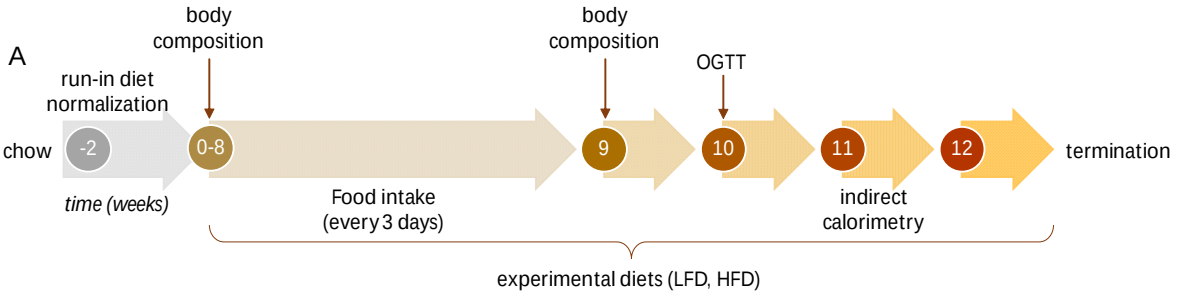
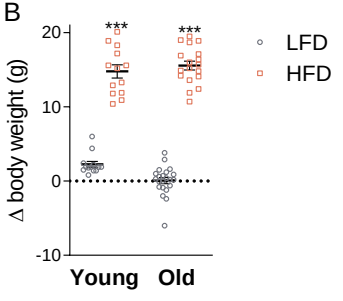
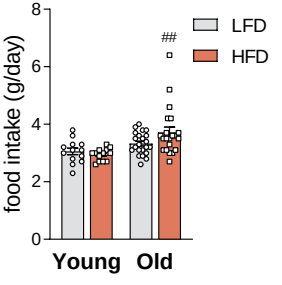
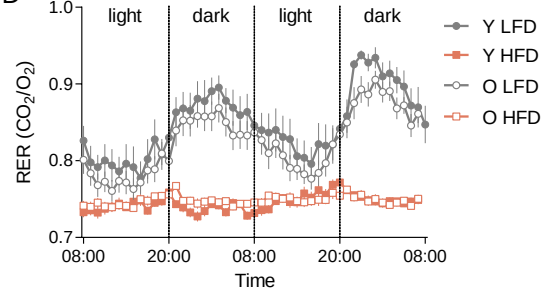
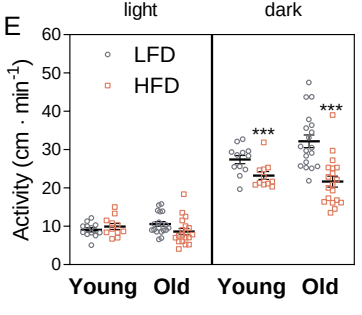
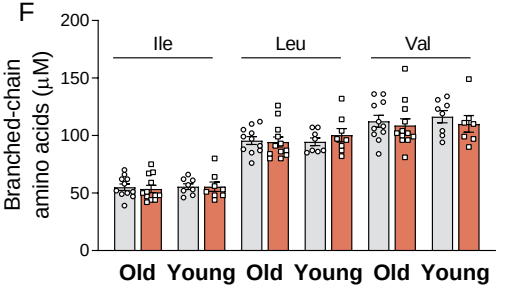

A
body composition
body composition
OGTT
run-in diet normalization
chow
-2
0-8
9
10
11
12
termination
time (weeks)
Food intake (every 3 days)
indirect calorimetry
experimental diets (LFD, HFD)


B
Δ body weight (g)
LFD
HFD
***
***
Young
Old

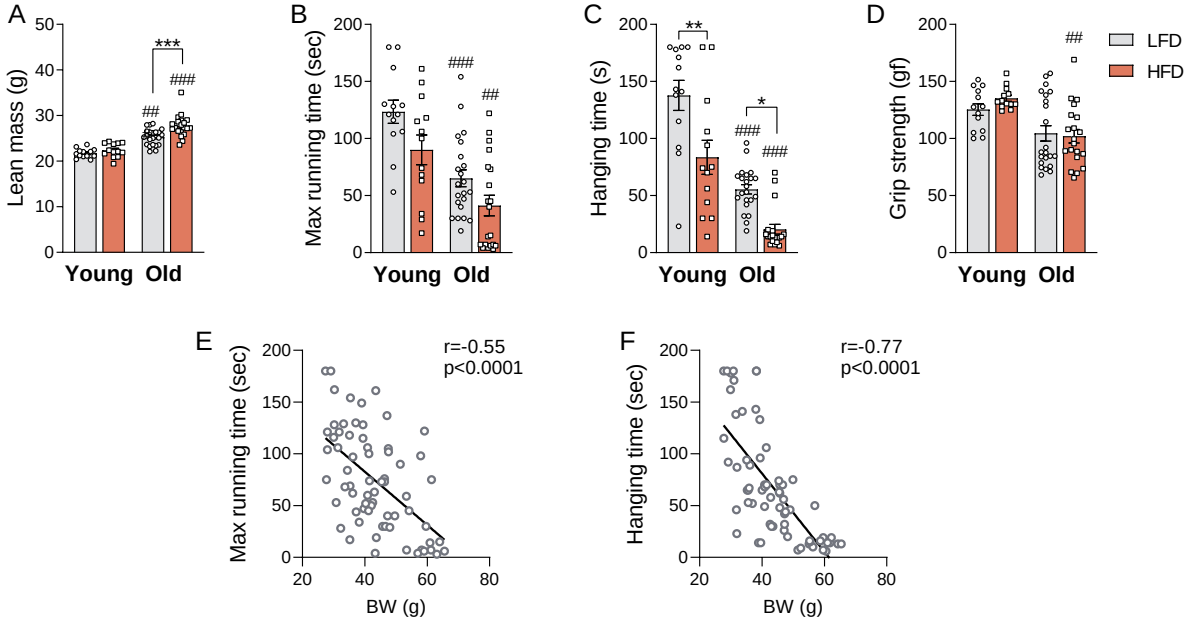

C
food intake (g/day)
LFD
HFD
##
Young
Old


D
RER (CO2/O2)
light
dark
light
dark
Y LFD
Y HFD
O LFD
O HFD
08:00
20:00
08:00
20:00
08:00
Time


E
light
dark
Activity (cm · min-1)
LFD
HFD
***
***
Young
Old
Young
Old

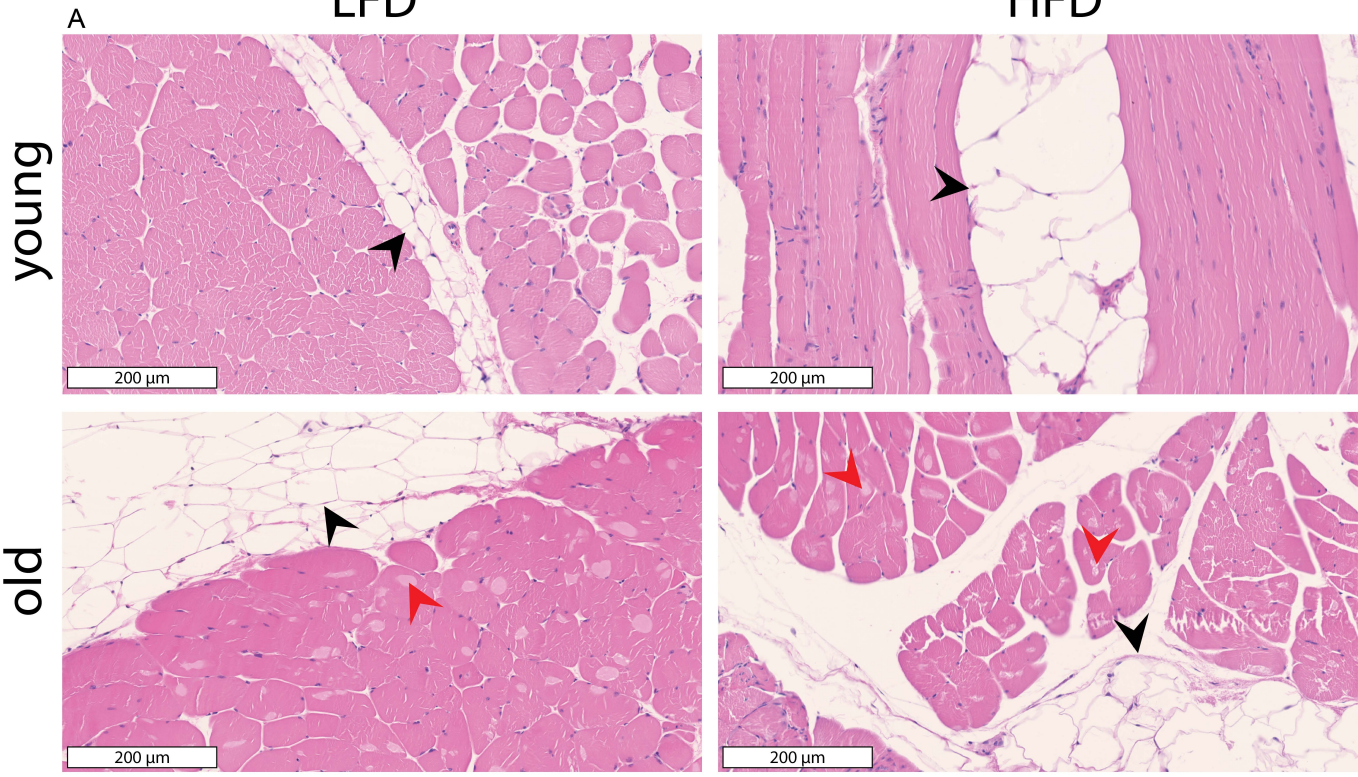

F
Branched-chain amino acids (μM)
Ile
Leu
Val
Old
Young
Old
Young
Old
Young

Supp. Fig. 1


A
Lean mass (g)
***
##
###
Young
Old
B
Max running time (sec)
###
##
Young
Old
C
Hanging time (s)
**
*
###
###
Young
Old
D
Grip strength (gf)
##
LFD
HFD
Young
Old
E
Max running time (sec)
r=-0.55
p<0.0001
BW (g)
F
Hanging time (sec)
r=-0.77
p<0.0001
BW (g)

Supp. Fig. 2

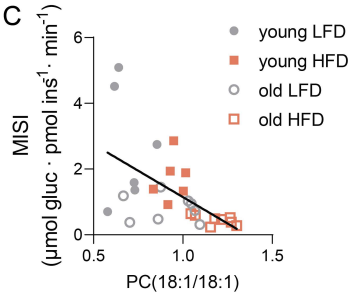
A



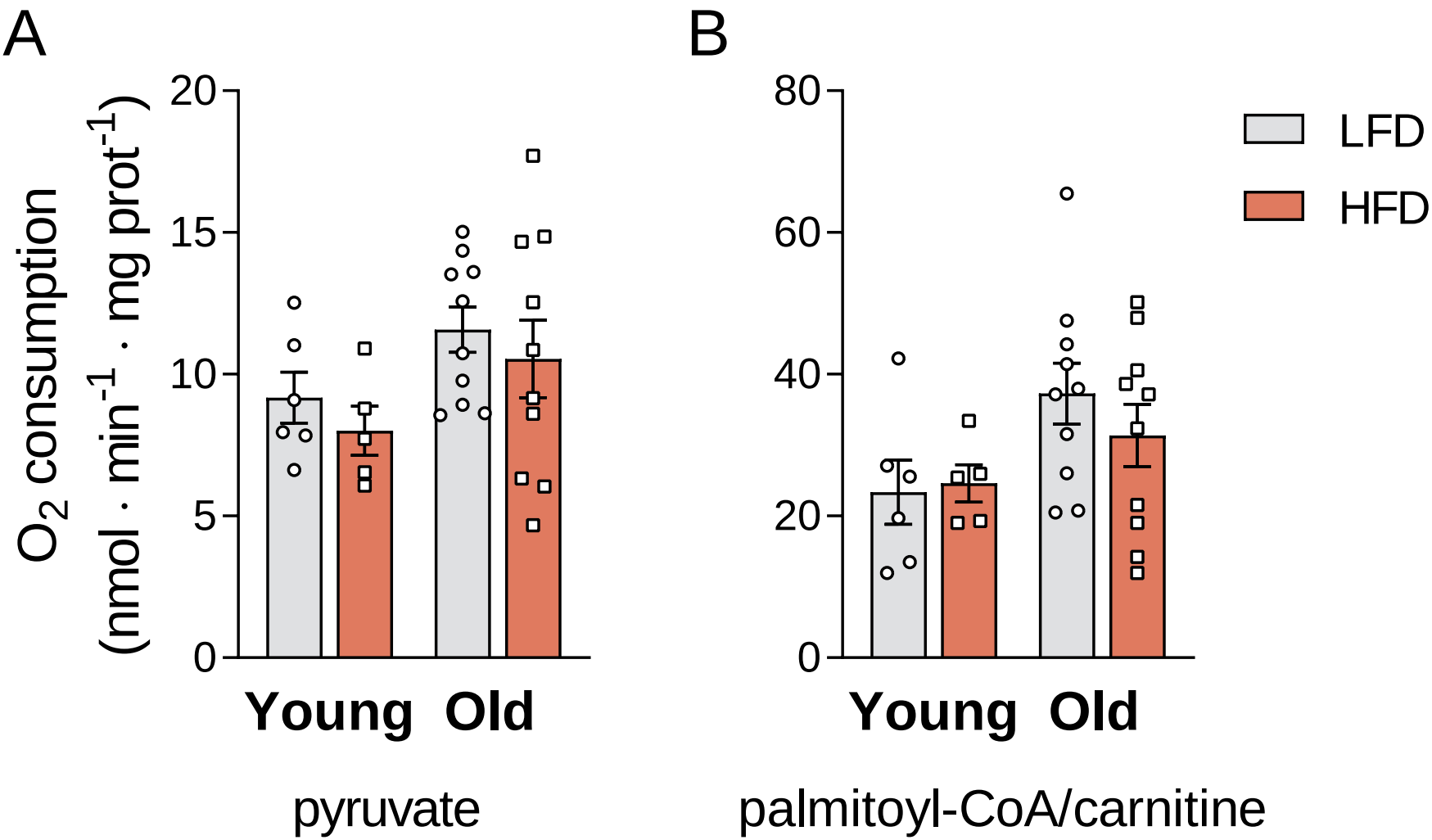
B

Group	Vacuolization (mean)	Fat infiltration (mean)
Young LFD	0	1
Young HFD	0	2
Old LFD	2	2
Old HFD	3	3

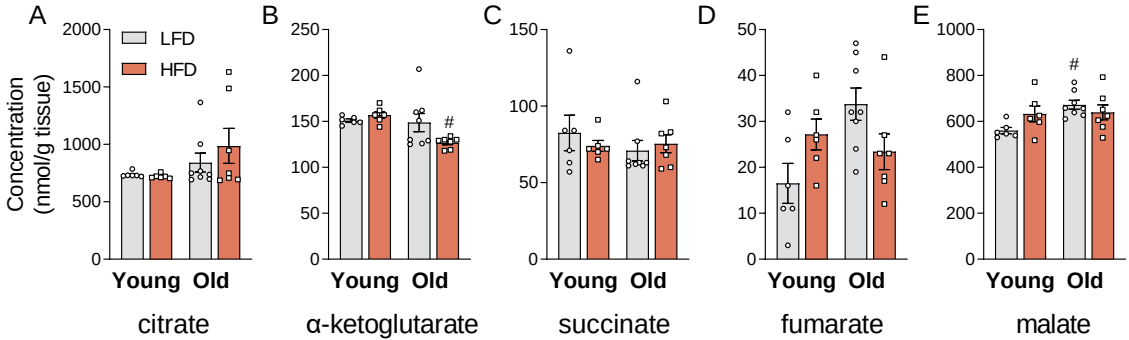
C



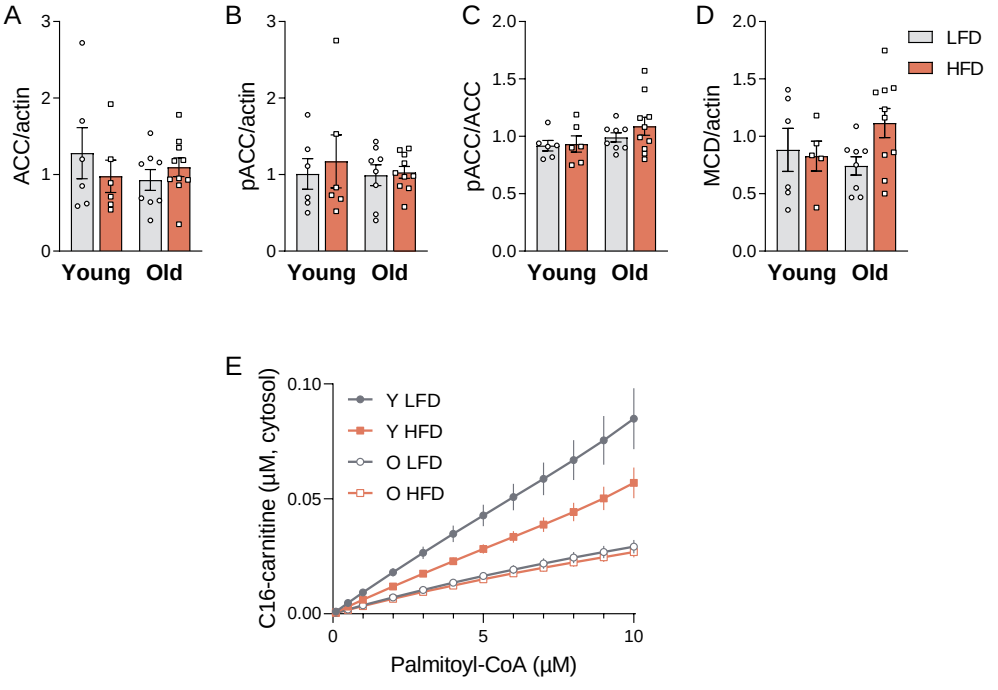
Supp. Fig. 3


A
B
O_2 consumption
(nmol · min^{-1} · mg prot^{-1})
20
15
10
5
0
80
60
40
20
0
LFD
HFD
Young Old
Young Old
pyruvate
palmitoyl-CoA/carnitine

Supp. Fig. 4


A
B
C
D
E
LFD
HFD
Concentration (nmol/g tissue)
2000
1500
1000
500
0
250
200
150
100
50
150
100
50
50
40
30
20
10
1000
800
600
400
200
#
Young
Old
citrate
α-ketoglutarate
succinate
fumarate
malate

Supp. Fig. 5


A
ACC/actin
3
2
1
0
Young
Old
B
pACC/actin
3
2
1
0
Young
Old
C
pACC/ACC
2.0
1.5
1.0
0.5
0.0
Young
Old
D
MCD/actin
2.0
1.5
1.0
0.5
0.0
Young
Old
LFD
HFD
E
C16-carnitine (µM, cytosol)
0.10
0.05
0.00
0
5
10
Palmitoyl-CoA (µM)
Y LFD
Y HFD
O LFD
O HFD

Supp. Fig. 6

Supplemental Figure - Legends

Supplemental Figure 1: A: Schematic design of animal experiments. B: Change in body weight for both age groups after 8 weeks on either a LFD or HFD. C: Average food intake per day from several measurements over the course of 8 weeks. D: 48h time course for respiratory exchange ratios (RER) in both light/dark phases following 10 weeks for each age/diet group. E: Activity of animals ($cm \cdot min^{-1}$) measured in metabolic cages with the use of lasers in parallel with indirect calorimetry. F: Plasma branched-chain amino acids measured at termination blood spots for all age/diet groups. Data are shown as mean ± SEM. n=13-22 per group, ***p<0.001 (LFD vs HFD, matched age), Two-Way-ANOVA followed by Tukey's post hoc test.

Supplemental figure 2: A: lean mass, B: maximal running time in seconds, C: hanging time in seconds, D: grip strength (gf). Spearman correlations between body weight (BW) in grams and either maximal running time (E) or hanging time (F). Data are shown as mean ± SEM. n=13-22 per group, *p<0.05, **p<0.01,***p<0.001 (LFD vs HFD, matched age), $^{\#\#}$p<0.01, $^{\#\#\#}$p<0.001 (old vs young, matched diet), Two-Way-ANOVA followed by Tukey's post hoc test.

Supplemental figure 3: A: H&E staining of quadriceps samples. Representative images per experimental group. Black arrows represent fat infiltration and red arrows show vacuolization. B: quantification of fat infiltration and vacuolization per group (n=3-4). C: Linear regression between phosphatidylcholine PC(18:1/18:1) and MISI (Pearson r = -0.56, p = 0.002).

Supplemental figure 4: Maximal ADP-stimulated O_2 consumption in liver corrected for total tissue protein. A: pyruvate and B: palmitoyl-CoA and carnitine (D) were used as substrates, all in the presence of malate.

Supplemental Figure 5: Absolute quantification of TCA cycle intermediates in nmol/g tissue. A: citrate, B: α-ketoglutarate, C: succinate, D: fumarate, E: malate. Data are shown as mean ± SEM. n=6-9 per group, $^{\#}$p<0.05 (old vs young, matched diet), Two-Way-ANOVA followed by Tukey's post hoc test.

Supplemental figure 6: A-D: Total ACC, pACC (S79), pACC/ACC and MCD levels in quadriceps homogenates normalized by actin levels. E: predicted cytosolic C16-carnitine concentrations, n=5-10 per group.