

Supplemental Data

Supplementary Table 1: Association between plasma glucagon concentrations and liver fat content (multiple linear regression)

Glucagon (log)			
	Model glucagon 1	Model glucagon 2	Model glucagon 3
(Intercept)	3.62 *** (1.61 to 5.62)	3.37 ** (1.28 to 5.46)	2.68 * (0.46 to 4.91)
Liver fat content (log)	0.11 * (0.01 to 0.21)	0.11 * (0.01 to 0.21)	0.10 (-0.01 to 0.21)
Age	-0.01 (-0.02 to 0.01)	-0.01 (-0.02 to 0.01)	-0.00 (-0.02 to 0.02)
Male gender	0.06 (-0.20 to 0.32)	0.07 (-0.19 to 0.33)	0.01 (-0.27 to 0.30)
BMI	-0.02 (-0.05 to 0.01)	-0.02 (-0.05 to 0.01)	-0.02 (-0.05 to 0.02)
eGFR	-0.00 (-0.01 to 0.00)	-0.00 (-0.01 to 0.00)	-0.00 (-0.01 to 0.01)
Fasting glucose		0.02 (-0.02 to 0.06)	0.02 (-0.02 to 0.06)
Insulin			-0.05 (-0.31 to 0.21)
Metformin			0.22 (-0.10 to 0.54)
DDP-4 inhibitor			0.06 (-0.24 to 0.37)
GLP-1RA			0.01 (-0.26 to 0.28)
SGLT-1 inhibitor			0.22 (-0.11 to 0.56)
Sulfonylurea			-0.13 (-0.55 to 0.29)

The estimated difference/beta (CI) in glucagon (log) by a unit increase in liver fat content (log) (multiple linear regression). Model glucagon 1 with adjustment for age, gender, BMI, and eGFR; Model glucagon 2 with further adjustment for fasting glucose; and Model glucagon 3 with further adjustment for diabetes medication. Variables with a 95% CI not including zero are considered significantly associated with glucagon. *** $p < 0.001$; ** $p < 0.01$; * $p < 0.05$.

Abbreviations: BMI, body mass index; DDP-4, dipeptidyl peptidase 4; eGFR, estimated glomerular filtration rate; GLP-1RA, glucagon-like peptide 1 receptor agonist; SGLT-2, sodium glucose cotransporter 2.

Supplementary Table 2: Association between plasma amino acid concentrations and liver fat content (multiple linear regression)

Alanine		
	Model amino acid 1	Model amino acid 2
(Intercept)	542.79 ** (149.51 to 936.07)	566.67 ** (148.03 to 985.32)
Liver fat content (log)	19.64 (-0.21 to 39.49)	20.34 (-0.01 to 40.69)
Age	1.51 (-1.78 to 4.79)	1.46 (-1.85 to 4.78)
Gender (male)	-27.40 (-78.30 to 23.50)	-27.00 (-78.18 to 24.18)
BMI	-3.94 (-10.22 to 2.33)	-4.07 (-10.42 to 2.27)
eGFR	-1.06 (-2.61 to 0.49)	-1.08 (-2.65 to 0.48)
Glucagon (log)		-6.60 (-44.95 to 31.74)
Arginine		
	Model 1	Model 2
(Intercept)	90.94 ** (34.01 to 147.86)	99.51 ** (39.09 to 159.93)
Liver fat content (log)	-1.22 (-4.10 to 1.65)	-0.97 (-3.91 to 1.97)
Age	-0.48 (-0.95 to 0.00)	-0.49 * (-0.97 to -0.01)
Gender (male)	3.03 (-4.34 to 10.40)	3.18 (-4.21 to 10.56)
BMI	0.69 (-0.22 to 1.59)	0.64 (-0.28 to 1.56)
eGFR	-0.10 (-0.33 to 0.12)	-0.11 (-0.34 to 0.11)
Glucagon (log)		-2.37 (-7.90 to 3.16)

Asparagine		
	Model 1	Model 2
(Intercept)	60.73 *** (33.38 to 88.08)	64.13 *** (35.07 to 93.19)
Liver fat content (log)	-0.77 (-2.15 to 0.61)	-0.67 (-2.08 to 0.74)
Age	-0.16 (-0.39 to 0.07)	-0.16 (-0.39 to 0.07)
Gender (male)	1.98 (-1.56 to 5.52)	2.04 (-1.52 to 5.59)
BMI	-0.05 (-0.49 to 0.38)	-0.07 (-0.51 to 0.37)
eGFR	-0.08 (-0.19 to 0.03)	-0.09 (-0.19 to 0.02)
Glucagon (log)		-0.94 (-3.60 to 1.72)
Aspartate		
	Model 1	Model 2
(Intercept)	5.88 (-3.84 to 15.60)	2.73 (-7.45 to 12.92)
Liver fat content (log)	-0.12 (-0.61 to 0.37)	-0.21 (-0.70 to 0.29)
Age	-0.04 (-0.12 to 0.04)	-0.03 (-0.11 to 0.05)
Gender (male)	0.41 (-0.85 to 1.67)	0.36 (-0.89 to 1.60)
BMI	0.07 (-0.09 to 0.22)	0.08 (-0.07 to 0.24)
eGFR	0.01 (-0.03 to 0.05)	0.01 (-0.03 to 0.05)
Glucagon (log)		0.87 (-0.06 to 1.80)

Glutamate		
	Model 1	Model 2
(Intercept)	273.99 *	201.56
	(34.35 to 513.63)	(-50.08 to 453.20)
Liver fat content (log)	8.19	6.06
	(-3.90 to 20.29)	(-6.17 to 18.29)
Age	-1.01	-0.88
	(-3.01 to 0.99)	(-2.87 to 1.11)
Gender (male)	6.56	5.33
	(-24.46 to 37.58)	(-25.43 to 36.10)
BMI	-2.42	-2.04
	(-6.25 to 1.40)	(-5.85 to 1.78)
eGFR	-0.41	-0.33
	(-1.36 to 0.53)	(-1.27 to 0.61)
Glucagon (log)		20.03
		(-3.02 to 43.08)

Glutamine		
	Model 1	Model 2
(Intercept)	484.61 *	678.56 **
	(49.94 to 919.28)	(229.97 to 1127.16)
Liver fat content (log)	-5.06	0.65
	(-26.99 to 16.88)	(-21.16 to 22.45)
Age	0.35	-0.00
	(-3.28 to 3.98)	(-3.55 to 3.55)
Gender (male)	-13.20	-9.91
	(-69.46 to 43.06)	(-64.75 to 44.93)
BMI	4.25	3.22
	(-2.69 to 11.18)	(-3.58 to 10.02)
eGFR	-0.64	-0.85
	(-2.35 to 1.07)	(-2.53 to 0.83)
Glucagon (log)		-53.63 *
		(-94.72 to -12.54)

Glycine		
	Model 1	Model 2
(Intercept)	154.07 (-43.07 to 351.20)	234.19 * (29.61 to 438.77)
Liver fat content (log)	-8.03 (-17.98 to 1.92)	-5.67 (-15.62 to 4.27)
Age	0.17 (-1.48 to 1.82)	0.03 (-1.59 to 1.64)
Gender (male)	-4.76 (-30.28 to 20.75)	-3.40 (-28.41 to 21.61)
BMI	1.26 (-1.88 to 4.41)	0.84 (-2.26 to 3.94)
eGFR	0.27 (-0.51 to 1.05)	0.19 (-0.58 to 0.95)
Glucagon (log)		-22.15 * (-40.89 to -3.42)

Histidine		
	Model 1	Model 2
(Intercept)	88.54 *** (50.10 to 126.98)	88.81 *** (47.87 to 129.75)
Liver fat content (log)	0.82 (-1.12 to 2.76)	0.83 (-1.16 to 2.82)
Age	-0.14 (-0.47 to 0.18)	-0.14 (-0.47 to 0.18)
Gender (male)	2.06 (-2.92 to 7.03)	2.06 (-2.94 to 7.07)
BMI	0.14 (-0.48 to 0.75)	0.13 (-0.49 to 0.76)
eGFR	-0.10 (-0.25 to 0.05)	-0.10 (-0.25 to 0.06)
Glucagon (log)		-0.07 (-3.82 to 3.68)

Isoleucine		
	Model 1	Model 2
(Intercept)	105.66 *** (46.90 to 164.42)	63.76 * (6.26 to 121.25)
Liver fat content (log)	2.01 (-0.95 to 4.98)	0.78 (-2.02 to 3.57)
Age	-0.39 (-0.88 to 0.11)	-0.31 (-0.76 to 0.15)
Gender (male)	2.94 (-4.66 to 10.55)	2.23 (-4.80 to 9.26)
BMI	-0.37 (-1.31 to 0.57)	-0.15 (-1.02 to 0.73)
eGFR	0.00 (-0.23 to 0.23)	0.05 (-0.17 to 0.26)
Glucagon (log)		11.59 *** (6.32 to 16.85)
Leucine		
	Model 1	Model 2
(Intercept)	178.29 *** (75.08 to 281.51)	121.31 * (16.64 to 225.98)
Liver fat content (log)	1.87 (-3.34 to 7.08)	0.20 (-4.89 to 5.28)
Age	-0.57 (-1.44 to 0.29)	-0.47 (-1.30 to 0.36)
Gender (male)	7.63 (-5.73 to 20.99)	6.66 (-6.13 to 19.46)
BMI	-0.29 (-1.93 to 1.36)	0.02 (-1.57 to 1.60)
eGFR	-0.07 (-0.48 to 0.34)	-0.01 (-0.40 to 0.38)
Glucagon (log)		15.76 ** (6.17 to 25.34)

Lysine		
	Model 1	Model 2
(Intercept)	193.73 *** (83.22 to 304.25)	192.08 ** (74.37 to 309.78)
Liver fat content (log)	-2.11 (-7.69 to 3.46)	-2.16 (-7.88 to 3.56)
Age	-0.48 (-1.40 to 0.45)	-0.47 (-1.40 to 0.46)
Gender (male)	8.53 (-5.77 to 22.84)	8.50 (-5.89 to 22.89)
BMI	0.91 (-0.85 to 2.68)	0.92 (-0.86 to 2.71)
eGFR	-0.05 (-0.49 to 0.38)	-0.05 (-0.49 to 0.39)
Glucagon (log)		0.46 (-10.32 to 11.24)
Methionine		
	Model 1	Model 2
(Intercept)	23.22 ** (8.09 to 38.35)	17.90 * (2.09 to 33.71)
Liver fat content (log)	-0.14 (-0.90 to 0.63)	-0.29 (-1.06 to 0.47)
Age	-0.06 (-0.19 to 0.07)	-0.05 (-0.18 to 0.07)
Gender (male)	1.21 (-0.75 to 3.17)	1.12 (-0.82 to 3.05)
BMI	0.07 (-0.17 to 0.31)	0.10 (-0.14 to 0.34)
eGFR	0.00 (-0.06 to 0.06)	0.01 (-0.05 to 0.07)
Glucagon (log)		1.47 * (0.02 to 2.92)

Phenylalanine		
	Model 1	Model 2
(Intercept)	47.79 ** (17.05 to 78.54)	35.44 * (3.52 to 67.36)
Liver fat content (log)	-0.80 (-2.36 to 0.75)	-1.17 (-2.72 to 0.38)
Age	-0.05 (-0.30 to 0.21)	-0.02 (-0.28 to 0.23)
Gender (male)	3.08 (-0.90 to 7.06)	2.87 (-1.04 to 6.77)
BMI	0.50 * (0.01 to 0.99)	0.56 * (0.08 to 1.05)
eGFR	-0.03 (-0.15 to 0.10)	-0.01 (-0.13 to 0.11)
Glucagon (log)		3.42 * (0.49 to 6.34)

Proline		
	Model 1	Model 2
(Intercept)	251.43 * (31.87 to 470.99)	256.06 * (22.23 to 489.90)
Liver fat content (log)	8.39 (-2.69 to 19.47)	8.53 (-2.84 to 19.89)
Age	0.54 (-1.30 to 2.37)	0.53 (-1.32 to 2.38)
Gender (male)	-4.55 (-32.97 to 23.87)	-4.47 (-33.06 to 24.12)
BMI	-0.83 (-4.33 to 2.68)	-0.85 (-4.39 to 2.69)
eGFR	-0.59 (-1.46 to 0.27)	-0.60 (-1.47 to 0.28)
Glucagon (log)		-1.28 (-22.70 to 20.14)

Serine		
	Model 1	Model 2
(Intercept)	101.25 ** (30.51 to 171.98)	115.51 ** (40.63 to 190.38)
Liver fat content (log)	-2.57 (-6.14 to 1.00)	-2.15 (-5.79 to 1.49)
Age	-0.01 (-0.60 to 0.58)	-0.04 (-0.63 to 0.55)
Gender (male)	5.29 (-3.86 to 14.45)	5.53 (-3.62 to 14.69)
BMI	-0.48 (-1.60 to 0.65)	-0.55 (-1.69 to 0.58)
eGFR	0.18 (-0.10 to 0.46)	0.16 (-0.12 to 0.44)
Glucagon (log)		-3.94 (-10.80 to 2.91)

Threonine		
	Model 1	Model 2
(Intercept)	138.03 ** (49.88 to 226.18)	140.33 ** (46.45 to 234.21)
Liver fat content (log)	-0.37 (-4.82 to 4.08)	-0.30 (-4.87 to 4.26)
Age	-0.45 (-1.18 to 0.29)	-0.45 (-1.19 to 0.29)
Gender (male)	2.94 (-8.47 to 14.35)	2.98 (-8.49 to 14.46)
BMI	0.24 (-1.17 to 1.65)	0.23 (-1.20 to 1.65)
eGFR	-0.08 (-0.42 to 0.27)	-0.08 (-0.43 to 0.27)
Glucagon (log)		-0.64 (-9.24 to 7.96)

Tryptophan		
	Model 1	Model 2
(Intercept)	51.16 ** (19.86 to 82.45)	42.35 * (9.43 to 75.27)
Liver fat content (log)	0.12 (-1.46 to 1.69)	-0.14 (-1.74 to 1.46)
Age	-0.16 (-0.42 to 0.10)	-0.14 (-0.40 to 0.12)
Gender (male)	4.46 * (0.41 to 8.51)	4.31 * (0.28 to 8.33)
BMI	0.17 (-0.33 to 0.67)	0.22 (-0.28 to 0.72)
eGFR	0.07 (-0.06 to 0.19)	0.07 (-0.05 to 0.20)
Glucagon (log)		2.43 (-0.58 to 5.45)

Tyrosine		
	Model 1	Model 2
(Intercept)	31.30 (-6.91 to 69.50)	20.70 (-19.51 to 60.91)
Liver fat content (log)	-1.17 (-3.10 to 0.76)	-1.48 (-3.44 to 0.47)
Age	-0.03 (-0.35 to 0.29)	-0.01 (-0.33 to 0.31)
Gender (male)	2.50 (-2.44 to 7.45)	2.32 (-2.59 to 7.24)
BMI	0.72 * (0.11 to 1.33)	0.78 * (0.17 to 1.39)
eGFR	0.05 (-0.10 to 0.20)	0.07 (-0.08 to 0.22)
Glucagon (log)		2.93 (-0.75 to 6.61)

Valine		
	Model 1	Model 2
(Intercept)	334.77 *** (168.35 to 501.19)	239.19 ** (71.15 to 407.24)
Liver fat content (log)	5.67 (-2.73 to 14.06)	2.85 (-5.31 to 11.02)
Age	-1.06 (-2.45 to 0.33)	-0.89 (-2.22 to 0.44)
Gender (male)	6.01 (-15.53 to 27.55)	4.39 (-16.15 to 24.94)
BMI	-1.20 (-3.86 to 1.45)	-0.69 (-3.24 to 1.85)
eGFR	0.14 (-0.52 to 0.80)	0.24 (-0.39 to 0.87)
Glucagon (log)		26.43 *** (11.04 to 41.82)

The estimated difference/beta (CI) in amino acid concentration by a unit increase in the liver fat content (log) (multiple linear regression). Model amino acid 1 with adjustment for age, gender, BMI, and eGFR; Model amino acid 2 with further adjustment for fasting glucagon concentration (log). Variables with a 95% CI not including zero are considered significantly associated with glucagon. *** $p < 0.001$; ** $p < 0.01$; * $p < 0.05$.

Abbreviations: BMI, body mass index; eGFR, estimated glomerular filtration rate.