Methods of Oral Glucose Tolerance Test and Laboratory measurements

All subjects underwent a seven-point oral glucose tolerance test (OGTT) at the department of clinical diabetology in the Academic Medical Center. During the OGTT subjects ingested a standardized amount of 75 g of glucose dissolved in 300 ml water, at time point 0. Blood samples were collected at time points 0 (before the administration of the glucose), 10, 20, 30, 60, 90 and 120 minutes. Data were analysed using a glucose-insulin model to derive an OGTT-based insulin sensitivity (oral glucose insulin sensitivity [OGIS]) index to predict glucose clearance.(*1*) Plasma glucose levels were measured using a HemoCue Glucose 201 RT Analyzer. Plasma insulin levels were measured using sandwich enzyme immunoassay(Roche Diagnostics, Rotzkreuz, Switzerland). Furthermore we assessed HbA1c (ion-exchange chromatography on a Tosoh-G8 analyser; Tosoh Bioscience, Tokyo, Japan), levels of total cholesterol, HDL-cholesterol, LDL-cholesterol, triacylglycerol (enzymatic colorimetric method for all cholesterol and triacylglycerol measurements, Roche Diagnostics) and creatinine (colorimetric). Free fatty acids were measured using spectrophotometry.

Results

The obese were significantly more insulin resistant, as measured with HOMA-IR (homeostatic model assessment – insulin resistance) and OGIS than the lean young (table 1). Measurements for insulin sensitivity did not correlate with any of the scintigraphic BAT parameters.

Supplemental Table 1. Characteristics of the male volunteers

	Obese young	IQR	Lean young	IQR	Lean old	IQR
N	10		14		11	
Age (years)	25.5	21.0-31.3	25.5	20.8-28.0	54***	50.0-60.0
BMI (kg/m2)	32.2***	30.8-38.9	22.0	20.9-23.2	23.1	21.7-23.8
Waist circumference (cm)	117.5***	110.5-144.5	83.0	82.0-85.0	90.0**	85.0-93.0
Body surface area (m^2)	2.4***	2.3-2.5	2.1	2.0-2.1	2.0	1.9-2.0
Laboratory values						
HbA1c (mmol/mol)	35.0	32.8-37.8	33.5	31.8-35.0	39.0***	36.0-40.0
Creatinine (μmol/L)	77.5	69.8-87.3	76.0	71.5-82.5	82.0	76.0-90.0
Fasting plasma glucose (mmol/L)	3.9	3.7-4.4	4.2	4.0-4.6	4.2	3.7-4.5
Fasting plasma insulin (pmol/L)	86**	52-132	27	15-42	41	17-52
Total Cholesterol (mmol/L)	4.2	4.1-5.4	4.0	3.6-4.5	5.8***	5.2-6.0
HDL Cholesterol (mmol/L)	1.0**	0.9-1.3	1.5	1.3-1.7	1.4	1.3-1.8
LDL Cholesterol (mmol/L)	2.6	2.2-3.1	2.1	1.6-2.6	4.0***	2.9-4.1
Cholesterol ratio	4.4**	3.4-5.0	2.6	2.1-3.4	4.1*	3.0-4.7
Triglycerides (mmol/L)	1.1*	0.8-1.7	0.6	0.5-1.1	0.9	0.8-1.1
TSH (mE/L)	2.4	1.5-3.3	1.8	1.0-2.4	1.7	1.4-2.2
Free Fatty Acids (mmol/L)	0.6	0.6-0.8	0.5	0.4-0.7	0.5	0.3-0.8
HOMA-IR	2.4**	1.3-3.5	0.7	0.4-1.3	1.0	0.4-1.4
OGIS (ml/min/m^2)	440*	395-505	530	455-571	495	429-545
Energy Expenditure in Cold (kcal/24h)	2421	1499-2530	2294	1807-2666	1484	1033-2294

BMI, body mass index; HDL, high density lipoprotein; LDL, low density lipoprotein; TSH, thyroid stimulating hormone. HOMA-IR, homeostatic model assessment – insulin resistance, calculated as ([glucose(mmol/L) x insulin(mU/L)]/22.5). OGIS (oral glucose insulin sensitivity).
*Data are presented as n or median [interquartile range] Significant different as compared to the lean young males * p<0.05, **p<0.01, ***p<0.001.

Supplemental Table 2. Correlations between BAT parameters and insulin sensitivity tests

	SUV_{max}	SUV_{mean}	$SQUV_{max}$	$SQUV_{mean}$	BAT volume	BAT volume
HOMA-IR	ρ=-0.10	ρ=-0.11	ρ=-0.10	ρ=-0.15	ρ=-0.15	ρ=-0.07
OGIS (ml/min/m^2)	ρ=-0.00	ρ=-0.02	ρ=-0.27	ρ=0.01	ρ=0.02	ρ=0.14

Spearman's correlations for BAT activity parameters and HOMA-IR (homeostatic model assessment – insulin resistance, calculated as ([glucose(mmol/L) x insulin(mU/L)]/22.5) and OGIS (oral glucose insulin sensitivity). * p<0.05, **p<0.01.

References

1.   Mari A, Pacini G, Murphy E, Ludvik B, Nolan JJ. A model-based method for assessing insulin sensitivity from the oral glucose tolerance test. *Diabetes Care.* 2001;24:539-548.