

Supplementary Materials for  
**High-throughput proteomics uncovers exercise training and type 2 diabetes-  
induced changes in human white adipose tissue**

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*Sci. Adv.* **9**, eadi7548 (2023)  
DOI: 10.1126/sciadv.adi7548

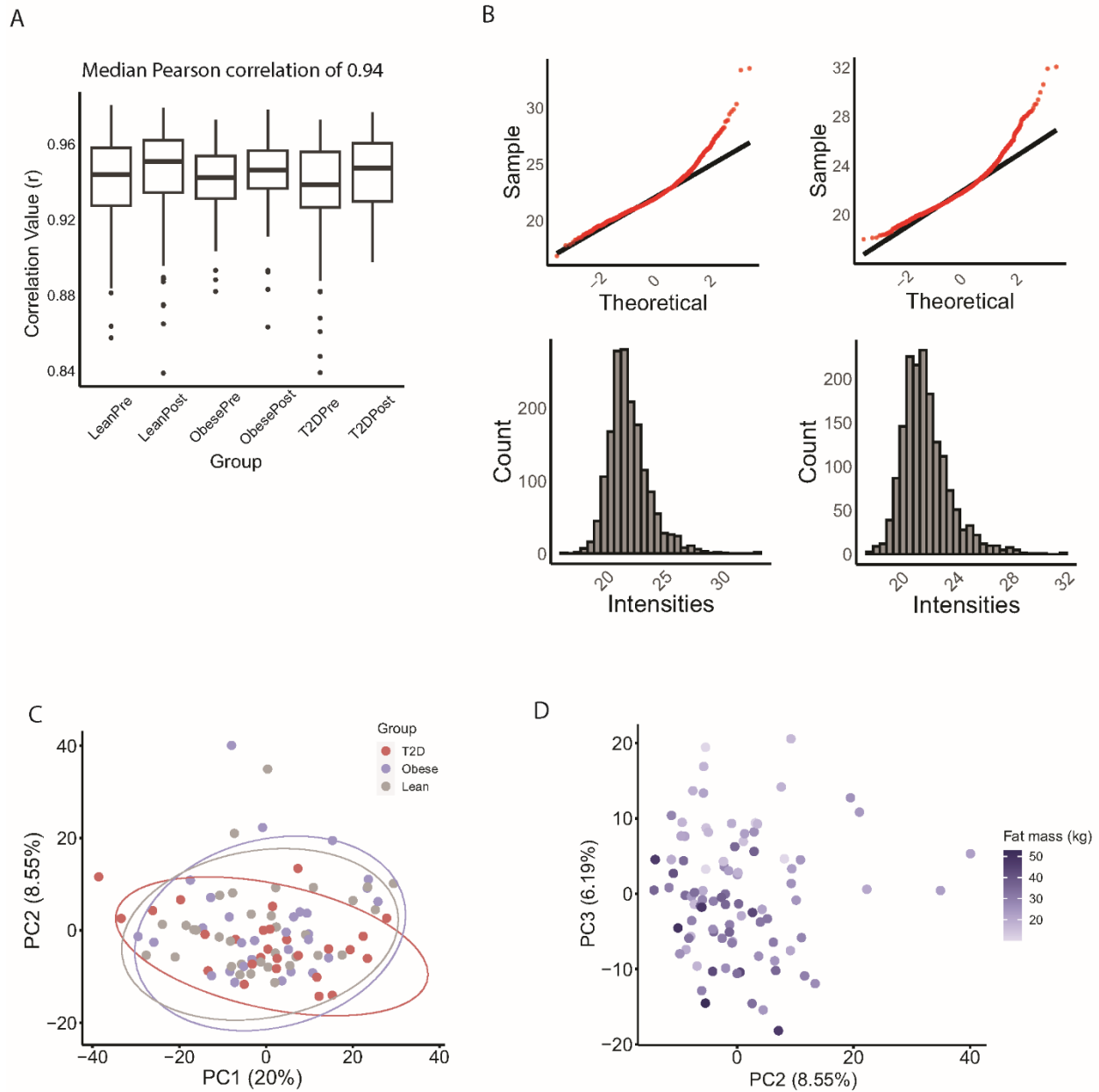
**The PDF file includes:**

Figs. S1 to S3  
Tables S1 and S2  
Legend for data S1

**Other Supplementary Material for this manuscript includes the following:**

Data S1

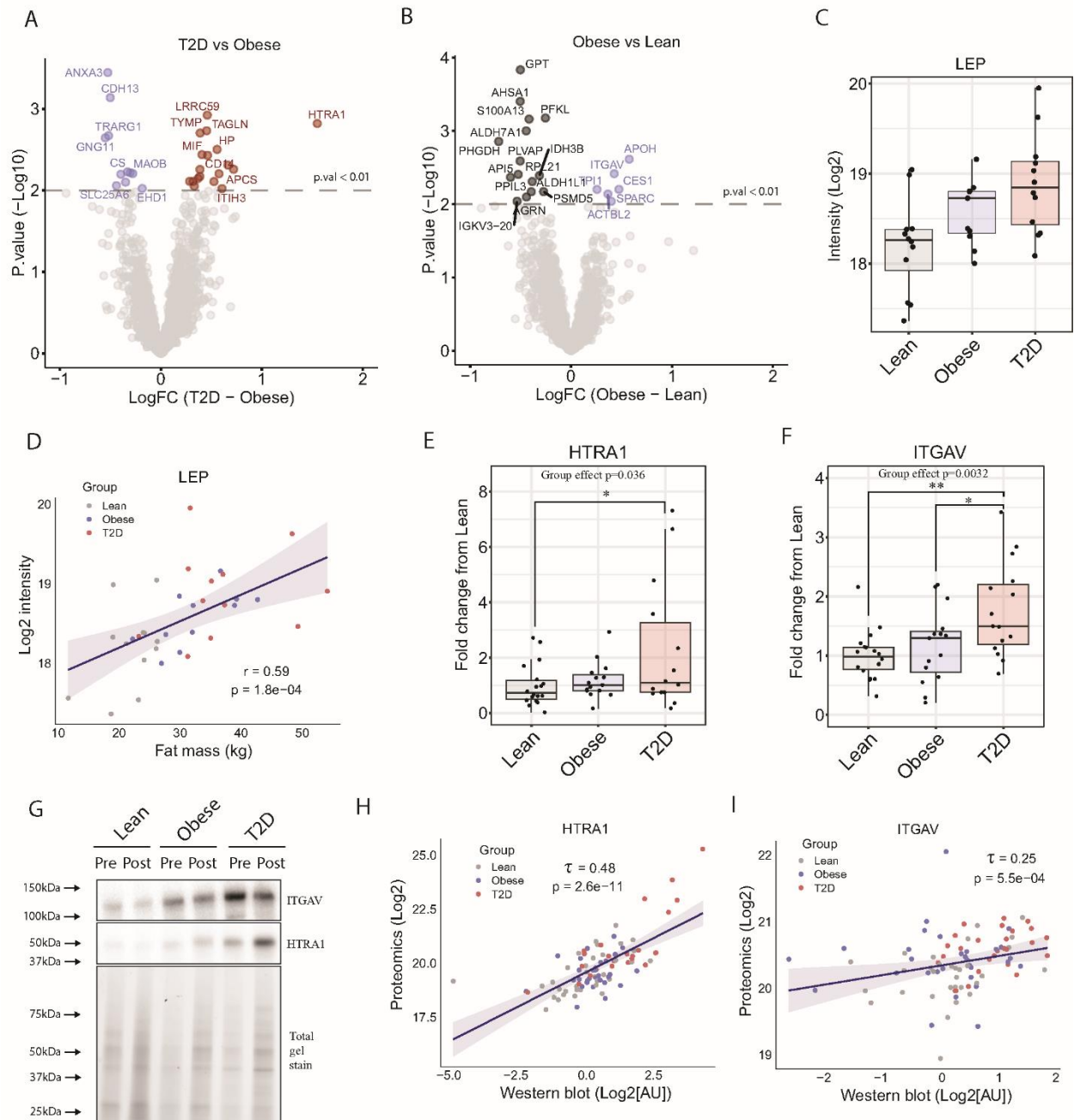
Figure S1



**Fig. S1.**

**High-throughput proteome analysis of human white adipose tissue.** A) Boxplot of Pearson correlation between runs within each experimental group. B) QQ-plot and histogram of protein log<sub>2</sub>-intensity distribution of two samples, C) PCA plot of component 1 (x-axis) and 2 (y-axis) colored by group. D) PCA plot of component 2 (x-axis) and 3 (y-axis) colored by fat mass (kg).

Figure S2

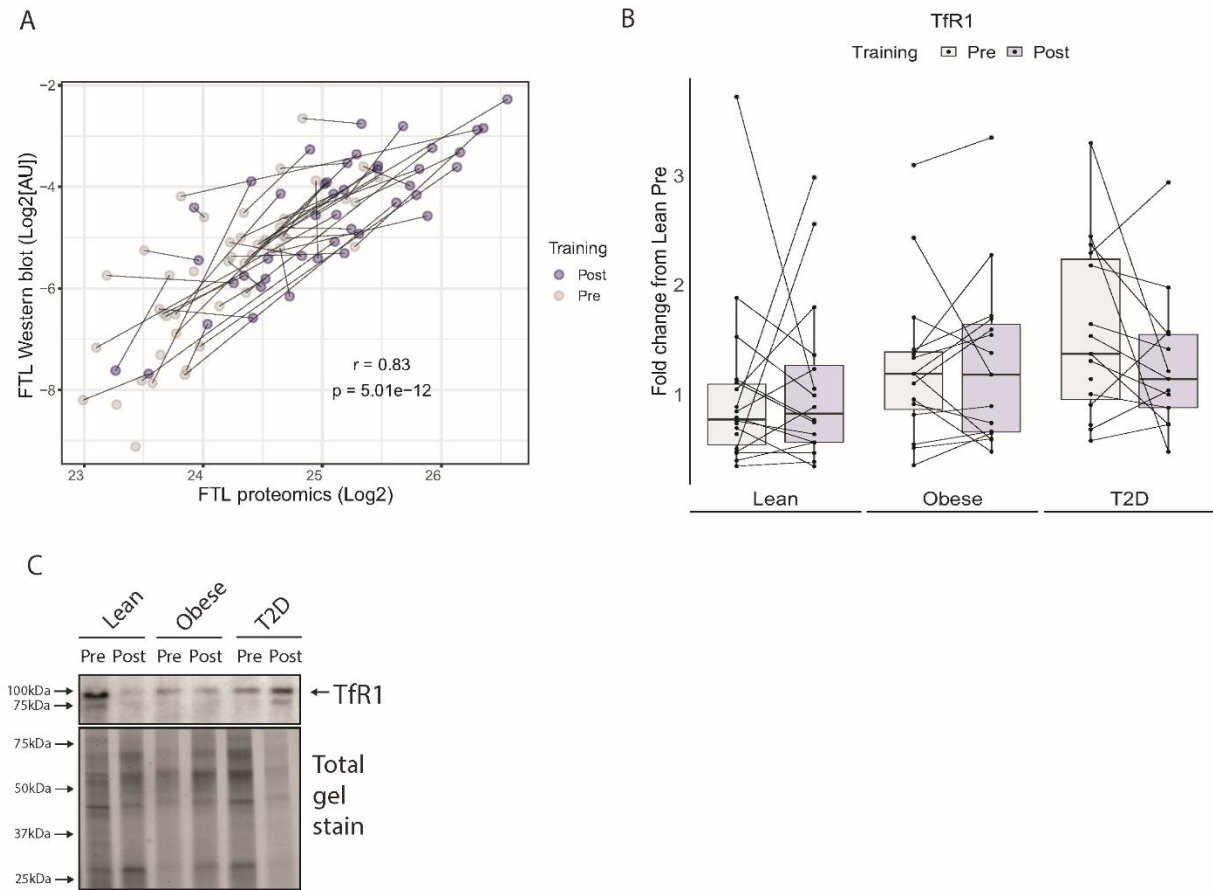


**Fig. S2.**

**Baseline proteome differences in WAT from lean obese and T2D individuals.** A-B) Volcano plot of baseline T2D-Obese and Obese-Lean comparison ( $p < 0.01$ ). C) Boxplot of baseline log2-abundance of Leptin. D) Pearson's correlation of Leptin abundance and fat mass. E-F) Boxplot of baseline HTRA1 and ITGAV abundance in scWAT measured by western blot. G) Representative western blots. H-I) Kendall's rank correlation of HTRA1 and ITGAV intensities measured by proteomics and western blot, respectively. One-way ANOVA was applied to test for group-

differences. Tukey's multiple comparisons test was used to test differences between individual groups. \* $p < 0.05$  and \*\* $p < 0.01$  versus T2D.

Figure S3



**Fig. S3.**

**Effects of exercise training on FTL and TfR1 in human scWAT.** A) Repeated measures correlation analysis of quantified FTL abundance by proteomics (x-axis) and western blot (y-axis). B) TfR1 abundance pre and post HIIT as quantified by western blot. C) Representative western blots. The upper band at ~ 100 kDa was quantified as TfR1.

**Table S1.****Clinical and metabolic characteristics, before and after 8 weeks high-intensity interval training (HIIT)**

Characteristics	Lean individuals		Obese individuals		T2D individuals	
	Pre	Post	Pre	Post	Pre	Post
<i>N</i>	18	16	15	15	15	13
Age (years)	56.2±1.5		53.8±1.8		55.2±1.7	
Weight (kg)	78.9±2.0	77.3±2.2*	100.0±2.9 <sup>††</sup>	98.5±2.6* <sup>††</sup>	103.1±3.7 <sup>††</sup>	102.5±4.1* <sup>††</sup>
BMI (kg/m <sup>2</sup> )	24.0±0.4	23.7±0.4*	30.8±0.7 <sup>††</sup>	30.3±0.6* <sup>††</sup>	31.2±0.8 <sup>††</sup>	30.8±0.9* <sup>††</sup>
Waist (cm)	90±1	87±2**	106±2 <sup>††</sup>	102±2** <sup>††</sup>	109±2 <sup>††</sup>	106±2** <sup>††</sup>
Total fat mass (kg)	20.1±1.0	18.2±1.2**	32.0±1.9 <sup>††</sup>	29.7±1.8** <sup>††</sup>	34.8±2.3 <sup>††</sup>	33.0±2.5** <sup>††</sup>
Total lean body mass (kg)	56.9±1.3	57.1±1.4*	65.3±1.3 <sup>††</sup>	66.2±1.2* <sup>††</sup>	64.8±1.7 <sup>††</sup>	66.8±2.0* <sup>††</sup>
VO <sub>2</sub> max (ml O <sub>2</sub> /kg/min)	38.0±1.5	42.8±1.7**	33.4±1.8 <sup>†</sup>	36.6±1.2* <sup>†</sup>	25.8±0.9 <sup>†††</sup>	30.5±1.0* <sup>†††</sup>
HbA1c (mmol/mol)	35±1	34±1*	35±1	34±1*	54±4 <sup>†††</sup>	51±4* <sup>†††</sup>
Fasting plasma glucose (mmol/l)	5.2±0.1	5.4±0.1	5.6±0.1	5.4±0.1*	9.6±0.7 <sup>†††</sup>	8.8±0.7* <sup>†††</sup>
Fasting serum insulin (pmol/l)	61±9	62±12	71±8	65±7	117±19 <sup>†</sup>	98±15* <sup>†</sup>
Plasma triglycerides (mmol/l)	1.52±0.23	1.43±0.31	1.59±0.19	1.35±0.13	2.47±0.40 <sup>†</sup>	2.08±0.37 <sup>†</sup>
GIR (mg/min/m <sup>2</sup> )	325±6	442±33**	325±22	439±33**	189±28 <sup>†</sup>	300±36* <sup>††</sup>

Data are means±SEM. \**P*<0.05 and \*\**P*<0.001 vs. before (Pre) HIIT, <sup>†</sup>*P*<0.05 and <sup>††</sup>*P*<0.001 vs. lean, <sup>‡</sup>*P*<0.05 and <sup>‡‡</sup>*P*<0.001 vs. obese. T2D, type 2 diabetes; WHR, waist-to-hip ratio; Pre, pre-training; Post, post-training; GIR, insulin-stimulated glucose infusion rate; HIIT, high-intensity interval training. The clinical and metabolic data were reported previously (6).

**Table S2.****Key resources table**

REAGENT or RESOURCE	SOURCE	IDENTIFIER
Antibodies		
FTL	LifeTechnologies	Cat #PA5-83567
FTH1	FischerScientific	Cat #PA5-27500
Transferin receptor	Abcam	Cat #Ab214039
Oxphos cocktail	Abcam	Cat #Ab110411
HTRA1	Abcam	Cat #Ab274322
ITGAV	Invitrogen	Cat #MA5-32195
Anti-rabbit	Jackson Immuno	Cat #111-036-045
Anti-mouse	Jackson Immuno	Cat #315-036-045
Biological Samples		
Human subcutaneous fat biopsies	This study	N/A
Chemicals, Peptides, and Recombinant Proteins		
2-Chloroacetamide	Sigma	Cat# 22790
SDS	Sigma	Cat# 71736/74255
Tris-HCl	Roche	Cat# 10812846001
Acetonitrile	Thermo	Cat# A9554
Trifluoroacetic acid	Sigma	Cat# 808260
NH4OH	Merck	Cat# 105428
Benzonase	Merck	Cat# 71205
4-20% Mini-Protean TGX stain-free protein gels	BioRad	Cat #4568096
8-16% Mini-Protean TGX stain-free protein gels	BioRad	Cat #4568106
Bovine serum albumin	Merck	Cat #A7906
Skim milk powder	Merck	Cat #70166
Trisma	Sigma	Cat #T6066
NaCl	Sigma	Cat #31434
Tween20	Sigma	Cat #P7949
Glycine	Sigma	Cat #G7126
Precision plus protein all blue standard	BioRad	Cat #1610393
ECL luminata forte western HRP substrat	Merck	Cat #WBLUF0500
Super ECL	Fischer Scientific	Cat #34094
Deposited Data		
Mass spectrometry proteomics data	This paper	PRIDE: PXD040434.
Software and Algorithms		
R version v4.2.1	R Development Core Team, 2016	<a href="https://www.R-projects.org/">https://www.R-projects.org/</a>
FragPipe v19.0	(37)	<a href="https://github.com/Nesvilab/FragPipe">https://github.com/Nesvilab/FragPipe</a>

DIA-NN v1.8.2	(41)	<a href="https://github.com/vdemichev/DiaNN">https://github.com/vdemichev/DiaNN</a>
SigmaPlot 12.5	Systat Software	
ImageLab 6.1	BioRad	
ChemiDoc VRS+ Imaging system	BioRad	

**Captions for the supplementary Data File S1:**

- A. Overview
- B. Complete matrix of all 3773 quantified proteins
- C. Filtered matrix (2016 proteins) for differential expression (DE) analysis
- D. Overview of 48 significant proteins in T2D-Lean baseline comparison
- E. Baseline correlation of 48 significant proteins with clinical parameters
- F. Diabetes-driven GeneSet enrichment analysis of GOBP and GOCC terms
- G. Obesity-driven GeneSet enrichment analysis of GOBP and GOCC terms
- H. Overlap of HIIT-induced changes in GOCC terms in lean and obese group
- I. HIIT-induced changes in GOCC and GOBP terms in T2D group