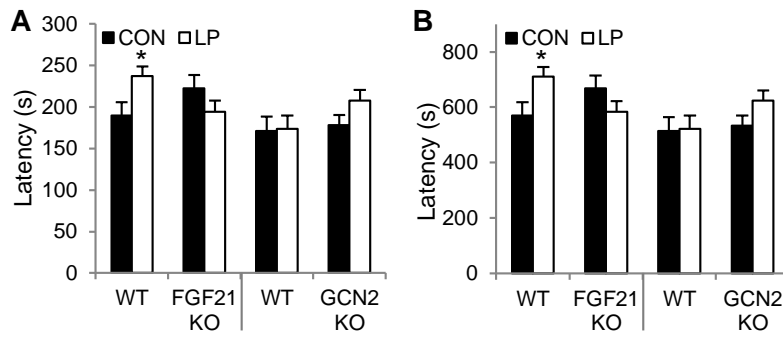
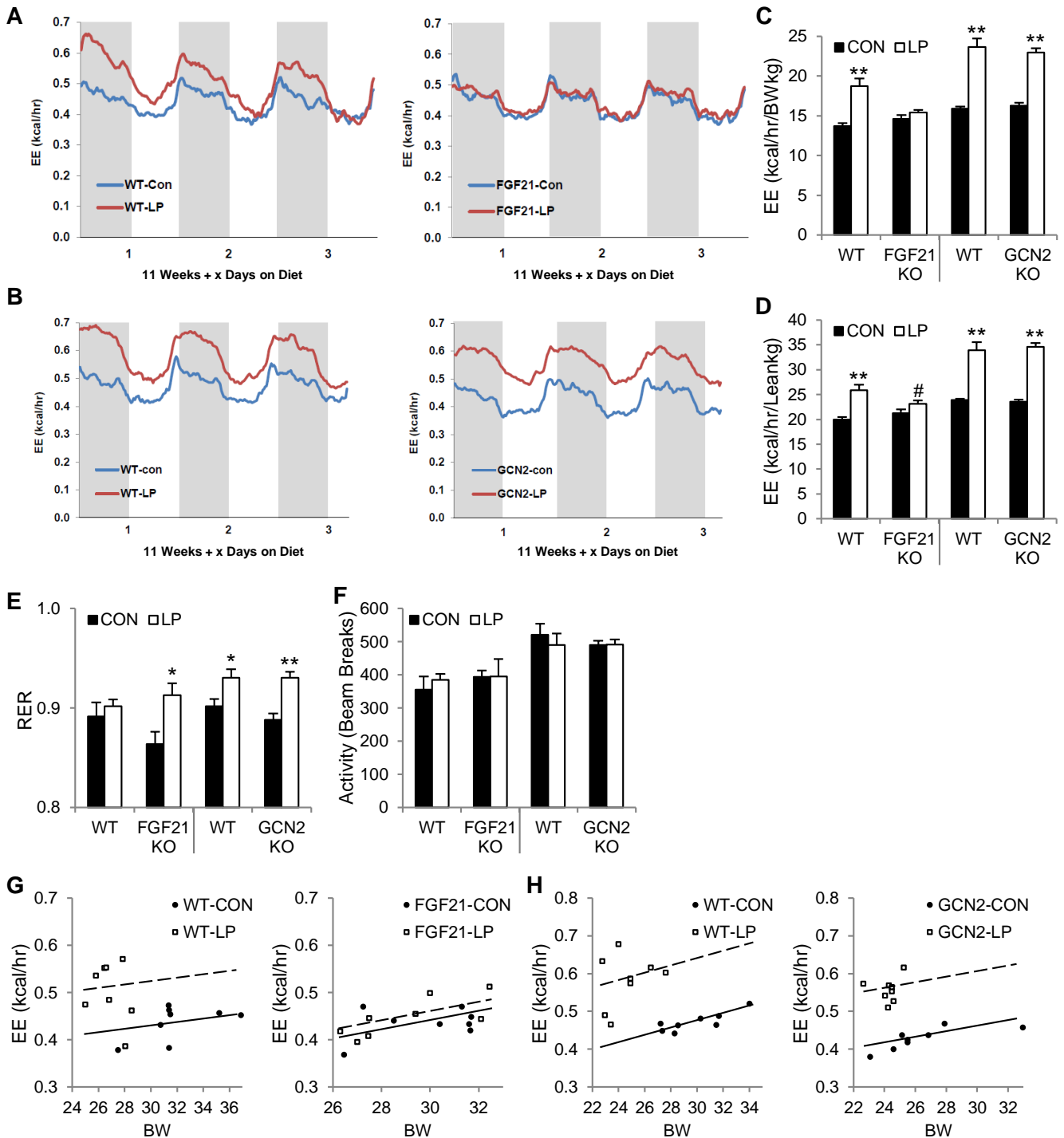


**Table S1. Related to Figure 1; Composition of diets**

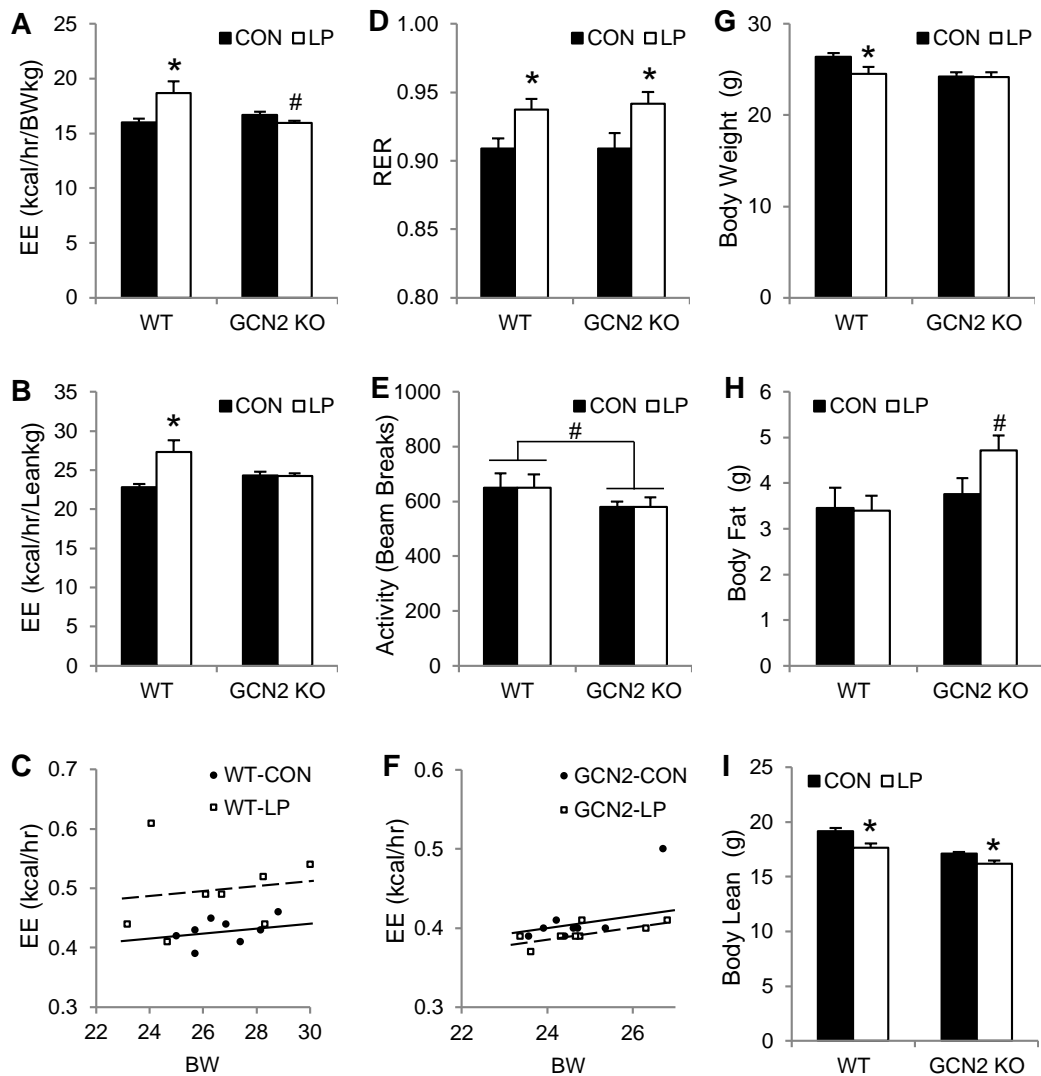
Product # <b>Ingredient (g)</b>	D11092301 <b>5% Casein</b>	D11051801 <b>20% Casein</b>
Casein	50	200
L-Cystine	0.75	3
Corn Starch	485	375.7
Maltodextrin 10	150	125
Sucrose	107	107
Cellulose	50	50
Soybean Oil	25	25
Lard	75	75
Mineral Mix S10022C	3.5	3.5
Calcium Carbonate	8.7	12.5
Calcium Phosphate, Dibasic	5.3	0
Potassium Citrate, 1 H <sub>2</sub> O	2.5	2.5
Potassium Phosphate, Monobasic	6.9	6.9
Sodium Chloride	2.6	2.6
Vitamin Mix V10037	10	10
Choline Bitartrate	2.5	2.5
FD&C Yellow Dye #5	0	0.05
FD&C Red Dye #40	0.05	0
FD&C Blue Dye #1	0	0
kcal/gram	4.1	4.1
Protein (gm%)	5	18
Carbohydrate (gm%)	76	62
Fat (gm%)	10	10
Protein (kcal%)	4	18
Carbohydrate (kcal%)	74	60
Fat (kcal%)	22	22



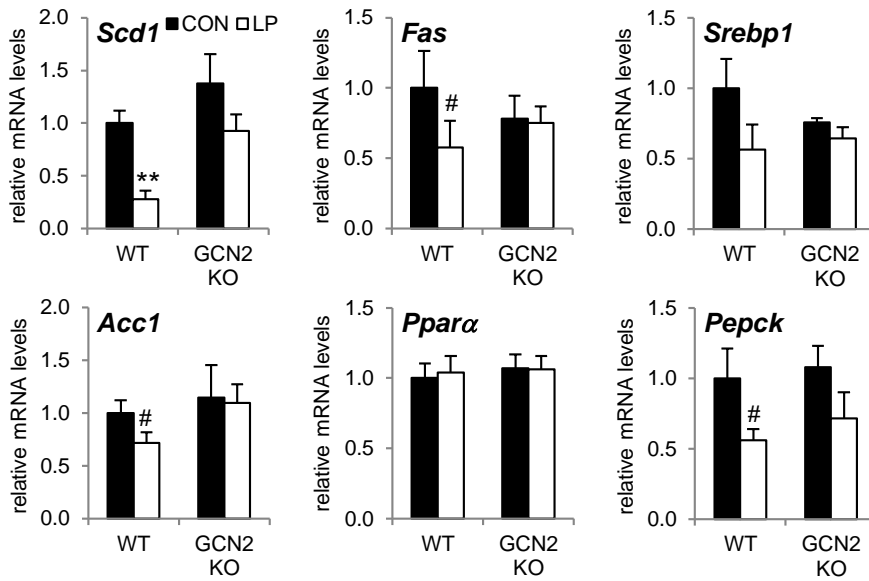
**Figure S1. Related to Figure 1 and Figure 5; Long-term low protein diet does not impair motor function and coordination.** Latency to fall (average trial 1 to 3, **A**; cumulative trial 1 to 3, **B**) on an accelerating rod was recorded in WT, *FGF21*-KO, and *GCN2*-KO mice on isocaloric control or LP diet for 20 weeks.  $n = 8-10/\text{group}$ ,  $*P < 0.05$ .



**Figure S2. Related to Figure 1 and Figure 5; Energy expenditure, RER, and physical activity in wild-type, *FGF21*-KO, and *GCN2*-KO mice on low protein for 11 weeks.** Energy expenditure (EE) in WT and *FGF21*-KO mice (A) and WT and *GCN2*-KO mice (B) consuming isocaloric control or LP diet for 11 weeks. Average energy expenditure (EE) normalized to body weight (C), EE normalized to lean mass (D), respiratory exchange ratio (RER, E), and activity (F) in WT, *FGF21*-KO, and *GCN2*-KO mice over 3 days period of EE measurement consuming isocaloric control or LP diet for 11 weeks. Scatter plots illustrating the relationship between body weight and EE, based on ANCOVA, in WT and *FGF21*-KO (G), and WT and *GCN2*-KO (H) mice.  $n = 8-10$ /group,  $\#0.1 > P > 0.05$ ; \* $P < 0.05$ ; \*\* $P < 0.01$ .



**Figure S3. Related to Figure 4; Energy expenditure, RER, physical activity, final body weight, and final body composition in wild-type and GCN2-KO mice on low protein for 2 weeks.** Average energy expenditure (EE) normalized to body weight (A), EE normalized to lean mass (B), respiratory exchange ratio (RER, D), and activity (E) in WT and GCN2-KO mice over days 5 to 7 consuming isocaloric control or LP diet. Scatter plots illustrating the relationship between body weight and EE, based on ANCOVA, in WT (C) and GCN2-KO (F) mice. Final body weight (G), final body fat (H), and final body lean mass (I).  $n = 8/\text{group}$ , # $0.1 > P > 0.05$ ; \* $P < 0.05$ .



**Figure S4. Related to Figure 2 and Figure 4; GCN2 is required for effects of low protein diet on hepatic metabolism.** WT and GCN2-KO were placed on isocaloric control or LP diet for 2 weeks and mRNA expression was measured in liver.  $n = 8/\text{group}$ , # $0.1 > P > 0.05$ ; \* $P < 0.05$ ; \*\* $P < 0.01$ .