 14,086 participants for analysis. Among them, data for SCr were available for 11,681 subjects, and 398 subjects who had their creatinine measured with jaffe method were excluded, leaving 11,283 subjects (who had their creatitine measured with enzyme method) for the final analyses. Characteristics of the study subjects are shown in Table 1. Written informed consent was obtained from all participants. The protocol of this study was approved by the Ethics Review Committee of the Nagoya University Graduate School of Medicine (Approval No. 939-14), Aichi Cancer Center and all the participated institutions. All the research procedures were conducted according to the Ethical Guidelines for

Human Genome and Genetic Sequencing Research in Japan and the Declaration of Helsinki.

Questionnaire

Lifestyle-related information was collected using a self-administered questionnaire evaluated by trained staff. The questionnaire included items on smoking status, alcohol consumption, food consumption and medical history [11-14]. Information on medication and the presence/history of disease were also based on the self-reported questionnaire and the health checkup data at baseline. The presence of hypertension (HT) and/or diabetes mellitus (DM) was defined as having HT/DM as the present illness or taking medication for their treatment based on the questionnaire, or fulfilled the diagnostic criteria of HT/DM in the health checkup laboratory data (systolic blood pressure [BP] $\geq$ 140 mmHg or diastolic BP $\geq$ 90 mmHg for HT, and fasting blood glucose $\geq$ 126 mg/dL or hemoglobin A1c (HbA1c) $\geq$ 6.1% based on the Japan Diabetes Society criteria (equivalent to > 6.5% in National Glycohemoglobin Standardization Program criteria) for DM [15].

Genotyping and quality control filtering

DNA was extracted from buffy coat with a BioRobot M48 Workstation (QIAGEN Group, Tokyo). The genotyping was conducted by the RIKEN institute (Yokohama, Japan) using an Illumina OmniExpressExome Array (Illumina, San Diego, CA, USA) for the 964,193 SNPs. 26 samples with inconsistent sex information between questionnaire and an estimate from genotype were excluded. The identity-by-descent method implemented in the PLINK 1.9 software (https://www.cog-genomics.org/plink2) found 388 close relationship

pairs (pi-hat > 0.1875) and one sample of each pair was excluded. PCA [16] with a 1000 Genomes reference panel (phase 3) (http://www.internationalgenome.org/category/phase-3/) detected 34 subjects whose estimated ancestries were outside of the Japanese population. The 34 samples were excluded. All the remaining 14,091 samples met a sample-wise genotype call rate criterion (≥ 0.99). SNPs with a genotype call rate <0.98 and/or a Hardy-Weinberg equilibrium exact test p value < 1×10^{-6}, a low minor allele frequency (MAF) < 0.01, or a departure from the allele frequency computed from the 1000 Genomes Phase 3 EAS (East Asian) samples were excluded. Quality control filtering resulted in 14,091 individuals and 570,162 SNPs.

Genotype imputation

Genotype imputation was conducted using SHAPEIT ver.2 (https://mathgen.stats.ox.ac.uk/genetics_software/shapeit/shapeit.html#home) and Minimac3 (http://genome.sph.umich.edu/wiki/Minimac3) software based on the 1000 Genomes Project cosmopolitan reference panel (phase 3). After the genotype imputation, variants with MAF < 0.05 and r^2 < 0.3 were excluded, to examine the substantial effects of common variants on renal functions in Japanese, resulting in 6,288,024 variants provided for the final analyses.

Estimated glomerular filtration rate and definitions of chronic kidney disease

SCr was measured in all participants analyzed using an enzymatic method. The eGFR of each participant was calculated based on SCr, age, and sex using the Japanese eGFR equation proposed by the Japanese Society of

Nephrology: eGFR (ml/min/1.73 m^2) = 194 × SCr (mg/dl)$^{-1.094}$ × age$^{-0.287}$ (× 0.739 if female), which is the calibrated version from the CKD-EPI (Chronic Kidney Disease Epidemiology Collaboration) [17]. Values of eGFR equal to or more than 120 (ml/min/1.73 m^2) were winsorized at 120. In those who had eGFR values of equal to or more than 120, SCr corresponding to eGFR of 120 were calculated based on the Japanese eGFR equation. The prevalence of CKD was determined for CKD stages 3–5 (eGFR <60 ml/min/1.73 m^2).

Replication study

We conducted the replication study with an independent data set from the BioBank Japan Project (BBJ), which is consisted of 148,829 subjects (81,629 males and 67,200 females) with 46 diseases (nephrosis excluded), to examine the validity of the SNPs found in the GWAS of the J-MICC Study [18]. The exclusion criteria used consisted of: 1) age < 18, 2) PCA outlier from the EastAsian cluster, 3) Closely related samples determined by identity by state (IBS) (visual inspection), and 4) Nephrotic syndrome. Genotyping was conducted by using Illumina OmniExpressExome Array and OmniExpress+HumanExome Array (Illumina, San Diego, CA, USA). Pre-phasing was conducted with Eagle (https://data.broadinstitute.org/alkesgroup/Eagle/). Genotype imputation was conducted using Minimac3 (http://genome.sph.umich.edu/wiki/Minimac3) software based on the 1000 Genomes Project cosmopolitan reference panel (phase 3). Association analyses were conducted using R (ver 3.3.2, https://cran.r-project.org/).

Statistical analysis and graphics

We applied genomic control correction where the genomic control parameter lambda was >1.0 [19]. Thereafter, we examined the associations of the SNPs with the risk of CKD and the quantitative traits of serum creatinine and eGFR using the EPACTS software (http://genome.sph.umich.edu/wiki/EPACTS). The association of SNPs with presence/absence of CKD as a binary trait was examined by logistic Wald test, and the associations of SNPs with serum creatinine and eGFR as continuous variables were tested by linear Wald test. With regard to the covariates to be adjusted, gender, age, the presence of hypertension and diabetes mellitus, and the first five principal components (PCs) were included. Variants with the MAF of more than (or equal to) 0.05 were considered. The Manhattan and Q-Q Plots were drawn with the 'qqman' function in R (https://cran.r-project.org/web/packages/qqman/index.html). The GWAS meta-analysis was conducted using the METAL (http://genome.sph.umich.edu/wiki/METAL_Documentation), and the regional plots were constructed using the LocusZoom software (http://genome.sph.umich.edu/wiki/LocusZoom_Standalone). The genome-wide significance levels were set at $p < 5\times10^{-8}$, and the genome-wide suggestive levels were set at $p < 1\times10^{-6}$ in all the analyses. In the replication phase, the significance threshold was defined as p values less than 0.05 devided by the number of comparisons based on the Bonferroni correction. Lead SNPs (or variants) were defined as the SNPs (or the variants) that reached the smallest p values in each genetic locus defined as the position on a chromosome identified by the cytogenetic banding of the chromosome [20].

Results

Variants identified in the discovery phase GWAS

In the present Japanese population-based (J-MICC) GWAS, we conducted the quantitative trait loci (QTL) analysis with the eGFR and SCr as the continuous dependent variable, and then examined the risk of CKD defined as eGFR <60 ml/min/1.73 m^2 as binary outcomes. From the analysis of eGFR QTL GWAS, we identified four loci with 28 variants suggestively associated with eGFR (on chr 2, 5, 17 and 18; $p < 1\times10^{-6}$) (Table 2), whereas in the analysis of SCr QTL GWAS, we also identified four loci with 28 variants demonstrated suggestively associated with eGFR (on chr 2, 5, 17 and 18; $p < 1\times10^{-6}$) (Table 3). From the analysis of GWAS of the presence/absence of CKD, one locus on chr 4 (with 2 SNPs) reached the suggestive level in a single-variant-based analysis (Table 4). The Manhattan Plots for the analyses above are shown in Fig. 1. The Q-Q plots for the *p* values in each of these analysis are shown in Fig. 2 (the Q-Q plots before genomic control correction are shown in online suppl. Fig. 1), all the genomic inflation factors (lambda values) of which were close to 1 (range from 0.99 - 1.01), suggesting that the population structure was fairly adjusted, given the larger sample size of the present study.

Replication of the detected variants in the independent data set

Next we conducted the replication study of the variants discovered in the J-MICC GWAS in an independent data set from BBJ GWASs using the variants that reached the suggestive level. In the replication phase, 25 variants within three loci (on chr 2 with seven variants including rs3770636 in the *LRP2* [*LDL receptor related protein 2*] gene locus, chr 17 with 15 variants including rs3785837 in the *BCAS3* gene locus, and chr 18 with

three variants including rs74183647 in the *NFATC1* [*nuclear factor of activated T-cells 1*] gene locus) became nominally statistically significant ($p < 0.05/28 = 0.0018$, with Bonferroni correction), of which 15 variants on chr 17 and 3 variants on chr 18 reached genome-wide significance of $p < 5\times10^{-8}$ in association with eGFR and SCr (Tables 2 and 3), all of which were previously reported loci for renal traits in the transethnic GWAS [21]. Meanwhile, the locus identified for the presence/absence of CKD on chr 4 did not reach genome-wide significance in the combined study, although it fulfilled the nominal statistical significance ($p < 0.05/2 = 0.025$, with Bonferroni correction) in the replication phase (Table 4). We also evaluated the LD (linkage disequilibrium) status of these variants in the detected loci by constructing the regional plots of each locus, which revealed the close LD between the variants in the corresponding loci (Fig. 3). Additionally, conditional analyses of the lead SNPs in each locus revealed that the associations were not significant for the other SNPs/variants in the corresponding loci (online suppl. Fig. 2).

Replicability of the previously reported variants in the J-MICC Study data

We also examined the associations of 41 SNPs reported to be associated [3] with the risk of CKD and eGFR as a continuous trait using the 1000 Genomes imputed data. Of these, there were 16 SNPs significantly associated with both of eGFR and SCr as quantitative traits, and 8 SNPs were significantly associated with risk of CKD as a binary trait (online suppl. Table 1).

Discussion

To our knowledge, this is the largest GWAS of CKD and renal function traits with the cross-sectional study in a population-based Japanese cohort published to date. Although the present study couldn't identify the novel loci for renal traits, it revealed the novel associations of three loci on chr 2 and 18 with renal function traits, which were previously unreported in the East Asian and Japanese populations. Additionally, we identified the locus on chr 17 as the significantly associated locus with renal functions, which was consistent with the previous finding in Japanese subjects [3]. Regarding the newly identified loci for the East Asian and Japanese populations, the *LRP2* gene on chr 2, also called *megalin*, was originally identified as the target antigen of the rat model of membranous glomerulonephritis, and is structurally similar to the low density lipoprotein receptor [22]. Megalin is abundantly found in kidney proximal tubules, and has been shown to mediate endocytic uptake of vitamin D3 filtered in the glomeruli [23]. Endocytosis mediated by this molecule is demonstrated to play key roles in the reabsorption of albumin in renal proximal tubule cells, and it is shown to work as an important sensor in cooperation with PKB (protein kinase B), determining the survival of renal proximal tubule cells under the existence of albumin, thus considered to play roles in the progression of CKD to end-stage renal disease (ESRD) [24]. A functional mutation in *LRP2* (*LRP2* Arg365Ter, rs80338744) has been demonstrated to cause Donnai-Barrow Syndrome, which is a congenital disease presented with facio-oculo-acoustico-renal syndrome with proteinuria [25]. NFATC1 is the cytoplasmic component of the nuclear factor of activated T-cells transcription complex, which is activated upon T-cell receptor (TCR) engagement, and is involved in the functions of T lymphocytes [26-29]. It has been demonstrated that NFAT-dependent injury-response gene,

DSCR1, is involved in the phenotype switching/remodeling of the vascular smooth muscle cells [30]. Furthermore, the rs74183647 SNP of *NFATC1* found in this study is shown to modulate its gene expression in human whole blood, according to the GTEx database (https://www.gtexportal.org/). Given its biological roles in vasculature in the context of renal injury, variations in the *NFATC1* gene may be considered potential susceptibility factors for human CKD. A recent experimental study also revealed the roles of NFAT1 within the context of salt sensitivity, an important process in the development of CKD [31], thereby supporting the involvement of this protein in the genesis of CKD. With respect to the previous reports, the same locus on chr 17 as that identified in the present study was found to be associated with renal functions in the East Asian cohort consortium data. This locus includes the *BCAS3* gene, which is reportedly a biologically important gene in controlling the directional cell migration and angiogenesis by facilitating the crosstalk between cytoskeletons [32], and is amplified in 9% of primary breast tumors, expressed tumor-derived cell lines [33]. *BCAS3* is also found to be expressed in human embryonic stem (ES) cells during their differentiation into blood vascular precursors, and highly expressed in the tumor cells and blood vessels of glioblastoma, hemangiopericytoma and brain abscess, suggesting that *BCAS3* can serve as a marker for both human ES cells and tumors [33]. Moreover, genetic variations in *BCAS3* gene, including the rs3785837 identified in the present study, have been shown to change significantly in expression levels significantly in human tibial arteries according to the GTEx. However, the roles of BCAS3 in human kidney functions impairments are poorly understood. Based on the factors discussed above, it is hypothesized to play a role in the formation or modulation of human renal vasculature. Therefore, further biological and epidemiological studies are warranted. Regarding the locus found to be

associated with the risk of CKD on chr 4 (4q23), *microsomal triglyceride transfer protein* (*MTTP*) gene encoded by this locus is reportedly associated with the risk of celiac disease in Caucasians, although the roles of genomic locus in the genesis of human CKD remain largely unknown [34]. *MTTP* reportedly catalyzes the transport of lipids between phospholipid surfaces and its genetic variation is associated with abetalipoproteinemia and glucose tolerance [35]. Given its roles in lipid and glucose metabolisms, analyzing the roles of this molecule in the development of CKD may be beneficial, although its genetic variation did not reach genome-wide significance in the combined study. Moreover, our GWAS analysis of the same data set without adjustment for PCA based on GWAS genotypes revealed another locus significantly associated with renal function trait located in chr 6, including *major histocompatibility complex* (*MHC*) loci, the statistical significance of which disappeared after adjusting for PCA. This suggested that the significant association of this locus with renal function traits may be attributable to population stratification, at least when Japanese population is concerned (data not shown).

It appears that distinct genetic loci for renal function traits exist for each race/ethnicity. In the GWAS-meta-analysis of the European population, genetic loci such as the one on chr 16 (including the *UMOD* gene), or those on chr 10 (including *CDH23* gene) and on chr 7 (including *solute carrier family 22, member 2* [*SLC22A2*] and *GALNTL5* gene) were shown to be significantly associated with renal functions [1, 20, 35-37]. In the present GWAS in Japanese population, these loci were not found to be genome-wide significant, suggesting the possibility that specific genetic mechanisms exist between the races. Of the four loci found to be genome-wide significant in the present study (discovery phase), the locus on chr 17 have already been reported in East Asians,

whereas other two loci on chr 2 and 18 are firstly reported in the East Asian and Japanese populations. However, they were reported in the GWAS of renal traits in European populations and/or those with African ancestries, as represented by the *LRP2* locus on chr 2 in African Americans [21, 39]. To the best of our knowledge, the association of the 4q23 locus with CKD risk is the novel finding, which warrants further investigations with independent data sets. Whereas two out of four top SNPs for each loci found to be genome-wide significant (rs3785837 on chr 17 and rs74183647 on chr 18) had functionality by itself as described above, the functionalities of the SNPs on chr 2 (rs3770636) and chr 4 (rs78351985 and rs555786707) remain unknown. The minor allele frequencies of the newly detected SNPs for Japanese (those with the top *p* values in each locus) were 0.2326 in Japanese, 0.0221 in Caucasians and 0.0974 in Africans for the *LRP2* rs3770636 *G* allele, 0.4355 in East Asians, 0.0924 in Europeans and 0.2095 in Africans for the *NFATC1* rs74183647 *C* allele, 0.0288 in East Asians, 0.0010 in Europeans and 0.0000 in Africans for the rs555786707 *A* allele on chr 4, 0.0317 in East Asians, 0.0000 in Europeans and 0.0000 in Africans for the rs78351985 *G* allele on chr 4 according to dbSNP (https://www.ncbi.nlm.nih/gov/SNP), all of which are more than 1%, suggesting the substantial involvement of these variants in the regulation of renal functions in Japanese and East Asians. Although world-wide GWAS consortiums of human chronic diseases are flourishing these days, such GWAS of each race/ethnicity may also be considered meaningful, given racially/ethnically specific gene-environment interaction could provide more effective ways for disease prevention in each race/ethnicity [7, 40]. The strength of the present study is the large sample size and the densely imputed genetic data, which lead to the discovery of as yet unidentified genetic loci associated with renal functions specifically in Japanese population.

In conclusion, the present GWAS of renal function traits included the largest sample of Japanese population published to date, and revealed genetic loci on chr 2, 4, and 18 as involved in the renal function in the Japanese population. Further investigations of these associations, including prospective validation within the present cohort, are expected to confirm our findings, for the potential establishment of personalized prevention of renal disease in the Japanese and East Asian populations.

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Disclosure Statement

The authors have no conflict of interest to disclose.

References

1. Gorski M, Tin A, Garnaas M, McMahon GM, Chu AY, Tayo BO, Pattaro C, Teumer A, Chasman DI, Chalmers J, Hamet P, Tremblay J, Woodward M, Aspelund T, Eiriksdottir G, Gudnason V, Harris TB, Launer LJ, Smith AV, Mitchell BD, O'Connell JR, Shuldiner AR, Coresh J, Li M, Freudenberger P, Hofer E, Schmidt H, Schmidt R, Holliday EG, Mitchell P, Wang JJ, de Boer IH, Li G, Siscovick DS, Kutalik Z, Corre T, Vollenweider P, Waeber G, Gupta J, Kanetsky PA, Hwang SJ, Olden M, Yang Q, de Andrade M, Atkinson EJ, Kardia SL, Turner ST, Stafford JM, Ding J, Liu Y, Barlassina C, Cusi D, Salvi E, Staessen JA, Ridker PM, Grallert H, Meisinger C, Müller-Nurasyid M, Krämer BK, Kramer H, Rosas SE, Nolte IM, Penninx BW, Snieder H, Fabiola Del Greco M, Franke A, Nöthlings U, Lieb W, Bakker SJ, Gansevoort RT, van der Harst P, Dehghan A, Franco OH, Hofman A, Rivadeneira F, Sedaghat S, Uitterlinden AG, Coassin S, Haun M, Kollerits B, Kronenberg F, Paulweber B, Aumann N, Endlich K, Pietzner M, Völker U, Rettig R, Chouraki V, Helmer C, Lambert JC, Metzger M, Stengel B, Lehtimäki T, Lyytikäinen LP, Raitakari O, Johnson A, Parsa A, Bochud M, Heid IM, Goessling W, Köttgen A, Kao WH, Fox CS, Böger CA: Genome-wide association study of kidney function decline in individuals of European descent. Kidney Int. 2015;87:1017-1029.

2. Pattaro C, Köttgen A, Teumer A, Garnaas M, Böger CA, Fuchsberger C, Olden M, Chen MH, Tin A, Taliun D, Li M, Gao X, Gorski M, Yang Q, Hundertmark C, Foster MC, O'Seaghdha CM, Glazer N, Isaacs A, Liu CT, Smith AV, O'Connell JR, Struchalin M, Tanaka T, Li G, Johnson AD, Gierman HJ, Feitosa M, Hwang SJ, Atkinson EJ, Lohman K, Cornelis MC, Johansson Å, Tönjes A, Dehghan A, Chouraki V, Holliday EG, Sorice R, Kutalik Z, Lehtimäki T, Esko T, Deshmukh H, Ulivi S, Chu AY, Murgia F, Trompet S, Imboden M, Kollerits B, Pistis G; CARDIoGRAM Consortium; ICBP Consortium; CARe Consortium; Wellcome Trust Case Control Consortium 2 (WTCCC2), Harris TB, Launer LJ, Aspelund T, Eiriksdottir G, Mitchell BD, Boerwinkle E, Schmidt H, Cavalieri M, Rao M, Hu FB, Demirkan A, Oostra BA, de Andrade M, Turner ST, Ding J, Andrews JS, Freedman BI, Koenig W, Illig T, Döring A, Wichmann HE, Kolcic I, Zemunik T, Boban M, Minelli C, Wheeler HE, Igl W, Zaboli G, Wild SH, Wright AF, Campbell H, Ellinghaus D, Nöthlings U, Jacobs G, Biffar R, Endlich K, Ernst F, Homuth G, Kroemer HK, Nauck M, Stracke S, Völker U, Völzke H, Kovacs P, Stumvoll M, Mägi R, Hofman A, Uitterlinden AG, Rivadeneira F, Aulchenko YS, Polasek O, Hastie N, Vitart V, Helmer C, Wang JJ, Ruggiero D, Bergmann S, Kähönen M, Viikari J, Nikopensius T, Province M, Ketkar S, Colhoun H, Doney A, Robino A, Giulianini F, Krämer BK, Portas L, Ford I, Buckley BM, Adam M, Thun GA, Paulweber B, Haun M, Sala C, Metzger M, Mitchell P, Ciullo M, Kim SK, Vollenweider P, Raitakari O, Metspalu A, Palmer C, Gasparini P, Pirastu M, Jukema JW, Probst-Hensch NM, Kronenberg F, Toniolo D, Gudnason V, Shuldiner AR, Coresh J, Schmidt R, Ferrucci L, Siscovick DS, van Duijn CM, Borecki I, Kardia SL, Liu Y, Curhan GC, Rudan I, Gyllensten U, Wilson JF, Franke A, Pramstaller PP, Rettig R, Prokopenko I, Witteman JC, Hayward C, Ridker P, Parsa A, Bochud M, Heid IM, Goessling W, Chasman DI, Kao WH, Fox CS: Genome-wide association and functional follow-up reveals new loci for kidney function. PLoS Genet. 2012;8:e1002584.

3. Okada Y, Sim X, Go MJ, Wu JY, Gu D, Takeuchi F, Takahashi A, Maeda S, Tsunoda T, Chen P, Lim SC,

Wong TY, Liu J, Young TL, Aung T, Seielstad M, Teo YY, Kim YJ, Lee JY, Han BG, Kang D, Chen CH, Tsai FJ, Chang LC, Fann SJ, Mei H, Rao DC, Hixson JE, Chen S, Katsuya T, Isono M, Ogihara T, Chambers JC, Zhang W, Kooner JS; KidneyGen Consortium.; CKDGen Consortium., Albrecht E; GUGC consortium., Yamamoto K, Kubo M, Nakamura Y, Kamatani N, Kato N, He J, Chen YT, Cho YS, Tai ES, Tanaka T: Meta-analysis identifies multiple loci associated with kidney function-related traits in east Asian populations. Nat Genet 2012;44:904-909.

4. Köttgen A, Glazer NL, Dehghan A, Hwang SJ, Katz R, Li M, Yang Q, Gudnason V, Launer LJ, Harris TB, Smith AV, Arking DE, Astor BC, Boerwinkle E, Ehret GB, Ruczinski I, Scharpf RB, Chen YD, de Boer IH, Haritunians T, Lumley T, Sarnak M, Siscovick D, Benjamin EJ, Levy D, Upadhyay A, Aulchenko YS, Hofman A, Rivadeneira F, Uitterlinden AG, van Duijn CM, Chasman DI, Paré G, Ridker PM, Kao WH, Witteman JC, Coresh J, Shlipak MG, Fox CS: Multiple loci associated with indices of renal function and chronic kidney disease. Nat Genet 2009;41:712-717.

5. Böger CA, Gorski M, Li M, Hoffmann MM, Huang C, Yang Q, Teumer A, Krane V, O'Seaghdha CM, Kutalik Z, Wichmann HE, Haak T, Boes E, Coassin S, Coresh J, Kollerits B, Haun M, Paulweber B, Köttgen A, Li G, Shlipak MG, Powe N, Hwang SJ, Dehghan A, Rivadeneira F, Uitterlinden A, Hofman A, Beckmann JS, Krämer BK, Witteman J, Bochud M, Siscovick D, Rettig R, Kronenberg F, Wanner C, Thadhani RI, Heid IM, Fox CS, Kao WH; CKDGen Consortium: Association of eGFR-Related Loci Identified by GWAS with Incident CKD and ESRD. PLoS Genet. 2011;7:e1002292.

6. Köttgen A, Pattaro C, Böger CA, Fuchsberger C, Olden M, Glazer NL, Parsa A, Gao X, Yang Q, Smith AV, O'Connell JR, Li M, Schmidt H, Tanaka T, Isaacs A, Ketkar S, Hwang SJ, Johnson AD, Dehghan A, Teumer A, Paré G, Atkinson EJ, Zeller T, Lohman K, Cornelis MC, Probst-Hensch NM, Kronenberg F, Tönjes A, Hayward C, Aspelund T, Eiriksdottir G, Launer LJ, Harris TB, Rampersaud E, Mitchell BD, Arking DE, Boerwinkle E, Struchalin M, Cavalieri M, Singleton A, Giallauria F, Metter J, de Boer IH, Haritunians T, Lumley T, Siscovick D, Psaty BM, Zillikens MC, Oostra BA, Feitosa M, Province M, de Andrade M, Turner ST, Schillert A, Ziegler A, Wild PS, Schnabel RB, Wilde S, Munzel TF, Leak TS, Illig T, Klopp N, Meisinger C, Wichmann HE, Koenig W, Zgaga L, Zemunik T, Kolcic I, Minelli C, Hu FB, Johansson A, Igl W, Zaboli G, Wild SH, Wright AF, Campbell H, Ellinghaus D, Schreiber S, Aulchenko YS, Felix JF, Rivadeneira F, Uitterlinden AG, Hofman A, Imboden M, Nitsch D, Brandstätter A, Kollerits B, Kedenko L, Mägi R, Stumvoll M, Kovacs P, Boban M, Campbell S, Endlich K, Völzke H, Kroemer HK, Nauck M, Völker U, Polasek O, Vitart V, Badola S, Parker AN, Ridker PM, Kardia SL, Blankenberg S, Liu Y, Curhan GC, Franke A, Rochat T, Paulweber B, Prokopenko I, Wang W, Gudnason V, Shuldiner AR, Coresh J, Schmidt R, Ferrucci L, Shlipak MG, van Duijn CM, Borecki I, Krämer BK, Rudan I, Gyllensten U, Wilson JF, Witteman JC, Pramstaller PP, Rettig R, Hastie N, Chasman DI, Kao WH, Heid IM, Fox CS: New loci associated with kidney function and chronic kidney disease. Nat Genet 2010;42:376-384.

7. Kashyap MV, Nolan M, Sprouse M, Chakraborty R, Cross D, Roby R, Vishwanatha JK: Role of genomics in eliminating health disparities. J Carcinog 2015;14:6.

8. Nakamura Y: The BioBank Japan Project. Clin Adv Hematol Oncol 2007;5:696-697.

9. Hamajima N; J-MICC Study Group: The Japan Multi-Institutional Collaborative Cohort Study (J-MICC Study) to detect gene-environment interactions for cancer. Asian Pac J Cancer Prev 2007;8:317-323.

10. Wakai K, Hamajima N, Okada R, Naito M, Morita E, Hishida A, Kawai S, Nishio K, Yin G, Asai Y, Matsuo K, Hosono S, Ito H, Watanabe M, Kawase T, Suzuki T, Tajima K, Tanaka K, Higaki Y, Hara M, Imaizumi T, Taguchi N, Nakamura K, Nanri H, Sakamoto T, Horita M, Shinchi K, Kita Y, Turin TC, Rumana N, Matsui K, Miura K, Ueshima H, Takashima N, Nakamura Y, Suzuki S, Ando R, Hosono A, Imaeda N, Shibata K, Goto C, Hattori N, Fukatsu M, Yamada T, Tokudome S, Takezaki T, Niimura H, Hirasada K, Nakamura A, Tatebo M, Ogawa S, Tsunematsu N, Chiba S, Mikami H, Kono S, Ohnaka K, Takayanagi R, Watanabe Y, Ozaki E, Shigeta M, Kuriyama N, Yoshikawa A, Matsui D, Watanabe I, Inoue K, Ozasa K, Mitani S, Arisawa K, Uemura H, Hiyoshi M, Takami H, Yamaguchi M, Nakamoto M, Takeda H, Kubo M, Tanaka H; J-MICC Study Group: Profile of Participants and Genotype Distributions of 108 Polymorphisms in a Cross-sectional Study to Elucidate Associations between Genotypes and Lifestyle and Clinical Factors: A Project in the Japan Multi-institutional Collaborative Cohort (J-MICC) Study. J Epidemiol 2010;21:223-235.

11. Tokudome S, Goto C, Imaeda N, Tokudome Y, Ikeda M, Maki S: Development of a data-based short food frequency questionnaire for assessing nutrient intake by middle-aged Japanese. Asian Pac J Cancer Prev 2004;5:40-43.

12. Tokudome Y, Goto C, Imaeda N, Hasegawa T, Kato R, Hirose K, Tajima K, Tokudome S: Relative validity of a short food frequency questionnaire for assessing nutrient intake versus three-day weighed diet records in middle-aged Japanese. J Epidemiol 2005;15:135-145.

13. Goto C, Tokudome Y, Imaeda N, Takekuma K, Kuriki K, Igarashi F, Ikeda M, Tokudome S: Validation study of fatty acid consumption assessed with a short food frequency questionnaire against plasma concentration in middle-aged Japanese people. Scand J Nutr 2006;50:77-82.

14. Imaeda N, Goto C, Tokudome Y, Hirose K, Tajima K, Tokudome S: Reproducibility of a short food frequency questionnaire for Japanese general population. J Epidemiol 2007;17:100-107.

15. International Expert Committee: International Expert Committee Report
on the Role of the A1C Assay in the Diagnosis of Diabetes. Diabetes Care 2009;32:1327-1334.

16. Ma S, Dai Y: Principal component analysis based methods in bioinformatics studies. Brief Bioinform. 2011;12:714-722.

17. Matsuo S, Imai E, Horio M, Yasuda Y, Tomita K, Nitta K, Yamagata K, Tomino Y, Yokoyama H, Hishida A; Collaborators developing the Japanese equation for estimated GFR: Revised equations for estimated GFR from serum creatinine in Japan. Am J Kidney Dis 2009;53:982-992.

18. Nagai A, Hirata M, Kamatani Y, Muto K, Matsuda K, Kiyohara Y, Ninomiya T, Tamakoshi A, Yamagata Z, Mushiroda T, Murakami Y, Yuji K, Furukawa Y, Zembutsu H, Tanaka T, Ohnishi Y, Nakamura Y; BioBank Japan Cooperative Hospital Group., Kubo M. Overview of the BioBank Japan Project: Study design and profile. J Epidemiol 2017;27:S2-S8.

19. Devlin B, Roeder K, Wasserman L. Genomic control, a new approach to genetic-based association studies. Theor Popul Biol. 2001;60:155-66.

20. Wood EJ: The encyclopedia of molecular biology. Biochemical Education 1995;23:105.

21. Pattaro C, Teumer A, Gorski M, Chu AY, Li M, Mijatovic V, Garnaas M, Tin A, Sorice R, Li Y, Taliun D, Olden M, Foster M, Yang Q, Chen MH, Pers TH, Johnson AD, Ko YA, Fuchsberger C, Tayo B, Nalls M, Feitosa MF, Isaacs A, Dehghan A, d'Adamo P, Adeyemo A, Dieffenbach AK, Zonderman AB, Nolte IM, van der Most PJ, Wright AF, Shuldiner AR, Morrison AC, Hofman A, Smith AV, Dreisbach AW, Franke A, Uitterlinden AG, Metspalu A, Tonjes A, Lupo A, Robino A, Johansson Å, Demirkan A, Kollerits B, Freedman BI, Ponte B, Oostra BA, Paulweber B, Krämer BK, Mitchell BD, Buckley BM, Peralta CA, Hayward C, Helmer C, Rotimi CN, Shaffer CM, Müller C, Sala C, van Duijn CM, Saint-Pierre A, Ackermann D, Shriner D, Ruggiero D, Toniolo D, Lu Y, Cusi D, Czamara D, Ellinghaus D, Siscovick DS, Ruderfer D, Gieger C, Grallert H, Rochtchina E, Atkinson EJ, Holliday EG, Boerwinkle E, Salvi E, Bottinger EP, Murgia F, Rivadeneira F, Ernst F, Kronenberg F, Hu FB, Navis GJ, Curhan GC, Ehret GB, Homuth G, Coassin S, Thun GA, Pistis G, Gambaro G, Malerba G, Montgomery GW, Eiriksdottir G, Jacobs G, Li G, Wichmann HE, Campbell H, Schmidt H, Wallaschofski H, Völzke H, Brenner H, Kroemer HK, Kramer H, Lin H, Leach IM, Ford I, Guessous I, Rudan I, Prokopenko I, Borecki I, Heid IM, Kolcic I, Persico I, Jukema JW, Wilson JF, Felix JF, Divers J, Lambert JC, Stafford JM, Gaspoz JM, Smith JA, Faul JD, Wang JJ, Ding J, Hirschhorn JN, Attia J, Whitfield JB, Chalmers J, Viikari J, Coresh J, Denny JC, Karjalainen J, Fernandes JK, Endlich K, Butterbach K, Keene KL, Lohman K, Portas L, Launer LJ, Lyytikäinen LP, Yengo L, Franke L, Ferrucci L, Rose LM, Kedenko L, Rao M, Struchalin M, Kleber ME, Cavalieri M, Haun M, Cornelis MC, Ciullo M, Pirastu M, de Andrade M, McEvoy MA, Woodward M, Adam M, Cocca M, Nauck M, Imboden M, Waldenberger M, Pruijm M, Metzger M, Stumvoll M, Evans MK, Sale MM, Kähönen M, Boban M, Bochud M, Rheinberger M, Verweij N, Bouatia-Naji N, Martin NG, Hastie N, Probst-Hensch N, Soranzo N, Devuyst O, Raitakari O, Gottesman O, Franco OH, Polasek O, Gasparini P, Munroe PB, Ridker PM, Mitchell P,

Muntner P, Meisinger C, Smit JH; ICBP Consortium; AGEN Consortium; CARDIOGRAM; CHARGe-Heart Failure Group; ECHOGen Consortium, Kovacs P, Wild PS, Froguel P, Rettig R, Mägi R, Biffar R, Schmidt R, Middelberg RP, Carroll RJ, Penninx BW, Scott RJ, Katz R, Sedaghat S, Wild SH, Kardia SL, Ulivi S, Hwang SJ, Enroth S, Kloiber S, Trompet S, Stengel B, Hancock SJ, Turner ST, Rosas SE, Stracke S, Harris TB, Zeller T, Zemunik T, Lehtimäki T, Illig T, Aspelund T, Nikopensius T, Esko T, Tanaka T, Gyllensten U, Völker U, Emilsson V, Vitart V, Aalto V, Gudnason V, Chouraki V, Chen WM, Igl W, März W, Koenig W, Lieb W, Loos RJ, Liu Y, Snieder H, Pramstaller PP, Parsa A, O'Connell JR, Susztak K, Hamet P, Tremblay J, de Boer IH, Böger CA, Goessling W, Chasman DI, Köttgen A, Kao WH, Fox CS: Genetic associations at 53 loci highlight cell types and biological pathways relevant for kidney function. Nat Commun 2016;7:10023.

22. Raychowdhury R., Niles JL, McCluskey RT, Smith JA: Autoimmune target in Heymann nephritis is a glycoprotein with homology to the LDL receptor. Science 1989;244:1163-1165.

23. Nykjaer A, Dragun D, Walther D, Vorum H, Jacobsen C, Herz J, Melsen F, Christensen EI, Willnow TE: An endocytic pathway essential for renal uptake and activation of the steroid 25-(OH) vitamin D3. Cell 1999;96:507-515.

24. Caruso-Neves C, Pinheiro AA, Cai H, Souza-Menezes J, Guggino WB: PKB and megalin determine the survival or death of renal proximal tubule cells. Proc Natl Acad Sci U S A 2006;103:18810-18805.

25. Devriendt K, Standaert L, Van Hole C, Devlieger H, Fryns JP: Proteinuria in a patient with the diaphragmatic hernia-hypertelorism-myopia-deafness syndrome: further evidence that the facio-oculo-acoustico-renal syndrome represents the same entity. J Med Genet 1998;35:70-71.

26. Huang GN, Huso DL, Bouyain S, Tu J, McCorkell KA, May MJ, Zhu Y, Lutz M, Collins S, Dehoff M, Kang S, Whartenby K, Powell J, Leahy D, Worley PF: NFAT binding and regulation of T cell activation by the cytoplasmic scaffolding Homer proteins. Science 2008;319:476-481.

27. Li X, Ho SN, Luna J, Giacalone J, Thomas DJ, Timmerman LA, Crabtree GR, Francke U: Cloning and chromosomal localization of the human and murine genes for the T-cell transcription factors NFATc and NFATp. Cytogenet. Cell Genet 1995;68:185-191.

28. Peng SL, Gerth AJ, Ranger AM, Glimcher LH: NFATc1 and NFATc2 together control both T and B cell activation and differentiation. Immunity 2001;14: 13-20.

29. Zhou P, Sun LJ, Dotsch V, Wagner G, Verdine GL: Solution structure of the core NFATC1/DNA complex. Cell 1998;92:687-696.

30. Lee MY, Garvey SM, Baras AS, Lemmon JA, Gomez MF, Schoppee Bortz PD, Daum G, LeBoeuf RC, Wamhoff BR: Integrative genomics identifies DSCR1 (RCAN1) as a novel NFAT-dependent mediator of phenotypic modulation in vascular smooth muscle cells. Hum Mol Genet 2010; 19: 468-479.

31. Mahajan A, Rodan AR, Le TH, Gaulton KJ, Haessler J, Stilp AM, Kamatani Y, Zhu G, Sofer T, Puri S, Schellinger JN, Chu PL, Cechova S, van Zuydam N; SUMMIT Consortium; BioBank Japan Project, Arnlov J, Flessner MF, Giedraitis V, Heath AC, Kubo M, Larsson A, Lindgren CM, Madden PA, Montgomery GW, Papanicolaou GJ, Reiner AP, Sundström J, Thornton TA, Lind L, Ingelsson E, Cai J, Martin NG, Kooperberg C, Matsuda K, Whitfield JB, Okada Y, Laurie CC, Morris AP, Franceschini N: Trans-ethnic Fine Mapping Highlights Kidney-Function Genes Linked to Salt Sensitivity. Am J Hum Genet 2016; 99: 636-646.

32. Jain M, Bhat GP, Vijayraghavan K, Inamdar MS: Rudhira/BCAS3 is a cytoskeletal protein that controls Cdc42 activation and directional cell migration during angiogenesis. Exp Cell Res 2012;318:753-767.

33. Siva K, Venu P, Mahadevan A, S K S, Inamdar MS: Human BCAS3 expression in embryonic stem cells and vascular precursors suggests a role in human embryogenesis and tumor angiogenesis. PLoS One 2007;2:e1202.

34. Östensson M1, Montén C, Bacelis J, Gudjonsdottir AH, Adamovic S, Ek J, Ascher H, Pollak E, Arnell H, Browaldh L, Agardh D, Wahlström J, Nilsson S, Torinsson-Naluai Å. A possible mechanism behind autoimmune disorders discovered by genome-wide linkage and association analysis in celiac disease. PLoS One. 2013;8:e70174.

35. Online Mendelian Inheritance in Man, OMIM. Johns Hopkins University, Baltimore. MIM Number: {157147}:{09/08/2015}. Available at: http://www.ncbi.nlm.nih.gov/omim/. Accessed Mar. 1, 2018.

36. Koepsell H: The SLC22 family with transporters of organic cations, anions and zwitterions. Mol Aspects Med 2013;34:413-435.

37. Fujita T, Urban TJ, Leabman MK, Fujita K, Giacomini KM: Transport of drugs in the kidney by the human organic cation transporter, OCT2 and its genetic variants. J Pharm Sci 2006;95: 25-36.

38. Gorski M, van der Most PJ, Teumer A, Chu AY, Li M, Mijatovic V, Nolte IM, Cocca M, Taliun D, Gomez F, Li Y, Tayo B, Tin A, Feitosa MF, Aspelund T, Attia J, Biffar R, Bochud M, Boerwinkle E, Borecki I, Bottinger EP, Chen MH, Chouraki V, Ciullo M, Coresh J, Cornelis MC, Curhan GC, d'Adamo AP, Dehghan A, Dengler L, Ding J, Eiriksdottir G, Endlich K, Enroth S, Esko T, Franco OH, Gasparini P, Gieger C, Girotto G, Gottesman O, Gudnason V, Gyllensten U, Hancock SJ, Harris TB, Helmer C, Höllerer S, Hofer E, Hofman A,

Holliday EG, Homuth G, Hu FB, Huth C, Hutri-Kähönen N, Hwang SJ, Imboden M, Johansson Å, Kähönen M, König W, Kramer H, Krämer BK, Kumar A, Kutalik Z, Lambert JC, Launer LJ, Lehtimäki T, de Borst MH, Navis G, Swertz M, Liu Y, Lohman K, Loos RJF, Lu Y, Lyytikäinen LP, McEvoy MA, Meisinger C, Meitinger T, Metspalu A, Metzger M, Mihailov E, Mitchell P, Nauck M, Oldehinkel AJ, Olden M, Wjh Penninx B, Pistis G, Pramstaller PP, Probst-Hensch N, Raitakari OT, Rettig R, Ridker PM, Rivadeneira F, Robino A, Rosas SE, Ruderfer D, Ruggiero D, Saba Y, Sala C, Schmidt H, Schmidt R, Scott RJ, Sedaghat S, Smith AV, Sorice R, Stengel B, Stracke S, Strauch K, Toniolo D, Uitterlinden AG, Ulivi S, Viikari JS, Völker U, Vollenweider P, Völzke H, Vuckovic D, Waldenberger M, Jin Wang J, Yang Q, Chasman DI, Tromp G, Snieder H, Heid IM, Fox CS, Köttgen A, Pattaro C, Böger CA, Fuchsberger C: 1000 Genomes-based meta-analysis identifies 10 novel loci for kidney function. Sci Rep 2017;7:45040.

39. Ma J, Guan M, Bowden DW, Ng MC, Hicks PJ, Lea JP, Ma L, Gao C, Palmer ND, Freedman BI. Association Analysis of the Cubilin (CUBN) and Megalin (LRP2) Genes with ESRD in African Americans. Clin J Am Soc Nephrol. 2016; 11: 1034-1043.

40. Taylor AL: Making Medicine Precise and Personalized: What Can We Learn from the Past? Circ Heart Fail 2016;9:e002938.

Table 1. Characteristics of the study subjects.

	Discovery (J-MICC)	Replication (BBJ)
Number of analyzed samples	11,283	148,829
Sex: Male/Female	5,158/6,125	81,629/67,200
Age: mean(sd) (years)	54.9 (9.3)	63.6 (13.5)
SCr: mean (sd) (mg/dl)	0.72 (0.28)	0.91 (1.00)
eGFR: mean (sd) (ml/min/1.73m^2)	78.7 (15.1)	72.6 (25.0)
DM: N (%)	903 (8.0)	47,565 (32.0)
HT: N (%)	4,186 (37.1)	81,335 (54.6)
CKD: N (%)	939 (8.3)	38,143 (25.6)

SCr = serum creatinine; eGFR = estimated glomerular filtration rate; DM = diabetes mellitus; HT = hypertension; CKD = chronic kidney disease.

Table 2. Genetic variants suggestively associated with eGFR in the discovery phase ($p < 1\times10^{-6}$) and the results of association analyses in the replication and combined studies.

								Discovery (J-MICC)					Replication (BBJ)					Combined
rs ID	Geno/Imp	Cytoband	Position	Gene	Function	Effect allele	Ref. allele	EAF	r^2	β	SE	*p* value	EAF	r^2	β	SE	*p* value	*p* value
rs72878458	Imputed	2q31.1	170141086	*LRP2*	intronic	*T*	*C*	0.182	0.838	-0.0165	-0.0007	6.29×10^{-7}	0.18	0.848	-0.0003	0.0021	0.886	0.144
rs16856823	Imputed	2q31.1	170200452	*LRP2*	intronic	*T*	*A*	0.187	0.92	0.0183	0.0006	2.80×10^{-8}	0.181	0.917	0.0073	0.002	0.00029	6.71×10^{-07}
rs3770636	Imputed	2q31.1	170202833	*LRP2*	intronic	*G*	*T*	0.187	0.91	0.0184	0.0006	2.54×10^{-8}	0.181	0.91	0.0074	0.002	0.00026	5.79×10^{-07}
rs3770637	Imputed	2q31.1	170203104	*LRP2*	intronic	*C*	*T*	0.186	0.907	0.0183	0.0006	2.85×10^{-8}	0.181	0.907	0.0073	0.002	0.00028	6.34×10^{-07}
rs200309784	Imputed	2q31.1	170204098	*LRP2*	intronic	*GA*	*G*	0.208	0.829	0.0167	0.0006	1.37×10^{-7}	0.206	0.808	0.0077	0.002	0.00016	4.87×10^{-07}
rs2673171	Imputed	2q31.1	170204641	*LRP2*	intronic	*G*	*A*	0.185	0.887	0.0183	0.0006	3.50×10^{-8}	0.182	0.892	0.0072	0.002	0.00038	9.92×10^{-07}
rs3821129	Imputed	2q31.1	170204773	*LRP2*	intronic	*C*	*T*	0.182	0.884	0.0183	0.0006	3.59×10^{-8}	0.18	0.89	0.0074	0.002	0.00028	6.92×10^{-07}
rs2390793	Imputed	2q31.1	170205123	*LRP2*	intronic	*T*	*C*	0.186	0.867	0.0173	0.0006	1.71×10^{-7}	0.188	0.851	0.0076	0.0021	0.00022	7.44×10^{-07}
rs270188	Genotyped	5p15.32	5124149	*LINC01020,CTD-2297D10.2*	intergenic	*A*	*G*	0.432	1	-0.0126	-0.0005	9.91×10^{-7}	0.426	0.988	0.0018	0.0015	0.234	0.879
rs270184	Imputed	5p15.32	5124579	*LINC01020,CTD-2297D10.2*	intergenic	*T*	*C*	0.417	0.991	-0.013	-0.0005	4.94×10^{-7}	0.41	0.968	0.0015	0.0015	0.325	0.699
rs3785842	Imputed	17q23.2	59446530	*BCAS3*	intronic	*G*	*C*	0.428	0.943	0.015	0.0005	7.69×10^{-9}	0.438	0.931	0.0101	0.0015	8.21×10^{-11}	**6.35×10^{-15}**
rs3785841	Imputed	17q23.2	59446541	*BCAS3*	intronic	*G*	*C*	0.449	0.941	0.0155	0.0004	2.06×10^{-9}	0.455	0.933	0.0097	0.0015	3.44×10^{-10}	**2.12×10^{-14}**
rs11653176	Imputed	17q23.2	59447369	*BCAS3*	intronic	*C*	*T*	0.491	0.939	0.0148	0.0004	7.82×10^{-9}	0.515	0.868	0.0099	0.0016	5.91×10^{-10}	**6.24×10^{-14}**
rs398031258	Imputed	17q23.2	59447632	*BCAS3*	intronic	*CT*	*C*	0.469	0.846	0.0151	0.0004	5.35×10^{-9}	0.456	0.804	0.0107	0.0017	1.21×10^{-10}	**8.79×10^{-15}**
rs7217891	Imputed	17q23.2	59447984	*BCAS3*	intronic	*A*	*G*	0.512	0.934	0.0157	0.0004	1.03×10^{-9}	0.514	0.923	0.0103	0.0015	1.85×10^{-11}	**5.64×10^{-16}**
rs1010269	Imputed	17q23.2	59448945	*BCAS3*	intronic	*G*	*A*	0.485	0.929	0.0147	0.0005	1.12×10^{-8}	0.522	0.853	0.0118	0.0016	2.21×10^{-13}	**8.79×10^{-18}**
rs4968556	Imputed	17q23.2	59449082	*BCAS3*	intronic	*C*	*T*	0.484	0.949	-0.0149	-0.0004	7.16×10^{-9}	0.472	0.867	-0.0118	0.0016	1.07×10^{-13}	**3.28×10^{-18}**
rs11650989	Genotyped	17q23.2	59449636	*BCAS3*	intronic	*A*	*G*	0.284	0.995	0.0152	0.0005	1.02×10^{-7}	0.286	0.996	0.0087	0.0016	1.31×10^{-7}	**7.99×10^{-11}**
rs11657044	Imputed	17q23.2	59450105	*BCAS3*	intronic	*C*	*T*	0.513	0.979	0.0154	0.0004	2.65×10^{-9}	0.518	0.889	0.0117	0.0016	1.11×10^{-13}	**2.33×10^{-18}**
rs9905274	Imputed	17q23.2	59450441	*BCAS3*	intronic	*C*	*T*	0.494	0.997	0.0158	0.0004	8.28×10^{-10}	0.496	0.897	0.0106	0.0016	9.74×10^{-12}	**2.49×10^{-16}**
rs34754126	Imputed	17q23.2	59450453	*BCAS3*	intronic	*C*	*CT*	0.518	0.92	0.0141	0.0005	4.44×10^{-8}	0.525	0.839	0.0125	0.0016	1.51×10^{-14}	**7.72×10^{-19}**
rs9895661	Genotyped	17q23.2	59456589	*BCAS3*	intronic	*T*	*C*	0.456	0.999	0.0166	0.0004	1.55×10^{-10}	0.462	0.998	0.0117	0.0015	3.30×10^{-15}	**1.47×10^{-20}**
rs2079742	Imputed	17q23.2	59465697	*BCAS3*	intronic	*C*	*T*	0.464	0.855	-0.0167	-0.0004	1.16×10^{-10}	0.524	0.845	-0.0126	0.0016	5.93×10^{-15}	**2.56×10^{-20}**
rs11079428	Imputed	17q23.2	59466701	*BCAS3*	intronic	*A*	*T*	0.439	0.845	0.0167	0.0004	1.30×10^{-10}	0.451	0.83	0.0118	0.0016	5.10×10^{-13}	**4.33×10^{-18}**
rs3785837	Imputed	17q23.2	59468942	*BCAS3*	intronic	*A*	*G*	0.448	0.839	0.0171	0.0004	4.23×10^{-11}	0.455	0.829	0.0114	0.0016	2.75×10^{-12}	**2.07×10^{-17}**
rs74183647	Imputed	18q23	77156171	*NFATC1*	UTR5	*C*	*G*	0.482	0.714	0.0127	0.0005	6.56×10^{-7}	0.455	0.703	0.0098	0.0018	2.72×10^{-8}	**2.40×10^{-11}**
rs117314773	Imputed	18q23	77160066	*NFATC1*	intronic	*A*	*G*	0.497	0.767	0.0125	0.0005	9.47×10^{-7}	0.474	0.748	0.0092	0.0017	7.69×10^{-8}	**9.00×10^{-11}**
rs138901831	Imputed	18q23	77160067	*NFATC1*	intronic	*C*	*G*	0.497	0.767	0.0125	0.0005	9.47×10^{-7}	0.474	0.748	0.0092	0.0017	7.72×10^{-8}	**9.04×10^{-11}**

eGFR = estimated glomerular filtration rate; Geno/Imp = genotyped or imputed; EAF = effect allele frequency. Chromosomal locations were described based on hg19/GRCh37 coordinates. All the SNPs in the loci that reached the genome-wide suggestive level ($p < 1\times10^{-6}$) in the discovery phase are shown. *p* values that reached the genome-wide significance ($p < 5\times10^{-8}$) in the combined studies are indicated in boldface. "Combined" results indicate those for the GWAS meta-analysis of J-MICC and BBJ.

Table 3. Genetic variants suggestively associated with SCr in the discovery phase ($p < 1\times10^{-6}$) and the results of association analyses in the replication and combined studies.

								Discovery (J-MICC)					Replication (BBJ)					Combined
rs ID	Geno/Imp	Cytoband	Position	Gene	Function	Effect allele	Ref. allele	EAF	r^2	β	SE	*p* value	EAF	r^2	β	SE	*p* value	*p* value
rs72878458	Imputed	2q31.1	170141086	*LRP2*	intronic	*T*	*C*	0.182	0.838	0.0151	0.003	5.56×10^{-7}	0.18	0.848	0.0003	0.0019	0.883	0.141
rs16856823	Imputed	2q31.1	170200452	*LRP2*	intronic	*T*	*A*	0.187	0.92	-0.0168	0.003	2.24×10^{-8}	0.181	0.917	-0.0067	0.0018	0.000299	6.67×10^{-7}
rs3770636	Imputed	2q31.1	170202833	*LRP2*	intronic	*G*	*T*	0.187	0.91	-0.0169	0.003	2.04×10^{-8}	0.181	0.91	-0.0067	0.0018	0.00027	5.66×10^{-7}
rs3770637	Imputed	2q31.1	170203104	*LRP2*	intronic	*C*	*T*	0.186	0.907	-0.0169	0.003	2.26×10^{-8}	0.181	0.907	-0.0067	0.0018	0.00028	6.18×10^{-7}
rs200309784	Imputed	2q31.1	170204098	*LRP2*	intronic	*GA*	*G*	0.208	0.829	-0.0153	0.0029	1.14×10^{-7}	0.206	0.808	-0.007	0.0019	0.00017	4.82×10^{-7}
rs2673171	Imputed	2q31.1	170204641	*LRP2*	intronic	*G*	*A*	0.185	0.887	-0.0168	0.003	2.88×10^{-8}	0.182	0.892	-0.0066	0.0019	0.0004	9.76×10^{-7}
rs3821129	Imputed	2q31.1	170204773	*LRP2*	intronic	*C*	*T*	0.182	0.884	-0.0169	0.003	2.95×10^{-8}	0.18	0.89	-0.0068	0.0019	0.00029	6.73×10^{-7}
rs2390793	Imputed	2q31.1	170205123	*LRP2*	intronic	*T*	*C*	0.186	0.867	-0.0159	0.003	1.44×10^{-7}	0.188	0.851	-0.0069	0.0019	0.00027	7.37×10^{-7}
rs270188	Genotyped	5p15.32	5124149	*LINC01020,CTD-2297D10.2*	intergenic	*A*	*G*	0.432	1	0.0115	0.0024	9.90×10^{-7}	0.426	0.988	-0.0016	0.0014	0.256	0.838
rs270184	Imputed	5p15.32	5124579	*LINC01020,CTD-2297D10.2*	intergenic	*T*	*C*	0.417	0.991	0.0119	0.0024	4.77×10^{-7}	0.41	0.968	-0.0013	0.0014	0.3497	0.664
rs3785842	Imputed	17q23.2	59446530	*BCAS3*	intronic	*G*	*C*	0.428	0.943	-0.0138	0.0024	7.07×10^{-9}	0.438	0.931	-0.0093	0.0014	5.41×10^{-11}	**3.82×10^{-15}**
rs3785841	Imputed	17q23.2	59446541	*BCAS3*	intronic	*G*	*C*	0.449	0.941	-0.0142	0.0024	1.86×10^{-9}	0.455	0.933	-0.0089	0.0014	2.36×10^{-10}	**1.32×10^{-14}**
rs11653176	Imputed	17q23.2	59447369	*BCAS3*	intronic	*C*	*T*	0.509	0.939	-0.0135	0.0023	7.88×10^{-9}	0.515	0.868	-0.0091	0.0015	4.26×10^{-10}	**4.28×10^{-14}**
rs398031258	Imputed	17q23.2	59447632	*BCAS3*	intronic	*CT*	*C*	0.469	0.846	-0.0138	0.0024	5.16×10^{-9}	0.456	0.804	-0.0099	0.0015	8.22×10^{-11}	**5.53×10^{-15}**
rs7217891	Imputed	17q23.2	59447984	*BCAS3*	intronic	*A*	*G*	0.512	0.934	-0.0144	0.0023	9.51×10^{-10}	0.514	0.923	-0.0095	0.0014	1.29×10^{-11}	**3.61×10^{-16}**
rs1010269	Imputed	17q23.2	59448945	*BCAS3*	intronic	*G*	*A*	0.485	0.929	-0.0135	0.0024	1.15×10^{-8}	0.522	0.853	-0.0109	0.0015	1.41×10^{-13}	**5.40×10^{-18}**
rs4968556	Imputed	17q23.2	59449082	*BCAS3*	intronic	*C*	*T*	0.484	0.949	0.0136	0.0024	7.22×10^{-9}	0.472	0.867	0.0109	0.0015	7.02×10^{-14}	**2.05×10^{-18}**
rs11650989	Genotyped	17q23.2	59449636	*BCAS3*	intronic	*A*	*G*	0.284	0.995	-0.014	0.0026	8.51×10^{-8}	0.286	0.996	-0.008	0.0015	1.11×10^{-7}	**6.18×10^{-11}**
rs11657044	Imputed	17q23.2	59450105	*BCAS3*	intronic	*C*	*T*	0.513	0.979	-0.014	0.0024	2.70×10^{-9}	0.518	0.889	-0.0108	0.0014	7.04×10^{-14}	**1.40×10^{-18}**
rs9905274	Imputed	17q23.2	59450441	*BCAS3*	intronic	*C*	*T*	0.494	0.997	-0.0144	0.0023	8.61×10^{-10}	0.496	0.897	-0.0098	0.0014	6.51×10^{-12}	**1.59×10^{-16}**
rs34754126	Imputed	17q23.2	59450453	*BCAS3*	intronic	*C*	*CT*	0.482	0.92	-0.0129	0.0024	4.31×10^{-8}	0.525	0.839	-0.0115	0.0015	8.12×10^{-15}	**3.85×10^{-19}**
rs9895661	Genotyped	17q23.2	59456589	*BCAS3*	intronic	*T*	*C*	0.456	0.999	-0.0152	0.0024	1.42×10^{-10}	0.462	0.998	-0.0108	0.0014	1.97×10^{-15}	**7.91×10^{-21}**
rs2079742	Imputed	17q23.2	59465697	*BCAS3*	intronic	*C*	*T*	0.464	0.855	0.0153	0.0024	1.12×10^{-10}	0.524	0.845	0.0116	0.0015	3.36×10^{-15}	**1.32×10^{-20}**
rs11079428	Imputed	17q23.2	59466701	*BCAS3*	intronic	*A*	*T*	0.439	0.845	-0.0152	0.0024	1.37×10^{-10}	0.451	0.83	-0.0109	0.0015	3.56×10^{-13}	**2.92×10^{-18}**
rs3785837	Imputed	17q23.2	59468942	*BCAS3*	intronic	*A*	*G*	0.448	0.839	-0.0156	0.0024	4.56×10^{-11}	0.455	0.829	-0.0105	0.0015	2.06×10^{-12}	**1.51×10^{-17}**
rs74183647	Imputed	18q23	77156171	*NFATC1*	UTR5	*C*	*G*	0.482	0.714	-0.0116	0.0023	6.27×10^{-7}	0.455	0.703	-0.009	0.0016	2.97×10^{-8}	**2.61×10^{-11}**
rs117314773	Imputed	18q23	77160066	*NFATC1*	intronic	*A*	*G*	0.497	0.767	-0.0114	0.0023	9.81×10^{-7}	0.474	0.748	-0.0084	0.0016	7.37×10^{-8}	**8.67×10^{-11}**
rs138901831	Imputed	18q23	77160067	*NFATC1*	intronic	*C*	*G*	0.497	0.767	-0.0114	0.0023	9.81×10^{-7}	0.474	0.748	-0.0084	0.0016	7.39×10^{-8}	**8.71×10^{-11}**

SCr = serum creatinine; Geno/Imp = genotyped or imputed; EAF = effect allele frequency. Chromosomal locations were described based on hg19/GRCh37 coordinates. All the SNPs in the loci that reached the genome-wide suggestive level ($p < 1\times10^{-6}$) in the discovery phase are shown. *p* values that reached the genome-wide significance ($p < 5\times10^{-8}$) in the combined studies are indicated in boldface. "Combined" results indicate those for the GWAS meta-analysis of J-MICC and BBJ.

Table 4. Genetic variants suggestively associated with the risk of CKD in the discovery phase ($p < 1\times10^{-6}$) and the results of association analyses in the replication and combined studies.

								Discovery (J-MICC)					Replication (BBJ)					Combined
rs ID	Geno/Imp	Cytoband	Position	Gene	Function	Effect allele	Ref. allele	EAF	r^2	β	SE	*p* value	EAF	r^2	β	SE	*p* value	*p* value
rs78351985	Imputed	4q23	100576689	*MTTP,DAPP1*	intergenic	*G*	*A*	0.071	0.916	0.416	0.085	9.55×10^{-7}	0.071	0.929	0.036	0.015	0.0181	0.00034
rs555786707	Imputed	4q23	100969329	*LOC256880,DDIT4L*	intergenic	*A*	*G*	0.056	0.825	0.493	0.094	1.78×10^{-7}	0.056	0.835	0.077	0.031	0.0213	0.00031

CKD = chronic kidney disease, Geno/Imp = genotyped or imputed; EAF = effect allele frequency. Chromosomal locations were described based on hg19/GRCh37 coordinates. All the SNPs in the loci that reached the genome-wide suggestive level ($P < 1\times10^{-6}$) in the discovery phase are shown. "Combined" results indicate those for the GWAS meta-analysis of J-MICC and BBJ.

(Figure Legends)

Fig. 1. Manhattan plots for the GWAS of renal functions.

(Footnotes for Fig. 1)

Manhattan plots for the eGFR QTL (a), SCr QTL (b) and CKD binary (c) GWAS's. Gene names that reached the genome-wide significance ($P < 5x10^{-8}$) in the combined study are shown. GWAS, genome-wide-association study; eGFR, estimated glomerular filtration rate; SCr, serum creatinine, QTL, quantitative trait loci; CKD, chronic kidney disease.

Fig. 2. Q-Q plots for the GWAS of renal functions.

(Footnotes for Fig. 2)

Quantile-quantile (Q-Q) plots for the eGFR QTL (a), SCr QTL (b) and CKD binary (c) GWAS's (after genomic control correction).

Fig. 3. Regional plots of the detected loci.

(Footnotes for Fig. 3)

Detailed regional plots for the loci detected on *LRP2* on 2q31.1 (a), the *BCAS3* locus on 17q23.2 (c), and the *NFATC1* locus on 18q23 (e) in association with eGFR. Regional plots for the same set of loci as above in association with SCr (b, d and f), and the *MTTP-DDIT4L* locus on 4q23 (g) drawn based on the 1000 Genomes Phase 3 JPT (Japanese in Tokyo) data. The plots conditioned on the lead SNPs (variants) in each loci are shown. The p values are based on the discovery (J-MICC) data. eGFR = estimated glomerular filtration rate; SCr = serum creatinine. The x axis represents chromosome position, and the y axis represents the −log10 (*p* value) of association. The purple diamond represents the lead variant. Colors represent the degree of LD (r^2) between each variant and the lead variant. The LD (r^2) was calculated based on the 1000 genomes phase 3 JPT individuals.

- Original Report -

Genome-wide association study of renal function traits: Results from the J-MICC Study

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Key Words: Chronic kidney disease, genome-wide association study, population-based cohort

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Running Head: CKD imputed GWAS in Japanese participants

Abstract

Background: Chronic kidney disease (CKD) is a rapidly growing worldwide public health problem. Recent advances in genome-wide-association studies (GWAS) revealed several genetic loci associated with renal function traits worldwide. ***Methods:*** We investigated the association of genetic factors with the levels of serum creatinine (SCr) and estimated glomerular filtration rate (eGFR) in Japanese population-based cohorts analyzing the GWAS imputed data with 11,221 subjects and 12,617,569 variants, and replicated the findings with the 148,829 hospital-based Japanese subjects. ***Results:*** ~~Results:~~ In the discovery phase, 28 variants within four loci (chromosome [chr] 2 with eight variants including rs3770636 in the *LRP2* gene locus, on chr 5 with two variants including rs270184, chr 17 with 15 variants including rs3785837 in the *BCAS3* gene locus, and chr 18 with three variants including rs74183647 in the *NFATC1* gene locus) reached the suggestive level of $p < 1\times10^{-6}$ in association with eGFR and SCr, and 2 variants on chr 4 (including rs78351985 in the *MTTP* gene locus) fulfilled the suggestive level in association with the risk of CKD. In the replication phase, 25 variants within three loci (chr 2 with seven variants, chr17 with 15 variants and chr 18 with three variants) in association with eGFR and SCr, and two variants on chr 4 associated with the risk of CKD became nominally statistically significant after Bonferroni correction, among which 15 variants on chr 17 and three variants on chr 18 reached genome-wide significance of $p < 5\times10^{-8}$ in the combined study meta-analysis. The associations of the loci on chr 2 and 18 with eGFR and SCr as well as that on chr 4 with CKD risk have not been previously reported in the Japanese and East Asian populations. ***Conclusion:*** Although the present GWAS of renal function traits included the largest sample of Japanese participants to date, we did not

identify novel loci for renal traits. However, we identified the novel associations of the genetic loci on chr 2, 4, and 18 with renal function traits in the Japanese population, suggesting these are transethnic loci. Further investigations of these associations are expected to further validate our findings, for the potential establishment of personalized prevention of renal disease in the Japanese and East Asian populations.

Key Words

Chronic kidney disease, genome-wide association study, population-based cohort

Introduction

Chronic kidney disease (CKD) is a worldwide public health problem that is growing rapidly. Recent advances in genome-wide-association studies (GWASs) revealed several genetic loci associated with renal function traits in European populations [1, 2], Asian populations [3] and worldwide [4]. For example, recent GWASs investigating the genetic loci associated with kidney function identified the *glucokinase regulator* (*GCKR*) locus on chromosome (chr) 2 as highly significant loci [5, 6]. Additionally, genotype distributions and factors related to lifestyles are known to differ according to races, ethnicity and nations, which may lead to the existence of diverse population-specific gene-environmental interactions, making the nation-based reports of genetic associations still meaningful for the effective prevention of disease in each population [7]. Recently, a meta-analysis in an East Asian population revealed 17 loci that were newly associated with renal function traits. However, the participants of which were multi-ethnic and were mostly diseased with regard to the Japanese individuals [3, 8]. We conducted a nation-wide genome cohort study to find genetic factors for the possible establishment of personalized prevention of human chronic diseases in Japanese, named the Japan Multi-institutional Collaborative Cohort (J-MICC) Study. Regarding this study, 100,000 participants from 12 areas in Japan have been recruited, and the GWAS genotyping with 14,000 subjects for 1,000,000 single nucleotide polymorphisms (SNPs) have been completed [9, 10]. Herein, we systematically and comprehensively investigated the association of genetic factors with the levels of serum creatinine (SCr) and estimated glomerular filtration rate (eGFR) in Japanese population-based cohorts, using GWAS data from the J-MICC Study.

Subjects and Methods

Study subjects

We analyzed the data repository of J-MICC Study, launched in 2005 in 10 areas of Japan, in which about 100,000 volunteers aged 35-69 years old provided their blood and lifestyle data based on a questionnaire, after providing informed consent [9, 10].

The present study included 14,539 randomly selected J-MICC Study participants from ~~each of the~~ 12 areas (Chiba, Sakuragaoka, Shizuoka-Daiko, Okazaki, Aichi, Takashima, Kyoto, Tokushima, Fukuoka, Kagoshima and Kyushu-KOPS [Kyushu Okinawa Population Study]) where the J-MICC Study took place. As a sample quality check (sample QC), participants with a proportion of identity by descent (IBD) over 0.1875, and outliers in the principal component analysis (PCA) were excluded, resulting in 14,086 participants for analysis. Among them, data for SCr were available for 11,681 subjects, and 398 subjects who had their creatinine measured with jaffe method were excluded, leaving 11,283 subjects (who had their creatitine measured with enzyme method) for the final analyses. Characteristics of the study subjects are shown in Table 1. Written informed consent was obtained from all participants. The protocol of this study was approved by the Ethics Review Committee of the Nagoya University Graduate School of Medicine (Approval No. 939-14), Aichi Cancer Center and all the participated institutions. All the research procedures were conducted according to the Ethical Guidelines for

Human Genome and Genetic Sequencing Research in Japan and the Declaration of Helsinki.

Questionnaire

Lifestyle-related information was collected using a self-administered questionnaire evaluated by trained staff. The questionnaire included items on smoking status, alcohol consumption, food consumption and medical history [11-14]. Information on medication and the presence/history of disease were also based on the self-reported questionnaire and the health checkup data at baseline. The presence of hypertension (HT) and/or diabetes mellitus (DM) was defined as having HT/DM as the present illness or taking medication for their treatment based on the questionnaire, or fulfilled the diagnostic criteria of HT/DM in the health checkup laboratory data (systolic blood pressure [BP] $\geq$ 140 mmHg or diastolic BP $\geq$ 90 mmHg for HT, and fasting blood glucose $\geq$ 126 mg/dL or hemoglobin A1c (HbA1c) $\geq$ 6.1% based on the Japan Diabetes Society criteria (equivalent to > 6.5% in National Glycohemoglobin Standardization Program criteria) for DM [15].

Genotyping and quality control filtering

DNA was extracted from buffy coat with a BioRobot M48 Workstation (QIAGEN Group, Tokyo). The genotyping was conducted by the RIKEN institute (Yokohama, Japan) using an Illumina OmniExpressExome Array (Illumina, San Diego, CA, USA) for the 964,193 SNPs. 26 samples with inconsistent sex information between questionnaire and an estimate from genotype were excluded. The identity-by-descent method implemented in the PLINK 1.9 software (https://www.cog-genomics.org/plink2) found 388 close relationship

pairs (pi-hat > 0.1875) and one sample of each pair was excluded. PCA [16] with a 1000 Genomes reference panel (phase 3) (http://www.internationalgenome.org/category/phase-3/) detected 34 subjects whose estimated ancestries were outside of the Japanese population. The 34 samples were excluded. All the remaining 14,091 samples met a sample-wise genotype call rate criterion (≥ 0.99). SNPs with a genotype call rate <0.98 and/or a Hardy-Weinberg equilibrium exact test p value < 1×10^{-6}, a low minor allele frequency (MAF) < 0.01, or a departure from the allele frequency computed from the 1000 Genomes Phase 3 EAS (East Asian) samples were excluded. Quality control filtering resulted in 14,091 individuals and 570,162 SNPs.

Genotype imputation

Genotype imputation was conducted using SHAPEIT ver.2 (https://mathgen.stats.ox.ac.uk/genetics_software/shapeit/shapeit.html#home) and Minimac3 (http://genome.sph.umich.edu/wiki/Minimac3) software based on the 1000 Genomes Project cosmopolitan reference panel (phase 3). After the genotype imputation, variants with MAF < 0.05 and r^2 < 0.3 were excluded, to examine the substantial effects of common variants on renal functions in Japanese, resulting in 6,288,024 variants provided for the final analyses.

Estimated glomerular filtration rate and definitions of chronic kidney disease

SCr was measured in all participants analyzed using an enzymatic method. The eGFR of each participant was calculated based on SCr, age, and sex using the Japanese eGFR equation proposed by the Japanese Society of

Nephrology: eGFR (ml/min/1.73 m^2) = 194 × SCr (mg/dl)$^{-1.094}$ × age$^{-0.287}$ (× 0.739 if female), which is the calibrated version from the CKD-EPI (Chronic Kidney Disease Epidemiology Collaboration) [17]. Values of eGFR equal to or more than 120 (ml/min/1.73 m^2) were winsorized at 120. In those who had eGFR values of equal to or more than 120, SCr corresponding to eGFR of 120 were calculated based on the Japanese eGFR equation. The prevalence of CKD was determined for CKD stages 3–5 (eGFR <60 ml/min/1.73 m^2).

Replication study

We conducted the replication study with an independent data set from the BioBank Japan Project (BBJ), which is consisted of 148,829 subjects (81,629 males and 67,200 females) with 46 diseases (nephrosis excluded), to examine the validity of the SNPs found in the GWAS of the J-MICC Study [18]. The exclusion criteria used consisted of: 1) age < 18, 2) PCA outlier from the EastAsian cluster, 3) Closely related samples determined by identity by state (IBS) (visual inspection), and 4) Nephrotic syndrome. Genotyping was conducted by using Illumina OmniExpressExome Array and OmniExpress+HumanExome Array (Illumina, San Diego, CA, USA). Pre-phasing was conducted with Eagle (https://data.broadinstitute.org/alkesgroup/Eagle/). Genotype imputation was conducted using Minimac3 (http://genome.sph.umich.edu/wiki/Minimac3) software based on the 1000 Genomes Project cosmopolitan reference panel (phase 3). Association analyses were conducted using R (ver 3.3.2, https://cran.r-project.org/).

Statistical analysis and graphics

We applied genomic control correction where the genomic control parameter lambda was >1.0 [19]. Thereafter, we examined the associations of the SNPs with the risk of CKD and the quantitative traits of serum creatinine and eGFR using the EPACTS software (http://genome.sph.umich.edu/wiki/EPACTS). The association of SNPs with presence/absence of CKD as a binary trait was examined by logistic Wald test, and the associations of SNPs with serum creatinine and eGFR as continuous variables were tested by linear Wald test. With regard to the covariates to be adjusted, gender, age, the presence of hypertension and diabetes mellitus, and the first five principal components (PCs) were included. Variants with the MAF of more than (or equal to) 0.05 were considered. The Manhattan and Q-Q Plots were drawn with the 'qqman' function in R (https://cran.r-project.org/web/packages/qqman/index.html). The GWAS meta-analysis was conducted using the METAL (http://genome.sph.umich.edu/wiki/METAL_Documentation), and the regional plots were constructed using the LocusZoom software (http://genome.sph.umich.edu/wiki/LocusZoom_Standalone). The genome-wide significance levels were set at $p < 5\times10^{-8}$, and the genome-wide suggestive levels were set at $p < 1\times10^{-6}$ in all the analyses. In the replication phase, the significance threshold was defined as p values less than 0.05 devided by the number of comparisons based on the Bonferroni correction. Lead SNPs (or variants) were defined as the SNPs (or the variants) that reached the smallest p values in each genetic locus defined as the position on a chromosome identified by the cytogenetic banding of the chromosome [20].

Results

Variants identified in the discovery phase GWAS

In the present Japanese population-based (J-MICC) GWAS, we conducted the quantitative trait loci (QTL) analysis with the eGFR and SCr as the continuous dependent variable, and then examined the risk of CKD defined as eGFR <60 ml/min/1.73 m^2 as binary outcomes. From the analysis of eGFR QTL GWAS, we identified four loci with 28 variants suggestively associated with eGFR (on chr 2, 5, 17 and 18; $p < 1\times10^{-6}$) (Table 2), whereas in the analysis of SCr QTL GWAS, we also identified four loci with 28 variants demonstrated suggestively associated with eGFR (on chr 2, 5, 17 and 18; $p < 1\times10^{-6}$) (Table 3). From the analysis of GWAS of the presence/absence of CKD, one locus on chr 4 (with 2 SNPs) reached the suggestive level in a single-variant-based analysis (Table 4). The Manhattan Plots for the analyses above are shown in Fig. 1. The Q-Q plots for the *p* values in each of these analysis are shown in Fig. 2 (the Q-Q plots before genomic control correction are shown in online suppl. Fig. 1), all the genomic inflation factors (lambda values) of which were close to 1 (range from 0.99 - 1.01), suggesting that the population structure was fairly adjusted, given the larger sample size of the present study.

Replication of the detected variants in the independent data set

Next we conducted the replication study of the variants discovered in the J-MICC GWAS in an independent data set from BBJ GWASs using the variants that reached the suggestive level. In the replication phase, 25 variants within three loci (on chr 2 with seven variants including rs3770636 in the *LRP2* [*LDL receptor related protein 2*] gene locus, chr 17 with 15 variants including rs3785837 in the *BCAS3* gene locus, and chr 18 with

three variants including rs74183647 in the *NFATC1* [*nuclear factor of activated T-cells 1*] gene locus) became nominally statistically significant ($p < 0.05/28 = 0.0018$, with Bonferroni correction), of which 15 variants on chr 17 and 3 variants on chr 18 reached genome-wide significance of $p < 5\times10^{-8}$ in association with eGFR and SCr (Tables 2 and 3), all of which were previously reported loci for renal traits in the transethnic GWAS [21]. Meanwhile, the locus identified for the presence/absence of CKD on chr 4 did not reach genome-wide significance in the combined study, although it fulfilled the nominal statistical significance ($p < 0.05/2 = 0.025$, with Bonferroni correction) in the replication phase (Table 4). We also evaluated the LD (linkage disequilibrium) status of these variants in the detected loci by constructing the regional plots of each locus, which revealed the close LD between the variants in the corresponding loci (Fig. 3). Additionally, conditional analyses of the lead SNPs in each locus revealed that the associations were not significant for the other SNPs/variants in the corresponding loci (online suppl. Fig. 2).

Replicability of the previously reported variants in the J-MICC Study data

We also examined the associations of 41 SNPs reported to be associated [3] with the risk of CKD and eGFR as a continuous trait using the 1000 Genomes imputed data. Of these, there were 16 SNPs significantly associated with both of eGFR and SCr as quantitative traits, and 8 SNPs were significantly associated with risk of CKD as a binary trait (online suppl. Table 1).

Discussion

To our knowledge, this is the largest GWAS of CKD and renal function traits with the cross-sectional study in a population-based Japanese cohort published to date. Although the present study couldn't identify the novel loci for renal traits, it revealed the novel associations of three loci on chr 2 and 18 with renal function traits, which were previously unreported in the East Asian and Japanese populations. Additionally, we identified the locus on chr 17 as the significantly associated locus with renal functions, which was consistent with the previous finding in Japanese subjects [3]. Regarding the newly identified loci for the East Asian and Japanese populations, the *LRP2* gene on chr 2, also called *megalin*, was originally identified as the target antigen of the rat model of membranous glomerulonephritis, and is structurally similar to the low density lipoprotein receptor [22]. Megalin is abundantly found in kidney proximal tubules, and has been shown to mediate endocytic uptake of vitamin D3 filtered in the glomeruli [23]. Endocytosis mediated by this molecule is demonstrated to play key roles in the reabsorption of albumin in renal proximal tubule cells, and it is shown to work as an important sensor in cooperation with PKB (protein kinase B), determining the survival of renal proximal tubule cells under the existence of albumin, thus considered to play roles in the progression of CKD to end-stage renal disease (ESRD) [24]. A functional mutation in *LRP2* (*LRP2* Arg365Ter, rs80338744) has been demonstrated to cause Donnai-Barrow Syndrome, which is a congenital disease presented with facio-oculo-acoustico-renal syndrome with proteinuria [25]. NFATC1 is the cytoplasmic component of the nuclear factor of activated T-cells transcription complex, which is activated upon T-cell receptor (TCR) engagement, and is involved in the functions of T lymphocytes [26-29]. It has been demonstrated that NFAT-dependent injury-response gene,

DSCR1, is involved in the phenotype switching/remodeling of the vascular smooth muscle cells [30]. Furthermore, the rs74183647 SNP of *NFATC1* found in this study is shown to modulate its gene expression in human whole blood, according to the GTEx database (https://www.gtexportal.org/). Given its biological roles in vasculature in the context of renal injury, variations in the *NFATC1* gene may be considered potential susceptibility factors for human CKD. A recent experimental study also revealed the roles of NFAT1 within the context of salt sensitivity, an important process in the development of CKD [31], thereby supporting the involvement of this protein in the genesis of CKD. With respect to the previous reports, the same locus on chr 17 as that identified in the present study was found to be associated with renal functions in the East Asian cohort consortium data. This locus includes the *BCAS3* gene, which is reportedly a biologically important gene in controlling the directional cell migration and angiogenesis by facilitating the crosstalk between cytoskeletons [32], and is amplified in 9% of primary breast tumors, expressed tumor-derived cell lines [33]. *BCAS3* is also found to be expressed in human embryonic stem (ES) cells during their differentiation into blood vascular precursors, and highly expressed in the tumor cells and blood vessels of glioblastoma, hemangiopericytoma and brain abscess, suggesting that *BCAS3* can serve as a marker for both human ES cells and tumors [33]. Moreover, genetic variations in *BCAS3* gene, including the rs3785837 identified in the present study, have been shown to change significantly in expression levels significantly in human tibial arteries according to the GTEx. However, the roles of BCAS3 in human kidney functions impairments are poorly understood. Based on the factors discussed above, it is hypothesized to play a role in the formation or modulation of human renal vasculature. Therefore, further biological and epidemiological studies are warranted. Regarding the locus found to be

associated with the risk of CKD on chr 4 (4q23), *microsomal triglyceride transfer protein* (*MTTP*) gene encoded by this locus is reportedly associated with the risk of celiac disease in Caucasians, although the roles of genomic locus in the genesis of human CKD remain largely unknown [34]. *MTTP* reportedly catalyzes the transport of lipids between phospholipid surfaces and its genetic variation is associated with abetalipoproteinemia and glucose tolerance [35]. Given its roles in lipid and glucose metabolisms, analyzing the roles of this molecule in the development of CKD may be beneficial, although its genetic variation did not reach genome-wide significance in the combined study. Moreover, our GWAS analysis of the same data set without adjustment for PCA based on GWAS genotypes revealed another locus significantly associated with renal function trait located in chr 6, including *major histocompatibility complex* (*MHC*) loci, the statistical significance of which disappeared after adjusting for PCA. This suggested that the significant association of this locus with renal function traits may be attributable to population stratification, at least when Japanese population is concerned (data not shown).

It appears that distinct genetic loci for renal function traits exist for each race/ethnicity. In the GWAS-meta-analysis of the European population, genetic loci such as the one on chr 16 (including the *UMOD* gene), or those on chr 10 (including *CDH23* gene) and on chr 7 (including *solute carrier family 22, member 2* [*SLC22A2*] and *GALNTL5* gene) were shown to be significantly associated with renal functions [1, 20, 35-37]. In the present GWAS in Japanese population, these loci were not found to be genome-wide significant, suggesting the possibility that specific genetic mechanisms exist between the races. Of the four loci found to be genome-wide significant in the present study (discovery phase), the locus on chr 17 have already been reported in East Asians,

whereas other two loci on chr 2 and 18 are firstly reported in the East Asian and Japanese populations. However, they were reported in the GWAS of renal traits in European populations and/or those with African ancestries, as represented by the *LRP2* locus on chr 2 in African Americans [21, 39]. To the best of our knowledge, the association of the 4q23 locus with CKD risk is the novel finding, which warrants further investigations with independent data sets. Whereas two out of four top SNPs for each loci found to be genome-wide significant (rs3785837 on chr 17 and rs74183647 on chr 18) had functionality by itself as described above, the functionalities of the SNPs on chr 2 (rs3770636) and chr 4 (rs78351985 and rs555786707) remain unknown. The minor allele frequencies of the newly detected SNPs for Japanese (those with the top *p* values in each locus) were 0.2326 in Japanese, 0.0221 in Caucasians and 0.0974 in Africans for the *LRP2* rs3770636 *G* allele, 0.4355 in East Asians, 0.0924 in Europeans and 0.2095 in Africans for the *NFATC1* rs74183647 *C* allele, 0.0288 in East Asians, 0.0010 in Europeans and 0.0000 in Africans for the rs555786707 *A* allele on chr 4, 0.0317 in East Asians, 0.0000 in Europeans and 0.0000 in Africans for the rs78351985 *G* allele on chr 4 according to dbSNP (https://www.ncbi.nlm.nih/gov/SNP), all of which are more than 1%, suggesting the substantial involvement of these variants in the regulation of renal functions in Japanese and East Asians. Although world-wide GWAS consortiums of human chronic diseases are flourishing these days, such GWAS of each race/ethnicity may also be considered meaningful, given racially/ethnically specific gene-environment interaction could provide more effective ways for disease prevention in each race/ethnicity [7, 40]. The strength of the present study is the large sample size and the densely imputed genetic data, which lead to the discovery of as yet unidentified genetic loci associated with renal functions specifically in Japanese population.

In conclusion, the present GWAS of renal function traits included the largest sample of Japanese population published to date, and revealed genetic loci on chr 2, 4, and 18 as involved in the renal function in the Japanese population. Further investigations of these associations, including prospective validation within the present cohort, are expected to confirm our findings, for the potential establishment of personalized prevention of renal disease in the Japanese and East Asian populations.

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Disclosure Statement

The authors have no conflict of interest to disclose.

References

1. Gorski M, Tin A, Garnaas M, McMahon GM, Chu AY, Tayo BO, Pattaro C, Teumer A, Chasman DI, Chalmers J, Hamet P, Tremblay J, Woodward M, Aspelund T, Eiriksdottir G, Gudnason V, Harris TB, Launer LJ, Smith AV, Mitchell BD, O'Connell JR, Shuldiner AR, Coresh J, Li M, Freudenberger P, Hofer E, Schmidt H, Schmidt R, Holliday EG, Mitchell P, Wang JJ, de Boer IH, Li G, Siscovick DS, Kutalik Z, Corre T, Vollenweider P, Waeber G, Gupta J, Kanetsky PA, Hwang SJ, Olden M, Yang Q, de Andrade M, Atkinson EJ, Kardia SL, Turner ST, Stafford JM, Ding J, Liu Y, Barlassina C, Cusi D, Salvi E, Staessen JA, Ridker PM, Grallert H, Meisinger C, Müller-Nurasyid M, Krämer BK, Kramer H, Rosas SE, Nolte IM, Penninx BW, Snieder H, Fabiola Del Greco M, Franke A, Nöthlings U, Lieb W, Bakker SJ, Gansevoort RT, van der Harst P, Dehghan A, Franco OH, Hofman A, Rivadeneira F, Sedaghat S, Uitterlinden AG, Coassin S, Haun M, Kollerits B, Kronenberg F, Paulweber B, Aumann N, Endlich K, Pietzner M, Völker U, Rettig R, Chouraki V, Helmer C, Lambert JC, Metzger M, Stengel B, Lehtimäki T, Lyytikäinen LP, Raitakari O, Johnson A, Parsa A, Bochud M, Heid IM, Goessling W, Köttgen A, Kao WH, Fox CS, Böger CA: Genome-wide association study of kidney function decline in individuals of European descent. Kidney Int. 2015;87:1017-1029.

2. Pattaro C, Köttgen A, Teumer A, Garnaas M, Böger CA, Fuchsberger C, Olden M, Chen MH, Tin A, Taliun D, Li M, Gao X, Gorski M, Yang Q, Hundertmark C, Foster MC, O'Seaghdha CM, Glazer N, Isaacs A, Liu CT, Smith AV, O'Connell JR, Struchalin M, Tanaka T, Li G, Johnson AD, Gierman HJ, Feitosa M, Hwang SJ, Atkinson EJ, Lohman K, Cornelis MC, Johansson Å, Tönjes A, Dehghan A, Chouraki V, Holliday EG, Sorice R, Kutalik Z, Lehtimäki T, Esko T, Deshmukh H, Ulivi S, Chu AY, Murgia F, Trompet S, Imboden M, Kollerits B, Pistis G; CARDIoGRAM Consortium; ICBP Consortium; CARe Consortium; Wellcome Trust Case Control Consortium 2 (WTCCC2), Harris TB, Launer LJ, Aspelund T, Eiriksdottir G, Mitchell BD, Boerwinkle E, Schmidt H, Cavalieri M, Rao M, Hu FB, Demirkan A, Oostra BA, de Andrade M, Turner ST, Ding J, Andrews JS, Freedman BI, Koenig W, Illig T, Döring A, Wichmann HE, Kolcic I, Zemunik T, Boban M, Minelli C, Wheeler HE, Igl W, Zaboli G, Wild SH, Wright AF, Campbell H, Ellinghaus D, Nöthlings U, Jacobs G, Biffar R, Endlich K, Ernst F, Homuth G, Kroemer HK, Nauck M, Stracke S, Völker U, Völzke H, Kovacs P, Stumvoll M, Mägi R, Hofman A, Uitterlinden AG, Rivadeneira F, Aulchenko YS, Polasek O, Hastie N, Vitart V, Helmer C, Wang JJ, Ruggiero D, Bergmann S, Kähönen M, Viikari J, Nikopensius T, Province M, Ketkar S, Colhoun H, Doney A, Robino A, Giulianini F, Krämer BK, Portas L, Ford I, Buckley BM, Adam M, Thun GA, Paulweber B, Haun M, Sala C, Metzger M, Mitchell P, Ciullo M, Kim SK, Vollenweider P, Raitakari O, Metspalu A, Palmer C, Gasparini P, Pirastu M, Jukema JW, Probst-Hensch NM, Kronenberg F, Toniolo D, Gudnason V, Shuldiner AR, Coresh J, Schmidt R, Ferrucci L, Siscovick DS, van Duijn CM, Borecki I, Kardia SL, Liu Y, Curhan GC, Rudan I, Gyllensten U, Wilson JF, Franke A, Pramstaller PP, Rettig R, Prokopenko I, Witteman JC, Hayward C, Ridker P, Parsa A, Bochud M, Heid IM, Goessling W, Chasman DI, Kao WH, Fox CS: Genome-wide association and functional follow-up reveals new loci for kidney function. PLoS Genet. 2012;8:e1002584.

3. Okada Y, Sim X, Go MJ, Wu JY, Gu D, Takeuchi F, Takahashi A, Maeda S, Tsunoda T, Chen P, Lim SC,

Wong TY, Liu J, Young TL, Aung T, Seielstad M, Teo YY, Kim YJ, Lee JY, Han BG, Kang D, Chen CH, Tsai FJ, Chang LC, Fann SJ, Mei H, Rao DC, Hixson JE, Chen S, Katsuya T, Isono M, Ogihara T, Chambers JC, Zhang W, Kooner JS; KidneyGen Consortium.; CKDGen Consortium., Albrecht E; GUGC consortium., Yamamoto K, Kubo M, Nakamura Y, Kamatani N, Kato N, He J, Chen YT, Cho YS, Tai ES, Tanaka T: Meta-analysis identifies multiple loci associated with kidney function-related traits in east Asian populations. Nat Genet 2012;44:904-909.

4. Köttgen A, Glazer NL, Dehghan A, Hwang SJ, Katz R, Li M, Yang Q, Gudnason V, Launer LJ, Harris TB, Smith AV, Arking DE, Astor BC, Boerwinkle E, Ehret GB, Ruczinski I, Scharpf RB, Chen YD, de Boer IH, Haritunians T, Lumley T, Sarnak M, Siscovick D, Benjamin EJ, Levy D, Upadhyay A, Aulchenko YS, Hofman A, Rivadeneira F, Uitterlinden AG, van Duijn CM, Chasman DI, Paré G, Ridker PM, Kao WH, Witteman JC, Coresh J, Shlipak MG, Fox CS: Multiple loci associated with indices of renal function and chronic kidney disease. Nat Genet 2009;41:712-717.

5. Böger CA, Gorski M, Li M, Hoffmann MM, Huang C, Yang Q, Teumer A, Krane V, O'Seaghdha CM, Kutalik Z, Wichmann HE, Haak T, Boes E, Coassin S, Coresh J, Kollerits B, Haun M, Paulweber B, Köttgen A, Li G, Shlipak MG, Powe N, Hwang SJ, Dehghan A, Rivadeneira F, Uitterlinden A, Hofman A, Beckmann JS, Krämer BK, Witteman J, Bochud M, Siscovick D, Rettig R, Kronenberg F, Wanner C, Thadhani RI, Heid IM, Fox CS, Kao WH; CKDGen Consortium: Association of eGFR-Related Loci Identified by GWAS with Incident CKD and ESRD. PLoS Genet. 2011;7:e1002292.

6. Köttgen A, Pattaro C, Böger CA, Fuchsberger C, Olden M, Glazer NL, Parsa A, Gao X, Yang Q, Smith AV, O'Connell JR, Li M, Schmidt H, Tanaka T, Isaacs A, Ketkar S, Hwang SJ, Johnson AD, Dehghan A, Teumer A, Paré G, Atkinson EJ, Zeller T, Lohman K, Cornelis MC, Probst-Hensch NM, Kronenberg F, Tönjes A, Hayward C, Aspelund T, Eiriksdottir G, Launer LJ, Harris TB, Rampersaud E, Mitchell BD, Arking DE, Boerwinkle E, Struchalin M, Cavalieri M, Singleton A, Giallauria F, Metter J, de Boer IH, Haritunians T, Lumley T, Siscovick D, Psaty BM, Zillikens MC, Oostra BA, Feitosa M, Province M, de Andrade M, Turner ST, Schillert A, Ziegler A, Wild PS, Schnabel RB, Wilde S, Munzel TF, Leak TS, Illig T, Klopp N, Meisinger C, Wichmann HE, Koenig W, Zgaga L, Zemunik T, Kolcic I, Minelli C, Hu FB, Johansson A, Igl W, Zaboli G, Wild SH, Wright AF, Campbell H, Ellinghaus D, Schreiber S, Aulchenko YS, Felix JF, Rivadeneira F, Uitterlinden AG, Hofman A, Imboden M, Nitsch D, Brandstätter A, Kollerits B, Kedenko L, Mägi R, Stumvoll M, Kovacs P, Boban M, Campbell S, Endlich K, Völzke H, Kroemer HK, Nauck M, Völker U, Polasek O, Vitart V, Badola S, Parker AN, Ridker PM, Kardia SL, Blankenberg S, Liu Y, Curhan GC, Franke A, Rochat T, Paulweber B, Prokopenko I, Wang W, Gudnason V, Shuldiner AR, Coresh J, Schmidt R, Ferrucci L, Shlipak MG, van Duijn CM, Borecki I, Krämer BK, Rudan I, Gyllensten U, Wilson JF, Witteman JC, Pramstaller PP, Rettig R, Hastie N, Chasman DI, Kao WH, Heid IM, Fox CS: New loci associated with kidney function and chronic kidney disease. Nat Genet 2010;42:376-384.

7. Kashyap MV, Nolan M, Sprouse M, Chakraborty R, Cross D, Roby R, Vishwanatha JK: Role of genomics in eliminating health disparities. J Carcinog 2015;14:6.

8. Nakamura Y: The BioBank Japan Project. Clin Adv Hematol Oncol 2007;5:696-697.

9. Hamajima N; J-MICC Study Group: The Japan Multi-Institutional Collaborative Cohort Study (J-MICC Study) to detect gene-environment interactions for cancer. Asian Pac J Cancer Prev 2007;8:317-323.

10. Wakai K, Hamajima N, Okada R, Naito M, Morita E, Hishida A, Kawai S, Nishio K, Yin G, Asai Y, Matsuo K, Hosono S, Ito H, Watanabe M, Kawase T, Suzuki T, Tajima K, Tanaka K, Higaki Y, Hara M, Imaizumi T, Taguchi N, Nakamura K, Nanri H, Sakamoto T, Horita M, Shinchi K, Kita Y, Turin TC, Rumana N, Matsui K, Miura K, Ueshima H, Takashima N, Nakamura Y, Suzuki S, Ando R, Hosono A, Imaeda N, Shibata K, Goto C, Hattori N, Fukatsu M, Yamada T, Tokudome S, Takezaki T, Niimura H, Hirasada K, Nakamura A, Tatebo M, Ogawa S, Tsunematsu N, Chiba S, Mikami H, Kono S, Ohnaka K, Takayanagi R, Watanabe Y, Ozaki E, Shigeta M, Kuriyama N, Yoshikawa A, Matsui D, Watanabe I, Inoue K, Ozasa K, Mitani S, Arisawa K, Uemura H, Hiyoshi M, Takami H, Yamaguchi M, Nakamoto M, Takeda H, Kubo M, Tanaka H; J-MICC Study Group: Profile of Participants and Genotype Distributions of 108 Polymorphisms in a Cross-sectional Study to Elucidate Associations between Genotypes and Lifestyle and Clinical Factors: A Project in the Japan Multi-institutional Collaborative Cohort (J-MICC) Study. J Epidemiol 2010;21:223-235.

11. Tokudome S, Goto C, Imaeda N, Tokudome Y, Ikeda M, Maki S: Development of a data-based short food frequency questionnaire for assessing nutrient intake by middle-aged Japanese. Asian Pac J Cancer Prev 2004;5:40-43.

12. Tokudome Y, Goto C, Imaeda N, Hasegawa T, Kato R, Hirose K, Tajima K, Tokudome S: Relative validity of a short food frequency questionnaire for assessing nutrient intake versus three-day weighed diet records in middle-aged Japanese. J Epidemiol 2005;15:135-145.

13. Goto C, Tokudome Y, Imaeda N, Takekuma K, Kuriki K, Igarashi F, Ikeda M, Tokudome S: Validation study of fatty acid consumption assessed with a short food frequency questionnaire against plasma concentration in middle-aged Japanese people. Scand J Nutr 2006;50:77-82.

14. Imaeda N, Goto C, Tokudome Y, Hirose K, Tajima K, Tokudome S: Reproducibility of a short food frequency questionnaire for Japanese general population. J Epidemiol 2007;17:100-107.

15. International Expert Committee: International Expert Committee Report
on the Role of the A1C Assay in the Diagnosis of Diabetes. Diabetes Care 2009;32:1327-1334.

16. Ma S, Dai Y: Principal component analysis based methods in bioinformatics studies. Brief Bioinform. 2011;12:714-722.

17. Matsuo S, Imai E, Horio M, Yasuda Y, Tomita K, Nitta K, Yamagata K, Tomino Y, Yokoyama H, Hishida A; Collaborators developing the Japanese equation for estimated GFR: Revised equations for estimated GFR from serum creatinine in Japan. Am J Kidney Dis 2009;53:982-992.

18. Nagai A, Hirata M, Kamatani Y, Muto K, Matsuda K, Kiyohara Y, Ninomiya T, Tamakoshi A, Yamagata Z, Mushiroda T, Murakami Y, Yuji K, Furukawa Y, Zembutsu H, Tanaka T, Ohnishi Y, Nakamura Y; BioBank Japan Cooperative Hospital Group., Kubo M. Overview of the BioBank Japan Project: Study design and profile. J Epidemiol 2017;27:S2-S8.

19. Devlin B, Roeder K, Wasserman L. Genomic control, a new approach to genetic-based association studies. Theor Popul Biol. 2001;60:155-66.

20. Wood EJ: The encyclopedia of molecular biology. Biochemical Education 1995;23:105.

21. Pattaro C, Teumer A, Gorski M, Chu AY, Li M, Mijatovic V, Garnaas M, Tin A, Sorice R, Li Y, Taliun D, Olden M, Foster M, Yang Q, Chen MH, Pers TH, Johnson AD, Ko YA, Fuchsberger C, Tayo B, Nalls M, Feitosa MF, Isaacs A, Dehghan A, d'Adamo P, Adeyemo A, Dieffenbach AK, Zonderman AB, Nolte IM, van der Most PJ, Wright AF, Shuldiner AR, Morrison AC, Hofman A, Smith AV, Dreisbach AW, Franke A, Uitterlinden AG, Metspalu A, Tonjes A, Lupo A, Robino A, Johansson Å, Demirkan A, Kollerits B, Freedman BI, Ponte B, Oostra BA, Paulweber B, Krämer BK, Mitchell BD, Buckley BM, Peralta CA, Hayward C, Helmer C, Rotimi CN, Shaffer CM, Müller C, Sala C, van Duijn CM, Saint-Pierre A, Ackermann D, Shriner D, Ruggiero D, Toniolo D, Lu Y, Cusi D, Czamara D, Ellinghaus D, Siscovick DS, Ruderfer D, Gieger C, Grallert H, Rochtchina E, Atkinson EJ, Holliday EG, Boerwinkle E, Salvi E, Bottinger EP, Murgia F, Rivadeneira F, Ernst F, Kronenberg F, Hu FB, Navis GJ, Curhan GC, Ehret GB, Homuth G, Coassin S, Thun GA, Pistis G, Gambaro G, Malerba G, Montgomery GW, Eiriksdottir G, Jacobs G, Li G, Wichmann HE, Campbell H, Schmidt H, Wallaschofski H, Völzke H, Brenner H, Kroemer HK, Kramer H, Lin H, Leach IM, Ford I, Guessous I, Rudan I, Prokopenko I, Borecki I, Heid IM, Kolcic I, Persico I, Jukema JW, Wilson JF, Felix JF, Divers J, Lambert JC, Stafford JM, Gaspoz JM, Smith JA, Faul JD, Wang JJ, Ding J, Hirschhorn JN, Attia J, Whitfield JB, Chalmers J, Viikari J, Coresh J, Denny JC, Karjalainen J, Fernandes JK, Endlich K, Butterbach K, Keene KL, Lohman K, Portas L, Launer LJ, Lyytikäinen LP, Yengo L, Franke L, Ferrucci L, Rose LM, Kedenko L, Rao M, Struchalin M, Kleber ME, Cavalieri M, Haun M, Cornelis MC, Ciullo M, Pirastu M, de Andrade M, McEvoy MA, Woodward M, Adam M, Cocca M, Nauck M, Imboden M, Waldenberger M, Pruijm M, Metzger M, Stumvoll M, Evans MK, Sale MM, Kähönen M, Boban M, Bochud M, Rheinberger M, Verweij N, Bouatia-Naji N, Martin NG, Hastie N, Probst-Hensch N, Soranzo N, Devuyst O, Raitakari O, Gottesman O, Franco OH, Polasek O, Gasparini P, Munroe PB, Ridker PM, Mitchell P,

Muntner P, Meisinger C, Smit JH; ICBP Consortium; AGEN Consortium; CARDIOGRAM; CHARGe-Heart Failure Group; ECHOGen Consortium, Kovacs P, Wild PS, Froguel P, Rettig R, Mägi R, Biffar R, Schmidt R, Middelberg RP, Carroll RJ, Penninx BW, Scott RJ, Katz R, Sedaghat S, Wild SH, Kardia SL, Ulivi S, Hwang SJ, Enroth S, Kloiber S, Trompet S, Stengel B, Hancock SJ, Turner ST, Rosas SE, Stracke S, Harris TB, Zeller T, Zemunik T, Lehtimäki T, Illig T, Aspelund T, Nikopensius T, Esko T, Tanaka T, Gyllensten U, Völker U, Emilsson V, Vitart V, Aalto V, Gudnason V, Chouraki V, Chen WM, Igl W, März W, Koenig W, Lieb W, Loos RJ, Liu Y, Snieder H, Pramstaller PP, Parsa A, O'Connell JR, Susztak K, Hamet P, Tremblay J, de Boer IH, Böger CA, Goessling W, Chasman DI, Köttgen A, Kao WH, Fox CS: Genetic associations at 53 loci highlight cell types and biological pathways relevant for kidney function. Nat Commun 2016;7:10023.

22. Raychowdhury R., Niles JL, McCluskey RT, Smith JA: Autoimmune target in Heymann nephritis is a glycoprotein with homology to the LDL receptor. Science 1989;244:1163-1165.

23. Nykjaer A, Dragun D, Walther D, Vorum H, Jacobsen C, Herz J, Melsen F, Christensen EI, Willnow TE: An endocytic pathway essential for renal uptake and activation of the steroid 25-(OH) vitamin D3. Cell 1999;96:507-515.

24. Caruso-Neves C, Pinheiro AA, Cai H, Souza-Menezes J, Guggino WB: PKB and megalin determine the survival or death of renal proximal tubule cells. Proc Natl Acad Sci U S A 2006;103:18810-18805.

25. Devriendt K, Standaert L, Van Hole C, Devlieger H, Fryns JP: Proteinuria in a patient with the diaphragmatic hernia-hypertelorism-myopia-deafness syndrome: further evidence that the facio-oculo-acoustico-renal syndrome represents the same entity. J Med Genet 1998;35:70-71.

26. Huang GN, Huso DL, Bouyain S, Tu J, McCorkell KA, May MJ, Zhu Y, Lutz M, Collins S, Dehoff M, Kang S, Whartenby K, Powell J, Leahy D, Worley PF: NFAT binding and regulation of T cell activation by the cytoplasmic scaffolding Homer proteins. Science 2008;319:476-481.

27. Li X, Ho SN, Luna J, Giacalone J, Thomas DJ, Timmerman LA, Crabtree GR, Francke U: Cloning and chromosomal localization of the human and murine genes for the T-cell transcription factors NFATc and NFATp. Cytogenet. Cell Genet 1995;68:185-191.

28. Peng SL, Gerth AJ, Ranger AM, Glimcher LH: NFATc1 and NFATc2 together control both T and B cell activation and differentiation. Immunity 2001;14: 13-20.

29. Zhou P, Sun LJ, Dotsch V, Wagner G, Verdine GL: Solution structure of the core NFATC1/DNA complex. Cell 1998;92:687-696.

30. Lee MY, Garvey SM, Baras AS, Lemmon JA, Gomez MF, Schoppee Bortz PD, Daum G, LeBoeuf RC, Wamhoff BR: Integrative genomics identifies DSCR1 (RCAN1) as a novel NFAT-dependent mediator of phenotypic modulation in vascular smooth muscle cells. Hum Mol Genet 2010; 19: 468-479.

31. Mahajan A, Rodan AR, Le TH, Gaulton KJ, Haessler J, Stilp AM, Kamatani Y, Zhu G, Sofer T, Puri S, Schellinger JN, Chu PL, Cechova S, van Zuydam N; SUMMIT Consortium; BioBank Japan Project, Arnlov J, Flessner MF, Giedraitis V, Heath AC, Kubo M, Larsson A, Lindgren CM, Madden PA, Montgomery GW, Papanicolaou GJ, Reiner AP, Sundström J, Thornton TA, Lind L, Ingelsson E, Cai J, Martin NG, Kooperberg C, Matsuda K, Whitfield JB, Okada Y, Laurie CC, Morris AP, Franceschini N: Trans-ethnic Fine Mapping Highlights Kidney-Function Genes Linked to Salt Sensitivity. Am J Hum Genet 2016; 99: 636-646.

32. Jain M, Bhat GP, Vijayraghavan K, Inamdar MS: Rudhira/BCAS3 is a cytoskeletal protein that controls Cdc42 activation and directional cell migration during angiogenesis. Exp Cell Res 2012;318:753-767.

33. Siva K, Venu P, Mahadevan A, S K S, Inamdar MS: Human BCAS3 expression in embryonic stem cells and vascular precursors suggests a role in human embryogenesis and tumor angiogenesis. PLoS One 2007;2:e1202.

34. Östensson M1, Montén C, Bacelis J, Gudjonsdottir AH, Adamovic S, Ek J, Ascher H, Pollak E, Arnell H, Browaldh L, Agardh D, Wahlström J, Nilsson S, Torinsson-Naluai Å. A possible mechanism behind autoimmune disorders discovered by genome-wide linkage and association analysis in celiac disease. PLoS One. 2013;8:e70174.

35. Online Mendelian Inheritance in Man, OMIM. Johns Hopkins University, Baltimore. MIM Number: {157147}:{09/08/2015}. Available at: http://www.ncbi.nlm.nih.gov/omim/. Accessed Mar. 1, 2018.

36. Koepsell H: The SLC22 family with transporters of organic cations, anions and zwitterions. Mol Aspects Med 2013;34:413-435.

37. Fujita T, Urban TJ, Leabman MK, Fujita K, Giacomini KM: Transport of drugs in the kidney by the human organic cation transporter, OCT2 and its genetic variants. J Pharm Sci 2006;95: 25-36.

38. Gorski M, van der Most PJ, Teumer A, Chu AY, Li M, Mijatovic V, Nolte IM, Cocca M, Taliun D, Gomez F, Li Y, Tayo B, Tin A, Feitosa MF, Aspelund T, Attia J, Biffar R, Bochud M, Boerwinkle E, Borecki I, Bottinger EP, Chen MH, Chouraki V, Ciullo M, Coresh J, Cornelis MC, Curhan GC, d'Adamo AP, Dehghan A, Dengler L, Ding J, Eiriksdottir G, Endlich K, Enroth S, Esko T, Franco OH, Gasparini P, Gieger C, Girotto G, Gottesman O, Gudnason V, Gyllensten U, Hancock SJ, Harris TB, Helmer C, Höllerer S, Hofer E, Hofman A,

Holliday EG, Homuth G, Hu FB, Huth C, Hutri-Kähönen N, Hwang SJ, Imboden M, Johansson Å, Kähönen M, König W, Kramer H, Krämer BK, Kumar A, Kutalik Z, Lambert JC, Launer LJ, Lehtimäki T, de Borst MH, Navis G, Swertz M, Liu Y, Lohman K, Loos RJF, Lu Y, Lyytikäinen LP, McEvoy MA, Meisinger C, Meitinger T, Metspalu A, Metzger M, Mihailov E, Mitchell P, Nauck M, Oldehinkel AJ, Olden M, Wjh Penninx B, Pistis G, Pramstaller PP, Probst-Hensch N, Raitakari OT, Rettig R, Ridker PM, Rivadeneira F, Robino A, Rosas SE, Ruderfer D, Ruggiero D, Saba Y, Sala C, Schmidt H, Schmidt R, Scott RJ, Sedaghat S, Smith AV, Sorice R, Stengel B, Stracke S, Strauch K, Toniolo D, Uitterlinden AG, Ulivi S, Viikari JS, Völker U, Vollenweider P, Völzke H, Vuckovic D, Waldenberger M, Jin Wang J, Yang Q, Chasman DI, Tromp G, Snieder H, Heid IM, Fox CS, Köttgen A, Pattaro C, Böger CA, Fuchsberger C: 1000 Genomes-based meta-analysis identifies 10 novel loci for kidney function. Sci Rep 2017;7:45040.

39. Ma J, Guan M, Bowden DW, Ng MC, Hicks PJ, Lea JP, Ma L, Gao C, Palmer ND, Freedman BI. Association Analysis of the Cubilin (CUBN) and Megalin (LRP2) Genes with ESRD in African Americans. Clin J Am Soc Nephrol. 2016; 11: 1034-1043.

40. Taylor AL: Making Medicine Precise and Personalized: What Can We Learn from the Past? Circ Heart Fail 2016;9:e002938.

Table 1. Characteristics of the study subjects.

	Discovery (J-MICC)	Replication (BBJ)
Number of analyzed samples	11,283	148,829
Sex: Male/Female	5,158/6,125	81,629/67,200
Age: mean(sd) (years)	54.9 (9.3)	63.6 (13.5)
SCr: mean (sd) (mg/dl)	0.72 (0.28)	0.91 (1.00)
eGFR: mean (sd) (ml/min/1.73m^2)	78.7 (15.1)	72.6 (25.0)
DM: N (%)	903 (8.0)	47,565 (32.0)
HT: N (%)	4,186 (37.1)	81,335 (54.6)
CKD: N (%)	939 (8.3)	38,143 (25.6)

SCr = serum creatinine; eGFR = estimated glomerular filtration rate; DM = diabetes mellitus; HT = hypertension; CKD = chronic kidney disease.

Table 2. Genetic variants suggestively associated with eGFR in the discovery phase ($p < 1\times10^{-6}$) and the results of association analyses in the replication and combined studies.

								Discovery (J-MICC)					Replication (BBJ)					Combined
rs ID	Geno/Imp	Cytoband	Position	Gene	Function	Effect allele	Ref. allele	EAF	r^2	β	SE	*p* value	EAF	r^2	β	SE	*p* value	*p* value
rs72878458	Imputed	2q31.1	170141086	*LRP2*	intronic	*T*	*C*	0.182	0.838	-0.0165	-0.0007	6.29×10^{-7}	0.18	0.848	-0.0003	0.0021	0.886	0.144
rs16856823	Imputed	2q31.1	170200452	*LRP2*	intronic	*T*	*A*	0.187	0.92	0.0183	0.0006	2.80×10^{-8}	0.181	0.917	0.0073	0.002	0.00029	6.71×10^{-07}
rs3770636	Imputed	2q31.1	170202833	*LRP2*	intronic	*G*	*T*	0.187	0.91	0.0184	0.0006	2.54×10^{-8}	0.181	0.91	0.0074	0.002	0.00026	5.79×10^{-07}
rs3770637	Imputed	2q31.1	170203104	*LRP2*	intronic	*C*	*T*	0.186	0.907	0.0183	0.0006	2.85×10^{-8}	0.181	0.907	0.0073	0.002	0.00028	6.34×10^{-07}
rs200309784	Imputed	2q31.1	170204098	*LRP2*	intronic	*GA*	*G*	0.208	0.829	0.0167	0.0006	1.37×10^{-7}	0.206	0.808	0.0077	0.002	0.00016	4.87×10^{-07}
rs2673171	Imputed	2q31.1	170204641	*LRP2*	intronic	*G*	*A*	0.185	0.887	0.0183	0.0006	3.50×10^{-8}	0.182	0.892	0.0072	0.002	0.00038	9.92×10^{-07}
rs3821129	Imputed	2q31.1	170204773	*LRP2*	intronic	*C*	*T*	0.182	0.884	0.0183	0.0006	3.59×10^{-8}	0.18	0.89	0.0074	0.002	0.00028	6.92×10^{-07}
rs2390793	Imputed	2q31.1	170205123	*LRP2*	intronic	*T*	*C*	0.186	0.867	0.0173	0.0006	1.71×10^{-7}	0.188	0.851	0.0076	0.0021	0.00022	7.44×10^{-07}
rs270188	Genotyped	5p15.32	5124149	*LINC01020,CTD-2297D10.2*	intergenic	*A*	*G*	0.432	1	-0.0126	-0.0005	9.91×10^{-7}	0.426	0.988	0.0018	0.0015	0.234	0.879
rs270184	Imputed	5p15.32	5124579	*LINC01020,CTD-2297D10.2*	intergenic	*T*	*C*	0.417	0.991	-0.013	-0.0005	4.94×10^{-7}	0.41	0.968	0.0015	0.0015	0.325	0.699
rs3785842	Imputed	17q23.2	59446530	*BCAS3*	intronic	*G*	*C*	0.428	0.943	0.015	0.0005	7.69×10^{-9}	0.438	0.931	0.0101	0.0015	8.21×10^{-11}	**6.35×10^{-15}**
rs3785841	Imputed	17q23.2	59446541	*BCAS3*	intronic	*G*	*C*	0.449	0.941	0.0155	0.0004	2.06×10^{-9}	0.455	0.933	0.0097	0.0015	3.44×10^{-10}	**2.12×10^{-14}**
rs11653176	Imputed	17q23.2	59447369	*BCAS3*	intronic	*C*	*T*	0.491	0.939	0.0148	0.0004	7.82×10^{-9}	0.515	0.868	0.0099	0.0016	5.91×10^{-10}	**6.24×10^{-14}**
rs398031258	Imputed	17q23.2	59447632	*BCAS3*	intronic	*CT*	*C*	0.469	0.846	0.0151	0.0004	5.35×10^{-9}	0.456	0.804	0.0107	0.0017	1.21×10^{-10}	**8.79×10^{-15}**
rs7217891	Imputed	17q23.2	59447984	*BCAS3*	intronic	*A*	*G*	0.512	0.934	0.0157	0.0004	1.03×10^{-9}	0.514	0.923	0.0103	0.0015	1.85×10^{-11}	**5.64×10^{-16}**
rs1010269	Imputed	17q23.2	59448945	*BCAS3*	intronic	*G*	*A*	0.485	0.929	0.0147	0.0005	1.12×10^{-8}	0.522	0.853	0.0118	0.0016	2.21×10^{-13}	**8.79×10^{-18}**
rs4968556	Imputed	17q23.2	59449082	*BCAS3*	intronic	*C*	*T*	0.484	0.949	-0.0149	-0.0004	7.16×10^{-9}	0.472	0.867	-0.0118	0.0016	1.07×10^{-13}	**3.28×10^{-18}**
rs11650989	Genotyped	17q23.2	59449636	*BCAS3*	intronic	*A*	*G*	0.284	0.995	0.0152	0.0005	1.02×10^{-7}	0.286	0.996	0.0087	0.0016	1.31×10^{-7}	**7.99×10^{-11}**
rs11657044	Imputed	17q23.2	59450105	*BCAS3*	intronic	*C*	*T*	0.513	0.979	0.0154	0.0004	2.65×10^{-9}	0.518	0.889	0.0117	0.0016	1.11×10^{-13}	**2.33×10^{-18}**
rs9905274	Imputed	17q23.2	59450441	*BCAS3*	intronic	*C*	*T*	0.494	0.997	0.0158	0.0004	8.28×10^{-10}	0.496	0.897	0.0106	0.0016	9.74×10^{-12}	**2.49×10^{-16}**
rs34754126	Imputed	17q23.2	59450453	*BCAS3*	intronic	*C*	*CT*	0.518	0.92	0.0141	0.0005	4.44×10^{-8}	0.525	0.839	0.0125	0.0016	1.51×10^{-14}	**7.72×10^{-19}**
rs9895661	Genotyped	17q23.2	59456589	*BCAS3*	intronic	*T*	*C*	0.456	0.999	0.0166	0.0004	1.55×10^{-10}	0.462	0.998	0.0117	0.0015	3.30×10^{-15}	**1.47×10^{-20}**
rs2079742	Imputed	17q23.2	59465697	*BCAS3*	intronic	*C*	*T*	0.464	0.855	-0.0167	-0.0004	1.16×10^{-10}	0.524	0.845	-0.0126	0.0016	5.93×10^{-15}	**2.56×10^{-20}**
rs11079428	Imputed	17q23.2	59466701	*BCAS3*	intronic	*A*	*T*	0.439	0.845	0.0167	0.0004	1.30×10^{-10}	0.451	0.83	0.0118	0.0016	5.10×10^{-13}	**4.33×10^{-18}**
rs3785837	Imputed	17q23.2	59468942	*BCAS3*	intronic	*A*	*G*	0.448	0.839	0.0171	0.0004	4.23×10^{-11}	0.455	0.829	0.0114	0.0016	2.75×10^{-12}	**2.07×10^{-17}**
rs74183647	Imputed	18q23	77156171	*NFATC1*	UTR5	*C*	*G*	0.482	0.714	0.0127	0.0005	6.56×10^{-7}	0.455	0.703	0.0098	0.0018	2.72×10^{-8}	**2.40×10^{-11}**
rs117314773	Imputed	18q23	77160066	*NFATC1*	intronic	*A*	*G*	0.497	0.767	0.0125	0.0005	9.47×10^{-7}	0.474	0.748	0.0092	0.0017	7.69×10^{-8}	**9.00×10^{-11}**
rs138901831	Imputed	18q23	77160067	*NFATC1*	intronic	*C*	*G*	0.497	0.767	0.0125	0.0005	9.47×10^{-7}	0.474	0.748	0.0092	0.0017	7.72×10^{-8}	**9.04×10^{-11}**

eGFR = estimated glomerular filtration rate; Geno/Imp = genotyped or imputed; EAF = effect allele frequency. Chromosomal locations were described based on hg19/GRCh37 coordinates. All the SNPs in the loci that reached the genome-wide suggestive level ($p < 1\times10^{-6}$) in the discovery phase are shown. *p* values that reached the genome-wide significance ($p < 5\times10^{-8}$) in the combined studies are indicated in boldface. "Combined" results indicate those for the GWAS meta-analysis of J-MICC and BBJ.

Table 3. Genetic variants suggestively associated with SCr in the discovery phase ($p < 1\times10^{-6}$) and the results of association analyses in the replication and combined studies.

								Discovery (J-MICC)					Replication (BBJ)					Combined
rs ID	Geno/Imp	Cytoband	Position	Gene	Function	Effect allele	Ref. allele	EAF	r^2	β	SE	*p* value	EAF	r^2	β	SE	*p* value	*p* value
rs72878458	Imputed	2q31.1	170141086	*LRP2*	intronic	*T*	*C*	0.182	0.838	0.0151	0.003	5.56×10^{-7}	0.18	0.848	0.0003	0.0019	0.883	0.141
rs16856823	Imputed	2q31.1	170200452	*LRP2*	intronic	*T*	*A*	0.187	0.92	-0.0168	0.003	2.24×10^{-8}	0.181	0.917	-0.0067	0.0018	0.000299	6.67×10^{-7}
rs3770636	Imputed	2q31.1	170202833	*LRP2*	intronic	*G*	*T*	0.187	0.91	-0.0169	0.003	2.04×10^{-8}	0.181	0.91	-0.0067	0.0018	0.00027	5.66×10^{-7}
rs3770637	Imputed	2q31.1	170203104	*LRP2*	intronic	*C*	*T*	0.186	0.907	-0.0169	0.003	2.26×10^{-8}	0.181	0.907	-0.0067	0.0018	0.00028	6.18×10^{-7}
rs200309784	Imputed	2q31.1	170204098	*LRP2*	intronic	*GA*	*G*	0.208	0.829	-0.0153	0.0029	1.14×10^{-7}	0.206	0.808	-0.007	0.0019	0.00017	4.82×10^{-7}
rs2673171	Imputed	2q31.1	170204641	*LRP2*	intronic	*G*	*A*	0.185	0.887	-0.0168	0.003	2.88×10^{-8}	0.182	0.892	-0.0066	0.0019	0.0004	9.76×10^{-7}
rs3821129	Imputed	2q31.1	170204773	*LRP2*	intronic	*C*	*T*	0.182	0.884	-0.0169	0.003	2.95×10^{-8}	0.18	0.89	-0.0068	0.0019	0.00029	6.73×10^{-7}
rs2390793	Imputed	2q31.1	170205123	*LRP2*	intronic	*T*	*C*	0.186	0.867	-0.0159	0.003	1.44×10^{-7}	0.188	0.851	-0.0069	0.0019	0.00027	7.37×10^{-7}
rs270188	Genotyped	5p15.32	5124149	*LINC01020,CTD-2297D10.2*	intergenic	*A*	*G*	0.432	1	0.0115	0.0024	9.90×10^{-7}	0.426	0.988	-0.0016	0.0014	0.256	0.838
rs270184	Imputed	5p15.32	5124579	*LINC01020,CTD-2297D10.2*	intergenic	*T*	*C*	0.417	0.991	0.0119	0.0024	4.77×10^{-7}	0.41	0.968	-0.0013	0.0014	0.3497	0.664
rs3785842	Imputed	17q23.2	59446530	*BCAS3*	intronic	*G*	*C*	0.428	0.943	-0.0138	0.0024	7.07×10^{-9}	0.438	0.931	-0.0093	0.0014	5.41×10^{-11}	**3.82×10^{-15}**
rs3785841	Imputed	17q23.2	59446541	*BCAS3*	intronic	*G*	*C*	0.449	0.941	-0.0142	0.0024	1.86×10^{-9}	0.455	0.933	-0.0089	0.0014	2.36×10^{-10}	**1.32×10^{-14}**
rs11653176	Imputed	17q23.2	59447369	*BCAS3*	intronic	*C*	*T*	0.509	0.939	-0.0135	0.0023	7.88×10^{-9}	0.515	0.868	-0.0091	0.0015	4.26×10^{-10}	**4.28×10^{-14}**
rs398031258	Imputed	17q23.2	59447632	*BCAS3*	intronic	*CT*	*C*	0.469	0.846	-0.0138	0.0024	5.16×10^{-9}	0.456	0.804	-0.0099	0.0015	8.22×10^{-11}	**5.53×10^{-15}**
rs7217891	Imputed	17q23.2	59447984	*BCAS3*	intronic	*A*	*G*	0.512	0.934	-0.0144	0.0023	9.51×10^{-10}	0.514	0.923	-0.0095	0.0014	1.29×10^{-11}	**3.61×10^{-16}**
rs1010269	Imputed	17q23.2	59448945	*BCAS3*	intronic	*G*	*A*	0.485	0.929	-0.0135	0.0024	1.15×10^{-8}	0.522	0.853	-0.0109	0.0015	1.41×10^{-13}	**5.40×10^{-18}**
rs4968556	Imputed	17q23.2	59449082	*BCAS3*	intronic	*C*	*T*	0.484	0.949	0.0136	0.0024	7.22×10^{-9}	0.472	0.867	0.0109	0.0015	7.02×10^{-14}	**2.05×10^{-18}**
rs11650989	Genotyped	17q23.2	59449636	*BCAS3*	intronic	*A*	*G*	0.284	0.995	-0.014	0.0026	8.51×10^{-8}	0.286	0.996	-0.008	0.0015	1.11×10^{-7}	**6.18×10^{-11}**
rs11657044	Imputed	17q23.2	59450105	*BCAS3*	intronic	*C*	*T*	0.513	0.979	-0.014	0.0024	2.70×10^{-9}	0.518	0.889	-0.0108	0.0014	7.04×10^{-14}	**1.40×10^{-18}**
rs9905274	Imputed	17q23.2	59450441	*BCAS3*	intronic	*C*	*T*	0.494	0.997	-0.0144	0.0023	8.61×10^{-10}	0.496	0.897	-0.0098	0.0014	6.51×10^{-12}	**1.59×10^{-16}**
rs34754126	Imputed	17q23.2	59450453	*BCAS3*	intronic	*C*	*CT*	0.482	0.92	-0.0129	0.0024	4.31×10^{-8}	0.525	0.839	-0.0115	0.0015	8.12×10^{-15}	**3.85×10^{-19}**
rs9895661	Genotyped	17q23.2	59456589	*BCAS3*	intronic	*T*	*C*	0.456	0.999	-0.0152	0.0024	1.42×10^{-10}	0.462	0.998	-0.0108	0.0014	1.97×10^{-15}	**7.91×10^{-21}**
rs2079742	Imputed	17q23.2	59465697	*BCAS3*	intronic	*C*	*T*	0.464	0.855	0.0153	0.0024	1.12×10^{-10}	0.524	0.845	0.0116	0.0015	3.36×10^{-15}	**1.32×10^{-20}**
rs11079428	Imputed	17q23.2	59466701	*BCAS3*	intronic	*A*	*T*	0.439	0.845	-0.0152	0.0024	1.37×10^{-10}	0.451	0.83	-0.0109	0.0015	3.56×10^{-13}	**2.92×10^{-18}**
rs3785837	Imputed	17q23.2	59468942	*BCAS3*	intronic	*A*	*G*	0.448	0.839	-0.0156	0.0024	4.56×10^{-11}	0.455	0.829	-0.0105	0.0015	2.06×10^{-12}	**1.51×10^{-17}**
rs74183647	Imputed	18q23	77156171	*NFATC1*	UTR5	*C*	*G*	0.482	0.714	-0.0116	0.0023	6.27×10^{-7}	0.455	0.703	-0.009	0.0016	2.97×10^{-8}	**2.61×10^{-11}**
rs117314773	Imputed	18q23	77160066	*NFATC1*	intronic	*A*	*G*	0.497	0.767	-0.0114	0.0023	9.81×10^{-7}	0.474	0.748	-0.0084	0.0016	7.37×10^{-8}	**8.67×10^{-11}**
rs138901831	Imputed	18q23	77160067	*NFATC1*	intronic	*C*	*G*	0.497	0.767	-0.0114	0.0023	9.81×10^{-7}	0.474	0.748	-0.0084	0.0016	7.39×10^{-8}	**8.71×10^{-11}**

SCr = serum creatinine; Geno/Imp = genotyped or imputed; EAF = effect allele frequency. Chromosomal locations were described based on hg19/GRCh37 coordinates. All the SNPs in the loci that reached the genome-wide suggestive level ($p < 1\times10^{-6}$) in the discovery phase are shown. *p* values that reached the genome-wide significance ($p < 5\times10^{-8}$) in the combined studies are indicated in boldface. "Combined" results indicate those for the GWAS meta-analysis of J-MICC and BBJ.

Table 4. Genetic variants suggestively associated with the risk of CKD in the discovery phase ($p < 1\times10^{-6}$) and the results of association analyses in the replication and combined studies.

								Discovery (J-MICC)					Replication (BBJ)					Combined
rs ID	Geno/Imp	Cytoband	Position	Gene	Function	Effect allele	Ref. allele	EAF	r^2	β	SE	*p* value	EAF	r^2	β	SE	*p* value	*p* value
rs78351985	Imputed	4q23	100576689	*MTTP,DAPP1*	intergenic	*G*	*A*	0.071	0.916	0.416	0.085	9.55×10^{-7}	0.071	0.929	0.036	0.015	0.0181	0.00034
rs555786707	Imputed	4q23	100969329	*LOC256880,DDIT4L*	intergenic	*A*	*G*	0.056	0.825	0.493	0.094	1.78×10^{-7}	0.056	0.835	0.077	0.031	0.0213	0.00031

CKD = chronic kidney disease, Geno/Imp = genotyped or imputed; EAF = effect allele frequency. Chromosomal locations were described based on hg19/GRCh37 coordinates. All the SNPs in the loci that reached the genome-wide suggestive level ($P < 1\times10^{-6}$) in the discovery phase are shown. "Combined" results indicate those for the GWAS meta-analysis of J-MICC and BBJ.

(Figure Legends)

Fig. 1. Manhattan plots for the GWAS of renal functions.

(Footnotes for Fig. 1)

Manhattan plots for the eGFR QTL (a), SCr QTL (b) and CKD binary (c) GWAS's. Gene names that reached the genome-wide significance ($P < 5x10^{-8}$) in the combined study are shown. GWAS, genome-wide-association study; eGFR, estimated glomerular filtration rate; SCr, serum creatinine, QTL, quantitative trait loci; CKD, chronic kidney disease.

Fig. 2. Q-Q plots for the GWAS of renal functions.

(Footnotes for Fig. 2)

Quantile-quantile (Q-Q) plots for the eGFR QTL (a), SCr QTL (b) and CKD binary (c) GWAS's (after genomic control correction).

Fig. 3. Regional plots of the detected loci.

(Footnotes for Fig. 3)

Detailed regional plots for the loci detected on *LRP2* on 2q31.1 (a), the *BCAS3* locus on 17q23.2 (c), and the *NFATC1* locus on 18q23 (e) in association with eGFR. Regional plots for the same set of loci as above in association with SCr (b, d and f), and the *MTTP-DDIT4L* locus on 4q23 (g) drawn based on the 1000 Genomes Phase 3 JPT (Japanese in Tokyo) data. The plots conditioned on the lead SNPs (variants) in each loci are shown. The p values are based on the discovery (J-MICC) data. eGFR = estimated glomerular filtration rate; SCr = serum creatinine. The x axis represents chromosome position, and the y axis represents the −log10 (*p* value) of association. The purple diamond represents the lead variant. Colors represent the degree of LD (r^2) between each variant and the lead variant. The LD (r^2) was calculated based on the 1000 genomes phase 3 JPT individuals.

	Discovery (J-MICC)	Replication (BBJ)
Number of analyzed samples	11.283	148.829
Sex: Male/Female	5,158/6,125	81,629/67,200
Age: mean(sd) (years)	54.9 (9.3)	63.6 (13.5)
SCr: mean (sd) (mg/dl)	0.72 (0.28)	0.91 (1.00)
eGFR: mean (sd) (ml/min/1.73m^2	78.7 (15.1)	72.6 (25.0)
DM: N (%)	903 (8.0)	47,565 (32.0)
HT: N (%)	4,186 (37.1)	81,335 (54.6)
CKD: N (%)	939 (8.3)	38,143 (25.6)

rs ID	Geno/Imp	Cytoband	Position	Gene	Function
rs72878458	Imputed	2q31.1	170141086	*LRP2*	intronic
rs16856823	Imputed	2q31.1	170200452	*LRP2*	intronic
rs3770636	Imputed	2q31.1	170202833	*LRP2*	intronic
rs3770637	Imputed	2q31.1	170203104	*LRP2*	intronic
rs200309784	Imputed	2q31.1	170204098	*LRP2*	intronic
rs2673171	Imputed	2q31.1	170204641	*LRP2*	intronic
rs3821129	Imputed	2q31.1	170204773	*LRP2*	intronic
rs2390793	Imputed	2q31.1	170205123	*LRP2*	intronic
rs270188	Genotyped	5p15.32	5124149	*LINC01020,CTD-2297D10.2*	intergenic
rs270184	Imputed	5p15.32	5124579	*LINC01020,CTD-2297D10.2*	intergenic
rs3785842	Imputed	17q23.2	59446530	*BCAS3*	intronic
rs3785841	Imputed	17q23.2	59446541	*BCAS3*	intronic
rs11653176	Imputed	17q23.2	59447369	*BCAS3*	intronic
rs398031258	Imputed	17q23.2	59447632	*BCAS3*	intronic
rs7217891	Imputed	17q23.2	59447984	*BCAS3*	intronic
rs1010269	Imputed	17q23.2	59448945	*BCAS3*	intronic
rs4968556	Imputed	17q23.2	59449082	*BCAS3*	intronic
rs11650989	Genotyped	17q23.2	59449636	*BCAS3*	intronic
rs11657044	Imputed	17q23.2	59450105	*BCAS3*	intronic
rs9905274	Imputed	17q23.2	59450441	*BCAS3*	intronic
rs34754126	Imputed	17q23.2	59450453	*BCAS3*	intronic
rs9895661	Genotyped	17q23.2	59456589	*BCAS3*	intronic
rs2079742	Imputed	17q23.2	59465697	*BCAS3*	intronic
rs11079428	Imputed	17q23.2	59466701	*BCAS3*	intronic
rs3785837	Imputed	17q23.2	59468942	*BCAS3*	intronic
rs74183647	Imputed	18q23	77156171	*NFATC1*	UTR5
rs117314773	Imputed	18q23	77160066	*NFATC1*	intronic
rs138901831	Imputed	18q23	77160067	*NFATC1*	intronic

		Discovery (J-MICC)					
Effect allele	Ref. allele	EAF	r^2	β	SE	*p* value	EAF
T	*C*	0,182	0,838	-0,0165	-0,0007	6.29×10^{-7}	0,18
T	*A*	0,187	0,92	0,0183	0,0006	2.80×10^{-8}	0,181
G	*T*	0,187	0,91	0,0184	0,0006	2.54×10^{-8}	0,181
C	*T*	0,186	0,907	0,0183	0,0006	2.85×10^{-8}	0,181
GA	*G*	0,208	0,829	0,0167	0,0006	1.37×10^{-7}	0,206
G	*A*	0,185	0,887	0,0183	0,0006	3.50×10^{-8}	0,182
C	*T*	0,182	0,884	0,0183	0,0006	3.59×10^{-8}	0,18
T	*C*	0,186	0,867	0,0173	0,0006	1.71×10^{-7}	0,188
A	*G*	0,432	1	-0,0126	-0,0005	9.91×10^{-7}	0,426
T	*C*	0,417	0,991	-0,013	-0,0005	4.94×10^{-7}	0,41
G	*C*	0,428	0,943	0,015	0,0005	7.69×10^{-9}	0,438
G	*C*	0,449	0,941	0,0155	0,0004	2.06×10^{-9}	0,455
C	*T*	0,491	0,939	0,0148	0,0004	7.82×10^{-9}	0,515
CT	*C*	0,469	0,846	0,0151	0,0004	5.35×10^{-9}	0,456
A	*G*	0,512	0,934	0,0157	0,0004	1.03×10^{-9}	0,514
G	*A*	0,485	0,929	0,0147	0,0005	1.12×10^{-8}	0,522
C	*T*	0,484	0,949	-0,0149	-0,0004	7.16×10^{-9}	0,472
A	*G*	0,284	0,995	0,0152	0,0005	1.02×10^{-7}	0,286
C	*T*	0,513	0,979	0,0154	0,0004	2.65×10^{-9}	0,518
C	*T*	0,494	0,997	0,0158	0,0004	8.28×10^{-10}	0,496
C	*CT*	0,518	0,92	0,0141	0,0005	4.44×10^{-8}	0,525
T	*C*	0,456	0,999	0,0166	0,0004	1.55×10^{-10}	0,462
C	*T*	0,464	0,855	-0,0167	-0,0004	1.16×10^{-10}	0,524
A	*T*	0,439	0,845	0,0167	0,0004	1.30×10^{-10}	0,451
A	*G*	0,448	0,839	0,0171	0,0004	4.23×10^{-11}	0,455
C	*G*	0,482	0,714	0,0127	0,0005	6.56×10^{-7}	0,455
A	*G*	0,497	0,767	0,0125	0,0005	9.47×10^{-7}	0,474
C	*G*	0,497	0,767	0,0125	0,0005	9.47×10^{-7}	0,474

Replication (BBJ)				Combined
r^2	β	SE	*p* value	*p* value
0,848	-0,0003	0,0021	0,886	0,144
0,917	0,0073	0,002	0,00029	6.71×10^{-07}
0,91	0,0074	0,002	0,00026	5.79×10^{-07}
0,907	0,0073	0,002	0,00028	6.34×10^{-07}
0,808	0,0077	0,002	0,00016	4.87×10^{-07}
0,892	0,0072	0,002	0,00038	9.92×10^{-07}
0,89	0,0074	0,002	0,00028	6.92×10^{-07}
0,851	0,0076	0,0021	0,00022	7.44×10^{-07}
0,988	0,0018	0,0015	0,234	0,879
0,968	0,0015	0,0015	0,325	0,699
0,931	0,0101	0,0015	8.21×10^{-11}	$\mathbf{6.35\times10^{-15}}$
0,933	0,0097	0,0015	3.44×10^{-10}	$\mathbf{2.12\times10^{-14}}$
0,868	0,0099	0,0016	5.91×10^{-10}	$\mathbf{6.24\times10^{-14}}$
0,804	0,0107	0,0017	1.21×10^{-10}	$\mathbf{8.79\times10^{-15}}$
0,923	0,0103	0,0015	1.85×10^{-11}	$\mathbf{5.64\times10^{-16}}$
0,853	0,0118	0,0016	2.21×10^{-13}	$\mathbf{8.79\times10^{-18}}$
0,867	-0,0118	0,0016	1.07×10^{-13}	$\mathbf{3.28\times10^{-18}}$
0,996	0,0087	0,0016	1.31×10^{-7}	$\mathbf{7.99\times10^{-11}}$
0,889	0,0117	0,0016	1.11×10^{-13}	$\mathbf{2.33\times10^{-18}}$
0,897	0,0106	0,0016	9.74×10^{-12}	$\mathbf{2.49\times10^{-16}}$
0,839	0,0125	0,0016	1.51×10^{-14}	$\mathbf{7.72\times10^{-19}}$
0,998	0,0117	0,0015	3.30×10^{-15}	$\mathbf{1.47\times10^{-20}}$
0,845	-0,0126	0,0016	5.93×10^{-15}	$\mathbf{2.56\times10^{-20}}$
0,83	0,0118	0,0016	5.10×10^{-13}	$\mathbf{4.33\times10^{-18}}$
0,829	0,0114	0,0016	2.75×10^{-12}	$\mathbf{2.07\times10^{-17}}$
0,703	0,0098	0,0018	2.72×10^{-8}	$\mathbf{2.40\times10^{-11}}$
0,748	0,0092	0,0017	7.69×10^{-8}	$\mathbf{9.00\times10^{-11}}$
0,748	0,0092	0,0017	7.72×10^{-8}	$\mathbf{9.04\times10^{-11}}$

rs ID	Geno/Imp	Cytoband	Position	Gene	Function
rs72878458	Imputed	2q31.1	170141086	*LRP2*	intronic
rs16856823	Imputed	2q31.1	170200452	*LRP2*	intronic
rs3770636	Imputed	2q31.1	170202833	*LRP2*	intronic
rs3770637	Imputed	2q31.1	170203104	*LRP2*	intronic
rs200309784	Imputed	2q31.1	170204098	*LRP2*	intronic
rs2673171	Imputed	2q31.1	170204641	*LRP2*	intronic
rs3821129	Imputed	2q31.1	170204773	*LRP2*	intronic
rs2390793	Imputed	2q31.1	170205123	*LRP2*	intronic
rs270188	Genotyped	5p15.32	5124149	*LINC01020,CTD-2297D10.2*	intergenic
rs270184	Imputed	5p15.32	5124579	*LINC01020,CTD-2297D10.2*	intergenic
rs3785842	Imputed	17q23.2	59446530	*BCAS3*	intronic
rs3785841	Imputed	17q23.2	59446541	*BCAS3*	intronic
rs11653176	Imputed	17q23.2	59447369	*BCAS3*	intronic
rs398031258	Imputed	17q23.2	59447632	*BCAS3*	intronic
rs7217891	Imputed	17q23.2	59447984	*BCAS3*	intronic
rs1010269	Imputed	17q23.2	59448945	*BCAS3*	intronic
rs4968556	Imputed	17q23.2	59449082	*BCAS3*	intronic
rs11650989	Genotyped	17q23.2	59449636	*BCAS3*	intronic
rs11657044	Imputed	17q23.2	59450105	*BCAS3*	intronic
rs9905274	Imputed	17q23.2	59450441	*BCAS3*	intronic
rs34754126	Imputed	17q23.2	59450453	*BCAS3*	intronic
rs9895661	Genotyped	17q23.2	59456589	*BCAS3*	intronic
rs2079742	Imputed	17q23.2	59465697	*BCAS3*	intronic
rs11079428	Imputed	17q23.2	59466701	*BCAS3*	intronic
rs3785837	Imputed	17q23.2	59468942	*BCAS3*	intronic
rs74183647	Imputed	18q23	77156171	*NFATC1*	UTR5
rs117314773	Imputed	18q23	77160066	*NFATC1*	intronic
rs138901831	Imputed	18q23	77160067	*NFATC1*	intronic

		Discovery (J-MICC)					
Effect allele	Ref. allele	EAF	r^2	β	SE	*p* value	EAF
T	*C*	0,182	0,838	0,0151	0,003	5.56×10^{-7}	0,18
T	*A*	0,187	0,92	-0,0168	0,003	2.24×10^{-8}	0,181
G	*T*	0,187	0,91	-0,0169	0,003	2.04×10^{-8}	0,181
C	*T*	0,186	0,907	-0,0169	0,003	2.26×10^{-8}	0,181
GA	*G*	0,208	0,829	-0,0153	0,0029	1.14×10^{-7}	0,206
G	*A*	0,185	0,887	-0,0168	0,003	2.88×10^{-8}	0,182
C	*T*	0,182	0,884	-0,0169	0,003	2.95×10^{-8}	0,18
T	*C*	0,186	0,867	-0,0159	0,003	1.44×10^{-7}	0,188
A	*G*	0,432	1	0,0115	0,0024	9.90×10^{-7}	0,426
T	*C*	0,417	0,991	0,0119	0,0024	4.77×10^{-7}	0,41
G	*C*	0,428	0,943	-0,0138	0,0024	7.07×10^{-9}	0,438
G	*C*	0,449	0,941	-0,0142	0,0024	1.86×10^{-9}	0,455
C	*T*	0,509	0,939	-0,0135	0,0023	7.88×10^{-9}	0,515
CT	*C*	0,469	0,846	-0,0138	0,0024	5.16×10^{-9}	0,456
A	*G*	0,512	0,934	-0,0144	0,0023	9.51×10^{-10}	0,514
G	*A*	0,485	0,929	-0,0135	0,0024	1.15×10^{-8}	0,522
C	*T*	0,484	0,949	0,0136	0,0024	7.22×10^{-9}	0,472
A	*G*	0,284	0,995	-0,014	0,0026	8.51×10^{-8}	0,286
C	*T*	0,513	0,979	-0,014	0,0024	2.70×10^{-9}	0,518
C	*T*	0,494	0,997	-0,0144	0,0023	8.61×10^{-10}	0,496
C	*CT*	0,482	0,92	-0,0129	0,0024	4.31×10^{-8}	0,525
T	*C*	0,456	0,999	-0,0152	0,0024	1.42×10^{-10}	0,462
C	*T*	0,464	0,855	0,0153	0,0024	1.12×10^{-10}	0,524
A	*T*	0,439	0,845	-0,0152	0,0024	1.37×10^{-10}	0,451
A	*G*	0,448	0,839	-0,0156	0,0024	4.56×10^{-11}	0,455
C	*G*	0,482	0,714	-0,0116	0,0023	6.27×10^{-7}	0,455
A	*G*	0,497	0,767	-0,0114	0,0023	9.81×10^{-7}	0,474
C	*G*	0,497	0,767	-0,0114	0,0023	9.81×10^{-7}	0,474

Replication (BBJ)				Combined
r^2	β	SE	*p* value	*p* value
0,848	0,0003	0,0019	0,883	0,141
0,917	-0,0067	0,0018	0,000299	6.67×10^{-7}
0,91	-0,0067	0,0018	0,00027	5.66×10^{-7}
0,907	-0,0067	0,0018	0,00028	6.18×10^{-7}
0,808	-0,007	0,0019	0,00017	4.82×10^{-7}
0,892	-0,0066	0,0019	0,0004	9.76×10^{-7}
0,89	-0,0068	0,0019	0,00029	6.73×10^{-7}
0,851	-0,0069	0,0019	0,00027	7.37×10^{-7}
0,988	-0,0016	0,0014	0,256	0,838
0,968	-0,0013	0,0014	0,3497	0,664
0,931	-0,0093	0,0014	5.41×10^{-11}	$\mathbf{3.82\times10^{-15}}$
0,933	-0,0089	0,0014	2.36×10^{-10}	$\mathbf{1.32\times10^{-14}}$
0,868	-0,0091	0,0015	4.26×10^{-10}	$\mathbf{4.28\times10^{-14}}$
0,804	-0,0099	0,0015	8.22×10^{-11}	$\mathbf{5.53\times10^{-15}}$
0,923	-0,0095	0,0014	1.29×10^{-11}	$\mathbf{3.61\times10^{-16}}$
0,853	-0,0109	0,0015	1.41×10^{-13}	$\mathbf{5.40\times10^{-18}}$
0,867	0,0109	0,0015	7.02×10^{-14}	$\mathbf{2.05\times10^{-18}}$
0,996	-0,008	0,0015	1.11×10^{-7}	$\mathbf{6.18\times10^{-11}}$
0,889	-0,0108	0,0014	7.04×10^{-14}	$\mathbf{1.40\times10^{-18}}$
0,897	-0,0098	0,0014	6.51×10^{-12}	$\mathbf{1.59\times10^{-16}}$
0,839	-0,0115	0,0015	8.12×10^{-15}	$\mathbf{3.85\times10^{-19}}$
0,998	-0,0108	0,0014	1.97×10^{-15}	$\mathbf{7.91\times10^{-21}}$
0,845	0,0116	0,0015	3.36×10^{-15}	$\mathbf{1.32\times10^{-20}}$
0,83	-0,0109	0,0015	3.56×10^{-13}	$\mathbf{2.92\times10^{-18}}$
0,829	-0,0105	0,0015	2.06×10^{-12}	$\mathbf{1.51\times10^{-17}}$
0,703	-0,009	0,0016	2.97×10^{-8}	$\mathbf{2.61\times10^{-11}}$
0,748	-0,0084	0,0016	7.37×10^{-8}	$\mathbf{8.67\times10^{-11}}$
0,748	-0,0084	0,0016	7.39×10^{-8}	$\mathbf{8.71\times10^{-11}}$

rs ID	Geno/Imp	Cytoband	Position	Gene	Function	Effect allele
rs78351985	Imputed	4q23	100576689	*MTTP,DAPP1*	intergenic	*G*
rs555786707	Imputed	4q23	100969329	*LOC256880,DDIT4L*	intergenic	*A*

	Discovery (J-MICC)					Replication (BI		
Ref. allele	EAF	r^2	β	SE	p value	EAF	r^2	β
A	0,071	0,916	0,416	0,085	9.55×10^{-7}	0,071	0,929	0,036
G	0,056	0,825	0,493	0,094	1.78×10^{-7}	0,056	0,835	0,077

BJ)		Combined
SE	*p* value	*p* value
0,015	0,0181	0,00034
0,031	0,0213	0,00031

Fig.1

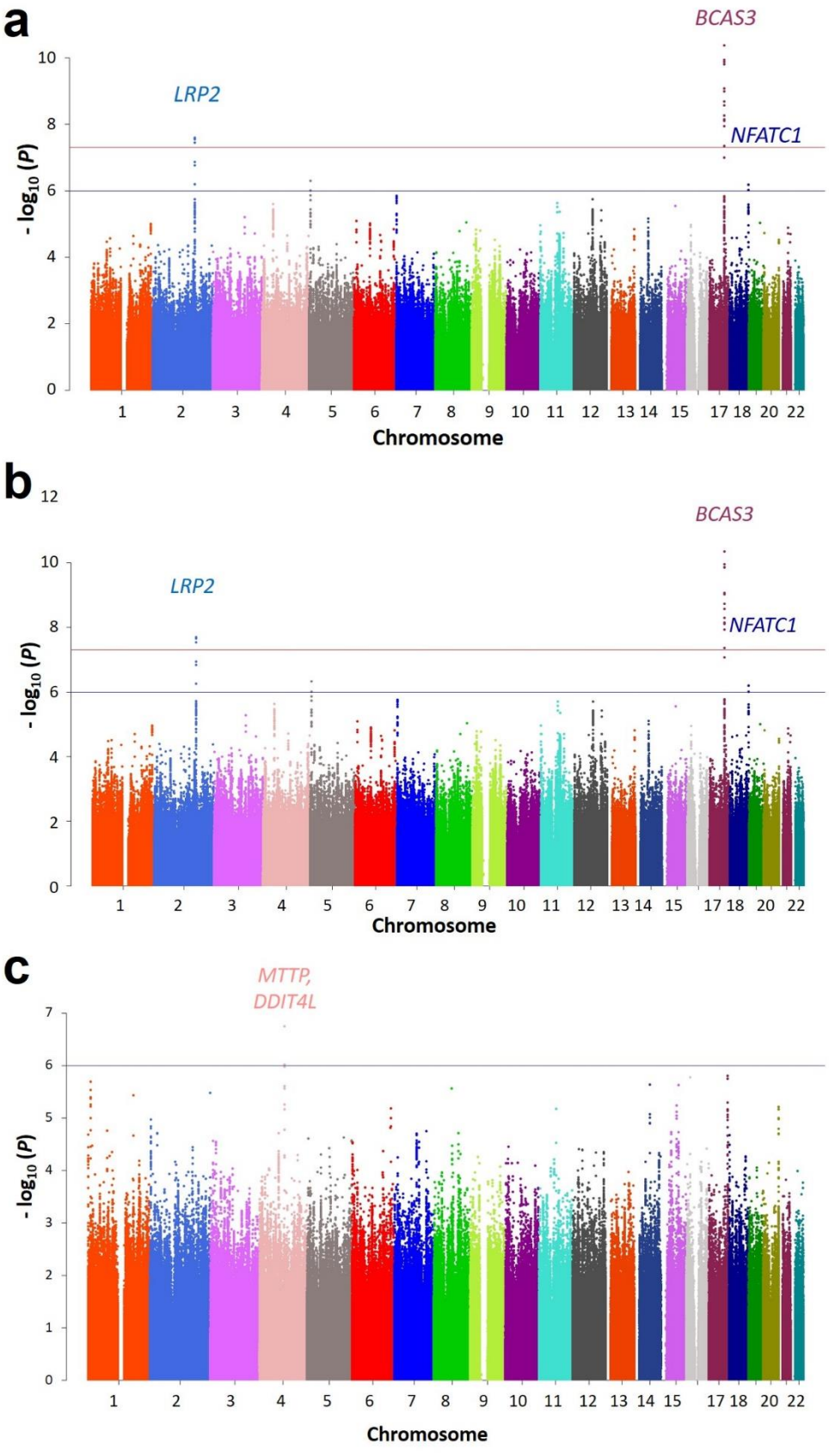

a
BCAS3
LRP2
NFATC1
- log10 (P)
Chromosome
b
BCAS3
LRP2
NFATC1
- log10 (P)
Chromosome
c
MTTP,
DDIT4L
- log10 (P)
Chromosome

Fig.2

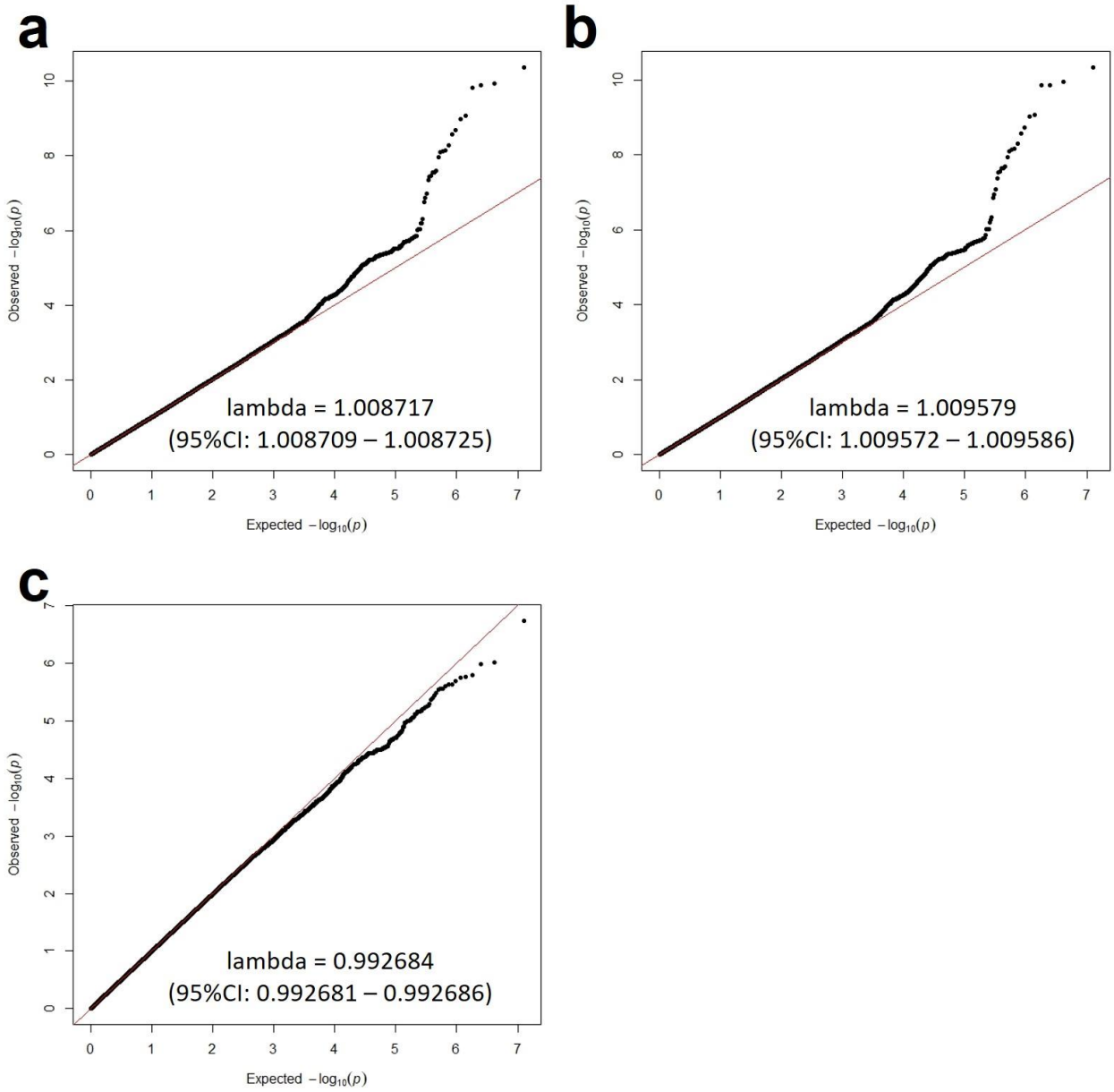
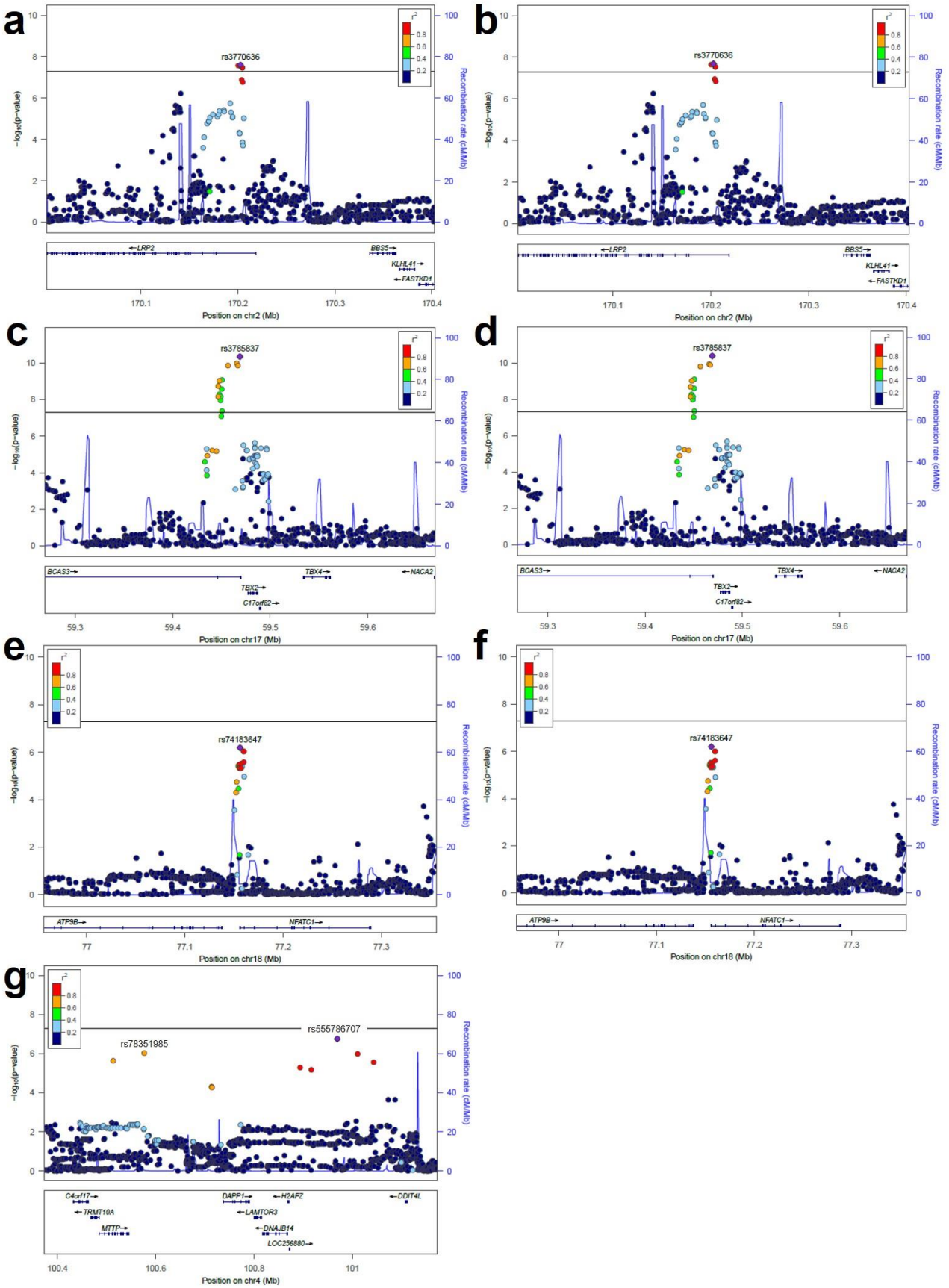

a
b
c
lambda = 1.008717
(95%CI: 1.008709 – 1.008725)
lambda = 1.009579
(95%CI: 1.009572 – 1.009586)
lambda = 0.992684
(95%CI: 0.992681 – 0.992686)
Observed $-\log_{10}(p)$
Expected $-\log_{10}(p)$

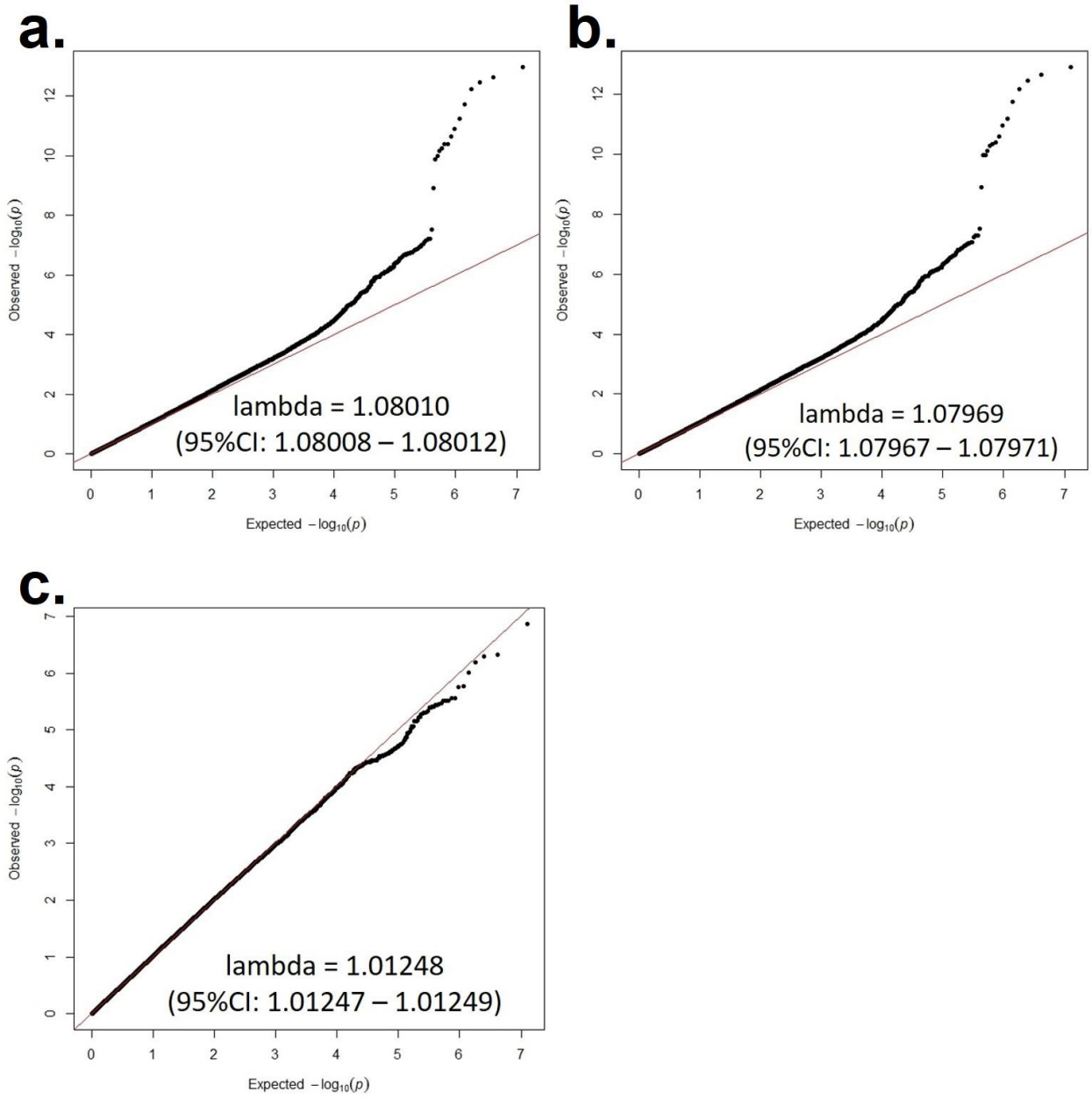
Fig.3


a
b
c
d
e
f
g
rs3770636
rs3785837
rs74183647
rs555786707
rs78351985
−log10(p-value)
Recombination rate (cM/Mb)
r²
0.8
0.6
0.4
0.2
LRP2
BBS5
KLHL41
FASTKD1
BCAS3
TBX4
NACA2
TBX2
C17orf82
ATP9B
NFATC1
C4orf17
TRMT10A
MTTP
DAPP1
H2AFZ
DDIT4L
LAMTOR3
DNAJB14
LOC256880
Position on chr2 (Mb)
Position on chr17 (Mb)
Position on chr18 (Mb)
Position on chr4 (Mb)

rs ID	Chr.	position (bp)	Cytoband	Gene	MAF	β
rs2049805	1	155194980	1q22	*MTX1-GBA*	0,182	0,0098
rs11123170	2	113978940	2q13	*PAX8*	0,349	-0,0058
rs1021916	2	213132822	2q34	*ERBB4*	0,138	-0,0002
rs13069000	3	66798950	3p14	*LRIG1-KBTBD8*	0,173	-0,0041
rs16853722	3	169150632	3q26	*MECOM*	0,281	-0,0044
rs10513699	3	171992912	3q26	*FNDC3B*	0,082	-0,0036
rs10937329	3	187713718	3q27	*BCL6-LPP*	0,315	-0,0021
rs7649443	3	196931821	3q29	*DLG1*	0,331	-0,0043
rs3775948	4	9995182	4p16	*SLC2A9*	0,418	-0,0034
rs13146355	4	77412140	4q21	*SHROOM3*	0,215	-0,0097
rs2725220	4	88959922	4q22	*ABCG2*	0,325	-0,0015
rs17663555	5	72432036	5q13	*TMEM171-TMEM174*	0,341	-0,0044
rs12654812	5	176794191	5q35	*RGS14*	0,358	-0,0076
rs2206271	6	31440669	6p12	*TFAP2B*	0,087	0,0087
rs17757177	6	50786008	6p12	*RBM16*	0,351	-0,0016
rs3828890	6	55295015	6p21	*MHC* region	0,198	0,0002
rs2797369	6	101674290	6q16	*GRIK2*	0,341	-0,0003
rs1936800	6	127436064	6q22	*RSPO3*	0,497	0,001
rs9397738	6	154986664	6q25	*CNKSR3-RBM16*	0,212	0,005
rs4870304	6	154991843	6q25	*CNKSR3-RBM16*	0,279	0,006
rs17169194	7	1273845	7p21	*RPA3*	0,331	-0,013
rs10275044	7	1285195	7p22	*UNCX*	0,316	-0,0123
rs10277115	7	7680697	7p22	*UNCX*	0,101	0,0011
rs10984991	9	123473242	9q33	*MEGF9*	0,113	-0,0026
rs10767873	11	30749090	11p14	*MPPED2-DCDC5*	0,341	0,0076
rs963837	11	30768678	11p14	*MPPED2-DCDC5*	0,318	0,0094
rs504915	11	64464085	11q13	*SLC22A12*	0,171	0,0001
rs2074356	12	112241766	12q24.13	*C12orf51*	0,257	-0,0098
rs671	12	112645401	12q24.2	*ALDH2*	0,233	-0,0108
rs17778975	13	109823110	13q33	*MYO16*	0,195	-0,0039
rs2470171	15	51666322	15q21	*GLDN*	0,37	0,0018
rs17730281	15	53907948	15q21	*WDR72*	0,387	0,0057
rs17730436	15	53942928	15q21	*WDR72*	0,383	0,0059
rs11864909	16	20400839	16p12	*UMOD*	0,179	0,0077
rs889472	16	79645989	16q23	*MAF*	0,443	-0,0034
rs438835	17	8804124	17p13	*PIK3R5*	0,251	0,0049
rs181533	17	8809832	17p13	*PIK3R5*	0,244	0,0036
rs11868441	17	59239221	17q23	*BCAS3*	0,259	-0,0101
rs9895661	17	59456589	17q23	*BCAS3*	0,455	0,0166
rs7227483	18	43187130	18q12	*SLC14A2*	0,195	0,005
rs6026584	20	57469073	20q13	*GNAS*	0,316	0,0046

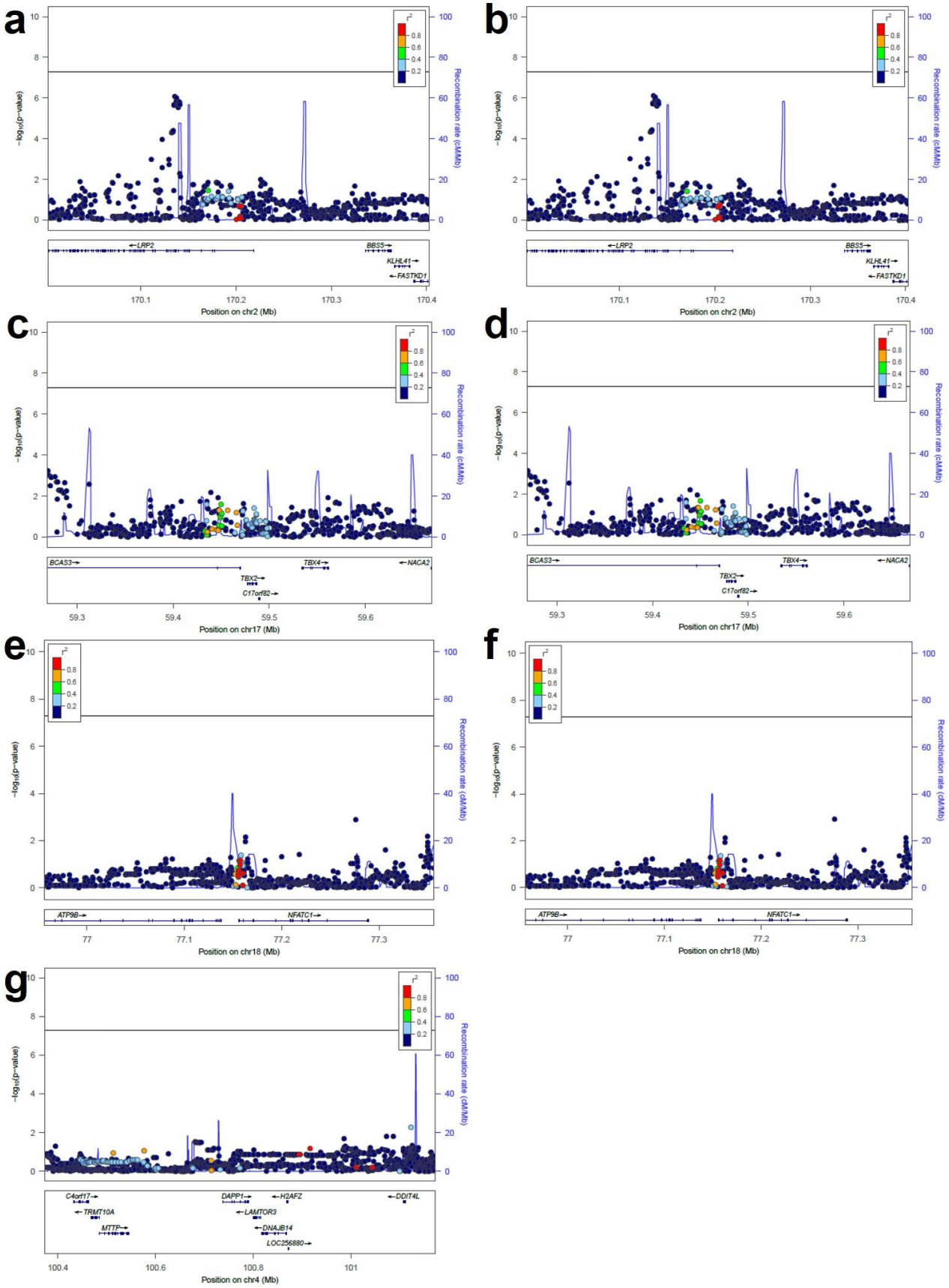
eGFR		SCr			CKD		
SE	*p value*	*β*	SE	*p value*	*β*	SE	*p value*
0,0032	0,0024	-0,0088	0,0029	0,0027	-0,1703	0,062	0,006
0,0026	0,0257	0,0053	0,0024	0,0274	0,1591	0,0512	0,0019
0,0036	0,9461	0,0001	0,0033	0,9663	-0,0213	0,071	0,764
0,0033	0,2108	0,0037	0,003	0,2163	-0,0408	0,0659	0,5357
0,0028	0,1076	0,0042	0,0025	0,0964	0,0194	0,0548	0,7237
0,0045	0,4227	0,0034	0,0041	0,4091	-0,0093	0,0902	0,9179
0,0027	0,4388	0,0019	0,0024	0,4259	0,0063	0,053	0,9047
0,0026	0,1084	0,0038	0,0024	0,1145	0,0899	0,0536	0,0931
0,0025	0,1797	0,003	0,0023	0,1972	0,1709	0,051	0,0008
0,003	0,0013	0,0088	0,0028	0,0013	0,0249	0,0596	0,6757
0,0027	0,5768	0,0013	0,0024	0,5953	-0,0165	0,0528	0,7542
0,0026	0,0935	0,004	0,0024	0,0924	0,0045	0,0519	0,9312
0,0026	0,0035	0,0069	0,0024	0,0035	0,0863	0,0513	0,0924
0,0071	0,2224	-0,0082	0,0065	0,2085	-0,2673	0,1492	0,0732
0,0026	0,5271	0,0015	0,0024	0,5353	0,0062	0,0518	0,9048
0,0031	0,954	-0,0001	0,0029	0,9647	-0,0048	0,062	0,938
0,0026	0,9094	0,0003	0,0024	0,9053	-0,0638	0,0519	0,2195
0,0025	0,6979	-0,0009	0,0023	0,6858	-0,0817	0,0492	0,0967
0,003	0,0971	-0,0046	0,0028	0,0994	-0,0512	0,0592	0,3865
0,0028	0,0298	-0,0055	0,0025	0,0302	-0,0659	0,0543	0,2245
0,0026	7.35×10^{-7}	0,0118	0,0024	9.14×10^{-7}	0,1883	0,0515	0,0003
0,0027	4.24×10^{-6}	0,0111	0,0024	5.28×10^{-6}	0,1822	0,0521	0,0005
0,0041	0,7943	-0,001	0,0038	0,7963	-0,0526	0,084	0,5315
0,0039	0,5114	0,0023	0,0036	0,515	-0,1415	0,0816	0,0829
0,0026	0,0036	-0,007	0,0024	0,0036	-0,1091	0,053	0,0395
0,0027	0,0004	-0,0086	0,0024	0,0004	-0,1347	0,0541	0,0128
0,0033	0,9797	-0,0001	0,003	0,9634	-0,0185	0,0658	0,779
0,0029	0,0006	0,009	0,0026	0,0007	0,0758	0,0564	0,1792
0,003	0,0003	0,0098	0,0027	0,0003	0,0625	0,0583	0,2837
0,0031	0,2149	0,0034	0,0029	0,2348	0,0186	0,0622	0,7655
0,0026	0,4843	-0,0016	0,0024	0,4996	0,0082	0,0513	0,873
0,0026	0,026	-0,0051	0,0023	0,029	-0,0699	0,051	0,1707
0,0026	0,0215	-0,0053	0,0023	0,0235	-0,0692	0,0512	0,1766
0,0032	0,0171	-0,0072	0,003	0,016	-0,1012	0,0663	0,1268
0,0025	0,1738	0,0032	0,0023	0,1644	-0,0153	0,0499	0,76
0,0029	0,0872	-0,0044	0,0026	0,0915	-0,109	0,0589	0,0639
0,0029	0,2088	-0,0033	0,0027	0,2173	-0,0716	0,0589	0,2243
0,0028	0,0004	0,0091	0,0026	0,0004	0,1084	0,057	0,057
0,0025	4.16×10^{-11}	-0,0152	0,0023	3.89×10^{-11}	-0,1023	0,0504	0,0426
0,0031	0,1086	-0,0046	0,0028	0,1076	0,07	0,0624	0,2623
0,0027	0,0864	-0,0042	0,0025	0,0896	-0,0283	0,0535	0,5962

Supplementary Fig. 1. Q-Q plots for the GWAS of renal functions before genomic control correction.



Quantile-quantile (Q-Q) plots for the eGFR QTL (a), SCr QTL (b) and CKD binary (c) GWAS's.

Supplementary Fig. 2. Conditional regional plots of the detected loci.



Conditional regional plots for loci detected on LRP2 on 2q31.1 (a), the *BCAS3* locus on 17q23.2 (c) and the *NFATC1* locus on 18q23 (e) in association with eGFR. Conditional regional plots for the same set of loci as above in association with SCr (b, d and f), and the *MTTP-DDIT4L* locus on 4q23 (g) drawn based on the 1000 Genomes Phase 3 JPT (Japanese in Tokyo) data. The plots conditioned on the lead SNPs (variants) in each loci are shown. The *p* values are based on the discovery (J-MICC) data. eGFR = estimated glomerular filtration rate; SCr = serum creatinine.