

## SUPPLEMENTARY DATA

**Supplementary Table S1.** Relation between glucose homeostasis parameters and plasma amino acid concentrations in bivariate linear regression analyses in a subgroup of fasting participants.

Amino acid	FPG	HbA <sub>1c</sub>	Ins	Cpep	β-cell	IR
Gly	-0.19	-0.01	-0.12	-0.20	-0.07	-0.21
Ser	0.01	-0.05	-0.12	-0.12	-0.08	-0.11
Gln	-0.16	0.02	-0.12	-0.08	-0.10	-0.10
Asn	-0.02	-0.03	-0.07	-0.01	0.04	-0.02
Thr	-0.01	-0.05	-0.02	0.03	0.10	0.01
tCys	0.09	0.04	0.13	0.16	0.15	0.16
Met	0.16	-0.05	0.12	0.21	0.21	0.20
Pro	0.13	-0.03	0.15	0.30	0.31	0.29
His	-0.04	-0.09	0.01	0.04	0.05	0.03
Orn	0.04	-0.04	0.07	0.08	0.11	0.08
Lys	0.13	-0.04	0.08	0.14	0.11	0.15
Arg	0.01	-0.10	0.01	-0.04	0.00	-0.06
Trp	0.12	-0.07	0.11	0.13	0.15	0.13
Glu	0.14	-0.03	0.07	0.20	0.15	0.20
Asp	0.10	-0.05	0.11	0.19	0.14	0.19
Tyr	0.18	-0.03	0.25	0.34	0.32	0.35
Ala	0.18	-0.05	0.19	0.35	0.26	0.35
Phe	0.22	-0.05	0.25	0.39	0.29	0.39
Leu	0.31	-0.04	0.21	0.34	0.22	0.37
Val	0.31	-0.03	0.25	0.38	0.30	0.40
Ile	0.31	-0.03	0.24	0.40	0.32	0.43

Standardized β coefficients obtained by bivariate linear regression analyses. Sample size of  $n = 607$ . Homeostasis Model Assessment (HOMA-2) was used to estimate both insulin resistance and β-cell function based on serum C-peptide. Abbreviations: Ala, alanine; Arg, arginine; Asn, asparagine; Asp, aspartic acid; β -cell, β-cell activity; Cpep, fasting serum C-peptide; FPG, fasting plasma glucose; Gln, glutamine; Glu, glutamic acid; Gly, glycine; HbA<sub>1c</sub>, glycated hemoglobin; His, histidine; Ile, isoleucine; Ins, fasting serum insulin; IR, insulin resistance; Leu, leucine; Lys, lysine; Met, methionine; Orn, ornithine; Phe, phenylalanine; Pro, proline; Ser, serine; tCys, total cysteine; Thr, threonine; Trp, tryptophan; Tyr, tyrosine; Val, valine.

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**Supplementary Table S2.** Association of baseline plasma amino acid concentrations with incident type 2 diabetes by Cox's regression.

Amino acid	Model 1		Model 2		Model 3		Model 4	
	HR	P	HR	P	HR	P	SHR	P
Val	1.56	0.001	1.21*	0.007	1.05*	0.51	1.07	0.33
Leu	1.45	0.001	1.20*	0.008	1.01*	0.91	1.02	0.74
Ile	1.41	0.001	1.15*	0.03	0.97*	0.66	0.97	0.66
Glu	1.36	0.001	1.16*	0.03	1.09*	0.24	1.10	0.17
Tyr	1.35	0.001	1.17*	0.03	1.04*	0.59	1.06	0.48
tCys	1.32	0.001	1.13*	0.08	1.06*	0.41	1.02	0.78
Phe	1.31	0.001	1.16*	0.03	0.99*	0.84	0.98	0.72
Asp	1.26	0.001	1.11*	0.13	1.05*	0.49	1.05	0.46
Trp	1.25	0.001	1.12*	0.08	0.98*	0.74	1.01	0.91
Arg	1.21	0.003	1.24	0.001	1.21	0.002	1.22	0.002
Ala	1.19	0.004	1.04*	0.58	0.88*	0.09	0.89	0.08
Lys	1.15	0.03	1.07	0.29	0.99*	0.87	1.01	0.93
Met	1.15	0.02	1.12	0.06	0.98*	0.78	0.99	0.87
Pro	1.06	0.34	0.95*	0.44	0.86*	0.04	0.86	0.04
Orn	1.00	0.96	0.97	0.69	0.96	0.57	0.94	0.37
His	0.93	0.26	0.94	0.36	0.93	0.23	0.95	0.39
Ser	0.87	0.02	1.02*	0.71	0.97*	0.65	0.98	0.72

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Gln	0.82	0.001	0.95*	0.42	0.96*	0.54	0.95	0.46
Asn	0.82	0.001	0.95*	0.42	0.87	0.04	0.87	0.03
Gly	0.66	0.001	0.83*	0.008	0.89*	0.09	0.90	0.10

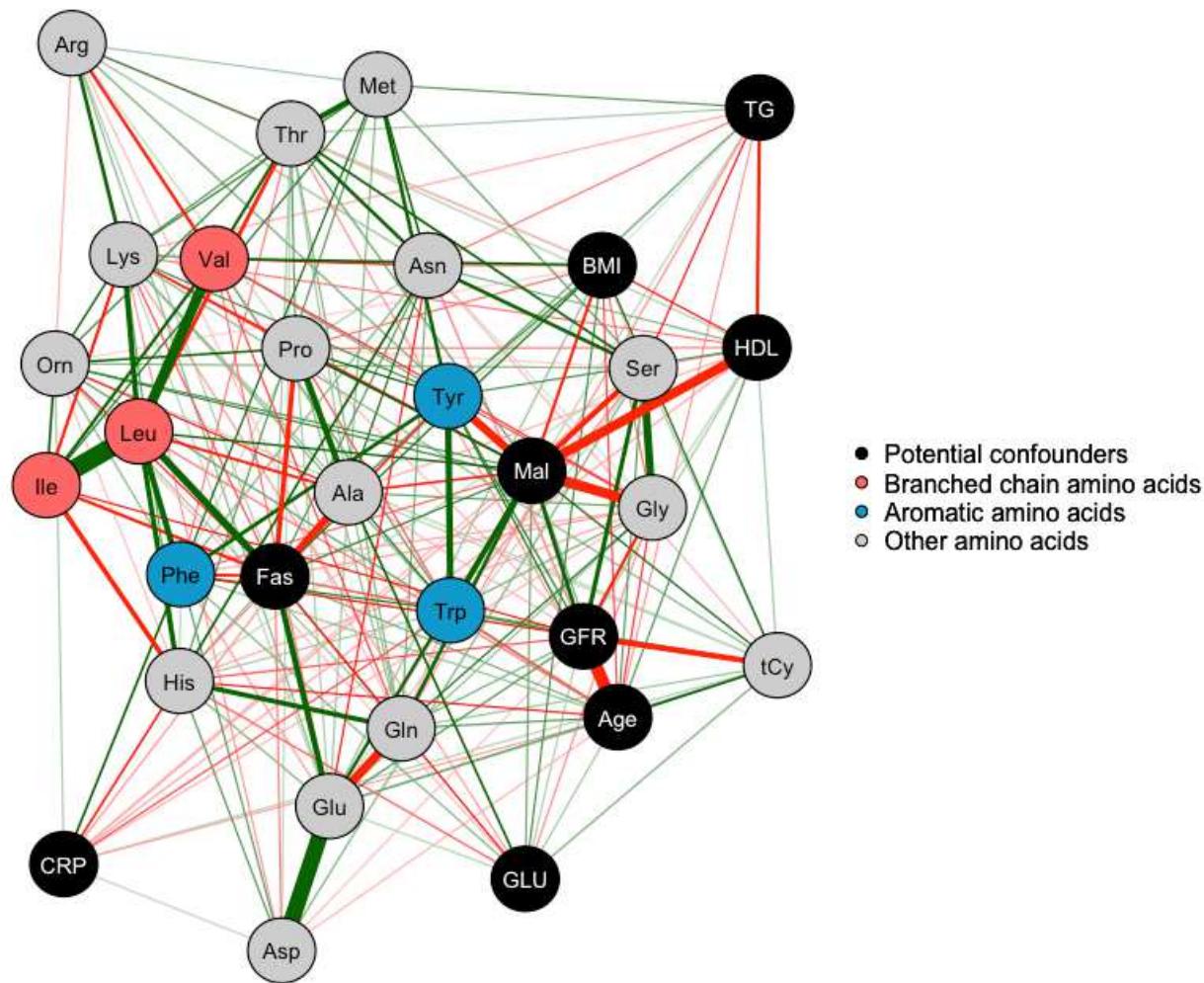
Observations for  $n = 2519$  (incident diabetes,  $n = 267$ ; mortality events,  $n = 464$ ). Cox's regression with adjustment for age & sex (Model 1), age, sex, GFR, BMI, HDL, TAG & CRP (Model 2), Model 2 plus plasma glucose (Model 3), and Model 3 plus mortality (Model 4). Change in estimate calculated as (HR crude - HR adjusted)/HR adjusted, with \* denoting change of  $\geq 10\%$ . Abbreviations: Ala, alanine; Arg, arginine; Asn, asparagine; Asp, aspartic acid; BMI, body mass index; CRP, C-reactive protein; GFR, estimated glomerular filtration rate; Gln, glutamine; Glu, glutamic acid; Gly, glycine; HDL, high-density lipoprotein; His, histidine; HR, hazard ratio; Ile, isoleucine; Leu, leucine; Lys, lysine; Met, methionine; Orn, ornithine; Phe, phenylalanine; Pro, proline; Ser, serine; SHR, standardized subdistribution hazard (risk of incident diabetes related to selected variable, given that participant has not died); tCys, total cysteine; TAG, triacylglycerol; Thr, threonine; Trp, tryptophan; Tyr, tyrosine; Val, valine.

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**Supplementary Figure S1.** Mixed Gaussian Modeling network plot illustrating weighted adjacencies between baseline plasma amino acid concentrations and confounders related to diabetes risk in diabetes-free participants at baseline.

Observations for  $n = 2519$ . Positive associations are displayed in green and negative associations in red. Abbreviations: Ala, alanine; Arg, arginine; Asn, asparagine; Asp, aspartic acid; BMI, body mass index; CRP, c-reactive protein; Fas, fasting status; GFR, estimated glomerular filtration rate; Gln, glutamine; GLU, glucose; Glu, glutamic acid; Gly, glycine; HDL, high-density lipoprotein; His, histidine; Ile, isoleucine; Leu, leucine; Lys, lysine; Mal, sex with males as reference; Met, methionine; Orn, ornithine; Phe, phenylalanine; Pro, proline; Ser, serine; tCys, total cysteine; TAG, triacylglycerol; Thr, threonine; Trp, tryptophan; Tyr, tyrosine; Val, valine.

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**Supplementary Figure S2.** Boxplot analyses showing baseline plasma arginine levels in relation to metformin use.

Analysis in  $n = 192$  metformin users compared to  $n = 192$  age and sex propensity score matched participants not using metformin medication.

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