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Supplementary Table 1. Mean (SD) concentrations of individual serum metabolites quantified by NMR and assayed for association with HOMA-IR.

Metabolite	Men	Women	P	$P_{adjusted}$
Alanine [cu]	13.3 (2.3)	13.0 (2.3)	2×10^{-9}	0.62
Glutamine [cu]	17.2 (2.9)	15.6 (2.9)	1×10^{-107}	2×10^{-99}
Histidine [cu]	2.2 (0.39)	2.1 (0.40)	8×10^{-14}	2×10^{-7}
Isoleucine [cu]	2.0 (0.53)	1.5 (0.40)	$<1\times10^{-324}$	1×10^{-123}
Leucine [cu]	3.1 (0.59)	2.6 (0.53)	1×10^{-273}	4×10^{-95}
Phenylalanine [cu]	2.6 (0.45)	2.5 (0.47)	9×10^{-5}	2×10^{-13}
Tyrosine [cu]	1.8 (0.38)	1.6 (0.38)	6×10^{-107}	5×10^{-19}
Valine [cu]	7.4 (1.4)	6.1 (1.2)	$<1\times10^{-324}$	5×10^{-135}
Lactate [cu]	33 (12)	33 (11)	0.22	0.17
Pyruvate [cu]	2.7 (0.87)	2.7 (0.84)	0.51	8×10^{-5}
Citrate [cu]	3.0 (0.72)	3.1 (0.74)	2×10^{-21}	6×10^{-6}
3-hydroxybutyrate [cu]	4.1 (3.1)	4.6 (3.8)	9×10^{-11}	0.01
Acetate [cu]	1.4 (0.84)	1.4 (0.67)	0.37	0.15
Acetoacetate [cu]	1.7 (1.3)	1.6 (1.2)	0.03	2×10^{-4}
Glycerol [cu]	2.6 (1.1)	2.7 (1.3)	8×10^{-9}	5×10^{-25}
Creatinine [cu]	2.2 (0.41)	1.8 (0.36)	$<1\times10^{-324}$	4×10^{-266}
Urea [cu]	2.1 (1.0)	1.8 (0.90)	5×10^{-32}	6×10^{-23}
α1-acid glycoprotein [cu]	43 (8.2)	41 (7.4)	2×10^{-10}	1×10^{-7}
Total fatty acids [mmol/l]	10 (2.5)	9.4 (2.3)	5×10^{-26}	0.51
Esterified cholesterol [mmol/l]	3.9 (0.85)	3.7 (0.78)	1×10^{-20}	1×10^{-3}
Free cholesterol [mmol/l]	1.4 (0.33)	1.4 (0.33)	5×10^{-12}	0.02
Total phosphoglycerides [mmol/l]	0.88 (0.20)	0.91 (0.23)	3×10^{-9}	2×10^{-9}
Phosphatidylcholine and other cholines [mmol/l]	2.0 (0.41)	2.1 (0.51)	4×10^{-24}	2×10^{-19}
Sphingomyelins and other sphingolipids [mmol/l]	0.34 (0.083)	0.34 (0.085)	6×10^{-4}	1×10^{-8}
Total cholines [mmol/l]	2.2 (0.43)	2.3 (0.49)	3×10^{-10}	8×10^{-10}
Linoleic acid [mmol/l]	3.3 (0.71)	3.2 (0.69)	6×10^{-8}	0.03
Docosahexaenoic acid [mmol/l]	0.11 (0.049)	0.13 (0.054)	6×10^{-30}	3×10^{-24}
Other polyunsaturated fatty acids than linoleic acid [mmol/l]	2.1 (0.57)	2.0 (0.54)	4×10^{-12}	0.66
ω-3 fatty acids [mmol/l]	0.30 (0.14)	0.28 (0.12)	7×10^{-11}	0.65
ω-6 and ω-7 fatty acids [mmol/l]	3.8 (0.80)	3.6 (0.77)	3×10^{-13}	0.57
ω-9 and saturated FA [mmol/l]	6.0 (1.7)	5.5 (1.5)	4×10^{-30}	0.53
ω-3/ω-6 [%]	8.1 (3.4)	7.9 (3.1)	0.001	0.65
ω-3/ω-9 [%]	5.1 (2.0)	5.2 (1.9)	0.67	0.77
Average number of methylene groups per fatty acid chain	11 (0.71)	11 (0.69)	0.36	0.73
Average number of methylene groups per double bond	7.8 (0.66)	7.7 (0.56)	2×10^{-14}	0.67
Average number of double bonds in a fatty acid chain	1.4 (0.12)	1.5 (0.11)	1×10^{-9}	0.79
Bisallylic groups/double bond [%]	53 (3.1)	53 (2.8)	2×10^{-12}	0.41

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Bisallylic groups/total fatty acids [%]	76 (9.8)	78 (8.7)	6×10^{-12}	0.87
Average fatty acid chain length	20 (0.92)	20 (0.90)	0.13	0.69

Mean (SD) concentrations of metabolites assayed in this study. P-values are for 2-tailed t-tests comparing concentrations for men and women with and without adjustment for age and waist circumference. cu: standardized concentration units.

Supplementary Table 2. Genetic variants affecting metabolite levels tested for association with HOMA-insulin resistance.

Gene candidate	**SNP ID**	**Chromosome position**	**Lead metabolite trait***	**R^2 (%)**	**Alleles (coded / other)**	**CAF (%)**	**HWE**
GCKR	rs1260326	2p23	Isoleucine	0.78	T/C	65	0.35
SLC1A4	rs2160387	2p15-p13	Valine	0.17	C/T	42	0.74
PPM1K	rs1440581	4q22.1	Valine	0.45	C/T	47	0.45
F12	rs2545801	5q33-qter	Phenylalanine	0.25	T/C	73	0.09
ALB	rs115136538	4q13.3	Phenylalanine	0.38	C/T	2	0.99
TAT	rs4788815	16q22.1	Tyrosine	0.56	A/T	35	0.41
SLC16A10	rs6900341	6q21-q22	Tyrosine	0.41	T/G	62	0.92
DHDPSL	rs2297644	10q24.2	Glutamine	0.41	C/T	20	0.38
GLS2	rs2638315	12q13	Glutamine	1.14	G/C	84	0.09
FADS1-2-3	rs174547	11q12.2	ω-3	0.86	T/C	42	0.22
ANGPTL3	rs1168029	1p31.1-p22.3	ω-6 and ω-7	0.36	A/G	70	0.91
APOA1-C3-A4-A5	rs651821	11q23-q24	Total fatty acids	0.33	T/C	91	0.43

Genetic associations were adjusted sex, age, waist and the 10 first principal components accounting for population structure. Associations were meta-analyzed from the Northern Finland Birth Cohort 1966 and the Cardiovascular Risk in Young Finns Study (total n=6,404).

*: Several SNPs were associated with multiple metabolites and the column indicates the top association.

R^2 indicates the variance explained by the SNP for the lead metabolite trait.

CAF: coded allele frequency; HWE: P-value for Hardy-Weinberg equilibirum.

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Supplementary Table 3. Backwards stepwise regression of the metabolites associated with HOMA-IR.

	Men		Women	
Metabolite	**β (*SE*)**	**P**	**β (*SE*)**	**P**
Leucine	–	–	-0.11 (0.023)	4×10^{-6}
Isoleucine	0.16 (0.023)	2×10^{-11}	0.080 (0.027)	0.003
Valine	–	–	0.092 (0.022)	4×10^{-5}
Phenylalanine	0.054 (0.021)	0.01	0.099 (0.018)	7×10^{-8}
Tyrosine	0.11 (0.021)	7×10^{-7}	–	–
Alanine	0.072 (0.023)	0.002	0.044 (0.021)	0.04
Glutamine	-0.18 (0.018)	4×10^{-25}	-0.19 (0.017)	2×10^{-27}
Lactate	0.063 (0.018)	4×10^{-4}	–	–
Pyruvate	0.083 (0.019)	2×10^{-5}	0.14 (0.017)	2×10^{-16}
3-hydroxybutyrate	–	–	0.083 (0.027)	0.003
Acetoacetate	-0.12 (0.016)	6×10^{-15}	-0.15 (0.027)	7×10^{-12}
α1-acid glycoprotein	–	–	–	–
Total fatty acids	–	–	0.441 (0.063)	3×10^{-12}
ω-3 fatty acids	0.051 (0.020)	0.009	–	–
ω-6 and ω-7 fatty acids	–	–	–	–
ω-9 and saturated fatty acids	–	–	–	–
Phosphocholines	–	–	-0.16 (0.055)	0.004
Phosphoglycerides	–	–	–	–
Av. methylene groups per double bond	0.098 (0.023)	2×10^{-5}	0.058 (0.023)	0.02
Av. number of double bonds per FA chain	–	–	0.10 (0.029)	4×10^{-4}

β-regression coefficients (standard error) in units of 1-SD change in HOMA-IR per 1-SD increase in metabolite concentration. The metabolites in the table were included in the initial stepwise regression model, and age, waist, total cholesterol, HDL cholesterol and triglycerides were forced in the model. Metabolites were excluded from the model in a stepwise manner if $P<0.05$. Associations were meta-analyzed for the two cohorts (n=7,098).

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Supplementary Figure 1. Associations of metabolites with HOMA-insulin resistance for men and women across tertiles of waist circumference. Linear regression models and ANCOVA were adjusted for age and waist circumference, total cholesterol, HDL cholesterol and triglycerides. The magnitude of association is in units of 1-SD increased HOMA-IR per 1-SD increase in the metabolite level and error bars indicate standard errors. The associations were analyzed separately in both cohorts and combined by inverse variance weighted meta-analysis. Associations for amino acids are shown in Figure 1.
*: P<0.05; ♦: P<0.0005 for association of metabolite with HOMA-IR.
◉: P<0.05; ⊗: P<0.0005 for metabolite×waist interaction by ANCOVA indicating different slopes for association between metabolite concentration and HOMA-IR across tertiles of waist circumference.