SUPPLEMENTAL MATERIALS

Supplemental Table 1. Published citations describing the functional characterization of human *MC4R* mutations.

MUTATION	N Pts	Defect	Citation
Pathogenic Mutations			
Het R7H	1		Xiang Z, Proneth B, Dirain ML, Litherland SA, Haskell-Luevano C. Pharmacological characterization of 30 human melanocortin-4 receptor polymorphisms with the endogenous proopiomelanocortin-derived agonists, synthetic agonists, and the endogenous agouti-related protein antagonist. 2010; Biochemistry Jun 8;49(22):4583-600
Het T11A	1	Partial	Farooqi IS, Keogh JM, Yeo GS, Lank EJ, Cheetham T, O'Rahilly S. Clinical spectrum of obesity and mutations in the melanocortin 4 receptor gene. 2003; N Engl J Med 348:1085-1095
Het S127L	1	Partial/full	Valli-Jaakola K, Lipsanen-Nyman M, Oksanen L, Hollenberg AN, Kontula K, Bjørbaek C, Schalin-Jäntti C. Identification and characterization of melanocortin-4 receptor gene mutations in morbidly obese Finnish children and adults. 2004; J Clin Endocrinol Metab. Feb;89(2):940-5.
Het I137T	2		Gu W, Tu Z, Kleyn PW, Kissebah A, Duprat L, Lee J, Chin W, Maruti S, Deng N, Fisher SL, Franco LS, Burn P, Yagaloff KA, Nathan J, Heymsfield S, Albu J, Pi-Sunyer FX, Allison DB. 1999; Identification and functional analysis of novel human melanocortin-4

			receptor variants. Diabetes 48:635–639
Het Q156P	1		Farooqi IS, Keogh JM, Yeo GS, Lank EJ, Cheetham T, O'Rahilly S. Clinical spectrum of obesity and mutations in the melanocortin 4 receptor gene. 2003; N Engl J Med 348:1085-1095
Het A175T	1	Partial	Yeo GS, Lank EJ, Farooqi IS, Keogh J, Challis BG, O'Rahilly S. Mutations in the human melanocortin-4 receptor gene associated with severe familial obesity disrupts receptor function through multiple molecular mechanisms. 2003; Hum Mol Genet Mar 1;12(5):561-74 Farooqi IS, Keogh JM, Yeo GS, Lank EJ, Cheetham T, O'Rahilly S. Clinical spectrum of obesity and mutations in the melanocortin 4 receptor gene. 2003; N Engl J Med 348:1085-1095
Het 597-599 delCAT, T199TdelM	1		Farooqi, unpublished observation
Het F202L	1		Xiang Z, Proneth B, Dirain ML, Litherland SA, Haskell-Luevano C. Pharmacological characterization of 30 human melanocortin-4 receptor polymorphisms with the endogenous proopiomelanocortin-derived agonists, synthetic agonists, and the endogenous agouti-related protein antagonist. 2010; Biochemistry Jun 8;49(22):4583-600

Het N240S	1		Xiang Z, Proneth B, Dirain ML, Litherland SA, Haskell-Luevano C. Pharmacological characterization of 30 human melanocortin-4 receptor polymorphisms with the endogenous proopiomelanocortin-derived agonists, synthetic agonists, and the endogenous agouti-related protein antagonist. 2010; Biochemistry Jun 8;49(22):4583-600
Het G252S	1		Xiang Z, Pogozheva ID, Sorenson NB, Wilczynski AM, Holder JR, Litherland SA, Millard WJ, Mosberg HI, Haskell-Luevano C. Peptide and small molecules rescue the functional activity and agonist potency of dysfunctional human melanocortin-4 receptor polymorphisms. 2007; Biochemistry Jul 17;46(28):8273-87. Epub 2007 Jun 23
Het V253I	1	Partial	Yeo GS, Lank EJ, Farooqi IS, Keogh J, Challis BG, O'Rahilly S. Mutations in the human melanocortin-4 receptor gene associated with severe familial obesity disrupts receptor function through multiple molecular mechanisms. 2003; Hum Mol Genet Mar 1;12(5):561-74 Farooqi IS, Keogh JM, Yeo GS, Lank EJ, Cheetham T, O'Rahilly S. Clinical spectrum of obesity and mutations in the melanocortin 4 receptor gene. 2003; N Engl J Med 348:1085-1095
Het L263V	1		Farooqi, unpublished observation
Het P299H	1		Lubrano-Berthelier C, Durand E, Dubern B, Shapiro A, Danzin P, Weill J, Ferron C,

			Frougel P, Vaisse C. Intracellular retention is a common characteristic of childhood obesity-associated MC4R mutations. 2003; Hum Mol Genet Jan 15; 12(2): 145-53
Het R331K	1		Farooqi, unpublished observation
Het 333T-334A insA, p.T112N fs10X	1		Yeo GS, Lank EJ, Farooqi IS, Keogh J, Challis BG, O'Rahilly S. Mutations in the human melanocortin-4 receptor gene associated with severe familial obesity disrupts receptor function through multiple molecular mechanisms. 2003; Hum Mol Genet Mar 1;12(5):561-74
Non-pathogenic Mutations			
Het S4S	1		No change in protein sequence
Het V103I	20		Gu W, Tu Z, Kleyn PW, Kissebah A, Duprat L, Lee J, Chin W, Maruti S, Deng N, Fisher SL, Franco LS, Burn P, Yagaloff KA, Nathan J, Heymsfield S, Albu J, Pi-Sunyer FX, Allison DB. 1999; Identification and functional analysis of novel human melanocortin-4 receptor variants. Diabetes 48:635–639
Het A135A	1		No change in protein sequence
Het T178T	1		No change in protein sequence
Het I198I	5		No change in protein sequence
Het I251L	18		Farooqi IS, Keogh JM, Yeo GS, Lank EJ, Cheetham T, O'Rahilly S. Clinical spectrum of obesity and mutations in the melanocortin 4 receptor gene. 2003; N Engl J Med

			348:1085-1095
Het P272P	1		No change in protein sequence
Het G324G	2		No change in protein sequence

Supplemental Table 2. Percent excess body weight loss (%EBWL) and percent weight change (%WC) at different time points by type of *MC4R* mutation.

	Type of *MC4R* Mutation				
		Single Copy Pathogenic		Single Copy Non-Pathogenic	
	None	Weight Loss	P vs. None*	Weight Loss	P vs. None*
%EBWL at 1 year	71.5	71.1	0.68	69.8	0.51
%EBWL at 2 years	75.5	72.7	0.44	77.5	0.42
%EBWL at weight nadir	79.6	87.2	0.41	78.2	0.52
%WC at 1 year	34.2	35.4	0.69	32.5	0.63
%WC at 2 years	37.3	39.0	0.58	36.6	0.36
%WC at weight nadir	39.1	42.1	0.53	37.4	0.23

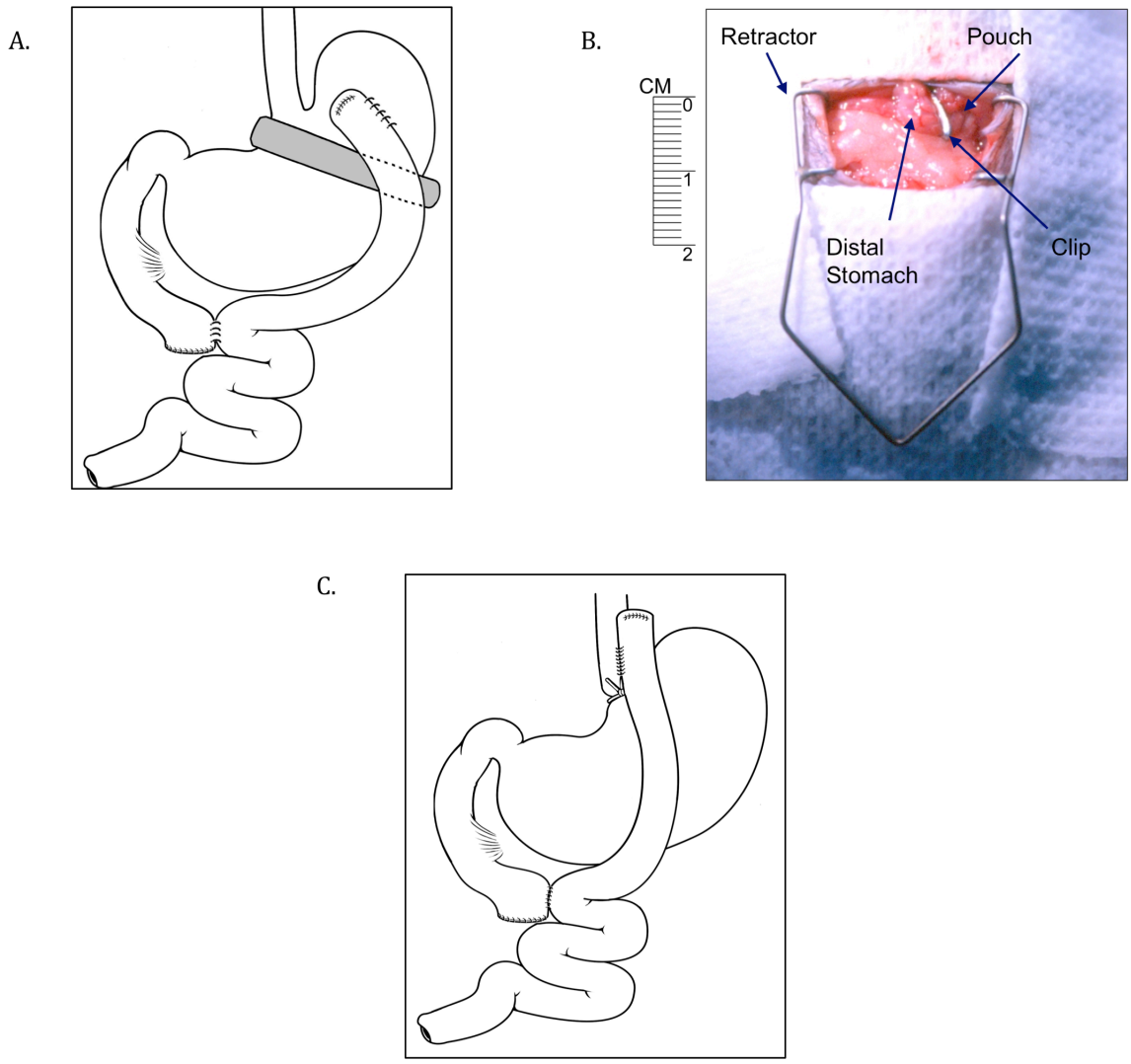
*P-values from multivariable regressions adjusted for age, sex, preoperative BMI, and type 2 diabetes status

Supplemental Table 3. Percent excess body weight loss (%EBWL) and measures of glucose homeostasis at different time points in patients heterozygous for the I251L or V103I mutation in *MC4R*.

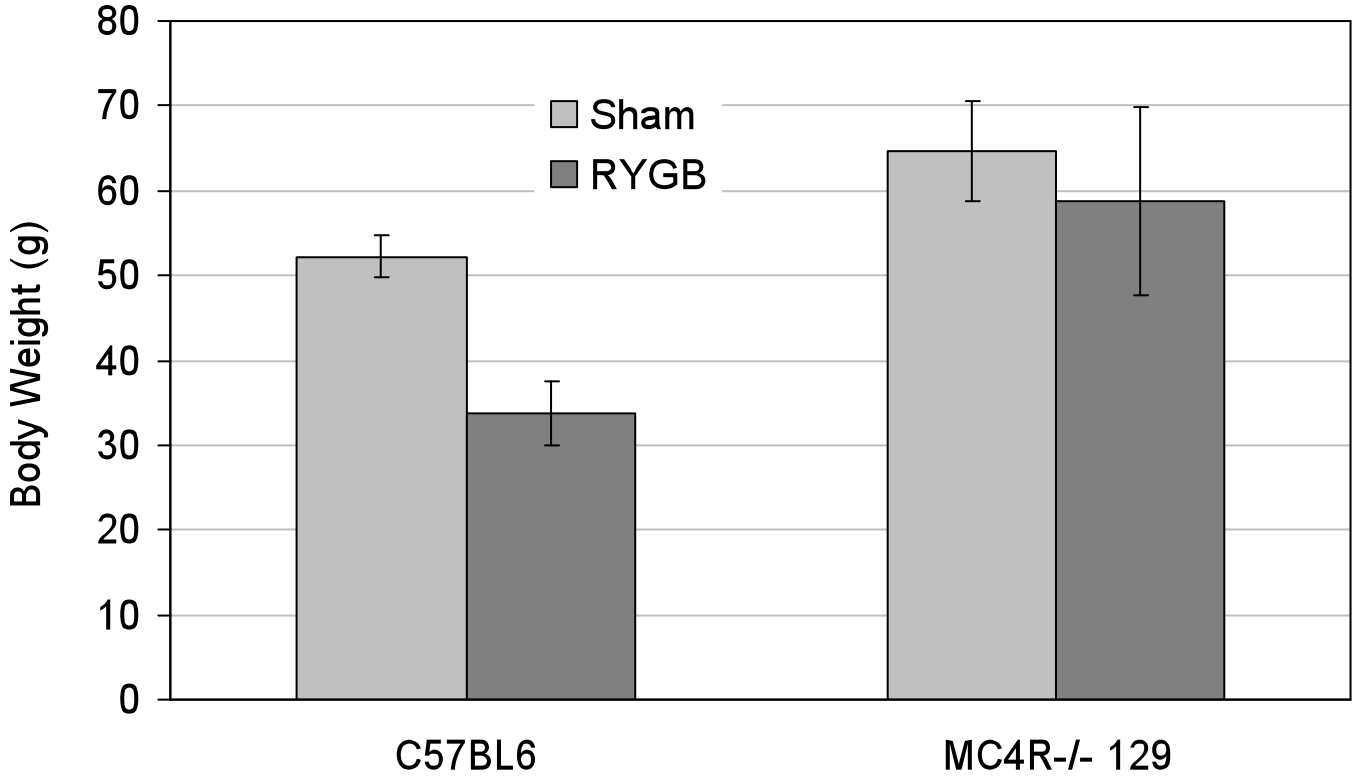
	MC4R Mutation				
		Single Copy I251L		Single Copy V103I	
	None	Characteristic	P vs. None	Characteristic	P vs. None
%EBWL at 1 year*	71.5	73.4	0.36	73.2	0.40
%EBWL at 2 years *	75.5	72.5	0.43	72.6	0.39
%EBWL at weight nadir*	79.6	73.7	0.27	81.4	0.56
Glucose (mg/dL) at baseline†	124.1	146.9	0.26	123.4	0.80
Glucose (mg/dL) at 1 year†	93.7	94.9	0.95	96.6	0.73
Insulin (μIU/mL) at baseline†	22.9	20.5	0.45	18.6	0.38
Insulin (μIU/mL) at 1 year†	7.2	5.2	0.24	5.3	0.32
HbA1c at baseline†	6.4	7.2	0.19	6.6	0.80
HbA1c at 1 year†	5.6	5.5	0.56	5.5	0.65

* P-values from multivariable regressions adjusted for age, sex, preoperative BMI and type 2 diabetes status

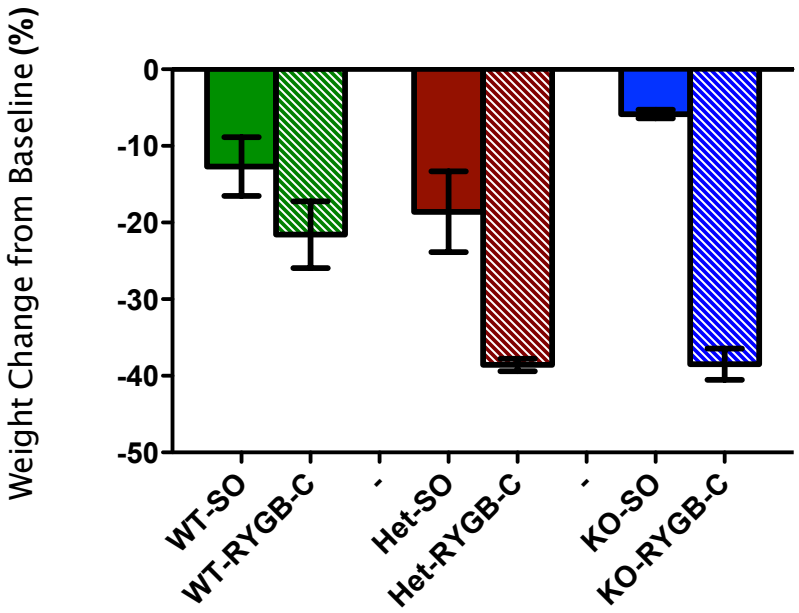
† P-values from multivariable regressions adjusted for age and sex



Supplemental Figure 1. **Gastric bypass in the mouse.** (A) Schematic and (B) photograph of the human-like RYGB (RYGB-H); the stomach is divided into a gastric pouch and distal stomach using a vascular clip, with a Roux limb length of approximately 6 cm, corresponding to 10-15% of the length of the small intestine. (C) Schematic of the complete gastric bypass (RYGB-C); the entire stomach is bypassed and the jejunum is connected to the esophagus, with an approximately 6 cm Roux limb.



Supplemental Figure 2. **Weight loss one year after RYGB-H in the mouse.** Body weight (grams) in WT mice on a C57BL/6 background (left) and *MC4R*−/− mice on a 129 background. Light gray bars represent sham-operated animals. Dark gray bars represent RYGB-operated animals. Error bars denote the standard error of the mean.



Supplemental Figure 3. **Percent weight change early after RYGB-C in wild-type (WT), MC4R $^{+/-}$ heterozygous (Het), and MC4R$^{-/-}$ null (KO) mice.** Change in body weight (percent) from postoperative week 0 to postoperative week 4 in RYGB-C and SO mice on a C57BL/6 background. Error bars denote the standard error of the mean.