Supplemental Data

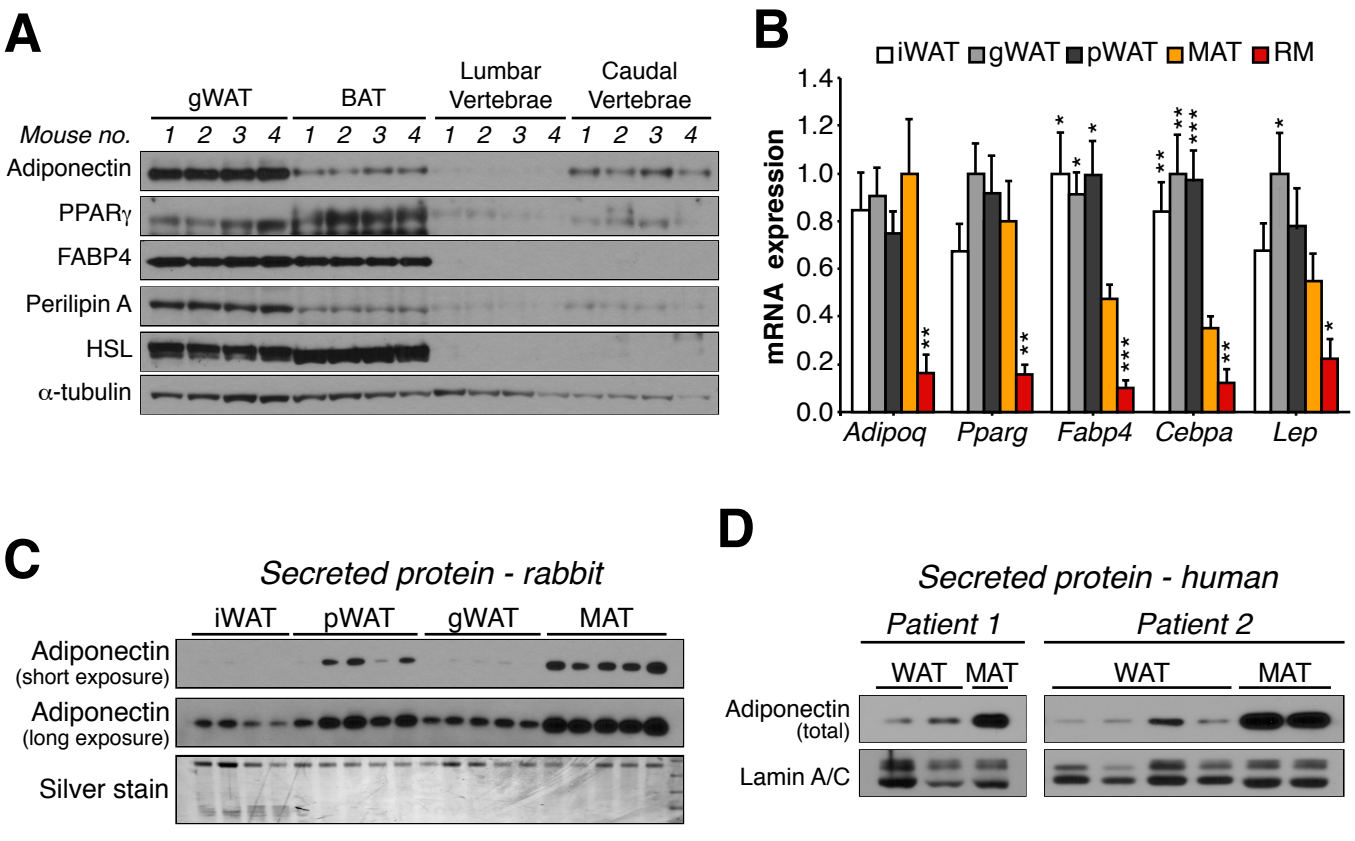

Figure S1, Related to Figure 1 – Adiponectin expression and secretion in MAT and WAT of mice, rabbits and humans. (A) gWAT and brown adipose tissue (BAT) were collected from male C3H mice, along with whole lumbar vertebrae for red marrow, and whole caudal vertebrae for MAT. Total protein was isolated and expression of the indicated proteins was determined by immunoblotting; α-tubulin was analyzed as a loading control. Blots of tissue lysates from four mice, representative of at least ten mice, are shown. **(B,C)** WAT, tibia MAT, and tibia red marrow (RM) were isolated from New Zealand White rabbits. **(B)** Total RNA was isolated from iWAT, gWAT, pWAT, tibia MAT and tibia RM and expression of the indicated transcripts was assessed by qPCR. Transcript expression was normalized to *Ppia* (cyclophilin A) and *Tbp* mRNA and is presented relative to the maximum expression of each transcript as mean +/- SEM of 8-9 rabbits. Statistically significant differences between expression in tibia MAT and other tissues are indicated as follows: * = P <0.05; ** = P <0.01; *** = P <0.001. **(C)** Immunoblots and silver stain of conditioned media from WAT and MAT from one rabbit, representative of seven rabbits. Silver staining was used to assess total protein content of media. **(D)** Immunoblots of conditioned media from explants of scWAT and MAT isolated from patients 1 and 2. Lamin A/C was analyzed as an estimate of explant breakdown.

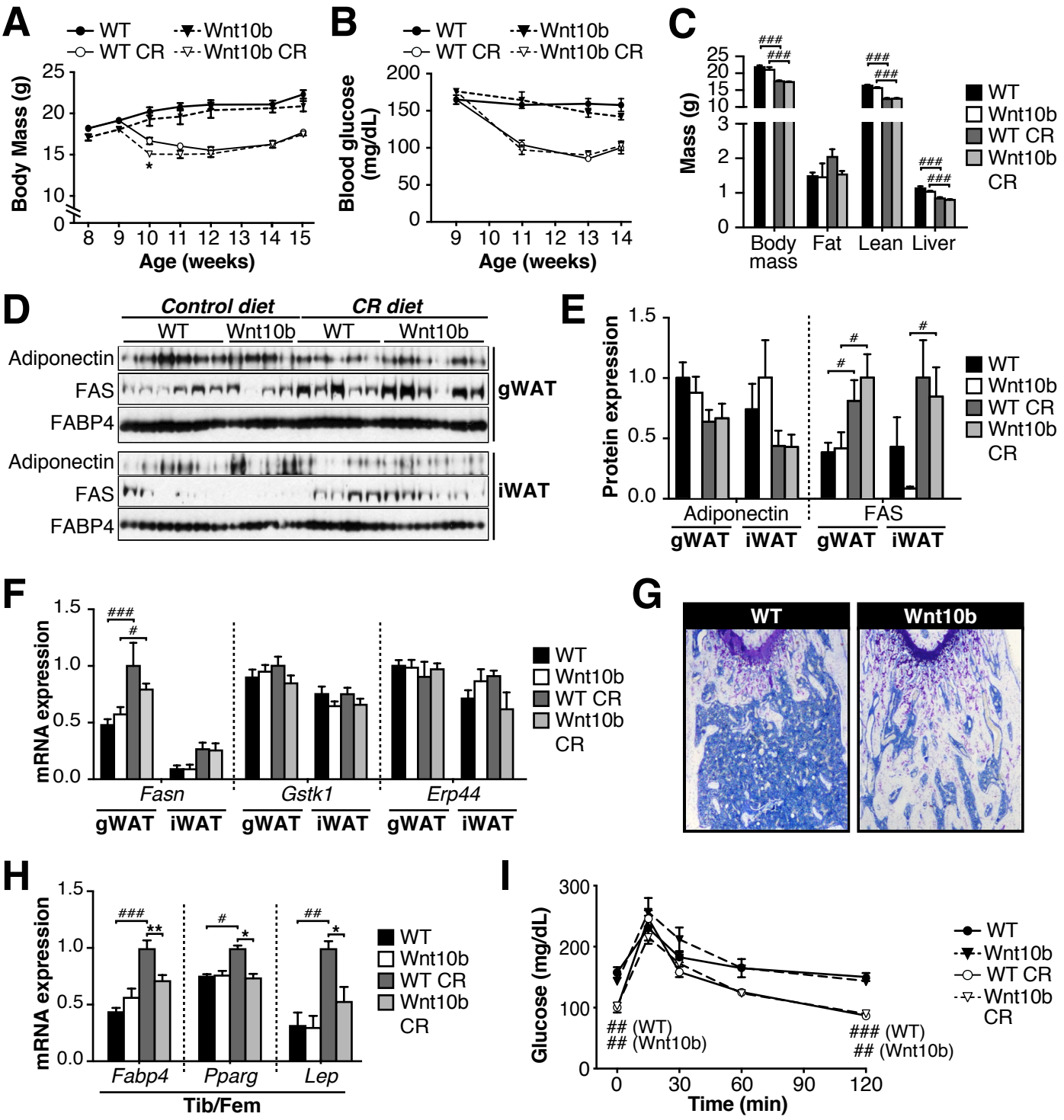

Figure S2, Related to Figure 2 – Additional characteristics of control- or CR-fed WT and Ocn-Wnt10b mice. Wild-type (WT) or Ocn-Wnt10b (Wnt10b) mice were fed *ad libitum* or a 30% CR diet from 9 to 15 weeks of age. **(A,B)** Body mass and blood glucose at the indicated ages. From 10 weeks of age, body mass and blood glucose of CR-fed mice of either genotype were significantly lower than in the control-fed counterparts; however, statistical significance is not indicated on these graphs to make them easier to view. **(C)** Body composition of 15-week-old live mice was determined by NMR; liver masses were recorded at necropsy. **(D-F)** Total RNA and/or protein was isolated from iWAT and gWAT of each mouse. Expression of the indicated proteins was determined by immunoblotting (D), with adiponectin and FAS quantified by densitometry (E). **(F)** Expression of the indicated transcripts, normalized to *Ppia* mRNA, was determined by qPCR. **(G)** Representative micrographs of Toluidine blue-stained sections of distal femurs of control-fed mice. **(H)** Total RNA was isolated from whole tibiae and femurs. Expression of *Fabp4, Pparg* and *Lep*, normalized to *Ppia* mRNA, was determined by qPCR to provide an alternative readout of BM adiposity. **(I)** Glucose tolerance tests in 14-week-old mice. Data and statistical significance are reported as described for Figure 2.

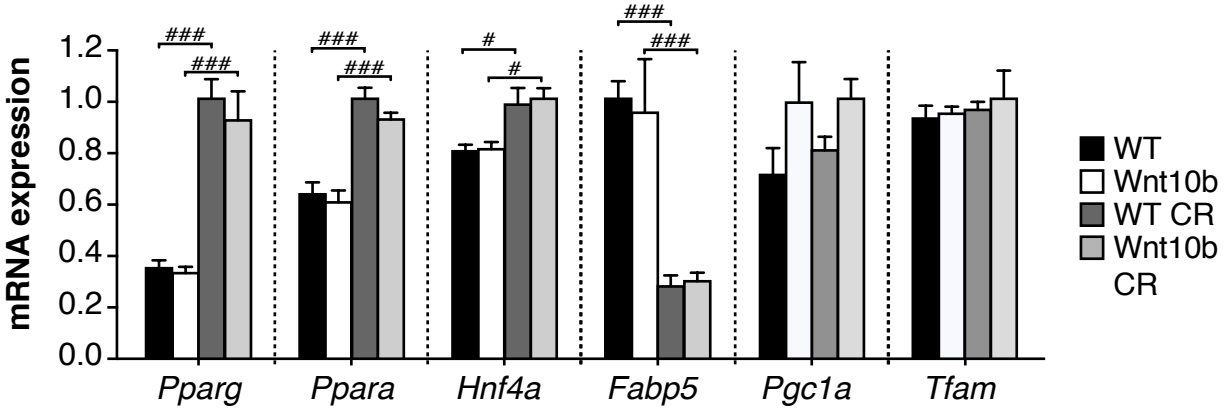

Figure S3, Related to Figure 2 – Transcript expression in livers of control- or CR-fed WT and Ocn-Wnt10b mice. Total RNA was isolated from the liver of each mouse. Expression of the indicated transcripts, normalized to *Ppia and Tbp* mRNA, was determined by qPCR. Data and statistical significance are reported as described for Figure 2.

Supplemental Data

Figure S1, Related to Figure 1 – MAT of rabbits and C3H/HeJ mice expresses adiponectin at higher levels than other adipocyte transcripts and proteins. (A) gWAT and brown adipose tissue (BAT) were collected from male C3H mice, along with whole lumbar vertebrae for red marrow, and whole caudal vertebrae for MAT. Total protein was isolated and expression of the indicated proteins was determined by immunoblotting; α-tubulin was analyzed as a loading control. Blots of tissue lysates from four mice, representative of at least ten mice, are shown. **(B)** iWAT, gWAT, pWAT, tibia MAT and tibia red marrow (RM) were isolated from New Zealand White rabbits. Total RNA was isolated from iWAT, gWAT, pWAT, tibia MAT and tibia RM, and expression of the indicated transcripts was assessed by qPCR. Transcript expression was normalized to *Ppia* (cyclophilin A) and *Tbp* mRNA and is presented relative to the maximum expression of each transcript as mean ±SEM of 8-9 rabbits. Statistically significant differences between expression in tibia MAT and other tissues are indicated as follows: * = P <0.05; ** = P <0.01; *** = P <0.001. **(C)** Immunoblots and silver stain of conditioned media from WAT and MAT from one rabbit, representative of seven rabbits. Silver staining was used to assess total protein content of media. **(D)** Immunoblots of conditioned media from explants of scWAT and MAT isolated from patients 1 and 2. Lamin A/C was analyzed as an estimate of explant breakdown.

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Supplemental Tables

						p-value			
		WT	Wnt10b	WT CR	Wnt10b CR	WT vs Wnt10b	WT CR vs Wnt10b CR	WT vs WT CR	Wnt10b vs Wnt10b CR
Trabecular	TV (mm^3)	1.598	1.640	1.643	1.742	0.575	0.208	0.530	0.217
	BV (mm^3)	0.360	0.578	0.335	0.558	**0.001**	**0.002**	0.556	0.737
	BVF (%)	22.4%	35.3%	20.3%	31.9%	**0.002**	**0.000**	0.238	0.212
	Conn. Dens.	103.0	132.8	155.0	139.0	**0.010**	0.358	**0.012**	0.500
	SMI	1.622	0.768	1.781	0.789	**0.002**	**0.000**	0.199	0.927
	Tb.N	4.48	5.12	4.88	5.32	**0.003**	**0.043**	**0.039**	0.305
	Tb.Th	0.069	0.080	0.056	0.071	**0.001**	**0.000**	**0.000**	**0.005**
	Tb.Sp	0.213	0.162	0.199	0.167	**0.001**	**0.004**	0.208	0.575
	BMC	200.0	303.1	159.7	259.2	**0.001**	**0.000**	**0.015**	0.052
Cortical	TV (mm^3)	0.485	0.522	0.476	0.492	**0.007**	0.439	0.616	0.092
	BV (mm^3)	0.264	0.288	0.229	0.256	**0.049**	**0.038**	**0.016**	**0.007**
	BVF (%)	54.4%	55.2%	48.1%	52.0%	0.582	**0.002**	**0.000**	**0.050**
	BMC	688.2	687.6	601.1	640.5	0.979	**0.007**	**0.000**	**0.050**
Marrow Volume (mm^3)		8.02	7.25	8.42	7.87	**0.024**	0.098	0.230	**0.046**

Table S1, Related to Figure 2 – Characteristics of tibiae of female WT and Ocn-Wnt10b mice on control or CR diets, as assessed by µCT.