



**University of Dundee**

**Adipocyte Integrin-Linked Kinase Plays a Key Role in the Development of Diet-Induced Adipose Insulin Resistance in Male Mice**

Bugler-Lamb, Aimée R.; Hasib, Annie; Weng, Xiong; Hennayake, Chandani K.; Lin, Chenshi; McCrimmon, Rory J.

*Published in:*  
Molecular Metabolism

*DOI:*  
[10.1016/j.molmet.2021.101197](https://doi.org/10.1016/j.molmet.2021.101197)

*Publication date:*  
2021

*Document Version*  
Other version

[Link to publication in Discovery Research Portal](#)

*Citation for published version (APA):*

Bugler-Lamb, A. R., Hasib, A., Weng, X., Hennayake, C. K., Lin, C., McCrimmon, R. J., Stimson, R. H., Ashford, M. L. J., Wasserman, D. H., & Kang, L. (2021). Adipocyte Integrin-Linked Kinase Plays a Key Role in the Development of Diet-Induced Adipose Insulin Resistance in Male Mice. *Molecular Metabolism*, 49, Article 101197. <https://doi.org/10.1016/j.molmet.2021.101197>

**General rights**

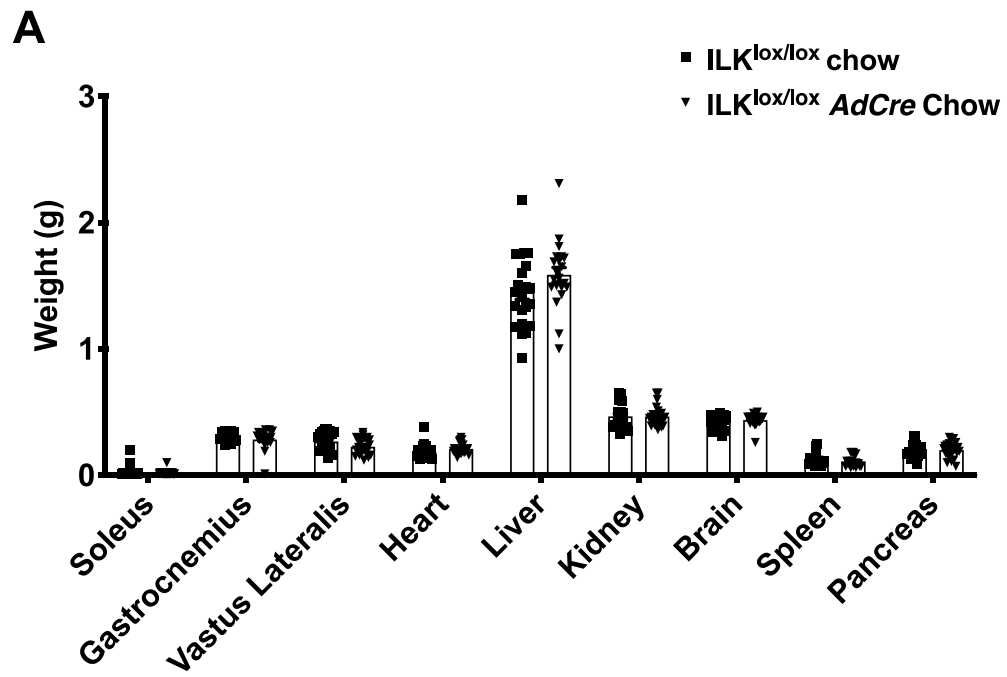
Copyright and moral rights for the publications made accessible in Discovery Research Portal are retained by the authors and/or other copyright owners and it is a condition of accessing publications that users recognise and abide by the legal requirements associated with these rights.

**Take down policy**

If you believe that this document breaches copyright please contact us providing details, and we will remove access to the work immediately and investigate your claim.

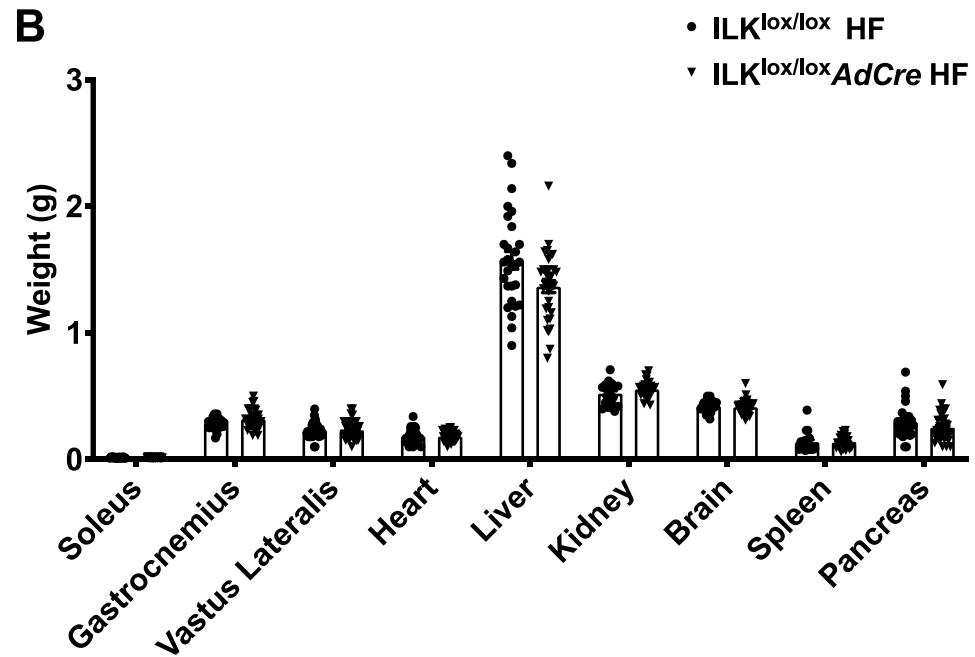
Subject number	Age	Sex	Height (m)	Weight (Kg)	BMI (kg/m <sup>2</sup> )	Waist (cm)	Hip (cm)	% Fat	Fat mass (kg)	Systolic BP	Diastolic BP
1	60	Female	1,65	65	23,9	90	100	36,3	23,6	121	68
2	39	Female	1,62	60	22,9	96	105	28,4	17	114	76
3	61	Female	1,58	58	23,2	81,5	95	37,4	21,7	132	97
4	69	Male	1,72	70	23,7	88	91	26,8	18,6	151	84
5	58	Female	1,6	74	28,9	103	107	38,6	28,6	137	92
6	40	Female	1,69	79	27,7	84,5	105	30,3	24,1	125	77
7	60	Female	1,778	87,8	27,8	82,5	112	38,8	34,1	118	78
8	67	Male	1,77	92,1	29,4	105	107	31,9	29,4	125	73
9	59	Female	1,47	88	40,7	112	125	47	41,4	159	95
10	34	Female	1,63	107,5	40,5	114	124	44,8	48,2	130	87
11	60	Female	1,54	104,8	44,2	119	130	49,8	52,2	120	77
12	64	Male	1,82	140	42,3	148	146	39,7	55,5	155	75

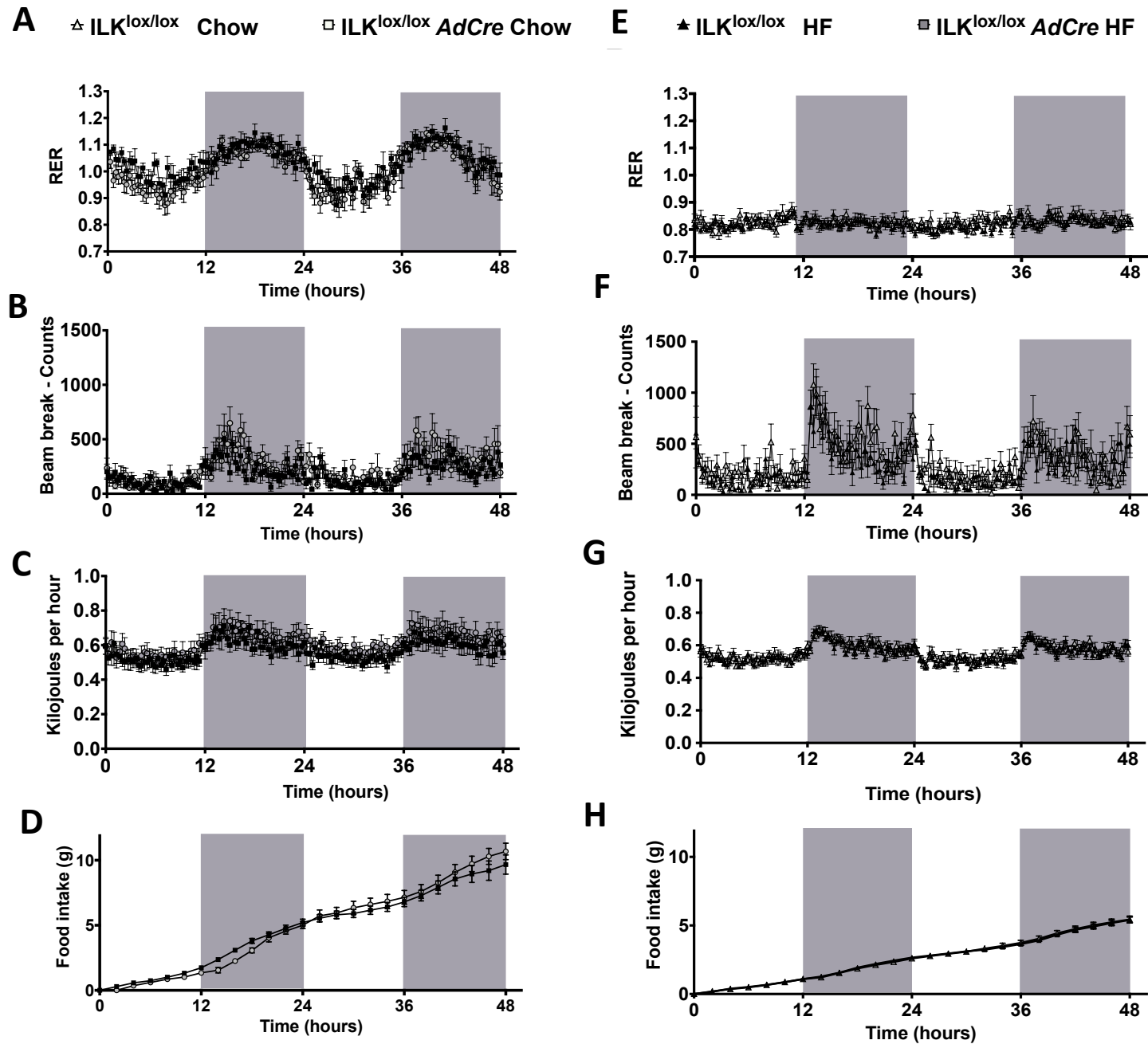
**Table S1:** Anthropometric data of the patients from which the visceral and subcutaneous adipose tissue samples were collected. To determine ILK protein expression, the patients were divided into three groups based on BMI: Lean BMI < 25; Overweight BMI < 30; Morbidly obese BMI > 40.



**Figure S1:**

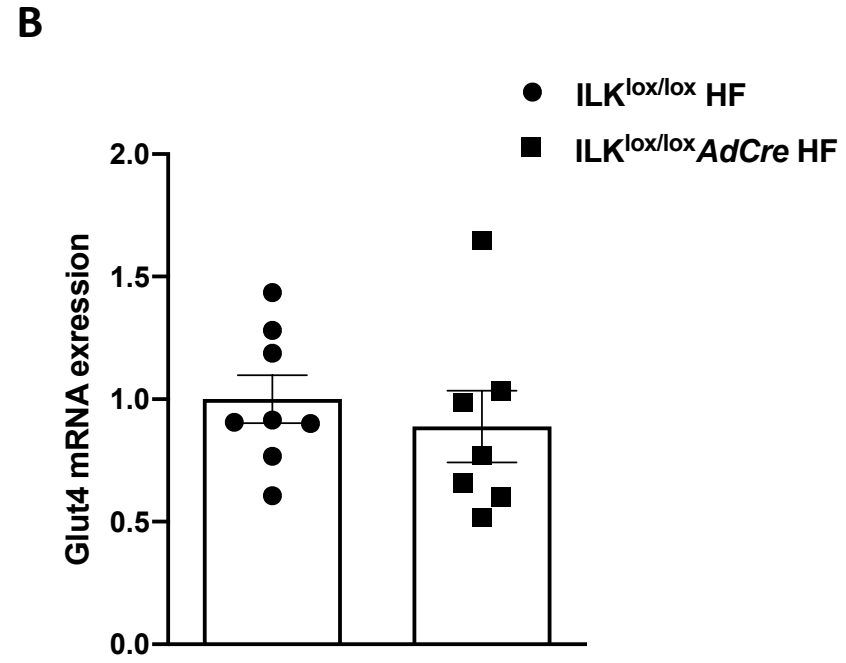
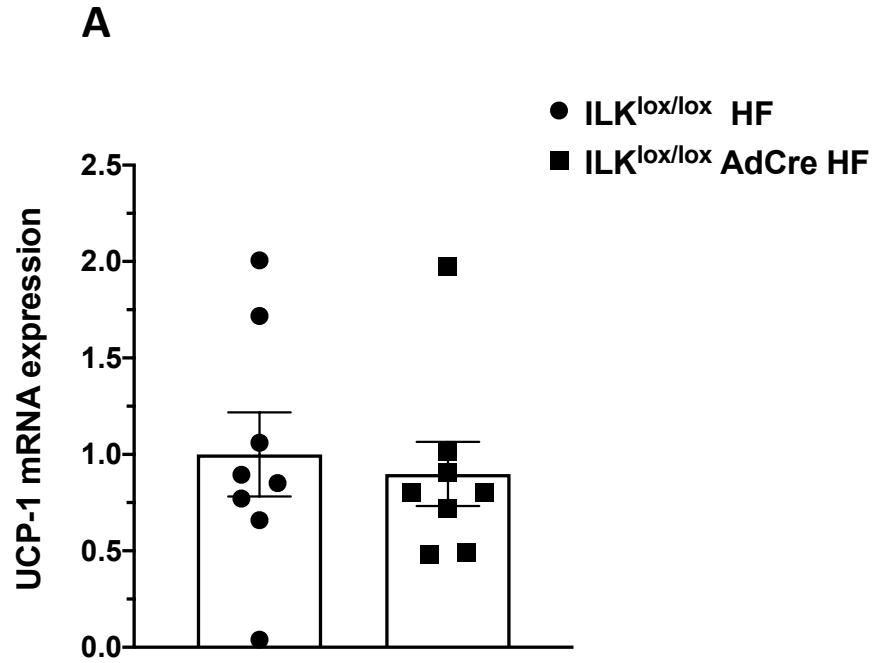
Wet weights of individual tissues from chow-  
(**A**) and HF-fed (**B**) mice at 22 weeks of age.  
N= 16-23.





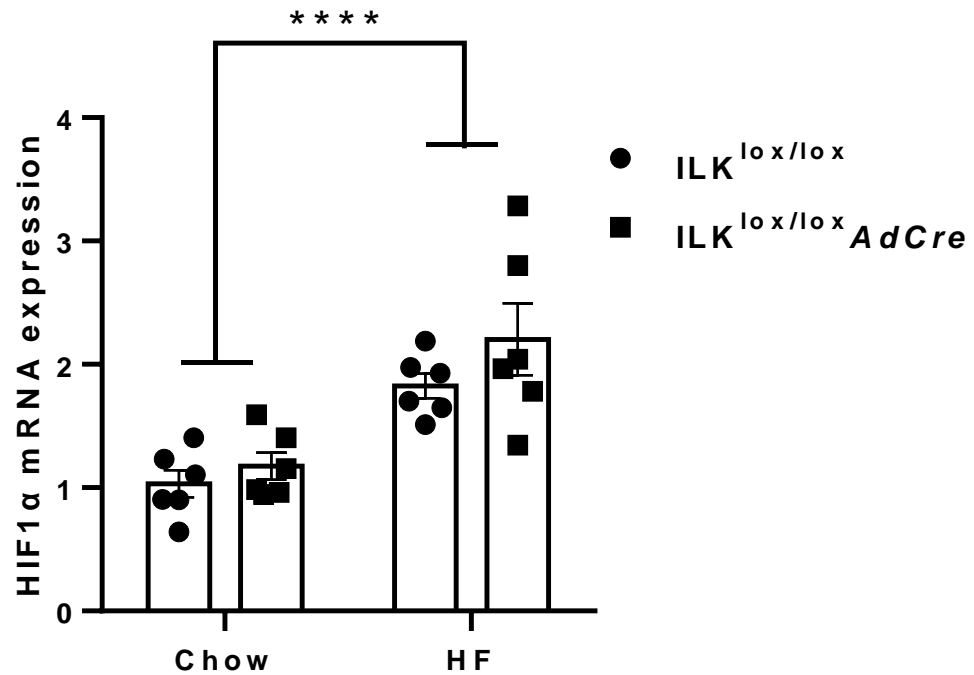
**Figure S2:**

Calorimetric data collected from mice at 20 weeks of age over the course of a 48 hour period. **A&E:** Respiratory exchange ratio (RER) in chow and HF diet-fed mice. **B&F:** Activity in the chow and HF diet-fed mice measured by number of beam breaks. **C&G:** Energy expenditure in chow and HF diet-fed mice. **D&H:** Accumulative food intake in chow and HF diet-fed mice. N=6-9.



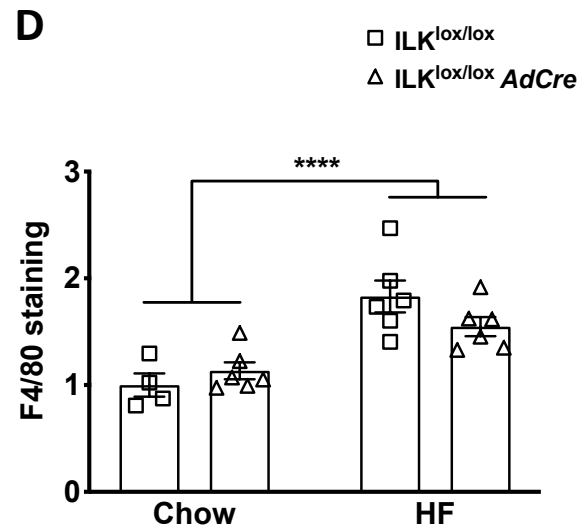
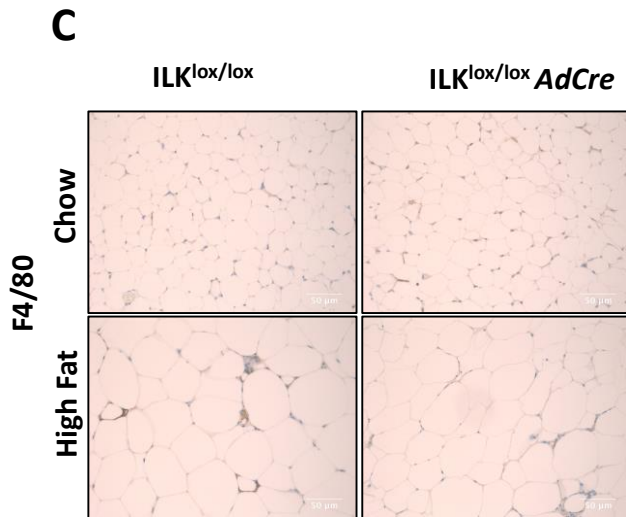
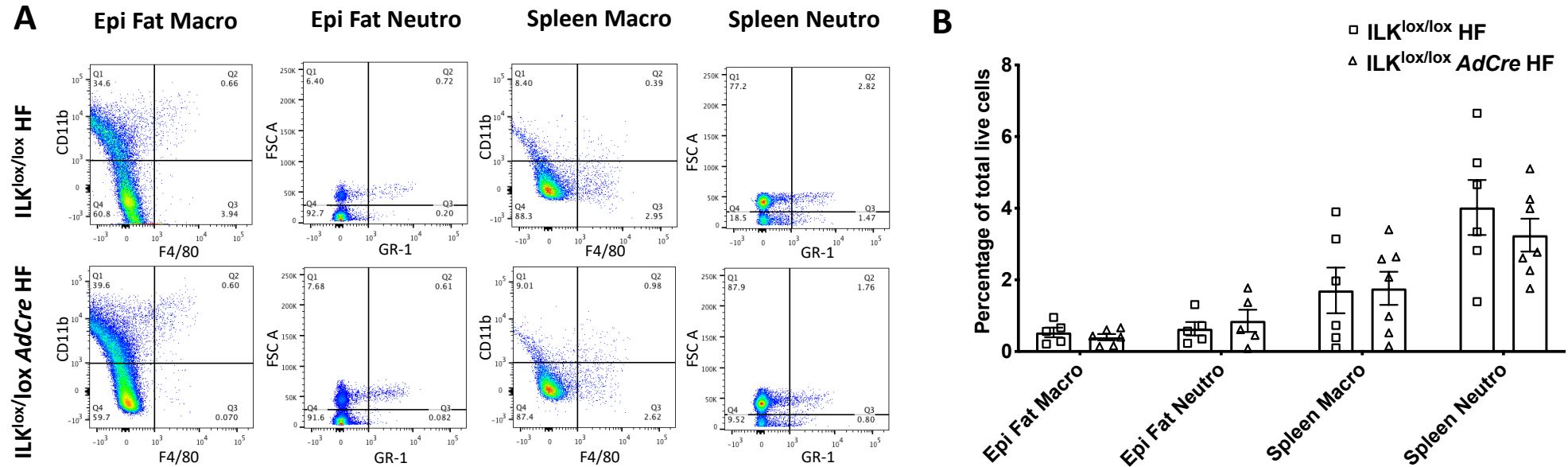
**Figure S3:**

Gene expression of UCP-1 (A) and GLUT4 (B) was determined by quantitative real-time PCR in BAT of HF-fed mice. Data were normalised to HF-fed  $ILK^{lox/lox}$  mice. N=7-8.



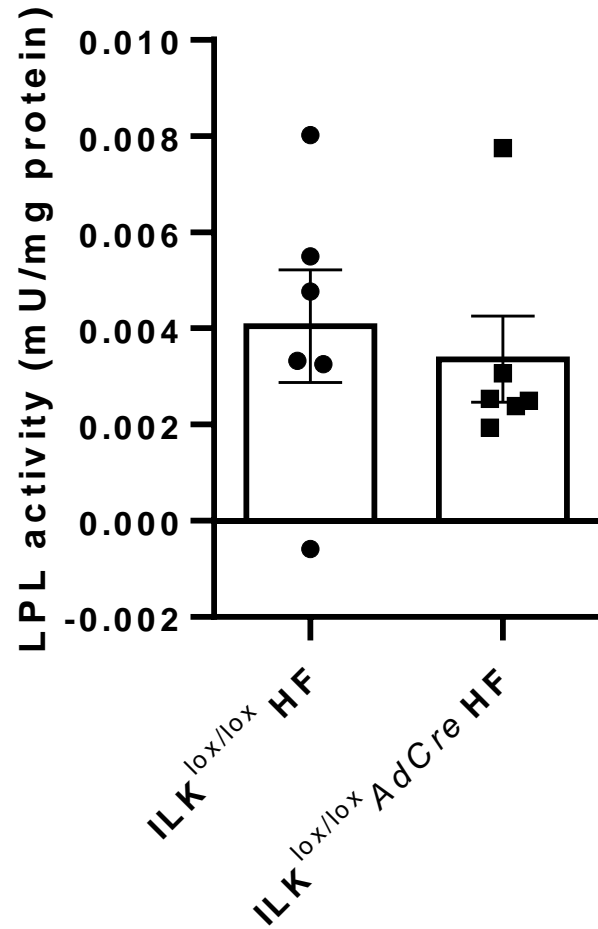
**Figure S4:**

Gene expression of HIF1 $\alpha$  was determined by quantitative real-time PCR in eWAT of chow- and HF-fed mice. Data were normalised to chow-fed  $ILK^{lox/lox}$  mice. N=6. \*\*\*\* $p$ <0.0001.



**Figure S5:**

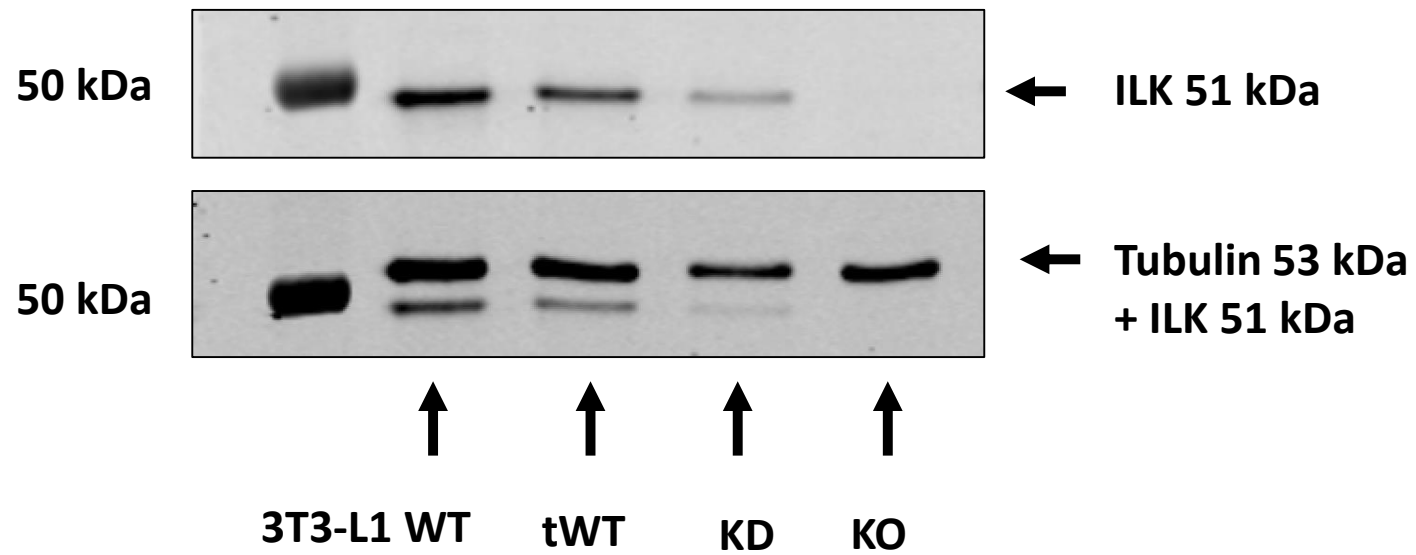
Macrophage infiltration in the eWAT and spleen of HF-fed ILK<sup>lox/lox</sup> and ILK<sup>lox/lox</sup>AdCre mice at 22 weeks of age. **A&B:** By flow cytometry, macrophages and neutrophils were detected and measured in both spleen and the eWAT of HF-fed ILK<sup>lox/lox</sup> and ILK<sup>lox/lox</sup>AdCre mice (n=5-7). **C&D:** Quantification of macrophage infiltration in eWAT through the use of F4/80 antibody (n=4-6). All data are represented by mean +/- SEM with significance \*\*\*\* p<0.001.



**Figure S6:**

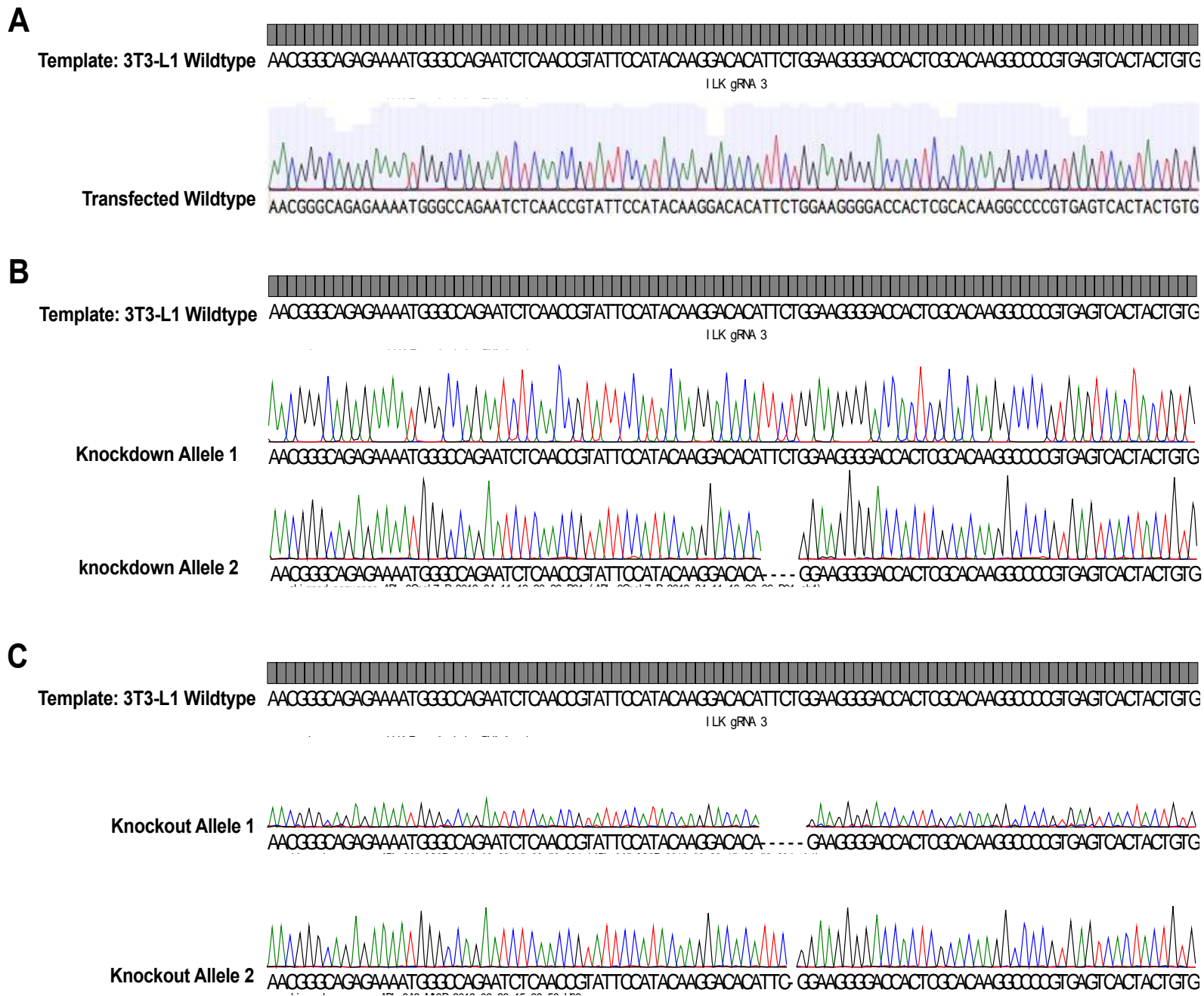
Lipoprotein lipase (LPL) activity was measured in the eWAT of HF-fed mice. N=6.





**Figure S7:**

ILK protein expression was measured by Western Blotting in the naïve 3T3-L1 wildtype (WT), transfected WT (tWT), knockdown (KD) and knockout (KO) 3T3-L1 cells. Representative blots from at least 2 independent biological replicates were shown.



**Figure S8:**

Genomic sequencing of the targeted exon of the ILK gene. **A:** Transfected wildtype cells vs the naïve 3T3-L1 wildtype cells. **B:** Mutation in one of the alleles in the knockdown cells compared to the 3T3-L1 template. **C:** Mutations in both alleles of the gene in the knockout cells compared to the 3T3-L1 template.