

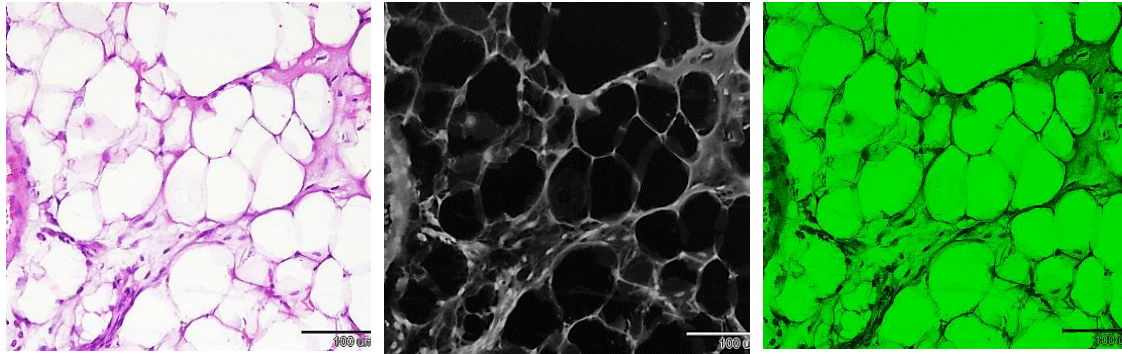
Supplementary information

Materials and methods:

Diet composition: Diet composition: ND (El-Salam Co., Cairo, Egypt) with the following nutritional ratios: 21% protein, 4.27% fat, and 3.55%, with a total energy of 3150 kcal/kg and main components of corn, soybean seeds, L-lysine, methionine, sodium bicarbonate, and sodium chloride. HFD: Each 100 g of the diet was prepared by mixing 35 g of beef tallow with 65 g of the normal chow diet to obtain the following composition: 13.65% protein, 37.78% fat, and 2.30% fiber (percentage of fat was reported previously) [1].

Supplementary Table 1: Sequences of primers used for qRT-PCR reactions, their annealing temperature, product size, and references used.

Gene	Sequences (5'→3')		Annealing Temp (°C)	Product size (bp)	Reference
<i>UCP-1</i>	F	ATGTACACCAAGGAAGGAC	57°C	147	[2]
	R	GGTACAATCCACTGTCTGTC			
<i>GAPDH</i>	F	ATGGCCTTCCGTGTTCTACCC	55°C	104	[3]
	R	GCCTGCTTCACCACCTTCTTGATG			

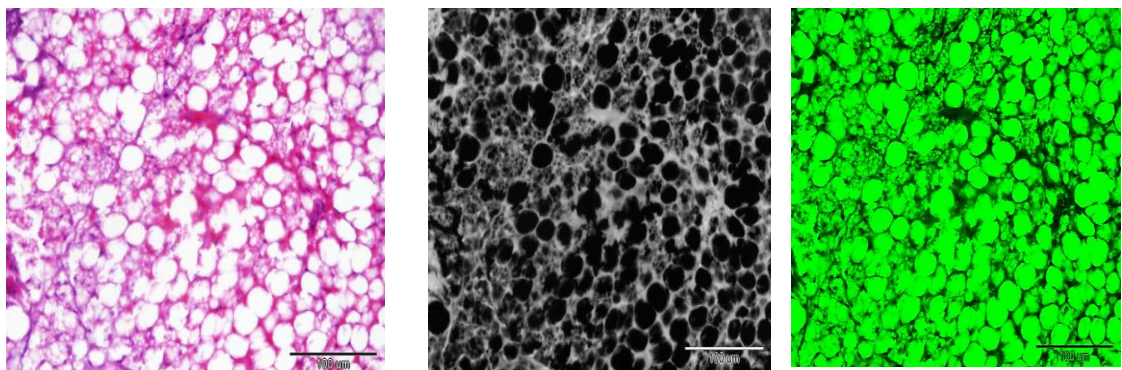


A: White fat from the female control group stained by H&E. The stored lipid appears as clear, unstained globules filling the unilocular adipocytes.

B: White fat from the female control group inverted by Digimizer software. Stored lipid appears as black areas within unilocular adipocytes.

C: White fat of the inverted picture colored by Digimizer software for analysis by Color Summarizer. Stored lipid appears as green-colored globules in unilocular adipocytes.

Supplementary Figure 1: Image processing stages of white fat sections to calculate the percentage of fat in the cells.

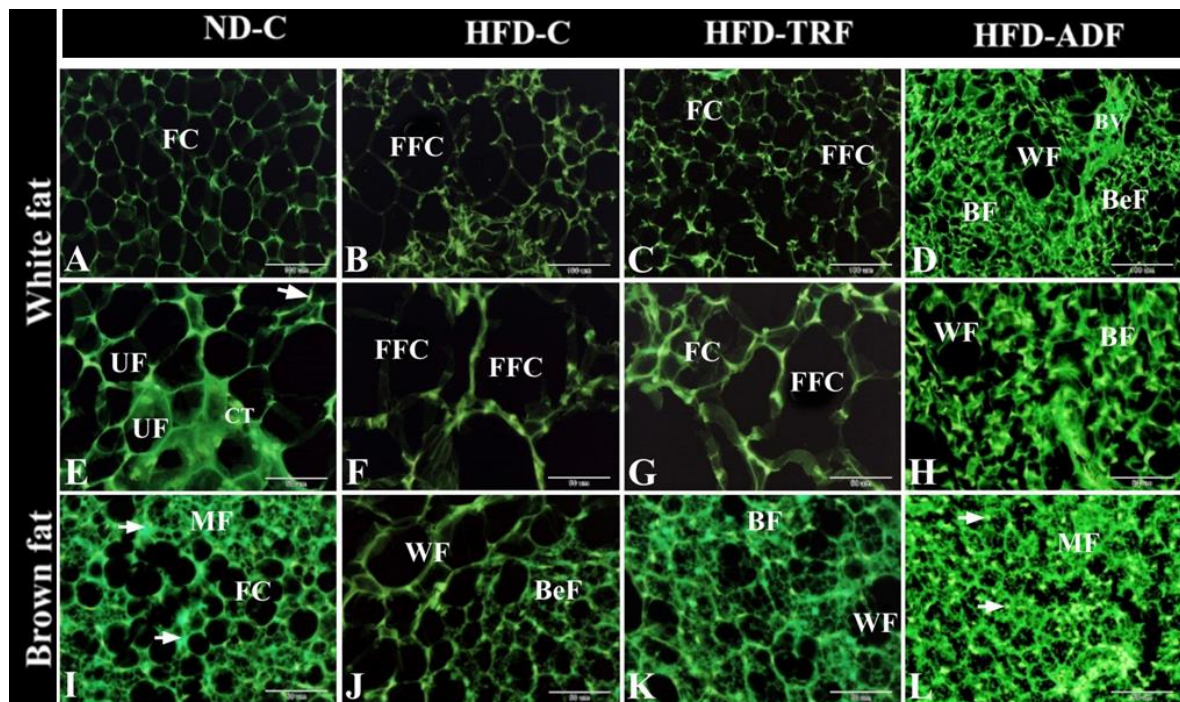


A: Brown fat from the female control group stained by H&E. The stored lipid appears as clear, unstained globules or particles in multilocular adipocytes.

B: Brown fat from the female control group inverted by Digimizer software. Stored lipid appears as black areas within adipocytes.

C: Brown fat of inverted picture colored by digimizer software for analysis by color summarizer. Stored lipid colored green.

Supplementary Figure 2: Image processing stages of brown fat sections to calculate the percentage of fat in the cells.



Supplementary Figure 3: Negative image of Fig. 6a, showing the size, amount, and distribution of fat droplets and connective tissue in different groups; fat droplets appear black and connective tissue appears green in color.

References:

1. Matias, A.M., et al., *Differential Effects of High Sugar, High Lard or a Combination of Both on Nutritional, Hormonal and Cardiovascular Metabolic Profiles of Rodents*. *Nutrients*, 2018. **10**(8): p. 1071.
2. Hiradate, R., et al., *A novel dual-targeted rosiglitazone-loaded nanoparticle for the prevention of diet-induced obesity via the browning of white adipose tissue*. *J Control Release*, 2021. **329**: p. 665-675.
3. Suenaga, K., et al., *Differential gene expression profiling between genotoxic and non-genotoxic hepatocarcinogens in young rat liver determined by quantitative real-time PCR and principal component analysis*. *Mutat Res*, 2013. **751**(1): p. 73-83.