Additional file 1 to:

An attenuated rate of leg muscle protein depletion and leg free amino acid efflux over time is seen in ICU long-stayers.

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Table S1. Individual patient characteristics.

Pat. no.	BMI[1] (kg/m^2)	ICU LOS (days)	SOFA median (IQR)	Kcal/24h mean±SD	Kcal/kg/24h mean±SD	AA (g)/24h mean±SD	AA (g)/kg/24h mean±SD	Insulin median (IQR)
1	27.9	42	6 (6-7)	1764±451	19±5	92±37	1.0±0.4	44 (22-80)
2	33.5	31	4 (3-6)	2096±460	19±4	98±25	0.9±0.2	179 (157-212)
3	27.0	30	8 (7-8)	1365±379	17±5	74±21	0.9±0.3	73 (52-99)
4	23.5	24	6 (5-8)	1479±306	20±4	83±17	1.1±0.2	51 (37-96)
5	25.4	36	4 (4-7)	1737±369	19±4	86±20	0.9±0.2	74 (57-101)
6	23.2	22	4 (3-6)	1513±403	20±5	76±25	1.0±0.3	70 (58-75)
7	30.5	17	4 (1-5)	1776±631	17±6	81±29	0.8±0.3	66 (26-73)
8	38.6	69	12 (10-13)	2138±401	14±3	105±23	0.7±0.1	87 (70-115)
9	24.7	30	5 (5-6)	1452±536	20±7	73±27	1.0±0.4	57 (32-78)
10	25.9	16	2 (2-3)	1314±491	17±6	74±29	0.9±0.4	31 (28-40)
11	28.4	27	8 (6-9)	1410±598	15±6	73±35	0.8±0.4	55 (24-85)
12	23.4	9	12 (11-13)	1149±526	19±9	64±32	1.0±0.5	27 (17-54)
13	23.7	21	7 (6-9)	1540±338	23±5	77±19	1.2±0.3	70 (34-90)
14	19.4	24	6 (4-9)	1152±374	22±7	65±32	1.2±0.6	5 (3-6)
15	22.0	27	4 (2-4)	1854±561	24±7	98±32	1.3±0.4	81 (72-101)
16	33.8	24	7 (3-7)	1925±678	18±6	99±35	0.9±0.3	39 (26-49)
17	15.7	14	6.5 (5.5-7)	1272±650	22±11	50±35	0.8±0.6	28 (22-47)
18	32.2	21	3 (3-3)	1764±493	17±5	81±25	0.8±0.2	43 (30-45)
19	26.1	68	3 (2-3)	2410±800	23±8	114±39	1.1±0.4	22 (18-26)
20	27.4	88	6 (4-9)	2269±519	27±6	99±24	1.2±0.3	24 (16-30)

[1]: mean of study days. AA: Amino acids; ICU: Intensive care unit; IQR: Inter-quartile range; LOS: Length of stay; SOFA: Sequential Organ Failure Assessment. Data is presented as mean ± SD for normally distributed data and as median (IQR) for non-normally distributed data.

Table S2. Patients characteristics at the 2 measurement periods illustrated in Figure 2.

	ICU day 10-20	ICU day 30-40
Weight (kg)	75 (64, 86)	78 (70, 91)
SOFA	7 (5, 8)	5 (4, 7)
Mechanical ventilation (%)	100 %	100%
CRRT (%)	30 %	30 %
Caloric intake[1] (kcal/kg/day)	21 (19, 25)	18 (18, 23)
Amino acid intake[1] (g/kg/day)	1.1 (1.0, 1.2)	1.0 (0.9-1.2)
Insulin IU/24 h	45 (27, 86)	26 (13, 69)

Values are given as n(%), medians (inter-quartile range). [1]: Median during 5 days before sampling.

Figure S1.

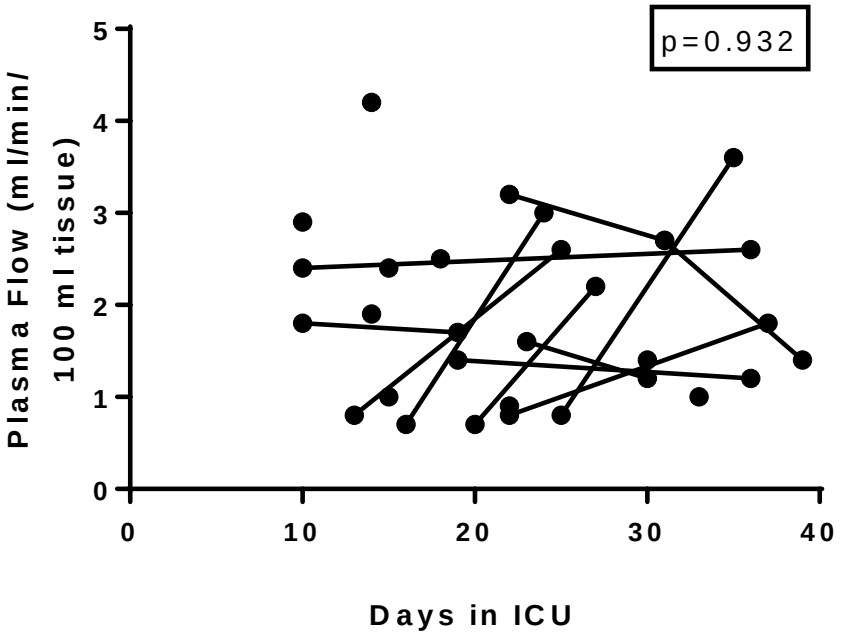
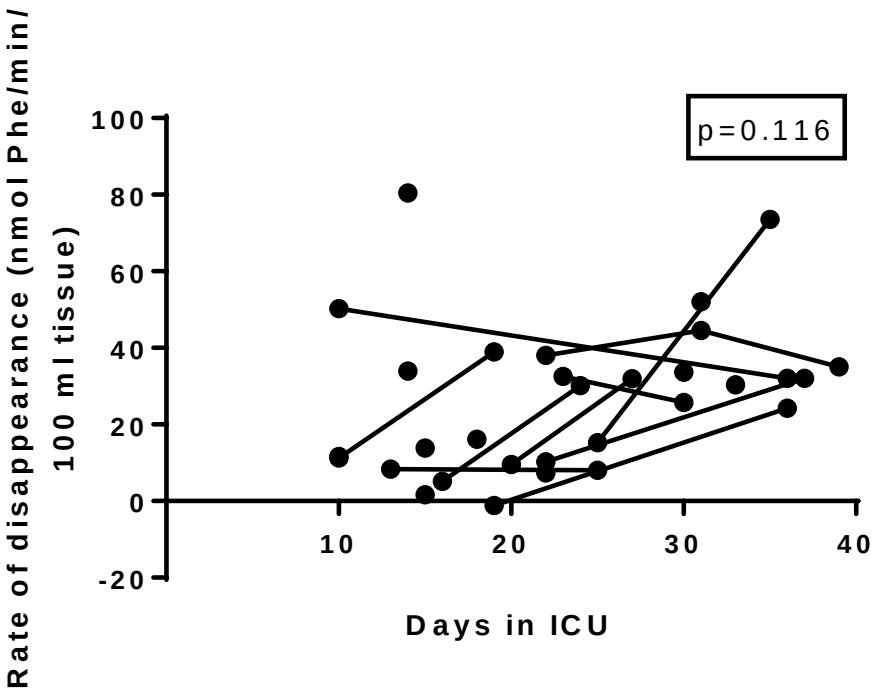


Figure S1. A longitudinal presentation of plasma flow across the leg on 30 occasions measured by venous occlusion plethysmography in long-staying ICU patients (n=20). The p-values from the linear mixed effect model against time in the ICU are indicated.

Figure S2.

A.



B.

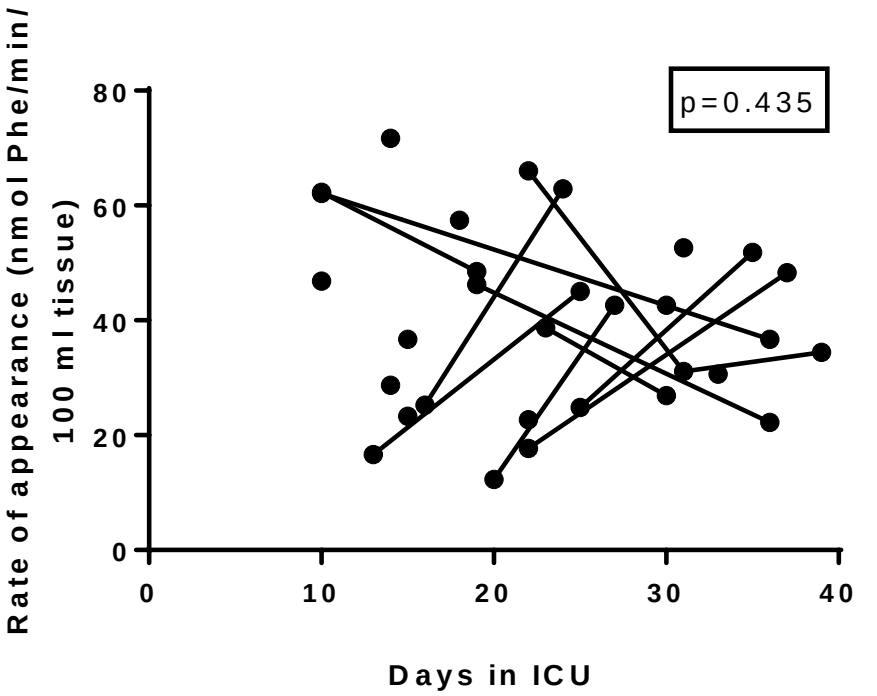
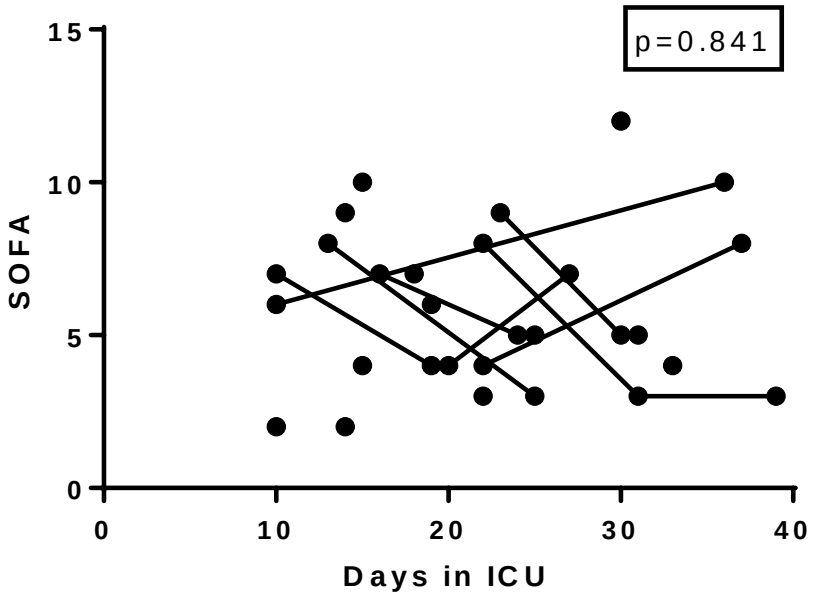


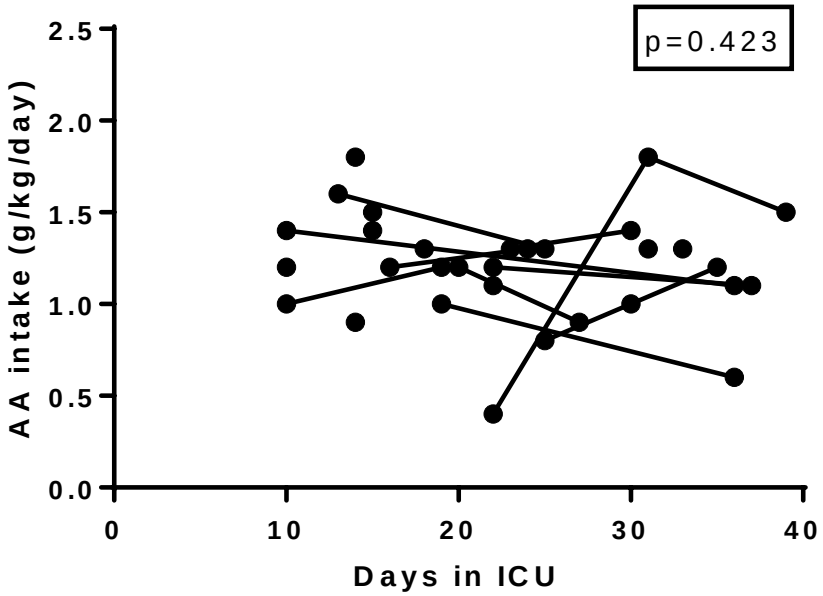
Figure S2. A longitudinal presentation of phenylalanine turnover in the leg on 30 occasions in long-staying ICU patients (n=20) assessed with a 2-pool model. The two panels depict; A) the rate of disappearance, and B) the rate of appearance. The p-values from the linear mixed effect model against time in the ICU are indicated.

Figure S3.

A.


p=0.841
SOFA
15
10
5
0
0
10
20
30
40
Days in ICU

B.


p=0.423
AA intake (g/kg/day)
2.5
2.0
1.5
1.0
0.5
0.0
0
10
20
30
40
Days in ICU

C.

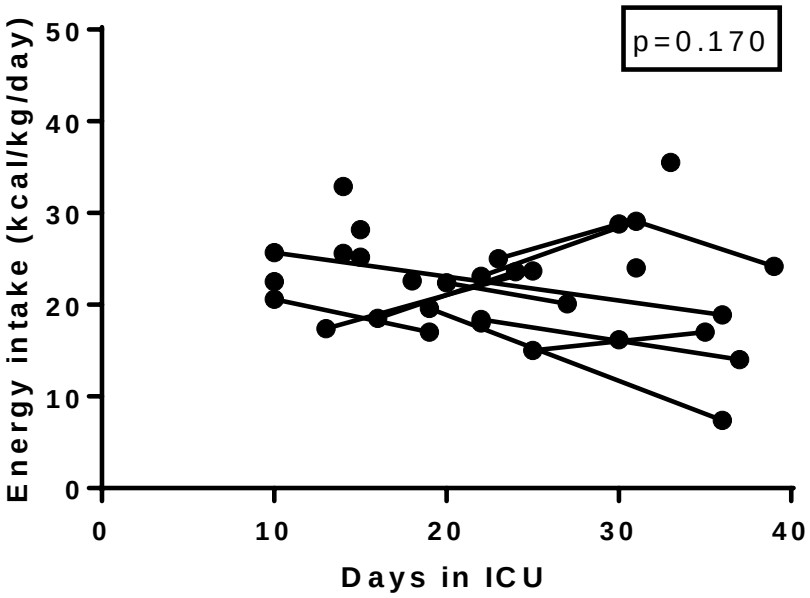
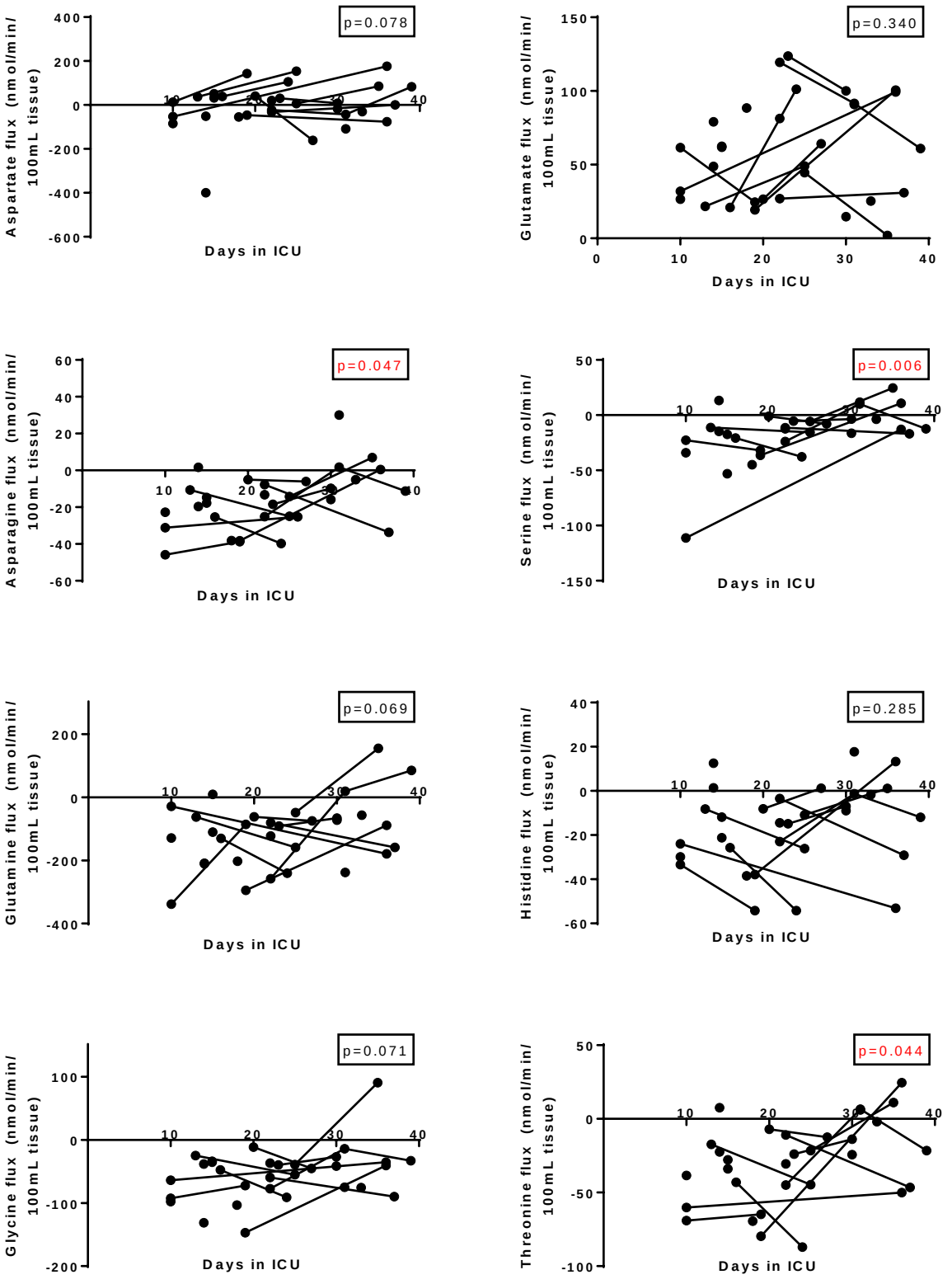
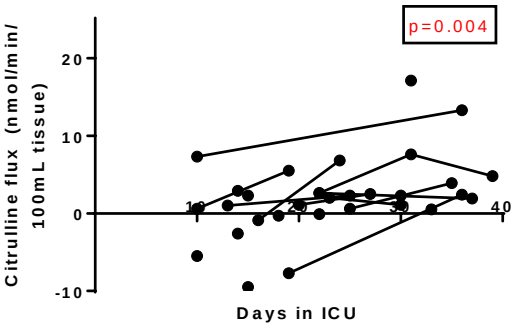
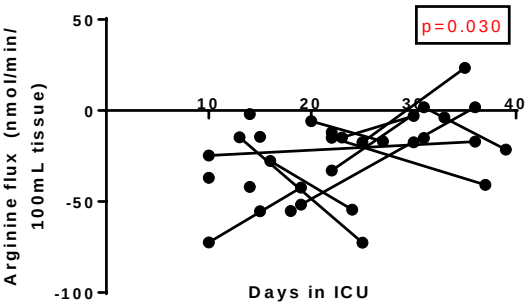


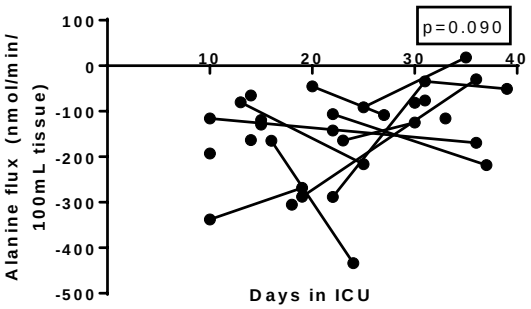
Figure S3. A longitudinal presentation of the SOFA score (panel A), the amino acid intake (panel B) and the energy intake (panel C) on 30 occasions measured in long-staying ICU patients (n=20). The p-values from the linear mixed effect model against time in the ICU are indicated.

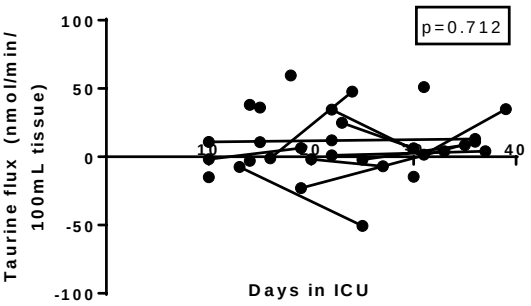
Figure S4. Longitudinal presentations of free amino acid fluxes across the leg on 30 occasions in longstaying ICU patients (n=20). Individual amino acids are displayed in 21 panels, in addition also the combined branched chain amino acids (BCAA) and the combined essential amino acids (EAA) are displayed in two additional piles. The p-values from the linear mixed effect model against time in the ICU are indicated. Significant correlations are indicated in red text.

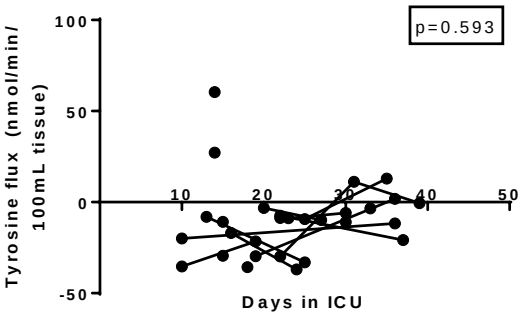


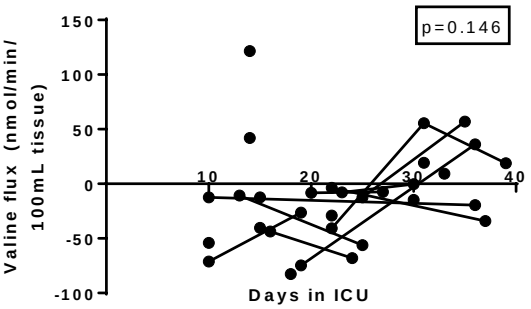

p=0.004
Citrulline flux (nmol/min/100mL tissue)
Days in ICU

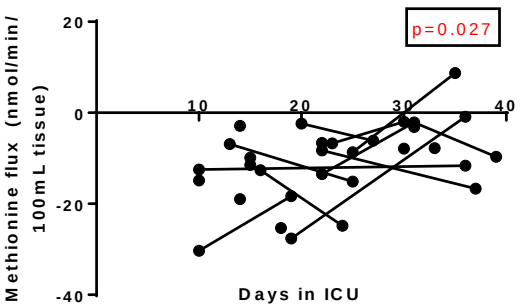

p=0.030
Arginine flux (nmol/min/100mL tissue)
Days in ICU

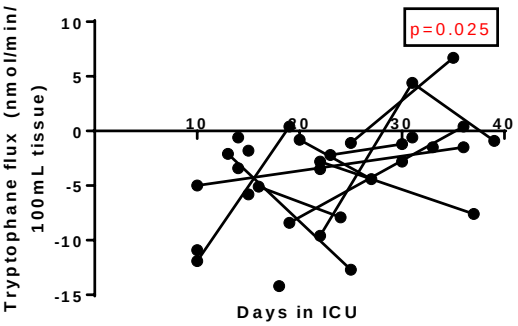

p=0.090
Alanine flux (nmol/min/100mL tissue)
Days in ICU

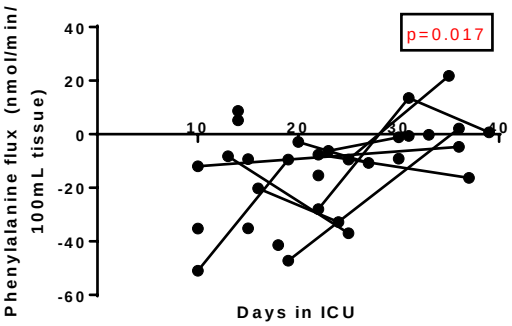

p=0.712
Taurine flux (nmol/min/100mL tissue)
Days in ICU

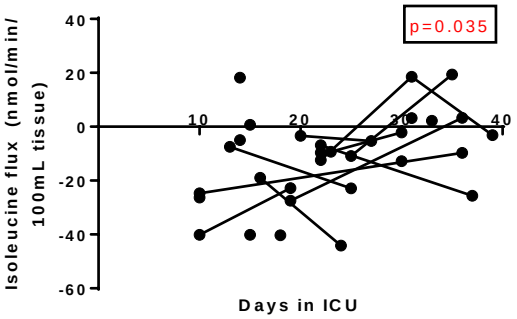

p=0.593
Tyrosine flux (nmol/min/100mL tissue)
Days in ICU

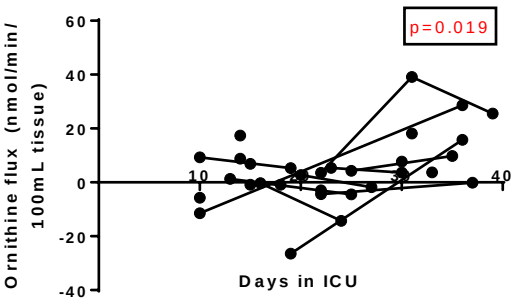

p=0.146
Valine flux (nmol/min/100mL tissue)
Days in ICU

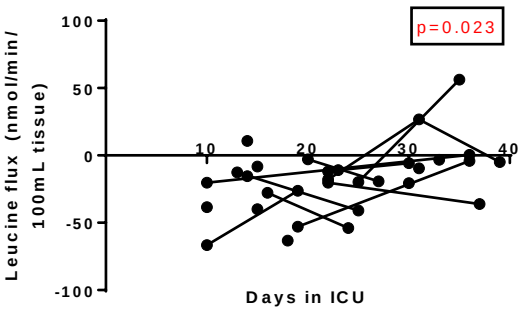

p=0.027
Methionine flux (nmol/min/100mL tissue)
Days in ICU

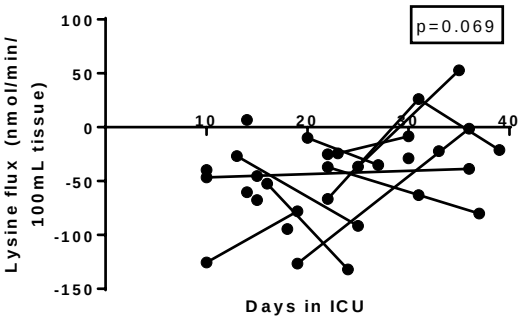

p=0.025
Tryptophane flux (nmol/min/100mL tissue)
Days in ICU

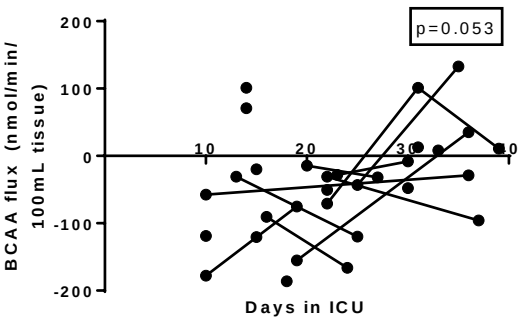

p=0.017
Phenylalanine flux (nmol/min/100mL tissue)
40
20
0
-20
-40
-60
10
20
30
40
Days in ICU


p=0.035
Isoleucine flux (nmol/min/100mL tissue)
40
20
0
-20
-40
-60
10
20
30
40
Days in ICU


p=0.019
Ornithine flux (nmol/min/100mL tissue)
60
40
20
0
-20
-40
10
20
30
40
Days in ICU


p=0.023
Leucine flux (nmol/min/100mL tissue)
100
50
0
-50
-100
10
20
30
40
Days in ICU


p=0.069
Lysine flux (nmol/min/100mL tissue)
100
50
0
-50
-100
-150
10
20
30
40
Days in ICU


p=0.053
BCAA flux (nmol/min/100mL tissue)
200
100
0
-100
-200
10
20
30
40
Days in ICU

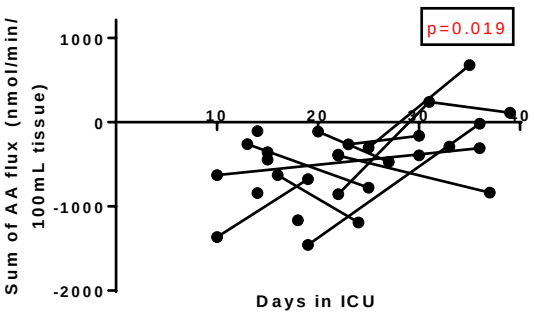
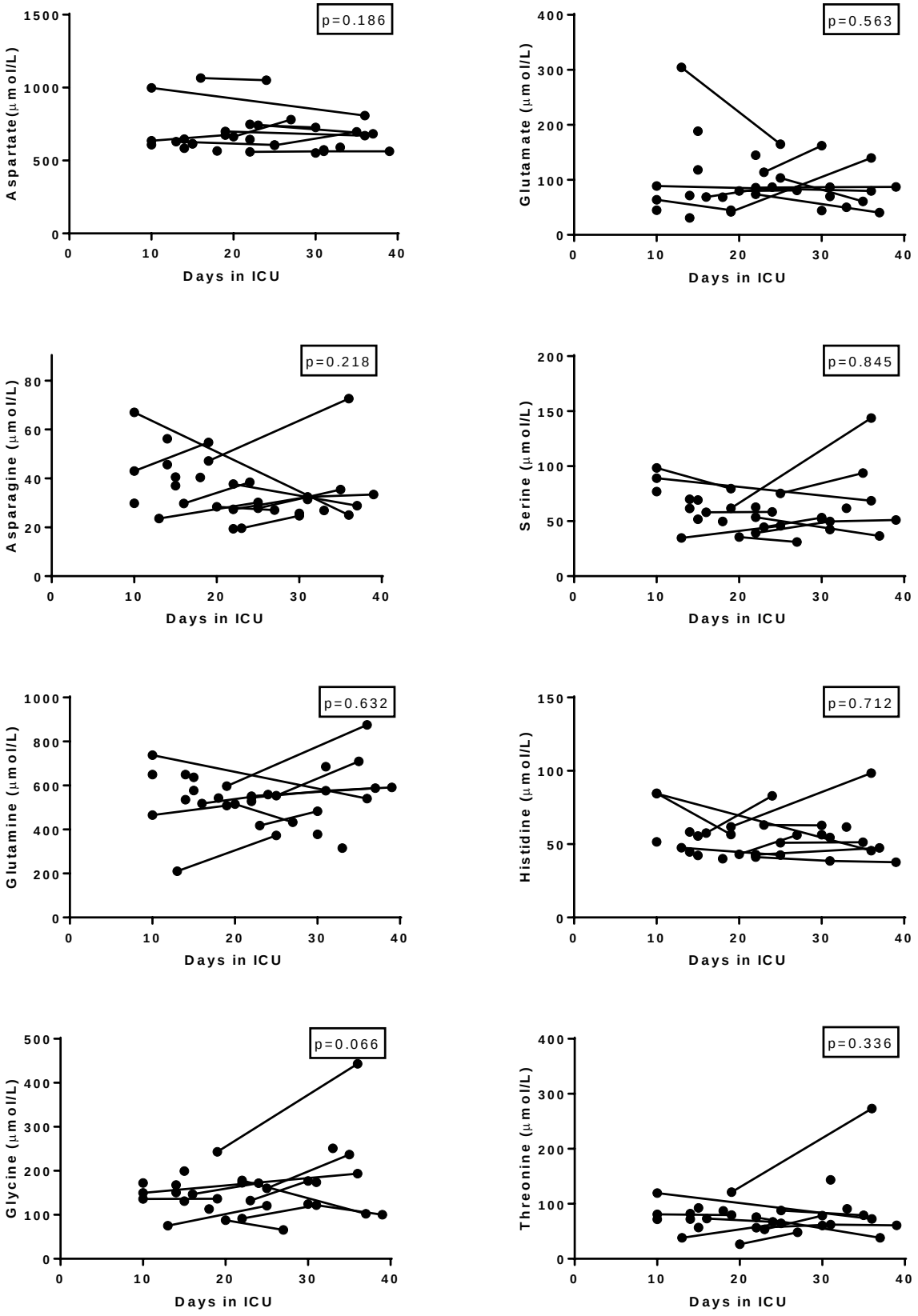
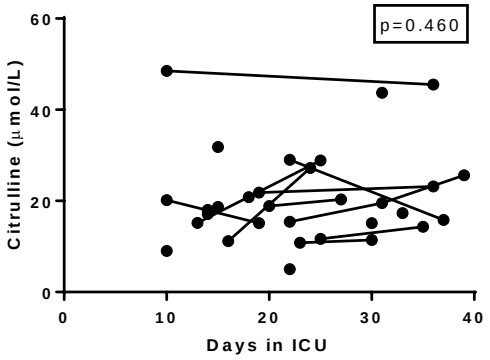
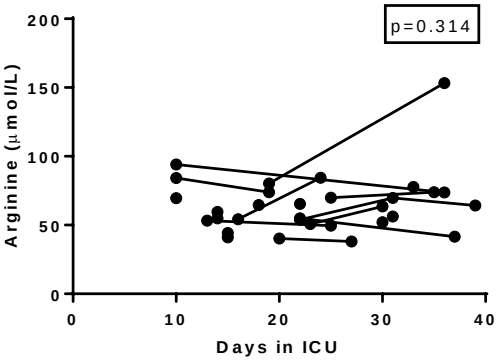
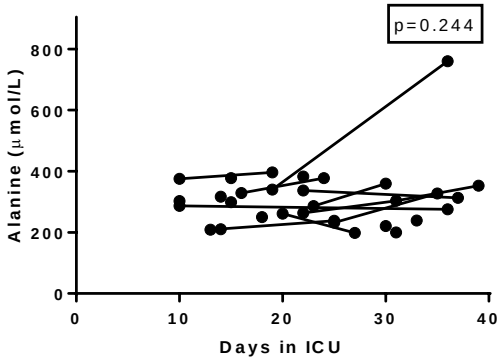

p=0.019
Sum of AA flux (nmol/min/100mL tissue)
1000
0
-1000
-2000
10
20
30
40
Days in ICU

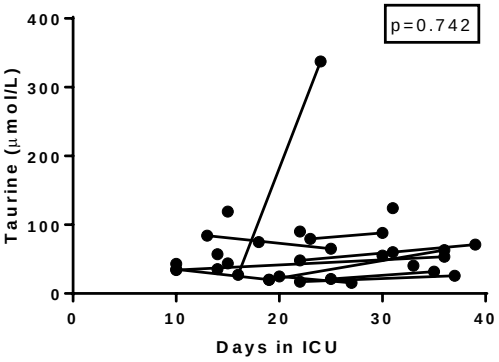
Figure S5. Longitudinal presentations of free amino acid concentrations in arterial plasma on 30 occasions in longstaying ICU patients (n=20). Individual amino acids are displayed in 21 panels, in addition also the combined branched chain amino acids (BCAA) and the combined essential amino acids (EAA) are displayed in two additional piles. The p-values from the linear mixed effect model against time in the ICU are indicated. Significant correlations are indicated in red text.

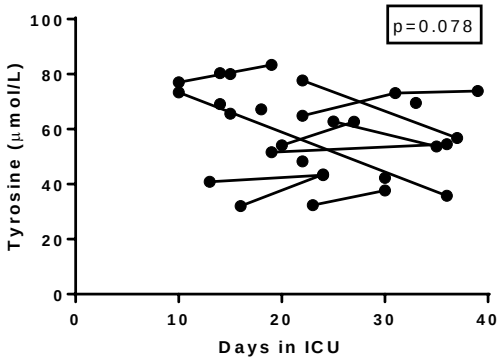


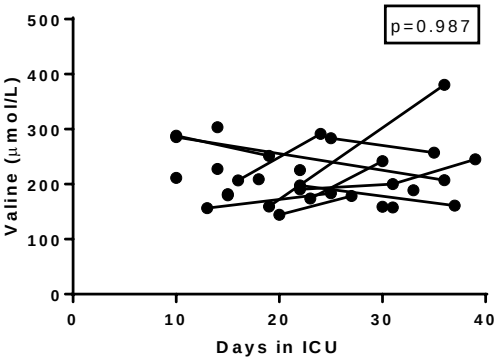

p=0.460
Citrulline (µmol/L)
Days in ICU

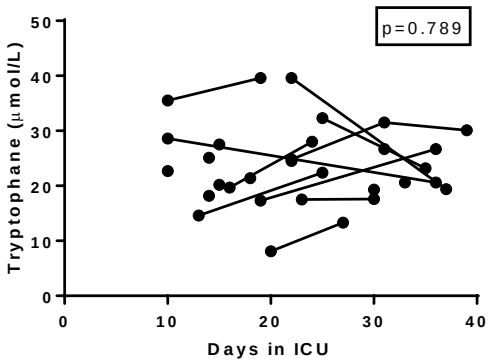

p=0.314
Arginine (µmol/L)
Days in ICU

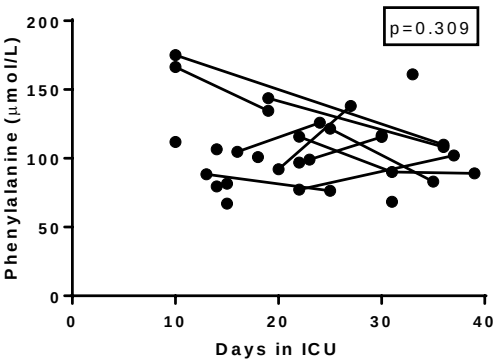

p=0.244
Alanine (µmol/L)
Days in ICU

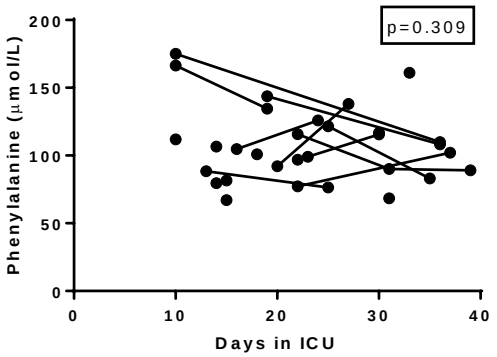

p=0.742
Taurine (µmol/L)
Days in ICU

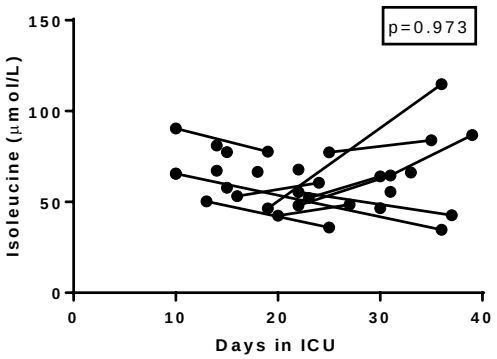

p=0.078
Tyrosine (µmol/L)
Days in ICU

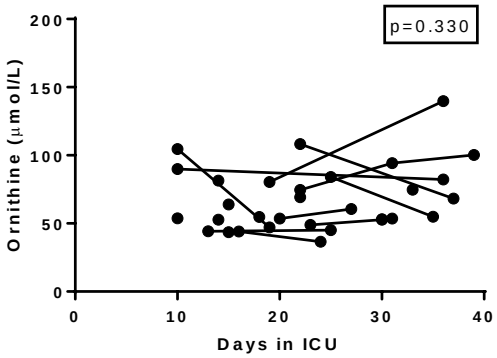

p=0.987
Valine (µmol/L)
Days in ICU

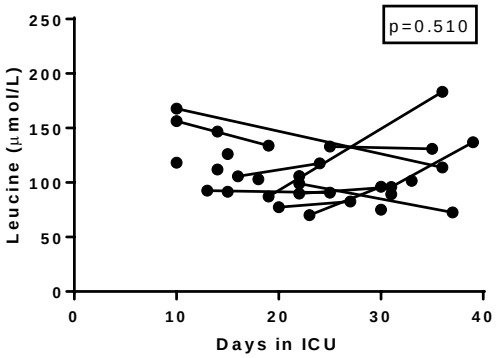

p=0.789
Tryptophane (µmol/L)
Days in ICU

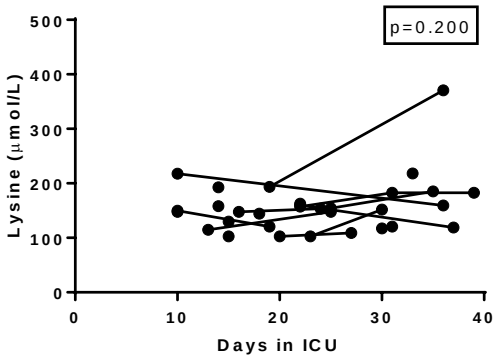

p=0.309
Phenylalanine (µmol/L)
Days in ICU

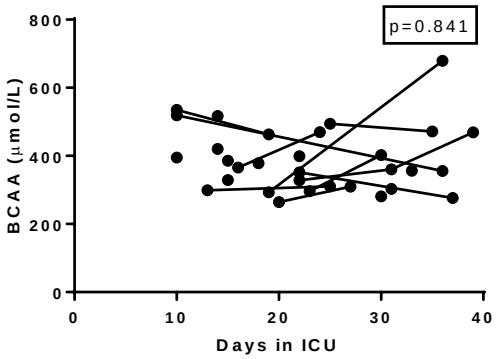

p=0.309
Phenylalanine (µmol/L)
200
150
100
50
0
0
10
20
30
40
Days in ICU

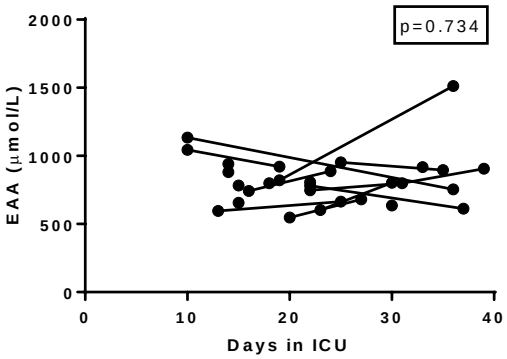

p=0.973
Isoleucine (µmol/L)
150
100
50
0
0
10
20
30
40
Days in ICU


p=0.330
Ornithine (µmol/L)
200
150
100
50
0
0
10
20
30
40
Days in ICU


p=0.510
Leucine (µmol/L)
250
200
150
100
50
0
0
10
20
30
40
Days in ICU


p=0.200
Lysine (µmol/L)
500
400
300
200
100
0
0
10
20
30
40
Days in ICU


p=0.841
BCAA (µmol/L)
800
600
400
200
0
0
10
20
30
40
Days in ICU


p=0.734
EAA (µmol/L)
2000
1500
1000
500
0
0
10
20
30
40
Days in ICU