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GLP-1R Responsiveness Predicts Individual Gastric Bypass Efficacy on Glucose Tolerance in Rats

Several bariatric operations are currently used to treat obesity and obesity-related comorbidities. These vary in efficacy, but most are more effective than current pharmaceutical treatments. Roux-en-Y gastric bypass (RYGB) produces substantial body weight (BW) loss and enhanced glucose tolerance, and is associated with increased secretion of the gut hormone glucagon-like peptide 1 (GLP-1). Given the success of GLP-1–based agents in lowering blood glucose levels and BW, we hypothesized that an individual sensitivity to GLP-1 receptor agonism could predict metabolic benefits of surgeries associated with increased GLP-1 secretion. One hundred ninety-seven high-fat diet–induced obese male Long-Evans rats were monitored for BW loss during exendin-4 (Ex4) administration. Stable populations of responders and nonresponders were identified based on Ex4-induced BW loss and GLP-1–induced improvements in glucose tolerance. Subpopulations of Ex4 extreme responders and nonresponders underwent RYGB surgery. After RYGB, responders and nonresponders showed similar BW loss compared with sham, but nonresponders retained impaired glucose tolerance. These data indicate that the GLP-1 response tests may predict some but not all of the improvements observed after RYGB. These findings present an opportunity to optimize the use of bariatric surgery based on an improved understanding of GLP-1 biology and suggest an opportunity for a more personalized therapeutic approach to the metabolic syndrome.

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Both developed and developing countries have seen increased rates of obesity over the last 30 years that has been paralleled by an unprecedented increase in the incidence of metabolic disturbances such as hypertension, dyslipidemia, and type 2 diabetes (T2D). The World Health Organization has described obesity as the greatest current threat to human health, based upon its association with numerous serious comorbidities (1,2). Conventional therapies, such as dietary and lifestyle changes, have proven to be largely ineffective (3). Furthermore, current pharmacotherapies are only mildly efficacious (4). At this time, surgical intervention stands alone in the context of sustained treatment for severe obesity (5).

Among these surgeries, adjustable gastric banding, vertical sleeve gastrectomy (VSG), and Roux-en-Y gastric bypass (RYGB) have become the most prevalent procedures (6). RYGB, the current gold standard, results in

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a loss of 60–70% excess body weight (BW) on average (7) and is associated with a surprisingly rapid resolution of T2D in 62–80% of patients (8,9). However, the precise mechanisms that underlie the robust effects of RYGB are poorly understood.

While a portion of the antidiabetic effects of RYGB are secondary to reduced BW, an altered gut hormone profile after RYGB is also likely to be a contributing factor (10). Clinical studies have identified substantial changes in multiple circulating factors, including glucagon-like peptide 1 (GLP-1), after RYGB (11). GLP-1 secretion is greatly elevated after RYGB, suggesting that it may function as a modulator of both BW and glucose homeostasis (12). Given that GLP-1 receptor (GLP-1R) signaling is a target for several approved diabetes therapies, modulation of the GLP-1 system may improve surgical therapies for weight loss and diabetes.

In this study, we investigated the hypothesis that GLP-1R agonists can be used to predict the efficacy of RYGB in a rat model of diet-induced obesity (DIO). We here show that sensitivity to GLP-1R agonists functions as a novel predictive biomarker for changes in glucose tolerance but not BW, food intake, or fat mass after RYGB. Our results suggest that a more personalized approach to bariatric surgery may be used to optimize the likelihood of beneficial effects such as improved glucose tolerance, while preventing unnecessary surgical intervention and side effect risk based on the use of a GLP-1 challenge as a novel predictive biomarker.

RESEARCH DESIGN AND METHODS

Animals

Male, Long-Evans rats (n = 197; 250–300 g) obtained from Harlan Laboratories (Indianapolis, IN) were individually housed and maintained on a 12-h light/dark cycle (lights off at 7:00 P.M.) at 25°C and 50–60% humidity. Rats were provided ad libitum access to water and a high-fat butter diet (HFD; 4.54 kcal/g, 41% fat; Research Diets, New Brunswick, NJ) previously shown to produce DIO and metabolic impairments (13). GLP-1R sensitivity studies were initiated after 8 weeks of high-fat feeding. All procedures for animal use were approved by the University of Cincinnati Institutional Animal Care and Use Committee.

Selection of Responders and Nonresponders for RYGB

Long-Evans rats (n = 197) were fed an HFD for 8 weeks. Upon reaching DIO (492.1 ± 2.6 g), all rats were administered exendin-4 (Ex4) (50 μg/kg/day i.p.) for 4 days. BW, food intake, and ad libitum blood glucose were measured as surrogates for GLP-1R agonist sensitivity. Response to GLP-1 challenge during an intraperitoneal glucose tolerance test (GTT) was also assessed. Rats were ranked first by BW and then by GLP-1 response. The 25 most responsive rats and 25 least responsive rats were then subjected to RYGB surgery and characterized over the next 130 days. Ten rats of intermediate response were administered a sham procedure to serve as controls to the RYGB rats.

RYGB

RYGB was performed in anesthetized rats (isoflurane) as previously described (14). Briefly, rats had a laparotomy and transection of the jejunum 30 cm from the ligament of Treitz, establishing two distinct jejunal limbs. At the level of the distal limb, a small longitudinal incision was made 10 cm distal to its proximal end and the proximal limb was anastomosed to it, end-to-side, with a running 7 × 0 Vicryl absorbable suture (Ethicon Endo-Surgery, Somerville, NJ) creating the "Y." The gastric pouch was then made using an ETS-Flex 35 mm staple gun (Ethicon Endo-Surgery) to resect most of the fundus and partition the gastric remnant across its waist, thereby creating a proximal gastric pouch ~10% of the original gastric size. After incision of the gastric pouch that spared its vascular architecture, the efferent jejunal limb (alimentary limb) was connected to the gastric pouch creating a side-to-end gastro-jejunal anastomosis with a running 8–0 prolene, nonabsorbable suture (Ethicon Endo-Surgery). The stomach and jejunum were then reintegrated into the peritoneal cavity, and the abdominal wall was closed in layers. For the sham surgery the jejunum was transected 30 cm distal to the ligament of Treitz and reanastomosed end-to-end with a running 7–0 Vicryl suture. Survival after this procedure yielded 7 sham, 12 responder, and 13 nonresponder rats.

Postoperative Care

The HFD was replaced with Ensure Plus liquid diet (1.41 kcal/g, 29% fat; Abbott Nutrition, Columbus, OH) 24 h prior to surgery. A liquid diet was continued for 120 h and then replaced with an HFD. Subcutaneous injections of Metacam (0.25 mg/100 g BW once daily for 4 days), gentamicin (0.8 mg/100 g BW on the day of surgery), Buprenex (0.3 mL 2 times per day for 5 days), and warm saline (10 and 5 mL 2 times per day for days 0–3 and 4–5, respectively) were given to all postoperative rats.

Body Composition Measurements

Echo magnetic resonance imaging whole-body composition analysis (fat and lean mass) was performed on all rats on postoperative days 28, 59, and 90 (EchoMRI, Houston, TX).

Peptides

Ex4 was obtained from American Peptide (Sunnyvale, CA). GLP-1 (7–36)-amide was obtained from PolyPeptide Group (San Diego, CA).

Insulin, Glucose, and Mixed-Meal Tolerance Tests

Insulin and glucose tolerance tests were performed by intraperitoneal injection of human insulin (1 unit/kg, 20% w/v D-glucose, in 0.9% w/v saline; Lilly Humalog) or glucose (2 g/kg, 20% w/v D-glucose, in 0.9% w/v saline; Sigma) after 6-h (GTT) or 16-h (insulin tolerance test [ITT]) fast. For mixed-meal tolerance tests, rats were

gavaged with 3 mL Ensure Plus Liquid diet. Blood was collected into tubes containing 20 μL antiproteolytic cocktail (4.65 g EDTA, 92 mg aprotinin, and 40,000 units heparin in 50 mL saline). Active and total GLP-1 (7–36) were measured by electrochemiluminescence assay (Meso Scale Discovery, Gaithersburg, MD). Blood samples were collected before and 15, 30, 60, 90, and 120 min after challenge. Blood glucose was determined by TheraSense Freestyle Glucometer.

Statistical Analyses

All data are represented as mean and SEM. Nonlinear correlation with a Gaussian fit was used to determine normal distribution of Ex4-stimulated effects throughout the population. One-way and two-way ANOVA with Bonferroni multiple-comparison post-test, or Student *t* test were performed to assess effects between study groups where appropriate. In all cases, statistical significance was assumed when $P < 0.05$ using GraphPad Prism software (San Diego, CA).

RESULTS

Diversity in Response to GLP-1R Agonism

GLP-1R signaling enhances glucose metabolism and decreases BW in both humans (15) and rodent models (15). However, whether the response to GLP-1R agonists varies among different subpopulations has yet to be systematically examined. To test this hypothesis, we assessed the sensitivity of an outbred population of DIO rats to Ex4 injections. Ex4 treatment stimulated BW loss (Fig. 1*A* and *B*) that followed a Gaussian distribution (Fig. 1*C* and *D*). Consistent with this effect on BW, food intake, final (day 4) blood glucose, and the change in ad libitum blood glucose (days 0–4) were marked by considerable variability within the population (Fig. 2*A*, *C*, and *E*).

The diversity within this population of outbred rats suggested a natural variation in individual responsiveness to GLP-1R agonism. Indeed, comparison of the upper and lower 15th percentiles for BW loss revealed a marked difference in the response to Ex4 treatment. Specifically, loss of BW in the upper 15th percentile (responders) was found to be much greater (7.05 ± 0.15%) than that observed in the lower 15th percentile (nonresponders, 1.68 ± 0.010%) or the total population (4.32 ± 0.12%) (Fig. 1*A*). To test the hypothesis that there is an innate variation in sensitivity to GLP-1R agonism; we analyzed food intake, glucose tolerance, and ad libitum blood glucose in subpopulations stratified according to change in BW. Food intake (Fig. 2*B*) and blood glucose (Fig. 2*D* and *F*) were decreased in responders compared with either the total population or nonresponders. Conversely, nonresponders had increased food intake and elevated ad libitum blood glucose compared with the other two populations (Fig. 2). After a 10-day washout period, Ex4 treatment was repeated in all rats to ensure that the phenomena

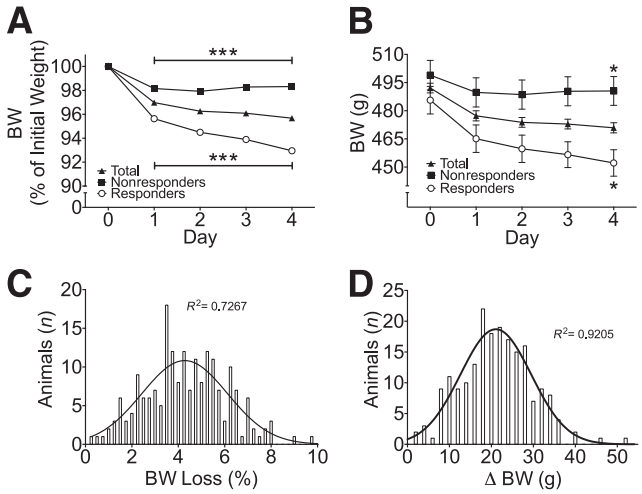

Figure 1—Relative (*A*) and absolute (*B*) BW response to Ex4 treatment in DIO rats. $n = 197$ for total (triangles) and $n = 30$ for both responder (circles) and nonresponder (squares) groups. Data are represented as the mean ± SEM. *$P < 0.05$, ***$P < 0.001$. Population distribution of Ex4 response during the 5-day treatment as a function of the percentage of BW loss (*C*) or BW change (*D*). Data are represented as the number of rats per bin. Solid line and R^2 denote nonlinear Gaussian fit of the data. All data obtained in male Long-Evans rats maintained on an HFD (40% butter fat) for 8 weeks.

were reproducible. As in the initial screen, rats were stratified into populations of responders and nonresponders based on BW loss (Supplementary Fig. 1*a*). Rats identified in the second screen were similar to those identified in the first study, with no crossover of groups observed. Decreased BW in the responder groups was associated with decreased ad libitum blood glucose, whereas in the nonresponder group there was a less pronounced weight loss and elevated blood glucose level (Supplementary Fig. 1*b* and *c*). However, unlike the initial screen, no significant difference in food intake was observed between the groups (Supplementary Fig. 1*d*).

To ensure that the biological response to Ex4 was based on GLP-1R signaling and not an off-target effect of the drug, glucose tolerance was assessed in the presence and absence of exogenous GLP-1 (7–36). After a 10-day washout, all rats were fasted for 6 h and then administered 2 g/kg i.p. glucose. As with other surrogates of GLP-1R signaling, we found that in regard to innate glucose tolerance there was considerable variability within the population (Fig. 3*A*). While fasting glycemia (Fig. 3*B*) was similar in all groups, responders exhibited greater glucose tolerance at 15 and 30 min ($P < 0.05$) (Fig. 3*C*) and a reduced area under the curve (AUC) ($P < 0.01$) (Fig. 3*D*). In the presence of exogenous GLP-1 (7–36), responders displayed a greater enhancement of glucose clearance (Fig. 3*C*) and a reduced AUC ($P < 0.01$) (Fig. 4*D*). Intriguingly, no effect of the GLP-1 was evident in the AUC of nonresponders.

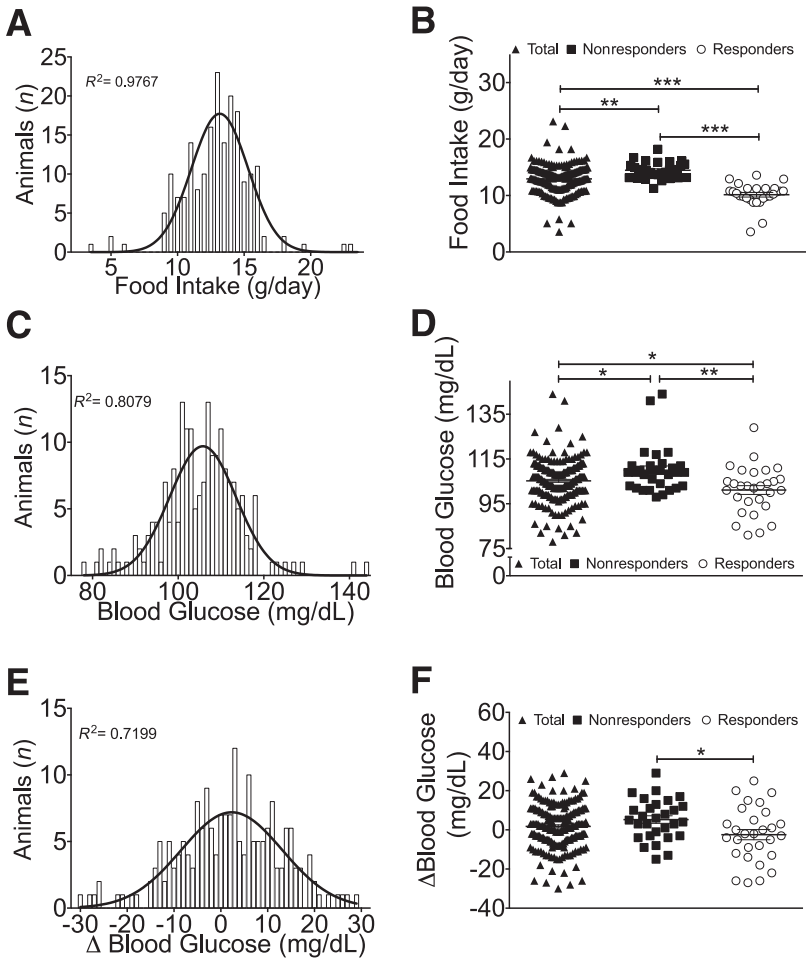

Figure 2—Population distribution of Ex4 response during the 5-day treatment as a function of food intake (*A*), day 4 ad libitum blood glucose levels (*C*), or days 0–4 change in ad libitum blood glucose (*E*). Data are represented as the number of rats per bin. Solid line and R^2 denote nonlinear Gaussian fit of the data. Food intake (*B*), final ad libitum blood glucose levels (*D*), and ad libitum blood glucose change (*F*) of responders and nonresponders identified by BW. n = 197 for total (triangles) and n = 30 for both responder (circles) and nonresponder (squares) groups. Data are represented as the mean ± SEM. *$P < 0.05$, **$P < 0.01$, ***$P < 0.001$.

GLP-1 secretion by an oral carbohydrate challenge is attenuated in obese individuals (16); thus, one possible explanation for the difference in sensitivity to GLP-1R agonists may be attributed to the animal's initial adiposity. To better understand the contribution of initial adiposity to Ex4 response, we analyzed the same variables according to the animal's BW prior to Ex4 treatment. We observed a significant correlation between BW loss and initial BW in the total population ($P < 0.01$, Supplementary Fig. 2*a*). However, this effect was lost when the comparison was restricted to either the responder or nonresponder groups (Supplementary Fig. 2*a*). Furthermore, animals from both the responder and nonresponder groups had initial BWs across the range of values observed in the total population (Supplementary Fig. 2*a*), suggesting that initial BW did not determine the BW response to Ex4. Food intake was also correlated with initial BW ($P < 0.01$, Supplementary Fig. 2*b*), such that animals with the greatest food intake also had the highest initial BW. This correlation was still observed when the analysis was restricted to the nonresponder group ($P < 0.05$) (Supplementary Fig. 2*b*). However, when analysis was restricted to the responder group, there was a slight but significant negative correlation between food-intake response and initial BW ($P < 0.05$) (Supplementary Fig. 2*b*), suggesting that the food-intake response may drive the overall BW response. Glycemic response to Ex4 was not correlated with initial BW in responders, nonresponders, or the population as a whole (Supplementary Fig. 2*c*). Taken together, these data suggest that there is an interindividual variation in innate GLP-1R sensitivity that is not explained by initial adiposity.

Effects of RYGB on BW, Food Intake, and Fat Mass

To test the hypothesis that sensitivity to GLP-1R agonists could be used to predict the outcome of RYGB, we compared the effects of RYGB in animals classed as

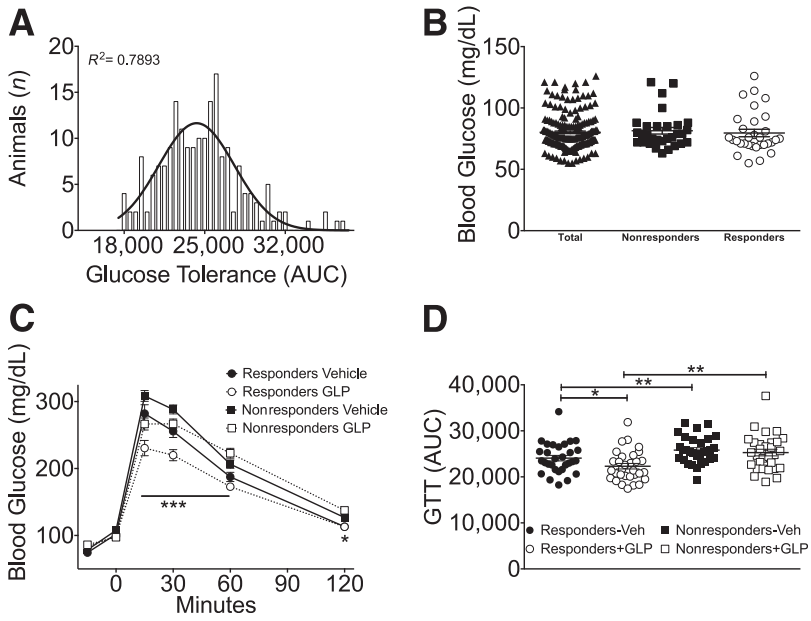

Figure 3—*A*: Population distribution (n = 197) as a function of glucose tolerance (AUC during intraperitoneal GTT). Data are represented as the number of rats per bin. Solid line and R^2 denote nonlinear Gaussian fit of the data. Fasting blood glucose before (*B*), and glucose excursion (*C*) and AUC (*D*) after glucose challenge in the presence (open symbols) or absence (closed symbols) of GLP-1 in responders and nonresponders identified by BW during both phases of the Ex4 response study. All data obtained in male Long-Evans rats maintained on an HFD (40% butter fat) for 12 weeks. n = 197 for total (triangles) and n = 30 for both responder (circles) and nonresponder (squares) groups. All data are represented as the mean ± SEM. *P < 0.05, **P < 0.01, ***P < 0.001. Veh, vehicle.

responders and nonresponders to Ex4. RYGB stimulated a characteristic decrease in BW in both responder and nonresponder groups compared with sham-operated control animals (Fig. 4*A*). Maximum weight loss was observed at day 13 in responders (19.1 ± 1.6%) and day 22 in nonresponders (20.8 ± 2.5%). In addition to this early weight loss, RYGB-treated animals displayed a sustained weight loss throughout the study, with neither group returning to its preoperative BW (Fig. 4*A*). A slight decrease in BW (6.5 ± 1.1%) was observed in sham-treated animals over the first 6 days, followed by a rapid and sustained rebound (Fig. 4*A*). Body composition assessed on days 28, 59, and 90 confirmed that the loss of BW was mirrored by similar effects on fat mass, with no difference between responder and nonresponder groups (Fig. 4*B*). Lean mass was initially reduced in the nonresponders ($P < 0.05$), with similar values observed at later time points in all groups (Fig. 4*C*). Analysis of food intake over the first 48 days of the study revealed reduced food intake in RYGB animals compared with sham controls. As with RYGB-induced loss of BW, no evidence of differential food intake was observed between the responder and nonresponder groups (Fig. 4*D*). Fasting-induced 24 h food intake was similar in responder-RYGB and sham groups, with a significant increase observed in the nonresponder-RYGB rats (Fig. 4*E*). Taken together, these data suggest that, regardless of prior GLP-1R sensitivity, RYGB stimulates loss of body and fat mass while reducing cumulative food intake in rats. Sensitivity to GLP-1R agonists therefore does not predict individual energy balance after RYGB surgery.

Effects of RYGB on Glucose Metabolism

In addition to its effects on food intake, BW, and fat mass; GLP-1 stimulates insulin secretion and increases insulin-independent glucose disposal (17), leading to subsequent enhancement of glucose homeostasis. We therefore addressed whether glucose homeostasis differed in responders and nonresponders after RYGB. When glucose homeostasis was challenged via glucose bolus (2 g/kg i.p.) 3 months after RYGB, we found that responders exhibited an enhanced glucose tolerance (Fig. 4*F*) and reduced AUC (Table 1) when compared with nonresponders. These rats displayed a similar trend for a reduced AUC (P = 0.09), when compared with shams. It should be noted that this enhanced tolerance was independent of BW or fat mass, as these parameters were identical in the two groups. Insulin-stimulated glucose clearance was assessed via intraperitoneal ITT (1 unit/kg) after a 16-h fast (Fig. 4*G*). This assessment elucidated a similar glucose clearance (Kd) over the initial 30 min in both RYGB groups that was enhanced compared with that of sham controls (Table 1).

To evaluate humoral regulation in the postprandial state, we analyzed plasma insulin, glucagon, and GLP-1 levels before and 30 min after a mixed-meal challenge.

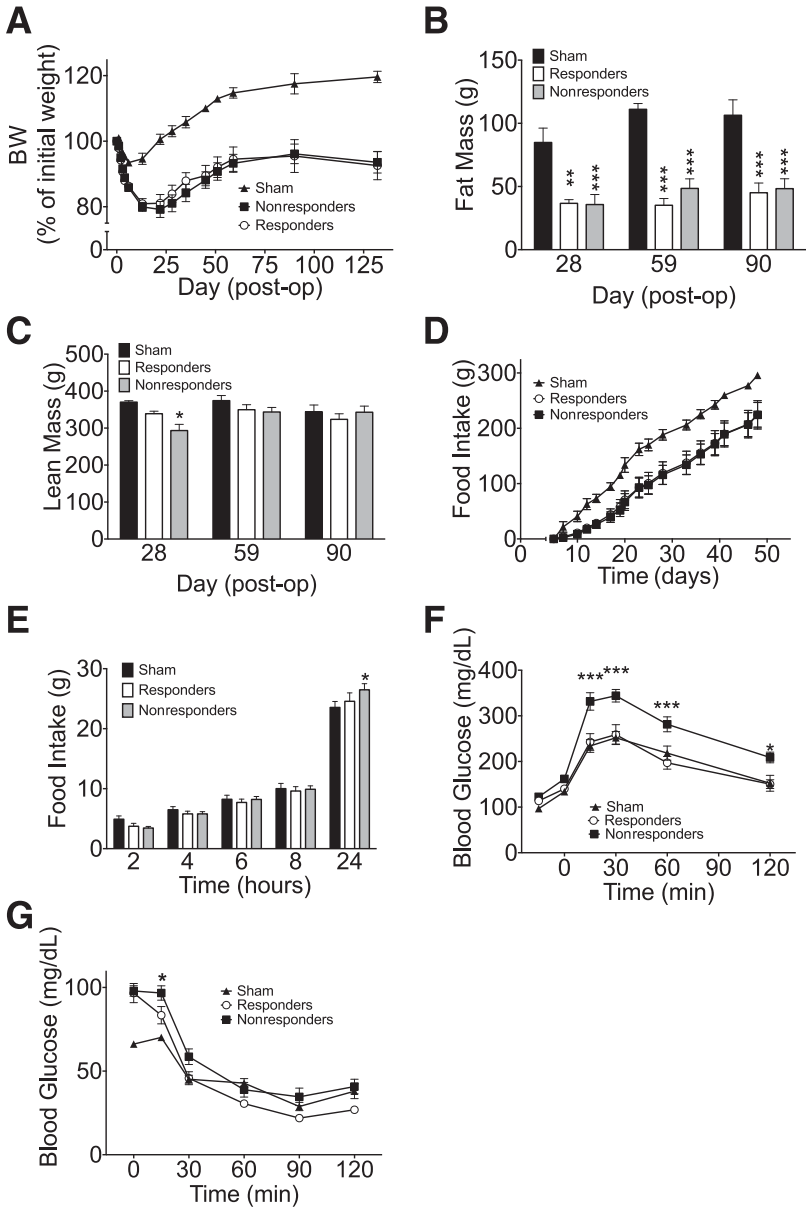

Figure 4—Relative BW loss (*A*), fat mass (*B*), lean mass (*C*), and food intake (*D*) after RYGB in previously identified responders and nonresponders. *E*: The 24-h fasting–induced food intake 19 weeks after RYGB in previously identified responders and nonresponders. Blood glucose excursion during intraperitoneal GTT (*F*) and intraperitoneal ITT (*G*) 12 and 19 weeks after RYGB in responders and nonresponders. $n = 7$ for sham (triangles), $n = 10$ for responder (circles), and $n = 13$ for nonresponder (squares) groups. All data are represented as the mean ± SEM. $^*P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$. post-op, postoperative.

At the time of the challenge, fasting glycemia was reduced in responders compared with nonresponders with similar levels of insulin (Table 1). Insulin resistance determined by homeostasis model assessment of insulin resistance tended to be lower ($P = 0.0775$) in responders compared with sham or nonresponder rats (Table 1). Total plasma GLP-1 levels were elevated after the challenge, although no difference was observed between the responder and nonresponder groups (Table 1). However, we observed greater levels of active GLP-1 30 min after the challenge (Table 1). This increase in plasma GLP-1 levels was associated with elevation of plasma insulin levels (Table 1). In addition, plasma glucagon was increased in both responder and nonresponder groups (Table 1). Taken together, these findings are consistent with the hypothesis that the level of GLP-1R sensitivity prior to surgical intervention predicts glucose tolerance after RYGB and suggests potential use of an Ex4 challenge test as a biomarker to predict metabolic benefits resulting from bariatric surgery. Furthermore, these data suggest that the enhancements in glucose metabolism are associated

Table 1—Circulating factors in RYGB rats

Factors	Sham	Responders	Nonresponders
Fasting insulin (pg/mL)	3,734.8 ± 959.6	2,523.9 ± 365.7	2,949.2 ± 344.1
Postprandial insulin (pg/mL)	4,294.8 ± 904.7	13,487.6 ± 4,318.4	7,688.5 ± 969.0
Fasting GLP-1 (pg/mL)	ND	16.3 ± 5.2	12.1 ± 3.3
Postprandial GLP-1 (pg/mL)	4.3 ± 2.7	74.3 ± 29.0*	56.2 ± 15.1
Fasting GLP-1^{active} (pg/mL)	10.2 ± 2.1	10.0 ± 2.9	6.3 ± 0.2
Postprandial GLP-1^{active} (pg/mL)	9.6 ± 1.8	26.6 ± 10.3†	9.9 ± 2.0
Fasting glucose (mg/dL)	96.6 ± 3.8	113.8 ± 3.1*	128.9 ± 4.4†,‡
HOMA-IR	0.91 ± 0.26	0.71 ± 0.11	0.95 ± 0.13
Fasting glucagon (pg/mL)	97.9 ± 31.9	78.1 ± 38.5	26.7 ± 5.3
Postprandial glucagon (pg/mL)	92.0 ± 31.6	162.0 ± 43.5	130.5 ± 27.0
ipGTT AUC	13,252 ± 1,530	10,464 ± 1,187	17,550 ± 1,625§
ipITT K_{d30} (mg/mL/min)	0.70 ± 0.06	1.70 ± 0.12‡	1.31 ± 0.16*

Data on fasting and postprandial plasma were collected 20 weeks after surgery in response to a mixed-meal challenge. All data are expressed as the mean ± SEM (n = 7–11), and were obtained from male Long-Evans rats maintained on an HFD (40% butter fat). ipGTT, intraperitoneal GTT; ipITT, intraperitoneal ITT; K_{d30}, rate of glucose disappearance calculated over the initial 30 min of ITT. *$P <$ 0.05 compared with sham. †$P < 0.05$ compared with responders. ‡$P < 0.01$ compared with sham via ANOVA. §$P < 0.01$ compared with responders.

with increased levels of active GLP-1 and subsequent plasma insulin levels in rats identified as responders.

DISCUSSION

This study shows that responsiveness to a GLP-1R agonist predicts populations of individuals who will not display some of the beneficial metabolic effects of RYGB surgery. Our findings raise the hope of a novel, tailored, and ultimately personalized, approach to the treatment of obesity. They also suggest that the prediction of treatment outcome and combinations of pharmacological and surgical interventions may be used to achieve maximum efficacy with minimal invasiveness in individual patients.

Current medical and lifestyle interventions offer modest efficacy in the treatment of obesity (3,4). Bariatric interventions, on the other hand, have proven to be highly efficacious and frequently carry the beneficial side effect of T2D resolution (8). However, not all of the currently available surgical options are equally effective (6). RYGB is the current gold standard in terms of both weight loss and resolution of T2D, and is associated with a similar mortality risk compared with adjustable gastric banding or VSG. However, 20–50% of patients fail to lose substantial weight, and 20–40% fail to achieve diabetes resolution after undergoing RYGB (18). Thus, identifying predictors of treatment outcome would reduce unnecessarily invasive interventions in patients who will not benefit from the surgery.

We hypothesized that the superior efficacy of RYGB is due to the humoral reprogramming observed after RYGB (19). Circulating levels of GLP-1, and therapies based upon those levels, modulate food intake, glucose homeostasis, BW (20), and fat mass (21). Furthermore, GLP-1–based therapies are unaffected by diabetic state (22). Thus, the variation in metabolic outcome after RYGB might involve variations in GLP-1 signaling that could be predicted by sensitivity to GLP-1R agonists before the procedure.

The hypothesis that GLP-1 response may hold a predictive value was based upon the observation that a sufficient and sustained degree of variability in response was clearly detectable within our model population. To our knowledge, this finding of such a variable response to GLP-1R agonism, across different agonist ligands and measured end points, is the first such in vivo example. Specifically, we found that subpopulations, which were either responsive or nonresponsive to Ex4-stimulated weight loss, could be identified from a large population of outbred rats. It is important to note that the response in BW change was associated with similar effects on food intake and glycemia, suggesting that the variable sensitivity was pervasive to some, if not all, GLP-1 actions. While we were unable to observe a significant increase in glucose tolerance in our responder cohort of RYGB rats, this may be attributed to our selection of sham animals. These sham animals were selected from rats of intermediate GLP-1R response and therefore were not matched to our specific responder and nonresponder groups. Importantly, we were able to identify a significant deficit in the glucose homeostasis of nonresponders. While the paradigm used here does not allow observation of clear differences between responder rats and sham rats, the data suggest that an individual's response to

GLP-1R agonism offers a novel, and currently the only, predictor of metabolic outcome after RYGB.

These observations are consistent with the finding that a single-nucleotide polymorphism of the GLP-1R (human GLP-1R Met^{149}) is associated with reduced ligand-receptor affinity and intracellular signaling (23). Likewise, the exchange of a single amino acid (E68) in GLP-1R markedly reduces receptor activity (24). Thus, it is tempting to speculate that GLP-1 responsive and nonresponsive subpopulations can also be identified in humans and that responsiveness to GLP-1R agonists would be similarly predictive.

Increased circulating levels of GLP-1 combined with enhanced insulin secretion are often suggested to play a role in the improved glucose metabolism observed after RYGB (19). Likewise, elevated plasma levels of GLP-1 have been described after VSG (14), ileal transposition (25), and duodenal-jejunal bypass (26). Consistent with the observed incretin hypersecretion, all of these procedures have been found to improve glucose tolerance (14,25,26). A recent study by our group has suggested that GLP-1R signaling is not necessary for the beneficial effects of VSG on food intake, BW, dietary fat preference, and glucose tolerance (27). While this finding leads us to reconsider the role of GLP-1 as the primary mediator of the metabolic benefits of bariatric surgery, its relevance to this study are not as clear. Several caveats prevent us from drawing a straight line between these surgeries. VSG and RYGB are not the same procedure, with the primary difference being the nutrient exclusion that occurs in RYGB. The study conducted by Wilson-Pérez et al. (27) used mice, whereas we have chosen the rat model. Thus, it is possible that a species-dependent effect is undermining the interpretation. Furthermore, the GLP-1–deficient mice used in this study develop in the absence of this important neuroendocrine factor. Thus, compensatory effects due to this deficiency (i.e., increased levels of GLP-1 and glucose-dependent insulinotropic peptide) may cloud the interpretation of these results. Moreover, an earlier report from our own group using the GLP-1R antagonist Ex9 demonstrated that surgery-induced improvements in glucose homeostasis are attenuated when GLP-1 action is blocked. Together, we interpret these findings to suggest that GLP-1 in not necessary for the effects of VSG on food intake, BW, dietary fat preference, and glucose tolerance. However, its sufficiency to drive these outcomes cannot be concluded from this study design. Thus, although the role of GLP-1 as the primary mediator of pleiotropic metabolic benefits of bariatric surgery requires reconsideration, its effect in the current study suggests that it may be predictive of outcome after RYGB.

While the work presented here awaits confirmation in humans, it undoubtedly offers translational potential for clinical diabetologists and obesity physicians observing therapy responder and nonresponder subpopulations. As differences in responsiveness have been reported for bariatric surgery (7), as well as incretin therapy (23,28), our findings provide one possible path toward more personalized treatment in metabolic medicine. Incretin challenge tests may allow us to begin individually stratifying risk when choosing the correct therapy for the metabolic syndrome.

In summary, our study in a rodent model of DIO indicates that distinct subpopulations exhibit differential responses to GLP-1R agonism and that these responses predict glucose metabolism after RYGB. Together, our results suggest that the GLP-1 system may offer untapped potential as a novel biomarker for personalized approaches to the treatment of T2D and obesity. Clinical studies in obese and T2D patients will be required to test whether this desirable novel biomarker shows the same, or even greater, promise in humans and can be used to predict the benefits of—as well as to prevent unnecessary risks in—bariatric surgeries.

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Author Contributions. K.M.Ha. and M.H.T. were responsible for study conception and design, analyses and interpretation of the data, and drafting of the manuscript. K.M.He., S.E.A., N.O., J.H., C.R., E.B., J.B., and M.T. generated experimental data. T.D.M., P.T.P., S.C.B., R.D.D., D.P.-T., D.A.D., and R.J.S. advised on the study concept, conducted specific analyses, and critically revised the manuscript. M.H.T. is the guarantor of this work and, as such, had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

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