Visceral adipose tissue bioenergetic varies according to individuals' obesity class

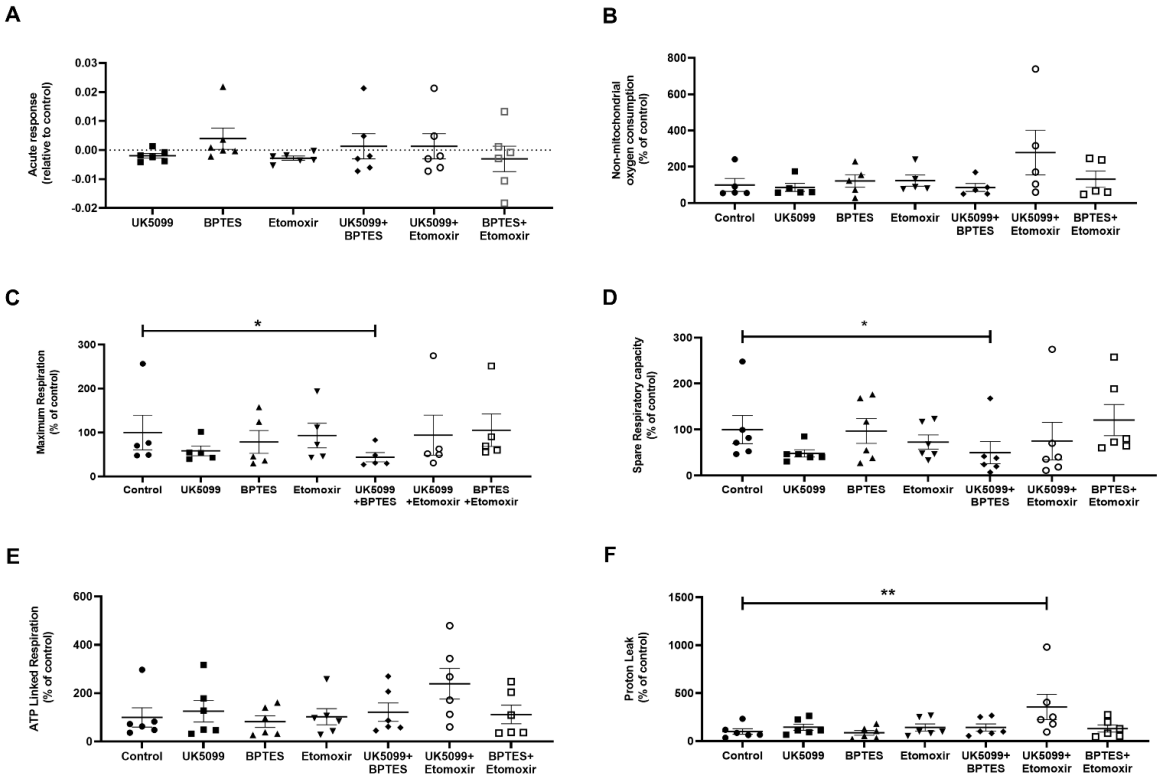


Figure S1. Influence of the inhibitors in (A) basal respiration (acute response); (B) non-mitochondrial oxygen consumption; (C) Maximum respiration oxygen consumption; (D) Spare respiratory capacity; (E) Proton Leak and (F) ATP link respiration in the VAT of patients with a class 2 obesity. Except for the acute response, all values are normalized to the control group. (Friedman test *$p<0.05$, **$p<0.01$).

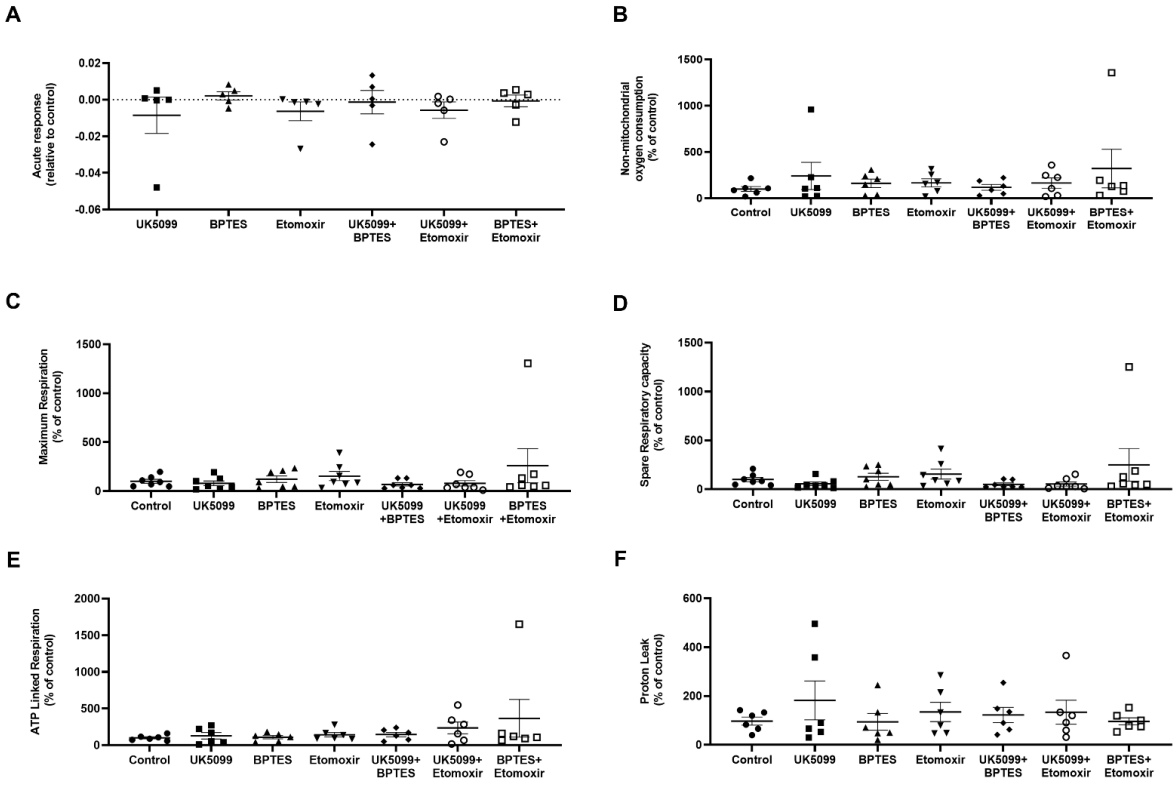


Figure S2. Influence of the inhibitors in (A) basal respiration (acute response); (B) non-mitochondrial oxygen consumption; (C) Maximum respiration oxygen consumption; (D) Spare respiratory capacity; (E) Proton Leak and (F) ATP link respiration in the VAT of patients with a class 3 obesity. Except for the acute response, all values are normalized to the control group.

Table S1. Correlation between Age (years) and visceral adipose tissue respiration parameters.

Parameter	Correlation (R value)	p value
Total Basal Oxygen Consumption	0.01	ns
Mitochondrial Basal Respiration	0.20	ns
Non-mitochondrial oxygen consumption	0.02	ns
ATP Link Respiration	0.31	ns
Proton Leak	0.28	ns
Maximum Respiration	0.21	ns
Spare Respiratory Capacity	0.13	ns

Table S2. Respiratory parameters calculation formulas.

Assay parameter	**Meaning/Equation**
Total Basal Oxygen Consumption	Last measure before the injection of the metabolic pathways` inhibitors
Non-mitochondrial oxygen consumption	Minimum rate measurement after Rotenone/Antimycin A injection
Mitochondrial Basal Respiration	(Last rate measurement before any injection) – (non-mitochondrial oxygen consumption)
ATP linked respiration	(Last rate measurement before Oligomycin injection) – (Minimum rate measurement after oligomycin injection)
Acute response	(Last rate measurement before oligomycin injection) – (Last rate measurement before acute inhibitor injection)
Maximum respiration	(Maximum rate measurement after FCCP injection) – (non-mitochondrial oxygen consumption)
Spare respiratory capacity	(Maximum respiration) – (Basal Respiration)
Proton leak	(Minimum rate measurement after oligomycin injection) – (non-mitochondrial oxygen consumption)

Table S3 Primers' sequences

Gene	Primer Sequence (Forward/Reverse)	T annealing (°C)	Size (bp)
ob	TCTTGTCCCCTCTTGACCCA AGCTTGTGTTGCTGGGAGTT	52.60	183
CPT1A	GCAGCGTTCTTTGTGACGTT AGGAGTGTTCAGCGTTGAGG	51.86	184
SLC1A5	TCTGGCTGGTAACCGCTACT GAGTTTCTCTGGGTGGGAGC	52.81	107
GLS1	TCTACAGGATTGCGAACGTCT CTTTGTCTAGCATGACACCATCT	50.31	100
MPC1	ATTTGCCTACAAGGTACAGCC AGTCATCTCGTGTTTGATAAGCC	50.05	109
SLC2A4	TCCACCAACAACACCGAGAC CGACCAGCATCTTCGAGACA	52.20	117
RPII	CTTCACGGTGCTGGGCATT GTGCGGCTGCTTCCATAA	52.20	239
LDHA	CGACCGCCCGACGTG TAAGGAAAAGGCTGCCATGTTG	52.94	264
FABP4	GTAGGAGTGGGCTTTGCCAC ACGCATTCCACCACCAGTTT	53.01	287
β-actin	CCAACCGCGAGAAGATGA CCAGAGGCGTACAGGGATAG	53.10	97

CPT1a – Carnitine Palmitoyl Transferase 1a; *FABP4* - Fatty Acid Binding-Protein 4; *GLS1* – Glutaminase 1; *LDHA* - Lactate Dehydrogenase A; *MPC1* – Mitochondrial Pyruvate Carrier 1; *ob* – Leptin; qRT-PCR - quantitative Real-Time Polymerase Chain reaction; *RPII* - RNA polymerase II; *SLC1A5* – Neutral amino acid transporter; *SLC2A4* – Glucose transporter 4; T – temperature

Table S4. - The three-step cycling qRT-PCR program used

Step	Tannealing (°C)	Time	N° of cycles
Ativation	95	2 min	1
Desnaturation	95	5 s	**35 cycles** (*CPT1a*, *GLS1*, *MPC1*) **37 cycles** (*SLC1A5*) **40 cycles** (*β-actin*, *RPII*, *FABP4*, *LDHA*) **41 cycles** (*SLC2A4*) **45 cycles** (*ob*)
Annealing	**50ºC** (*β-actin*, *FABP4*, *LDHA* and *RPII*) **56ºC** (*CPT1a*, *GLS1*, *MPC1*) **58ºC** (*SLC2A4* and *SLC1A5*) **60ºC** (*ob*)	30 s	
Extension	72	1 min	
Final Desnaturation	72	7 min	1

CPT1a – Carnitine Palmitoyl Transferase 1a; *FABP4* - Fatty Acid Binding-Protein 4; *GLS1* – Glutaminase 1; *LDHA* - Lactate Dehydrogenase A; *MPC1* – Mitochondrial Pyruvate Carrier 1; *ob* – Leptin; qRT-PCR - quantitative Real-Time Polymerase Chain reaction; *RPII* - RNA polymerase II; *SLC1A5* – Neutral amino acid transporter; *SLC2A4* – Glucose transporter 4; T – temperature