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SUPPLEMENTARY INFORMATION

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A catalog of genetic loci associated with kidney function from analyses of a million individuals

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A catalogue of genetic loci associated with kidney function from analyses of a million individuals

Supplementary Materials

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Supplementary Tables are provided separately as a spreadsheet.

Supplementary Note 1: Between-study heterogeneity and ancestry-related heterogeneity in the discovery meta-analysis

Before seeking replication, we evaluated results from the discovery meta-analysis for heterogeneity by design and heterogeneity related to ancestry. Most of the 308 SNPs showed homogeneous effects across studies (median I^2=5%, interquartile range: 0-13%; **Supplementary Table 3**; **Figure 2A**). Only one index SNP had I^2>50% (*UMOD-PDILT* locus, I^2=60%), where previously described heterogeneity[1,2] is suspected to be age-related.[3] We then investigated the heterogeneity of genetic effects that was correlated with ancestry using meta-regression[4] (Methods) and identified three index SNPs with significant ancestry-related heterogeneity at the *LINC01362*, *GATM*, and *PSD4* loci (ancestry heterogeneity p-value (p-anc-het) <0.05/308; **Figure 2A**, **Supplementary Table 3**). The index SNP at *UMOD-PDILT* did not show evidence for ancestry-related heterogeneity (p-anc-het=0.59). These results do not support large differences in estimated effects across ancestries for the majority of the identified SNPs. Ancestry-specific results for all 308 index SNPs are reported in **Supplementary Table 4**.

Supplementary Note 2: *Trans*-eQTL analysis

Trans-eQTL annotation of the index SNPs was only performed using whole blood and peripheral blood mononuclear cells, for which eQTL datasets with large sample size were available (Methods). Based on the analysis of 5 non-overlapping EA genome-wide eQTL studies (sample size range 1469 - 6645, **Supplementary Table 14**), we identified, among others, a reproducible link of rs10774625 (12q24.11) with several transcripts, including one for the calcium-binding protein gene *S100A10* (1q21.3) and *STAT1* (2q32.2). *S100A10* encodes a subunit of annexin A2, which co-localizes with *PLA2R* at the cell surface and in extracellular vesicles from podocytes.[5] Inhibition of *STAT1* has been reported to protect from glomerular mesangial cell senescence[6] and to ameliorate renal oxidative stress[7] (**Supplementary Table 15**).

Suppplementary Note 3: Details on the replication study: the Million Veteran's Program (MVP)

Study definition. The MVP[8] is an independent trans-ethnic study whose participants were recruited across 63 U.S. Veteran's Administration (VA) medical facilities. Written informed consent was obtained and all documents and protocols were approved by the VA Central Institutional Review Board.

Genotypes. DNA was genotyped using a customized Affymetrix Axiom Biobank Array chip with additional content added to provide coverage of African and Hispanic haplotypes, as well as markers for common diseases in the VA population. After QC, genotypes were pre-phased using EAGLE v2[9] and imputed based on the 1000Gp3v5 reference panel using minimac3.[10] Genotype PCs were estimated using FlashPCA.[11]

Phenotype. Serum creatinine was assessed up to one year prior to MVP enrollment using isotope dilution mass spectrometry. GFR was estimated using the CKD-EPI equation[12] after excluding subjects on dialysis, transplant patients, amputees, individuals on HIV medications, and those with creatinine values of <0.4 mg/dl.

Additional epidemiological information. Diabetes was defined as use of anti-diabetic medications or by assignment of an International Classification of Diseases 9 (ICD-9) code for diabetes during the baseline period. Hypertension was defined as having an ICD-9 code for hypertension, being on antihypertensive drug or having ≥2 measures of systolic or diastolic blood pressure >140 mmHg or >90 mmHg, respectively.

Genome-wide association study. GWAS of eGFR on SNP dosage levels were performed by fitting linear regression models adjusted for age at creatinine measurement, age^2, sex, body mass index, and the first 10 genetic PCs, using SNPTEST v2.5.4-beta.[13] All GWAS were stratified by self-reported ethnicity (79.6% White non-Hispanic; 20.4% Black non-Hispanic), diabetes, and hypertension status. Results were combined across strata using fixed effects inverse-variance weighted meta-analysis in METAL.[14] This analysis encompassed 280,722 individuals across all strata, of whom 216,518 were non-Hispanic Whites (EA).

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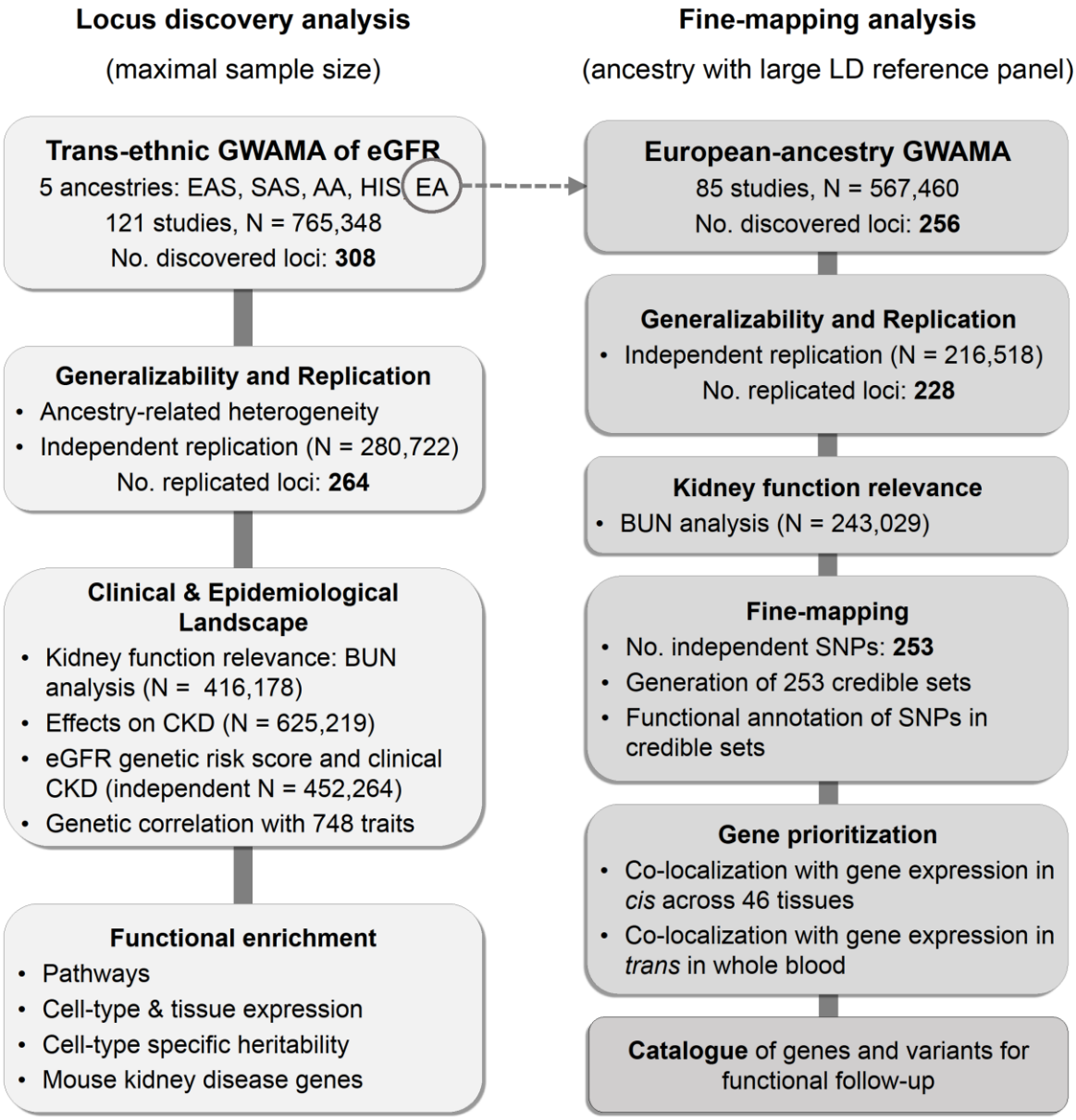
Supplementary Figure 1: Analysis Flowchart

Locus discovery analysis in the trans-ethnic sample and fine-mapping analysis in EA participants.



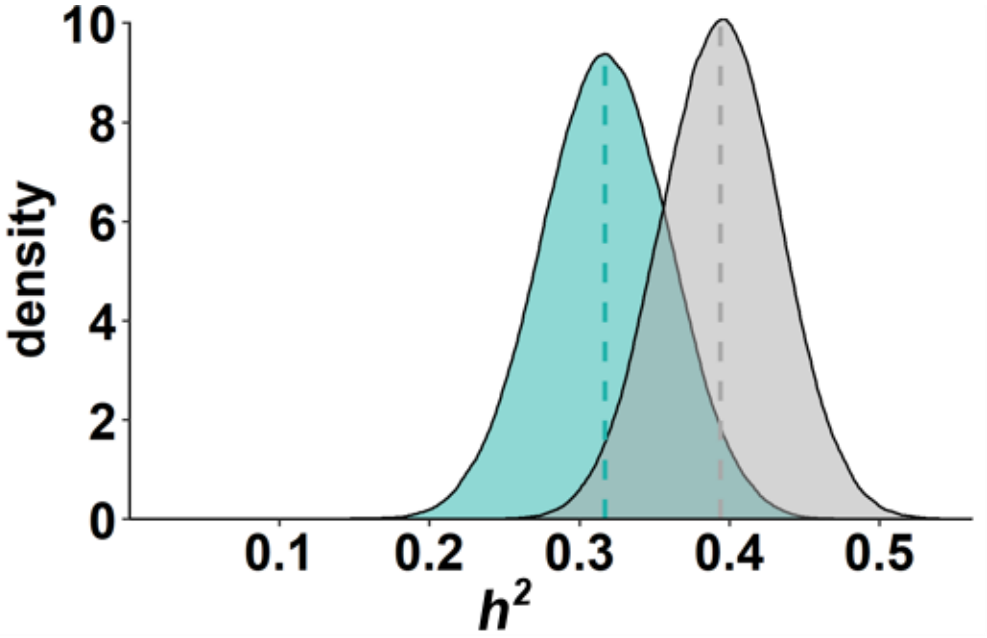
Supplementary Figure 2: Regional Association Plots

Regional Association Plot Booklet for all 308 loci identified in association with eGFR through trans-ethnic meta-analyses.

The PDF booklet is available online as a separate download with the Supplementary Information.

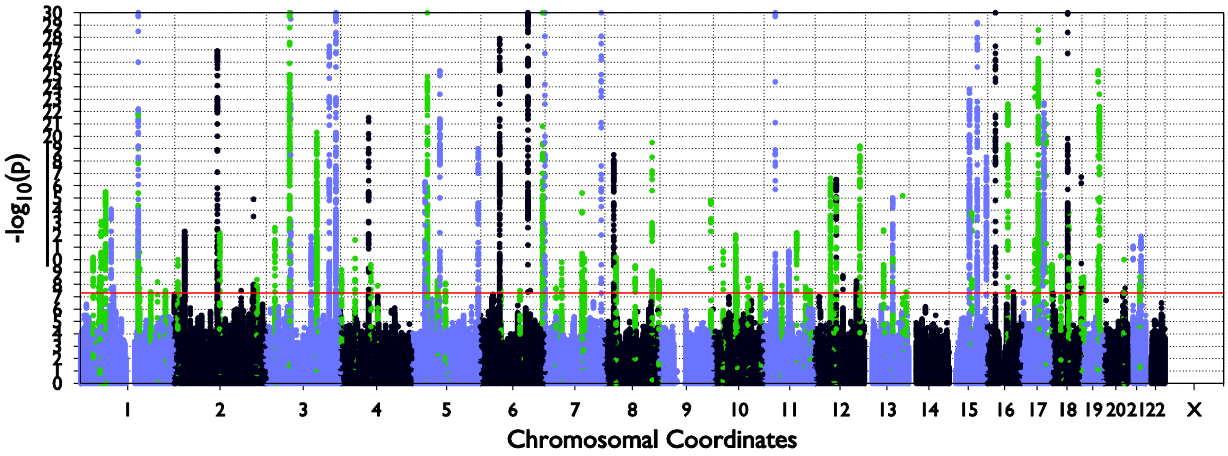
Supplementary Figure 3: Genetic Heritability

Distribution of the genetic heritability (h^2) estimates of age- and sex-adjusted log(eGFR) residuals in the Cooperative Health Research In South Tyrol (CHRIS) study, for index SNPs from the trans-ethnic GWAS. h^2 distribution is shown before (gray) and after (green) inclusion of the index SNPs into the model, with the shift representing the amount of h^2 explained by the index SNPs.



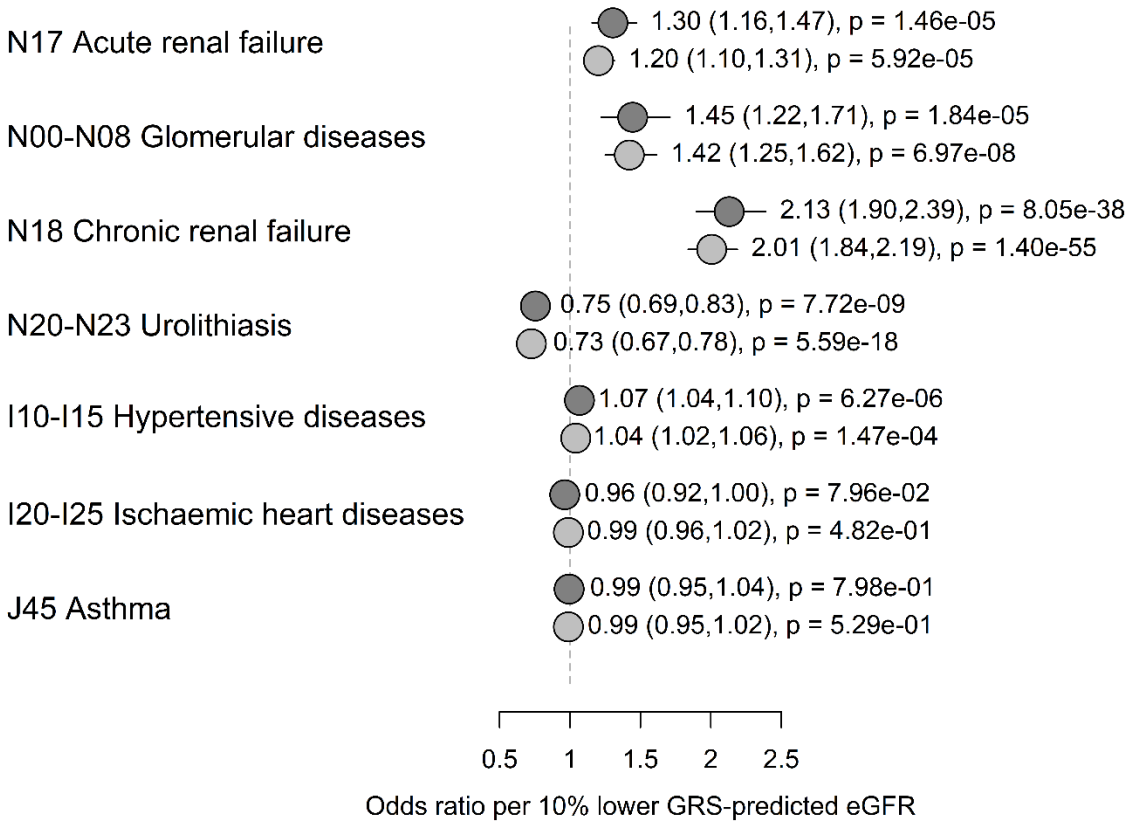
Supplementary Figure 4: BUN Manhattan plot

Manhattan plot of results from the GWAS meta-analysis of blood urea nitrogen (BUN).



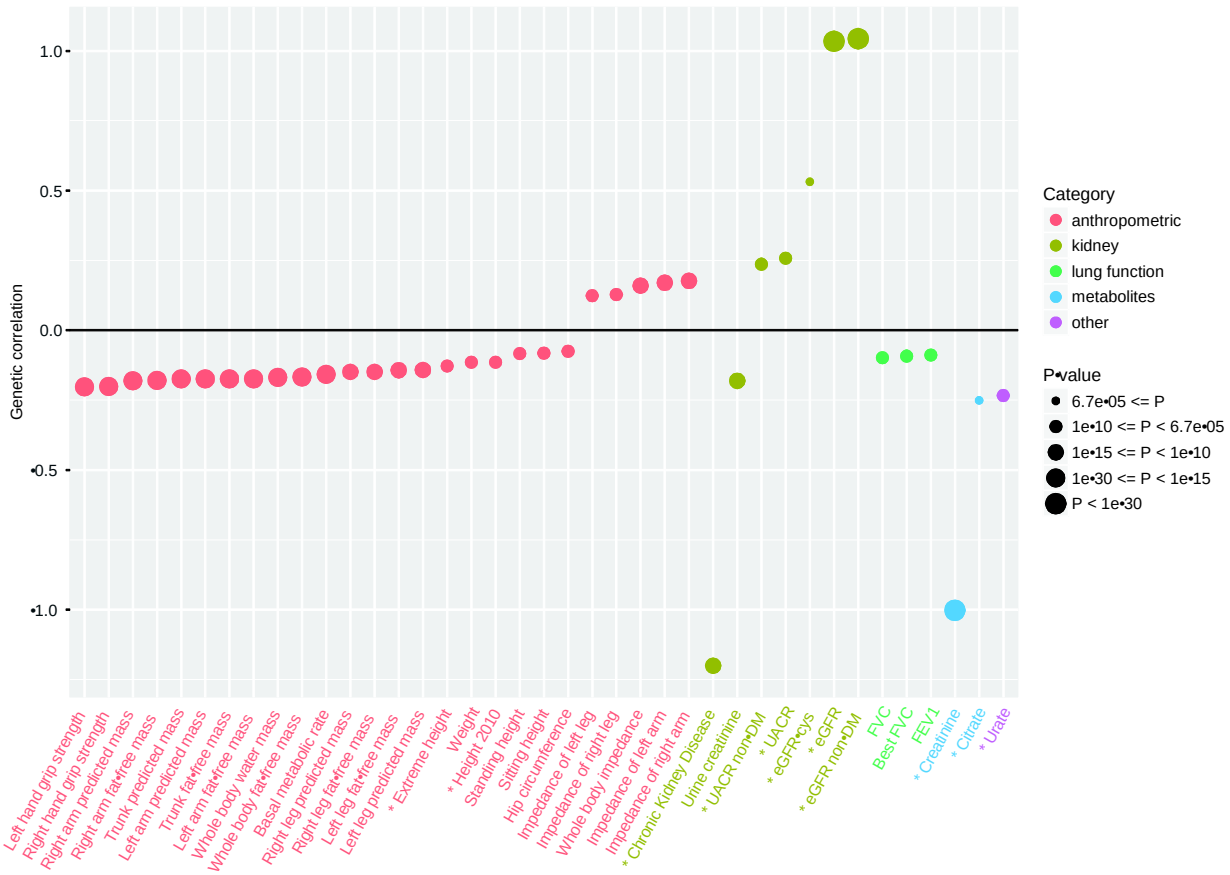
Supplementary Figure 5: Genetic Risk Score analysis

Association between lower genetically-predicted eGFR based on a genetic risk score (GRS) and ICD-10 based clinical diagnoses from 452,264 individuals from the UK Biobank. Asthma is included as a negative control. The GRS was derived as described in the Methods. Displayed are odds ratios and their 95% CIs per 10% lower GRS-predicted eGFR. Dark gray is used for results from the 147 SNPs likely to be most relevant for kidney function (same as in **Figure 2D**), light gray is used for results from all 264 replicated eGFR-associated index SNPs.



Supplementary Figure 6: Genetic correlation plot for eGFR

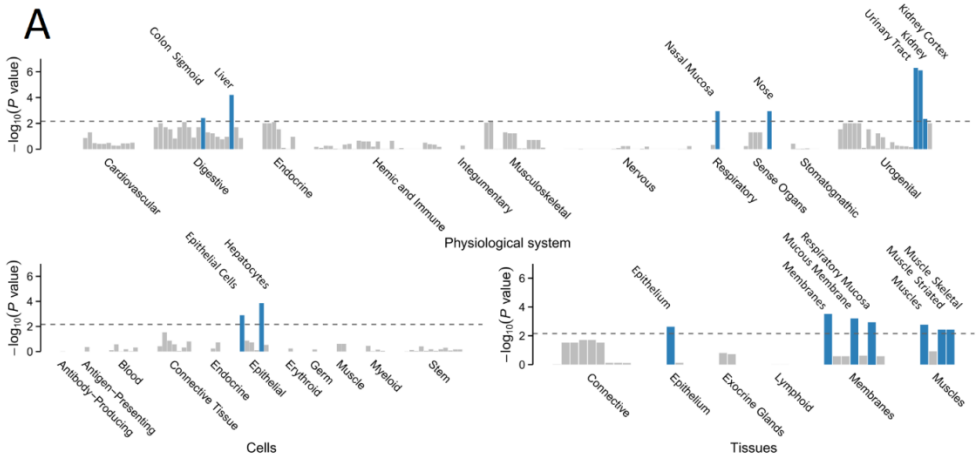
Genetic correlation plot based on the summary statistics from the trans-ethnic GWAS meta-analysis of eGFR and 748 other complex traits and diseases available through LD Hub.



The genetic correlations with citrate and cystatin C were not significant (P=6.0×10^{-4} and 4.0×10^{-4}, respectively, **Supplementary Table 7**), because these traits were measured in a limited number of studies, resulting in smaller GWAS sample sizes.

Supplementary Figure 7: Pathway and tissue enrichment analysis with DEPICT (eGFR)

Shown is the barplot of the results of the tissue and cell type enrichment analysis in **Panel A**. Cells, tissues and physiological systems are highlighted in blue if the association false discovery rate (FDR) was <0.05 and are summarized in the table in **Panel B**. The strongest enrichment was observed for urogenital and renal physiological systems and tissues: kidney, kidney cortex, and urinary tract. We additionally found significant enrichment for mucous membrane, respiratory mucosa, nasal mucosa, and nose (enrichment p-values from 3.1×10^{-4} to 1.2×10^{-3}), possibly reflecting epithelial cell processes including transport mechanisms shared with the kidney. **Panel C** illustrates the highly correlated and strongly associated meta gene sets (P <$1.x10^{-6}$, FDR<0.05) from the pathway and gene-set enrichment analysis clustered according to their biological relevance for kidney function, energy metabolism and signaling and transcription.

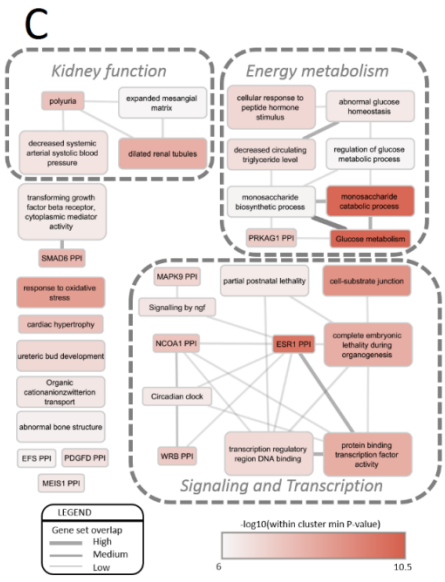


B

Enriched tissues and cell types identified by DEPICT. Shown are the tissues and cell types significantly (FDR<0.05) enriched in the meta-analysis results of subjects from all ethnicities.

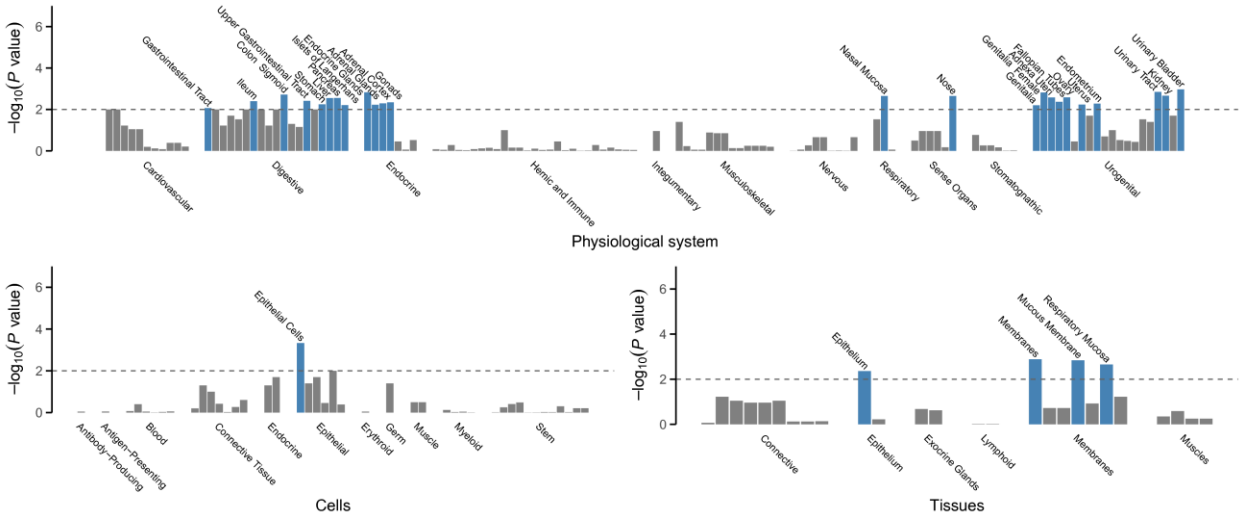
MeSH term	Name	MeSH 1st level term	MeSH 2nd level term	Nominal P-value
A05.810	Urinary Tract	Urogenital System	Urinary Tract	4.99×10^{-7}*
A05.810.453	Kidney	Urogenital System	Urinary Tract	7.84×10^{-7}*
A03.620	Liver	Digestive System	Liver	6.46×10^{-5}*
A11.436.348	Hepatocytes	Cells	Epithelial Cells	1.38×10^{-4}*
A10.615	Membranes	Tissues	Membranes	3.14×10^{-4}*
A10.615.550	Mucous Membrane	Tissues	Membranes	6.30×10^{-4}*
A10.615.550.760	Respiratory Mucosa	Tissues	Membranes	1.15×10^{-3}
A04.531.520	Nasal Mucosa	Respiratory System	Nose	1.15×10^{-3}
A09.531	Nose	Sense Organs	Nose	1.15×10^{-3}
A11.436	Epithelial Cells	Cells	Epithelial Cells	1.28×10^{-3}
A10.690	Muscles	Tissues	Muscles	1.72×10^{-3}
A10.272	Epithelium	Tissues	Epithelium	2.42×10^{-3}
A10.690.552	Muscle Striated	Tissues	Muscles	3.78×10^{-3}
A10.690.552.500	Muscle Skeletal	Tissues	Muscles	3.78×10^{-3}
A03.556.249.249.356.668	Colon Sigmoid	Digestive System	Gastrointestinal Tract	3.90×10^{-3}
A05.810.453.324	Kidney Cortex	Urogenital System	Urinary Tract	4.64×10^{-3}

MeSH = Medical Subject Heading, MeSH 1st level term is given as Cells, Tissues or as the respective annotation in the Physiological system, Nominal P-values with * indicate a False Discovery Rate < 0.01



Supplementary Figure 8: Pathway and tissue enrichment analysis with DEPICT (BUN)

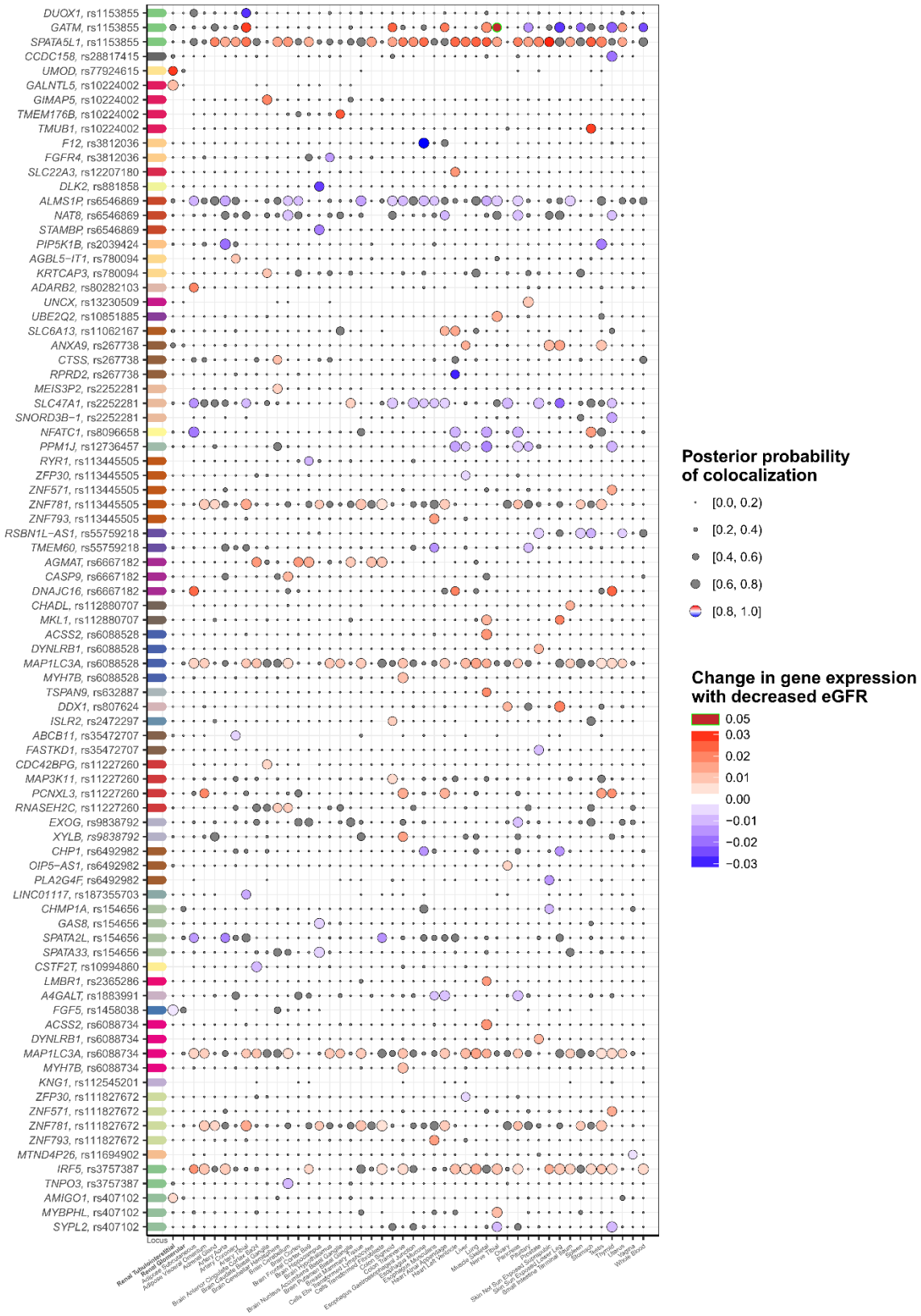
Shown is the barplot of the results of the tissue and cell type enrichment analysis. Cells, tissues and physiological systems are highlighted in blue if the association FDR was <0.05. Tissue and cell-type enrichment analysis of BUN-associated SNPs with $P<5\times10^{-8}$ highlighted a very similar pattern to the one observed for eGFR: the strongest enrichment was observed for urogenital and renal physiological systems and tissues, and significant enrichment was also observed for mucous membrane, respiratory mucosa, nasal mucosa, nose, epithelial cells and the epithelium.



Supplementary Figure 9: Co-localization of eGFR-association signals with gene expression across 44 GTEx tissues and two kidney tissues

All eGFR loci were tested for co-localization with all eQTLs where the eQTL cis-window overlapped (±100 kb) the sentinel genetic variants. Genes with at least one positive co-localization (posterior probability of one common causal variant, H4, ≥0.80) in any of the 44 tissues for which eQTL data was released by the GTEx Project or in two renal tissue are illustrated with the respective sentinel variants (Y-axis). Co-localizations across all tissues (X-axis) are illustrated as dots, where the size of the dots indicates the posterior probability of the co-localization. Negative co-localizations (posterior probability of H4 <0.80) are grey, while the positive co-localizations are color-coded based on the predicted change in expression relative to the allele associated with lower eGFR.

Supplementary Figure 9 – continued (1/4)



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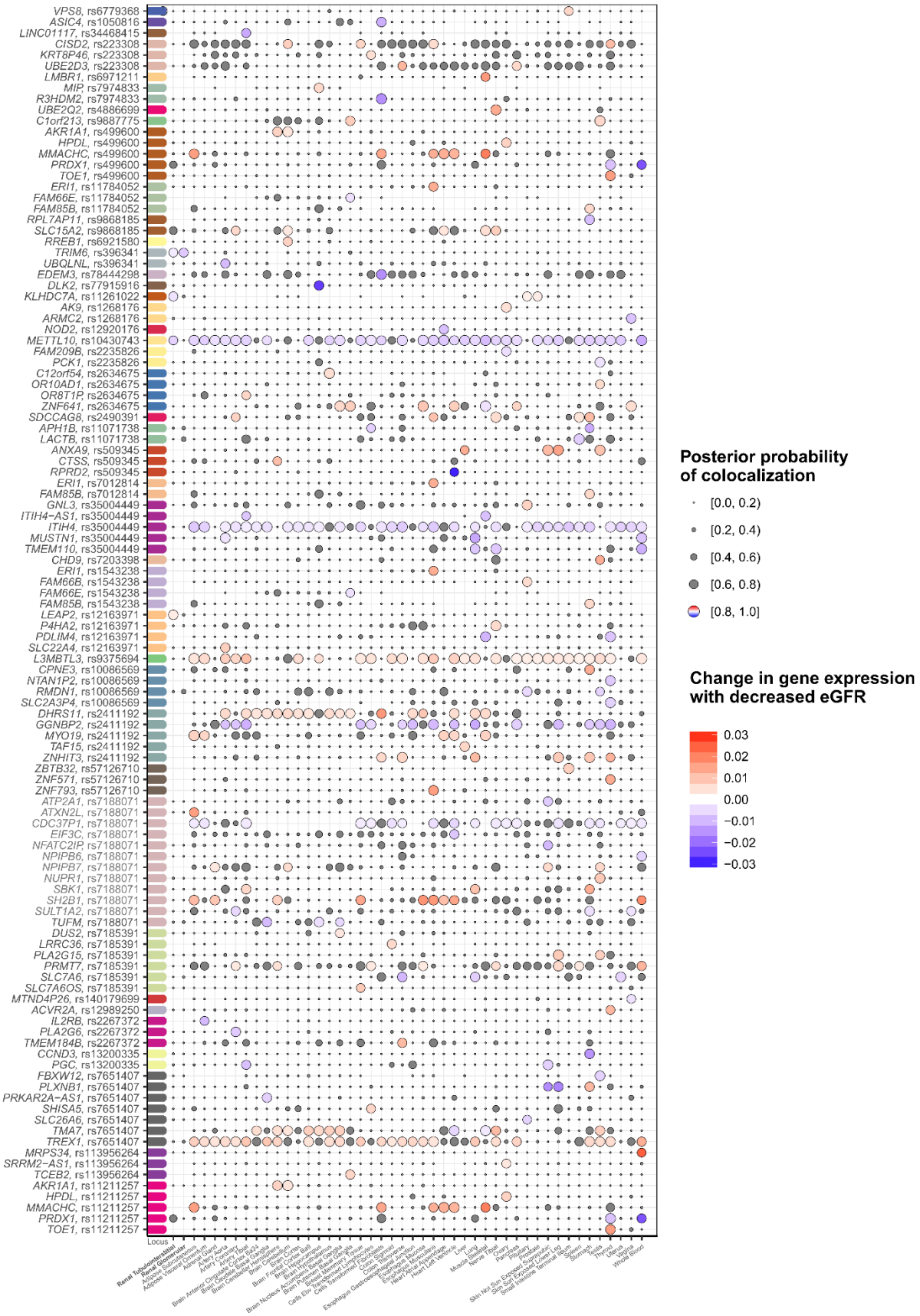

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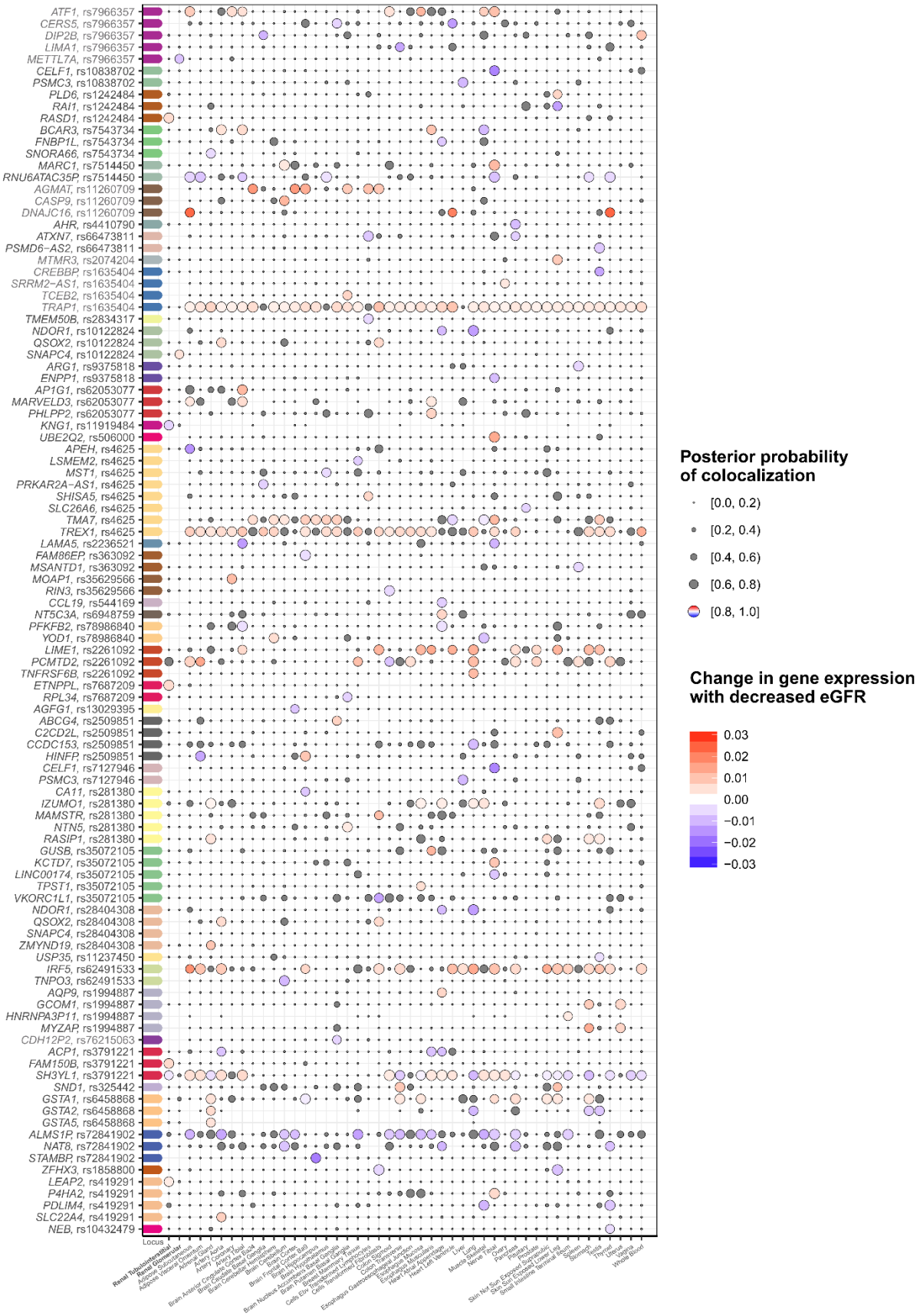

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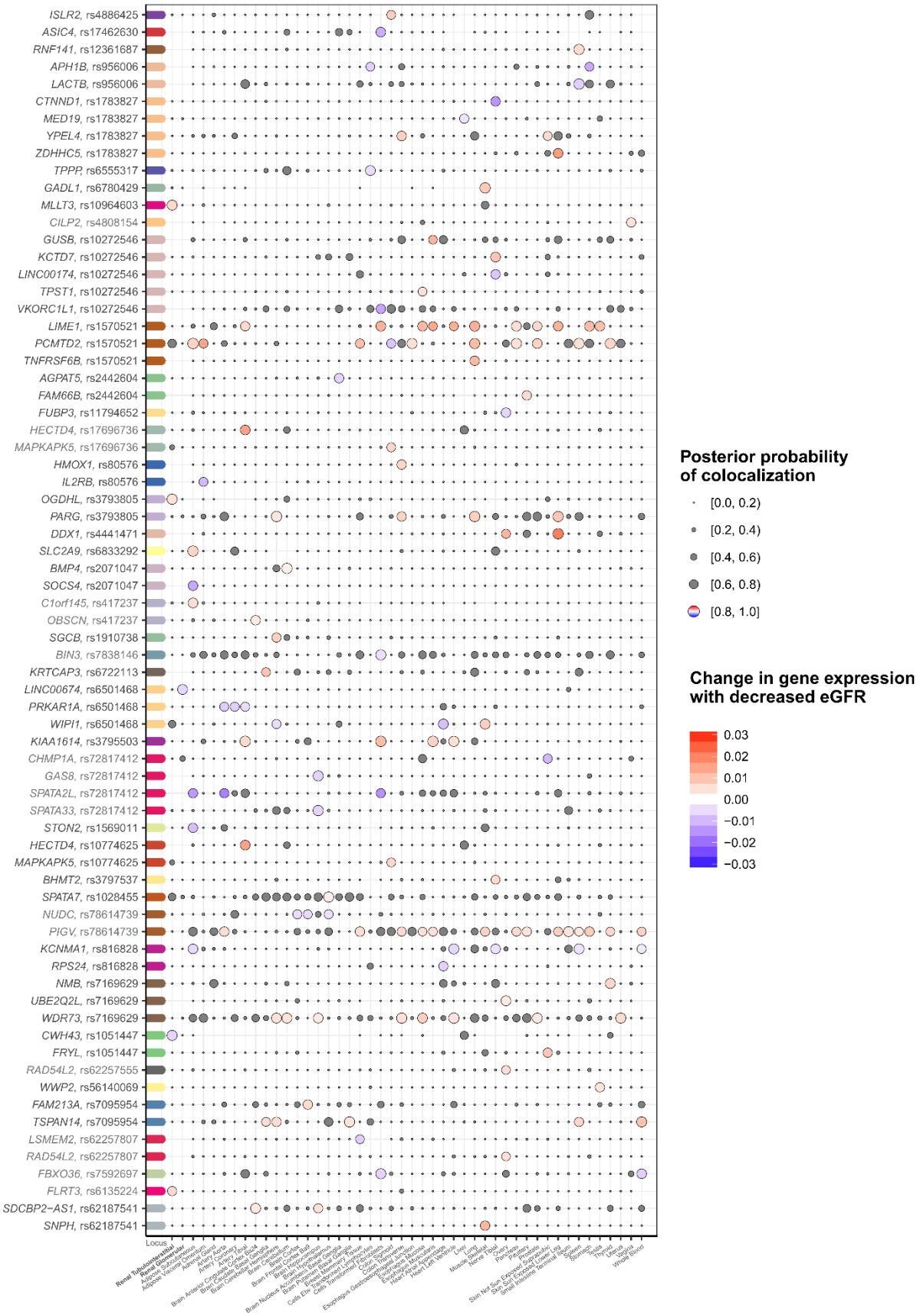

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