Lower Serum 6-Bromotryptophan Identified

as a Risk Factor for Chronic Kidney Disease Progression

Supplementary Tables 1 to 9, and Supplementary Figures 1 to 8

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Supplementary Table 1. Study population characteristics in AASK by number of *APOL1* risk alleles (n=588)

| | Number | of APOL1 risk allele | es | |
|---|---------------|----------------------|---------------|---------|
| | zero copies | one copy | two copies | P-value |
| N | 194 | 255 | 139 | |
| Age, mean (SD) | 54.93 (9.9) | 54.54 (10.4) | 52.71 (11.5) | 0.07 |
| Male, n (%) | 126 (64.9) | 144 (56.5) | 81 (58.3) | 0.18 |
| Body mass index, mean (SD) | 30.02 (6.14) | 31.51 (6.83) | 31.94 (7.13) | 0.01 |
| measured GFR, mL/min/1.73m ² Urine PCR, mg/g, median (25 th , 75 th | 48.13 (13.8) | 48.64 (13.0) | 43.9 (13.4) | 0.01 |
| percentile)* | 64 (26, 200) | 56 (25, 231) | 185 (42, 698) | < 0.001 |
| Coronary heart disease, n (%) | 99 (51) | 136 (53.3) | 59 (42.4) | 0.11 |
| Systolic blood pressure, mm Hg, mean (SD) | 148.04 (25.5) | 154.17 (24.65) | 145.5 (21.92) | 0.59 |
| Diastolic blood pressure, mm Hg, mean (SD) | 94.59 (14.6) | 98.02 (15.5) | 92.53 (13.9) | 0.39 |
| % European ancestry | 0.18 (0.15) | 0.16 (0.13) | 0.15 (0.12) | 0.04 |

Abbreviations. SD, standard deviation; GFR, glomerrular filtration rate.

*AASK sample size for proteinuria: 586.

Supplementary Table 2. Study population characteristics in Bio*Me* by number of *APOL1* risk alleles (n=678)

| | Numb | er of APOL1 risk al | leles | |
|--|----------------|---------------------|---------------|---------|
| | zero copies | one copy | two copies | P-value |
| Ν | 246 | 226 | 206 | |
| Age, mean (SD) | 60.27 (12.9) | 61.68 (13.0) | 58.92 (13.3) | 0.32 |
| Male, n (%) | 99 (40.2) | 47 (20.8) | 60 (29.1) | <0.001 |
| Body mass index, mean (SD)* | 31.21 (8.3) | 31.46 (7.6) | 31.96 (8.4) | 0.39 |
| eGFR, mL/min/1.73m ^{2*} | 58.84 (14.68) | 58.8 (12.59) | 58.72 (14.19) | 0.93 |
| Urine ACR, mg/g, median (1st, 3rd quartile)* | 15.5 (4.8, 50) | 13 (4.0, 62.5) | 11 (4.0, 112) | 0.97 |
| Coronary heart disease, n (%)* | 22 (8.9) | 13 (5.8) | 12 (5.8) | 0.30 |
| Systolic blood pressure, mm Hg, mean (SD)* | 133.9 (23.3) | 136.4 (23.7) | 133.9 (21.8) | 0.93 |
| Diastolic blood pressure, mm Hg, mean (SD)* | 76.7 (12.6) | 76.5 (12.3) | 77.4 (12.5) | 0.54 |
| Hypertension, n (%)* | 176 (71.5) | 181 (80.4) | 155 (75.2) | 0.08 |
| Diabetes, n (%)* | 99 (40.2) | 99 (44) | 80 (38.8) | 0.52 |
| % European ancestry* | 0.26 (0.22) | 0.16 (0.12) | 0.12 (0.08) | <0.001 |

Abbreviations. SD, standard deviation; eGFR, estimated glomerrular filtration rate.

*BioMe sample size. BMI (545), eGFR (658), ACR (177), SBP (673), DBP (677), CHD (677), hypertension (677), and diabetes (677); % European ancestry (478).

| Biochemical | Beta | SE | P-value | Superpathway | Subpathway | Platform | Mass | CAS ID | Pubchem ID | # missing |
|---|-------|------|----------|---------------------------|--|-----------------|--------|------------|---------------|-----------|
| 6-bromotryptophan | -0.23 | 0.05 | 4.70E-05 | Amino Acid | Tryptophan Metabolism | LC/MS Neg | 281.0* | | 96735 | 14 |
| methionine sulfone | 0.20 | 0.06 | 4.24E-04 | Amino Acid | Methionine, Cysteine, SAM and Taurine Metabolism | LC/MS Pos Early | 182.0 | 820-10-0 | 69961 | 1 |
| docosadioate | -0.18 | 0.05 | 9.30E-04 | Lipid | Fatty Acid, Dicarboxylate | LC/MS Neg | 369.3 | 505-56-6 | 244872 | 68 |
| guanidinosuccinate | 0.17 | 0.05 | 1.16E-03 | Amino Acid | Guanidino and Acetamido Metabolism | LC/MS Polar | 174.1 | 6133-30-8 | 97856 | 78 |
| 1-methylhistidine | 0.18 | 0.05 | 1.32E-03 | Amino Acid | Histidine Metabolism | LC/MS Pos Early | 170.1 | 332-80-9 | 92105 | 1 |
| N-acetylcarnosine | 0.16 | 0.05 | 1.39E-03 | Peptide | Dipeptide Derivative | LC/MS Neg | 267.1 | 56353-15-2 | 9903482 | 1 |
| retinol (Vitamin A) | 0.17 | 0.06 | 2.51E-03 | Cofactors and Vitamins | Vitamin A Metabolism | LC/MS Pos Late | 269.2 | 68-26-8 | 445354 | 1 |
| caprate (10:0) | -0.17 | 0.06 | 2.66E-03 | Lipid | Medium Chain Fatty Acid | LC/MS Neg | 171.1 | 112-37-8 | 2969 | 0 |
| taurocholate | 0.16 | 0.05 | 2.82E-03 | Lipid | Primary Bile Acid Metabolism | LC/MS Neg | 514.3 | 145-42-6 | 6675 | 63 |
| cortisone | -0.16 | 0.05 | 2.98E-03 | Lipid | Corticosteroids | LC/MS Neg | 359.2 | 53-06-5 | 222786 | 5 |
| guanidinoacetate | -0.15 | 0.05 | 3.32E-03 | Amino Acid | Creatine Metabolism | LC/MS Polar | 116.0 | 352-97-6 | 763 | 1 |
| pyroglutamine | 0.14 | 0.05 | 3.46E-03 | Amino Acid | Glutamate Metabolism | LC/MS Pos Early | 129.1 | 2353-44-8 | 134508 | 0 |
| eicosanodioate | -0.16 | 0.05 | 3.48E-03 | Lipid | Fatty Acid, Dicarboxylate | LC/MS Neg | 341.3 | 2424-92-2 | 75502 | 7 |
| creatinine | 0.14 | 0.05 | 4.41E-03 | Amino Acid | Creatine Metabolism | LC/MS Pos Early | 114.1 | 60-27-5 | 588 | 0 |
| 1-(1-enyl-palmitoyl)-2- arachidonoyl-GPC (P- 16:0/20:4) | 0.16 | 0.06 | 4.44E-03 | Lipid | Plasmalogen | LC/MS Pos Late | 766.6 | | | 1 |
| 4-hydroxychlorothalonil | -0.14 | 0.05 | 4.69E-03 | Xenobiotics | Chemical | LC/MS Neg | 244.9 | 28343-61-5 | 34217 | 113 |
| 1-stearoyl-2-arachidonoyl- GPC (18:0/20:4) | 0.15 | 0.05 | 4.77E-03 | Lipid | Phosphatidylcholine (PC) | LC/MS Pos Late | 810.6 | 35418-59-8 | 16219824 | 0 |
| serotonin | -0.16 | 0.06 | 4.82E-03 | Amino Acid | Tryptophan Metabolism | LC/MS Pos Early | 177.1 | 153-98-0 | 5202 | 16 |
| taurochenodeoxycholate | 0.15 | 0.05 | 5.92E-03 | Lipid | Primary Bile Acid Metabolism | LC/MS Neg | 498.3 | 6009-98-9 | 387316 | 7 |
| gulonate | 0.15 | 0.05 | 5.95E-03 | Cofactors and Vitamins | Ascorbate and Aldarate Metabolism | LC/MS Polar | 195.1 | 20246-53-1 | 9794176 | 10 |
| S-adenosylhomocysteine (SAH) | 0.15 | 0.05 | 6.40E-03 | Amino Acid | Methionine, Cysteine, SAM and Taurine Metabolism | LC/MS Neg | 383.1 | 979-92-0 | 439155 | 1 |

Supplementary Table 3. Serum metabolites associated with the number of *APOL1* risk alleles at p < 0.01 in AASK (n=588)

| cystathionine | 0.15 | 0.06 | 7.41E-03 | Amino Acid | Methionine, Cysteine, SAM and Taurine Metabolism | LC/MS Pos Early | 223.1 | 535-34-2 | 439258 | 19 |
|--|------|------|----------|---------------------------|--|-----------------|-------|-----------|--------|-----|
| homocitrulline | 0.15 | 0.06 | 7.65E-03 | Amino Acid | Urea cycle; Arginine and Proline Metabolism | LC/MS Polar | 188.1 | 1190-49-4 | 65072 | 2 |
| methylsuccinoylcarnitine (1) | 0.14 | 0.05 | 7.79E-03 | Amino Acid | Leucine, Isoleucine and Valine Metabolism | LC/MS Pos Early | 276.1 | | | 140 |
| 1-methylurate | 0.14 | 0.05 | 8.94E-03 | Xenobiotics | Xanthine Metabolism | LC/MS Pos Early | 183.1 | 708-79-2 | 69726 | 153 |
| N1-Methyl-2-pyridone-5- carboxamide | 0.14 | 0.06 | 9.82E-03 | Cofactors and Vitamins | Nicotinate and Nicotinamide Metabolism | LC/MS Neg | 151.1 | 701-44-0 | 69698 | 1 |

*The mass of 6-bromotryptophan in this table (281.0) is the monoisotopic mass of the negative charged ion of bromotryptophan containing ⁷⁹Br.

| Biochemical | Beta | SE | P-value | Super- pathway | Sub-pathway | Platform | Mass | CAS ID | Pubchem ID | # missing |
|---|-------|------|----------|-------------------|--|--------------------|-----------|---------------------------|---------------|--------------|
| choline phosphate | 0.15 | 0.05 | 1.36E-03 | Lipid | Phospholipid Metabolism | LC/MS Pos Early | 184.07332 | 72556-74-2 | 1014 | 0 |
| sarcosine | 0.15 | 0.05 | 1.64E-03 | Amino Acid | Glycine, Serine and Threonine Metabolism | LC/MS Pos Early | 90.05496 | 107-97-1 | 1088 | 0 |
| 7-alpha-hydroxy-3-oxo-4- cholestenoate (7-Hoca) | -0.15 | 0.05 | 1.65E-03 | Lipid | Sterol | LC/MS Neg | 429.30103 | 115538-85-7 | 3081085 | 0 |
| linoleoyl-arachidonoyl- glycerol (18:2/20:4) [2]* | 0.15 | 0.05 | 1.91E-03 | Lipid | Diacylglycerol | LC/MS Pos Late | 658.54051 | | | 0 |
| isoleucine | 0.14 | 0.05 | 2.56E-03 | Amino Acid | Leucine, Isoleucine and Valine Metabolism | LC/MS Pos Early | 132.10191 | 73-32-5 | 6306 | 0 |
| N-acetylleucine | 0.14 | 0.05 | 2.80E-03 | Amino Acid | Leucine, Isoleucine and Valine Metabolism | LC/MS Neg | 172.09791 | 1188-21-2 | 70912 | 0 |
| linoleoyl-arachidonoyl- glycerol (18:2/20:4) [1]* | 0.14 | 0.05 | 3.33E-03 | Lipid | Diacylglycerol | LC/MS Pos Late | 658.54051 | | | 2 |
| 1-(1-enyl-stearoyl)-GPE (P- 18:0)* | 0.14 | 0.05 | 4.01E-03 | Lipid | Lysoplasmalogen | LC/MS Pos Late | 466.3292 | | | 0 |
| cysteine sulfinic acid | 0.12 | 0.04 | 4.15E-03 | Amino Acid | Methionine, Cysteine, SAM and Taurine Metabolism | LC/MS Pos Early | 154.01686 | 207121-48-0 | 109 | 252 |
| pyrraline | 0.12 | 0.04 | 4.18E-03 | Xenobiotics | Food Component/Plant | LC/MS Pos Early | 255.13394 | 74509-14-1 | 122228 | 233 |
| 6-bromotryptophan | -0.13 | 0.05 | 5.73E-03 | Amino Acid | Tryptophan Metabolism | LC/MS Neg | 280.99311 | | 96735 | 0 |
| 1-(1-enyl-stearoyl)-2- arachidonoyl-GPE (P- 18:0/20:4)* | 0.13 | 0.05 | 6.35E-03 | Lipid | Plasmalogen | LC/MS Pos Late | 752.55887 | | 9547058 | 0 |
| isovalerylglycine | 0.13 | 0.05 | 6.46E-03 | Amino Acid | Leucine, Isoleucine and Valine Metabolism | LC/MS Neg | 158.08226 | 16284-60-9 | 546304 | 21 |
| 1-linoleoyl-2-arachidonoyl- GPC (18:2/20:4n6)* | 0.13 | 0.05 | 6.48E-03 | Lipid | Phosphatidylcholine (PC) | LC/MS Pos Late | 806.56944 | | | 3 |
| 2-hydroxyglutarate | 0.13 | 0.05 | 7.03E-03 | Lipid | Fatty Acid, Dicarboxylate | LC/MS Pos Early | 166.071 | 40951-21-1 | 43 | 0 |
| N6-carboxymethyllysine | 0.12 | 0.05 | 7.03E-03 | Carbohydrate | Advanced Glycation End-product | LC/MS Pos Early | 205.11829 | 4/3/5746 | 123800 | 13 |
| valine | 0.13 | 0.05 | 7.14E-03 | Amino Acid | Leucine, Isoleucine and Valine Metabolism | LC/MS Pos Early | 118.08626 | 72-18-4 | 6287 | 11 |
| sphingomyelin (d18:1/18:1, d18:2/18:0) | 0.12 | 0.04 | 8.04E-03 | Lipid | Sphingolipid Metabolism | LC/MS Pos Late | 729.5905 | 108392-10-5 | 6443882 | 0 |
| fumarate | -0.12 | 0.05 | 8.26E-03 | Energy | TCA Cycle | LC/MS Polar | 115.00368 | 100-17-8 | 444972 | 44 |
| stearoyl sphingomyelin (d18:1/18:0) | 0.12 | 0.05 | 8.82E-03 | Lipid | Sphingolipid Metabolism | LC/MS Pos Late | 731.60615 | 85187-10-6;85187- 10-6 | 6453725 | 0 |
| sphingomyelin (d18:2/21:0, d16:2/23:0)* | 0.11 | 0.04 | 9.58E-03 | Lipid | Sphingolipid Metabolism | LC/MS Pos Late | 771.63748 | | | 2 |

Supplementary Table 4. Serum metabolites associated with the number of *APOL1* risk alleles at p < 1e-2 in BioMe (n=678)

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| Biochemical | Spearman correlation | Superpathway | Subpathway | Platform | Mass | CAS ID | Pubchem ID | # missing |
|--|----------------------|---------------------------|--|--------------------|--------|---------------------------|---------------|--------------|
| tryptophan | 0.54 | Amino Acid | Tryptophan Metabolism | LC/MS Pos Early | 205.10 | 73-22-3 | 6305 | 0 |
| 3-methyl-2-oxovalerate | 0.30 | Amino Acid | Leucine, Isoleucine and Valine Metabolism | LC/MS Neg | 129.06 | 1460-34- 0;51829-07-3 | 47 | 1 |
| 2-hydroxystearate | 0.28 | Lipid | Fatty Acid, Monohydroxy | LC/MS Neg | 299.26 | 629-22-1 | 69417 | 1 |
| 4-methyl-2-oxopentanoate | 0.28 | Amino Acid | Leucine, Isoleucine and Valine Metabolism | LC/MS Neg | 129.06 | 816-66-0 | 70 | 1 |
| gamma-glutamyltyrosine | 0.27 | Peptide | Gamma-glutamyl Amino Acid | LC/MS Pos Early | 311.12 | 7432-23-7 | 94340 | 3 |
| tyrosine | 0.27 | Amino Acid | Tyrosine Metabolism | LC/MS Pos Farly | 182.08 | 60-18-4 | 6057 | 0 |
| glycerophosphorylcholine (GPC) | 0.27 | Lipid | Phospholipid Metabolism | LC/MS Pos Early | 258.11 | 28319-77-9 | 71920 | 0 |
| 2-hydroxypalmitate | 0.26 | Lipid | Fatty Acid, Monohydroxy | LC/MS Neg | 271.23 | 764-67-0 | 92836 | 0 |
| biliverdin | 0.26 | Cofactors and Vitamins | Hemoglobin and Porphyrin Metabolism | LC/MS Pos Late | 583.26 | 55482-27- 4;55482-27-4 | 5353439 | 3 |
| eicosanodioate | 0.24 | Lipid | Fatty Acid, Dicarboxylate | LC/MS Neg | 341.27 | 2424-92-2 | 75502 | 7 |
| docosadioate | 0.24 | Lipid | Fatty Acid, Dicarboxylate | LC/MS Neg | 369.30 | 505-56-6 | 244872 | 68 |
| homoarginine | 0.23 | Amino Acid | Urea cycle; Arginine and Proline Metabolism | LC/MS Pos Early | 189.13 | 156-86-5 | 9085 | 1 |
| linoleoylcarnitine (C18:2) | 0.23 | Lipid | Fatty Acid Metabolism(Acyl Carnitine) | LC/MS Pos Late | 424.34 | 36816-10-1 | 6450015 | 1 |
| 5-methyluridine (ribothymidine) | 0.23 | Nucleotide | Pyrimidine Metabolism, Uracil containing | LC/MS Neg | 257.08 | 1463-10-1 | 445408 | 1 |
| guanidinoacetate | 0.22 | Amino Acid | Creatine Metabolism | LC/MS Polar | 116.05 | 352-97-6 | 763 | 1 |
| 10-undecenoate (11:1n1) | 0.22 | Lipid | Medium Chain Fatty Acid | LC/MS Neg | 183.14 | 1333-28-4 | 14891 | 1 |
| linoleoylcholine* | 0.22 | Lipid | Fatty Acid Metabolism (Acyl Choline) | LC/MS Pos Late | 366.34 | | | 1 |
| 3beta,7alpha-dihydroxy-5- cholestenoate | 0.22 | Lipid | Sterol | LC/MS Neg | 431.32 | 115538-84-6 | 3081084 | 13 |
| alanine | 0.22 | Amino Acid | Alanine and Aspartate Metabolism | LC/MS Pos Early | 90.05 | 56-41-7 | 5950 | 0 |
| 7-alpha-hydroxy-3-oxo-4- cholestenoate (7-Hoca) | 0.22 | Lipid | Sterol | LC/MS Neg | 429.30 | 115538-85-7 | 3081085 | 1 |

Supplementary Table 5. Metabolites with Spearman correlation \leq -0.2 or \geq 0.2 with 6-bromotryptophan in AASK (n=588)

| alpha-hydroxyisocaproate | 0.22 | Amino Acid | Leucine, Isoleucine and Valine Metabolism | LC/MS Neg | 131.07 | 10303-64-7 | 83697 | 9 |
|---|-------|---------------------------|--|--------------------|--------|------------|----------|-----|
| fructose | 0.22 | Carbohydrate | Fructose, Mannose and Galactose Metabolism | LC/MS Polar | 225.06 | 57-48-7 | 5984 | 1 |
| lysine | 0.21 | Amino Acid | Lysine Metabolism | LC/MS Pos Early | 147.11 | 56-87-1 | 5962 | 0 |
| methionine | 0.21 | Amino Acid | Methionine, Cysteine, SAM and Taurine Metabolism | LC/MS Pos Early | 150.06 | 63-68-3 | 6137 | 0 |
| stearoylcholine* | 0.21 | Lipid | Fatty Acid Metabolism (Acyl Choline) | LC/MS Pos Late | 370.37 | | | 7 |
| bilirubin (E,E)* | 0.21 | Cofactors and Vitamins | Hemoglobin and Porphyrin Metabolism | LC/MS Pos Late | 585.27 | 114-25-0 | 5315454 | 4 |
| 1,5-anhydroglucitol (1,5-AG) | 0.21 | Carbohydrate | Glycolysis, Gluconeogenesis, and Pyruvate Metabolism | LC/MS Neg | 163.06 | 154-58-5 | 64960 | 1 |
| glycocholenate sulfate* | 0.21 | Lipid | Secondary Bile Acid Metabolism | LC/MS Neg | 254.62 | | | 1 |
| valine | 0.21 | Amino Acid | Leucine, Isoleucine and Valine Metabolism | LC/MS Pos Early | 118.09 | 72-18-4 | 6287 | 1 |
| thyroxine | 0.21 | Amino Acid | Tyrosine Metabolism | LC/MS Neg | 775.68 | 51-48-9 | 5819 | 3 |
| glycerate | 0.21 | Carbohydrate | Glycolysis, Gluconeogenesis, and Pyruvate Metabolism | LC/MS Polar | 105.02 | 600-19-1 | 752 | 0 |
| spermidine | 0.21 | Amino Acid | Polyamine Metabolism | LC/MS Pos Early | 146.17 | 124-20-9 | 1102 | 112 |
| palmitoylcholine | 0.21 | Lipid | Fatty Acid Metabolism (Acyl Choline) | LC/MS Pos Late | 342.34 | | 151731 | 1 |
| urea | -0.20 | Amino Acid | Urea cycle; Arginine and Proline Metabolism | LC/MS Pos Early | 121.07 | 57-13-6 | 1176 | 1 |
| arabonate/xylonate | -0.20 | Carbohydrate | Pentose Metabolism | LC/MS Polar | 165.04 | | | 2 |
| 3-methylglutaconate | -0.20 | Amino Acid | Leucine, Isoleucine and Valine Metabolism | LC/MS Polar | 99.05 | 5746-90-7 | 1551553 | 1 |
| N-acetyl-1-methylhistidine | -0.20 | Amino Acid | Histidine Metabolism | LC/MS Pos Early | 212.10 | | 193270 | 14 |
| arabitol/xylitol | -0.20 | Carbohydrate | Pentose Metabolism | LC/MS Polar | 151.06 | | 6912 | 1 |
| N-acetyl-3-methylhistidine | -0.21 | Amino Acid | Histidine Metabolism | LC/MS Pos Early | 212.10 | 37841-04-6 | 193270 | 101 |
| 1-palmitoyl-2-arachidonoyl- GPE (16:0/20:4)* | -0.21 | Lipid | Phosphatidylethanolamine (PE) | LC/MS Pos Late | 740.52 | | 9546800 | 1 |
| isobutyrylcarnitine (C4) | -0.22 | Amino Acid | Leucine, Isoleucine and Valine Metabolism | LC/MS Pos Early | 232.15 | 25518-49-4 | 168379 | 1 |
| indoleacetylglutamine | -0.22 | Amino Acid | Tryptophan Metabolism | LC/MS Neg | 302.11 | | 25200879 | 17 |
| p-cresol-glucuronide | -0.22 | Amino Acid | Tyrosine Metabolism | LC/MS Neg | 283.08 | 17680-99-8 | 154035 | 8 |
| 3-hydroxypyridine sulfate | -0.23 | Xenobiotics | Chemical | LC/MS Neg | 173.99 | 1955-23-3 | | 7 |

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| glucuronate | -0.28 | Carbohydrate | Aminosugar Metabolism | LC/MS Polar | 193.04 | 207300-70-7 | 444791 | 0 |
|------------------------------------|-------|---------------------------|--------------------------------------|--------------------|--------|-------------|----------|----|
| phenylacetylglutamine | -0.28 | Peptide | Acetylated Peptides | LC/MS Neg | 263.10 | 28047-15-6 | 92258 | 1 |
| phenylacetylglutamate | -0.27 | Peptide | Acetylated Peptides | LC/MS Neg | 264.09 | | 11579826 | 26 |
| phosphate | -0.26 | Energy | Oxidative Phosphorylation | LC/MS Pos Early | 98.98 | 7664-38-2 | 1061 | 0 |
| trimethylamine N-oxide | -0.26 | Lipid | Phospholipid Metabolism | LC/MS Pos Early | 76.08 | 1184-78-7 | 1145 | 1 |
| 4- hydroxyphenylacetylglutamine | -0.25 | Peptide | Acetylated Peptides | LC/MS Neg | 279.10 | | | 64 |
| N-acetylneuraminate | -0.25 | Carbohydrate | Aminosugar Metabolism | LC/MS Pos Early | 310.11 | 131-48-6 | 439197 | 1 |
| gulonate | -0.24 | Cofactors and Vitamins | Ascorbate and Aldarate Metabolism | LC/MS Polar | 195.05 | 20246-53-1 | 9794176 | 10 |

Supplementary Table 6. Association between APOL1 high-risk status and CKD progression with and without controlling for

serum 6-bromotyptophan in AASK (n=572) and BioMe (n=583)

| | Hazard ratio (95% co | onfidence interval) | | | |
|---------|---------------------------------------|-----------------------------------|--|--|--|
| AASK | Not controlling for 6-bromotryptophan | Controlling for 6-bromotryptophan | | | |
| Model 1 | 1.99 (1.51, 2.61) | 1.76 (1.33, 2.32) | | | |
| Model 2 | 1.80 (1.35, 2.38) | 1.68 (1.26, 2.24) | | | |
| BioMe | | | | | |
| Model 1 | 1.20 (0.78, 1.84) | 1.09 (0.71, 1.67) | | | |
| Model 2 | 1.14 (0.74, 1.76) | 1.05 (0.68, 1.62) | | | |

Unit of hazard ratio: doubling of 6-bromotryptophan

Covariates:

Model 1. AASK: age, sex, study assignment, percentages of European ancestry; BioMe: age, sex, baseline hypertension and diabetes status

Model 2: AASK: added measured GFR, coronary heart disease, and diastolic blood pressure; BioMe: added eGFR, coronary heart disease, and diastolic blood pressure.

Supplementary Table 7. Association between 6-bromotryptophan and CKD progression stratified by APOL1 high-risk status

| | Hazard ratio (95% confidence interval) | P-value | Hazard ratio (95% confidence interval) | P-value | P for interaction |
|---------|---|----------|---|---------|-------------------|
| AASK | APOL1 low-risk, n (event): 440 (160) | | APOL1 high-risk, n (event): 132 (76) | | |
| Model 1 | 0.61 (0.50, 0.74) | < 0.0001 | 0.84 (0.64, 1.09) | 0.19 | 0.05 |
| Model 2 | 0.67 (0.54, 0.84) | < 0.0001 | 0.93 (0.69, 1.26) | 0.65 | 0.11 |
| Model 3 | 0.72 (0.58, 0.90) | 0.004 | 0.97 (0.72, 1.32) | 0.85 | 0.12 |
| BioMe | APOL1 low-risk, n (event): 414 (69) | | APOL1 high-risk, n (event): 169 (31) | | |
| Model 1 | 0.56 (0.37, 0.83) | 0.004 | 0.64 (0.35, 1.14) | 0.13 | 0.90 |
| Model 2 | 0.58 (0.38, 0.87) | 0.009 | 0.67 (0.36, 1.25) | 0.21 | 0.81 |

Unit of hazard ratio: doubling of 6-bromotryptophan

Model 1. AASK: age, sex, study drug and blood pressure assignment; BioMe: age, sex, baseline diabetes, hypertension,

Model 2. AASK: Model 1 + measured GFR + coronary heart disease + diastolic blood pressure; BioMe: Model 1 + eGFR + coronary heart disease + diastolic blood pressure

Model 3. Model 2 + log proteinuria

Supplementary Table 8. Study population characteristics of Caucasian participants in MDRD: overall and stratified by tertile

of 6-bromotryptophan levels.

| | • • | | T (11 A | T (11 A | |
|---|---------------|-------------------|-------------------|----------------|---------|
| Variable | Overall | Tertile 1 | Tertile 2 | Tertile 3 | P-value |
| N | 390 | 130 | 130 | 130 | |
| Age | 52.3 (11.7) | 52.8 (12.0) | 51.4 (11.8) | 52.6 (11.3) | 0.89 |
| Male, n (%) | 252 (64.6) | 67 (51.5) | 92 (70.8) | 93 (71.5) | <0.001 |
| Study A, n (%) | 364 (93.3) | 121 (93.1) | 119 (91.5) | 124 (95.4) | 0.46 |
| Diet assignment, n (%) | | | | | 0.02 |
| Very low protein | 15 (3.8) | 5 (3.8) | 7 (5.4) | 3 (2.3) | |
| Low protein | 196 (50.3) | 52 (40.0) | 66 (50.8) | 78 (60.0) | |
| Usual protein | 179 (45.9) | 73 (56.2) | 57 (43.8) | 49 (37.7) | |
| Blood pressure assignment, moderate, n (%) | 191 (49) | 68 (52.3) | 61 (46.9) | 62 (47.7) | 0.64 |
| Cause of CKD, n (%) | | | | | 0.07 |
| Polycystic kidney disease | 185 (47.4) | 62 (47.7) | 61 (46.9) | 62 (47.7) | |
| Glomerular disease | 100 (25.6) | 29 (22.3) | 28 (21.5) | 43 (33.1) | |
| Other | 105 (26.9) | 39 (30) | 41 (31.5) | 25 (19.2) | |
| Coronary artery disease, n (%) | 40 (10.3) | 19 (14.6) | 12 (9.2) | 9 (6.9) | 0.11 |
| SBP, mm Hg | 128.9 (17.0) | 129.2 (18.1) | 129.5 (16.4) | 128.2 (16.5) | 0.61 |
| DBP, mm Hg | 79 (9.1) | 77.84 (9.6) | 79.9 (8.3) | 79.3 (9.3) | 0.21 |
| Estimated protein intake, g/kg/day, mean (SD) | 0.9 (0.3) | 1.0 (0.3) | 0.91 (0.3) | 0.86 (0.3) | <0.001 |
| Body mass index, kg/m ² | 27.3 (4.2) | 27 (4.6) | 27.49 (4.1) | 27.49 (3.7) | 0.34 |
| Measured GFR, mL/min/1.73m ² | 35.1 (10.2) | 33.19 (9.6) | 34.38 (9.8) | 37.88 (10.6) | <0.001 |
| Urine protein (mg/day), median (25th, 75th percentile) | 120 (50, 720) | 170 (50, 1185) | 160 (60, 1230) | 75 (40, 295) | <0.001 |
| History of diabetes, n (%) | 16 (4.1) | 8 (6.2) | 4 (3.1) | 4 (3.1) | 0.35 |
| History of hypertension, n (%) | 325 (83.3) | 108 (83.1) | 106 (81.5) | 111 (85.4) | 0.70 |

Abbreviations. SBP, systolic blood pressure; DBP, diastolic blood pressure; SD, standard deviation; GFR, glomerular filtration rate.

Supplementary Table 9. Association between serum 6-bromotryptophan and CKD progression in Caucasian participants in

MDRD (n=390, events=75)

| | Hazard ratio (95% Confidence Interval) | P-value |
|---------|---|---------|
| Model 1 | 0.44 (0.29, 0.66) | <0.001 |
| Model 2 | 0.49 (0.32, 0.75) | 0.001 |
| Model 3 | 0.52 (0.34, 0.79) | 0.002 |

Unit of hazard ratio: doubling of 6-bromotryptophan

Model 1. age, sex, study, diet, and blood pressure assignment, hypertension, diabetes, estimated protein intake, cause of CKD Model 2. Model 1 + measured GFR + diastolic blood pressure + coronary artery disease

Model 3: Model 2 + log(PCR)

Supplementary Figure 1. Flow diagram of participant inclusion in the AASK study population



Supplementary Figure 2. Flow diagram of participant inclusion in the BioMe study population



Supplementary Figure 3. Flow diagram of participant inclusion in the MDRD study population



Supplementary Figure 4. Experimental versus theoretical isotope pattern of bromotryptophan in negative ion mode (m-H)⁻: mass to charge ratio versus relative abundance



*Bromine has two significant and stable isotopes, one at 78.91834 and the other at 80.9163. In the neutral state, ⁷⁹BrTrp has a mass of 282.00039, and ⁸¹BrTrp has a mass of 283.99835. The figure depicts the m/z of the experimental metabolite which resolved with the negative ion method, and thus depicts the negative ion (which would have a mass of 280.99311 for ⁷⁹BrTrp and 282.99107 for ⁸¹BrTrp, respectively). The 281.99647 and 283.99442 represent the isomers with ¹³C.

Supplementary Figure 5. Comparison of fragmentation spectrum in negative ion mode (m-H)⁻ of experimental metabolite

and authentic reference standard of 6-bromotryptophan, with partial fragmentation ion annotation



*The bottom half of the graph represents the fragmentation pattern of the neat authentic standard in negative ion mode, which matches well the experimental fragmentation pattern.

Supplementary Figure 6. Co-elution of experimental metabolite in unspiked plasma with authentic reference standard of 6bromotryptophan spiked into plasma in negative ion mode (m-H)⁻



*Extracted Ion Chromatogram (XIC) for the negative ion mass of bromotryptophan 280.99277 (m-H)-. An unspiked plasma sample was extracted and analyzed 3 times on the primary LC/MS method of detection for bromotryptophan, specifically the RP/LC/MS Neg method. The endogenous peak for bromotryptophan can be seen in the lower three traces. In separate injections, the same plasma was extracted and, post extraction, an aliquot of neat 6-bromotryptophan was spiked into the plasma. This spiked plasma was run two times. The resulting increase in intensity of the bromotryptophan peak in the spiked traces shows complete co-elution with the endogenous peak. The x-axis represents retention time and the y-axis represents intensity.

Supplementary Figure 7. Distinctive elution patterns of four authentic reference standards of bromotryptophan in negative ion mode (m-H)⁻





Supplementary Figure 8. Correlations of experimental measure of 6-bromotryptophan from the negative ion method with the positive early and positive late ion methods.

* Note, the two separate reverse phase UPLC RP/UPLC-MS/MSⁿ methods using positive ion mode ESI methods are not as sensitive for 6-bromotryptophan as the negative ion method and therefore more samples had non-detected values. These were imputed in the study and shown in red on charts below.