ORIGINAL RESEARCH

CURRENT DEVELOPMENTS IN NUTRITION

Nutrition and Exercise Physiology

Rats Selectively Bred for High Voluntary Physical Activity Behavior are Not Protected from the Deleterious Metabolic Effects of a Western Diet When Sedentary

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ABSTRACT

Background: Physical activity and diet are well-established modifiable factors that influence chronic disease risk. We developed a selectively bred, polygenic model for high and low voluntary running (HVR and LVR, respectively) distances. After 8 generations, large differences in running distance were noted. Despite these inherent behavioral differences in physical activity levels, it is unknown whether HVR rats would be inherently protected from diet-induced metabolic dysfunction.

Objectives: The aim of this study was to determine whether HVR rats without voluntary running wheels would be inherently protected from diet-induced metabolic dysfunction.

Methods: Young HVR, LVR, and a wild-type (WT) control group were housed with no running wheel access and fed either a normal diet (ND) or a high-sugar/fat Western diet (WD) for 8 wk. Body weight, percentage body fat (by dual-energy X-ray absorptiometry scan), blood lipids [total cholesterol, low-density lipoprotein (LDL) cholesterol, high-density lipoprotein (HDL) cholesterol, triglycerides (TGs), nonesterified fatty acids], and hepatic TG content were measured, and indices of insulin sensitivity were determined via an intravenous glucose tolerance test. Additionally, weekly energy intake and feed efficiency were calculated.

Results: After 8 wk, significant differences in body weight and body fat percentage were noted in all WD animals compared with ND animals, with the LVR-WD exhibiting the greatest increase due, in part, to their enhanced feed efficiency. Lipid dysregulation was present in all WD rat lines compared with ND counterparts. Furthermore, LVR-WD rats had higher total cholesterol, HDL cholesterol, and TG concentrations, and higher areas under the curve (AUC) for insulin than HVR-WD and WT-WD, although HVR-WD animals had higher $AUC_{glucose}$ than both LVR-WD and WT-WD and higher LDL than WT-WD.

Conclusions: In the absence of high voluntary running behavior, the genetic predisposition for high running in HVR did not largely protect them from the deleterious effects of a WD compared with LVR, suggesting genetic factors influencing physical activity levels may, in part, be independent from genes influencing metabolism. *Curr Dev Nutr* 2019;3:nzz017.

Introduction

Within the last few decades, diet– and physical inactivity–related chronic diseases have become pandemic. In the United States, it is estimated that >30% of the population is obese (1–3), >115 million have diabetes or prediabetes (4), and ~29% of the population dies from cardiovascular diseases (5). The Western diet (WD), high in refined-carbohydrates and fats,

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Abbreviations used: HCR, highest capacity to run; HVR, high voluntary runner; IVGTT, intravenous glucose tolerance test, LVR, low voluntary runner; ND, normal diet; NEFA, nonesterified fatty acid; TG, triglyceride; WD, Western diet; WT, wild-type.

combined with a sedentary lifestyle, promotes increased fat mass and induces chronic diseases (6). Not surprisingly, diet– and physical inactivity–related chronic diseases represent the single largest cause of morbidity and mortality in the United States (7, 8).

Genes that predispose an individual to risk factors for chronic disease interact with the environment to influence phenotypic responses (6, 9). For example, physical exercise has been shown to have a genetic component that induces various levels of exercise behavior (10–13). We have developed an animal model in which rats were bred for high and low voluntary running (HVR and LVR, respectively) distances (14). After 8 generations in LVR and HVR lines, large differences in distance, time, and speed of running between the 2 lines were recorded (14). For example, in generation 8, total 6-d running distances were 3.6 and 33.7 km, whereas running times were 121 and 1071 min for LVR and HVR, respectively (15). Thus, this polygenic model supports a genetic inheritance of physical activity behavior and is a valuable tool for understanding the effects of inherent physical activity behavior as well as the development and progression of chronic diseases.

In the present study, we investigated the effects of a WD designed to be analogous to WDs consumed in modern societies, consisting of a high refined-carbohydrate and high-fat combination, in both LVR and HVR animals, as well as in a wild-type (WT) control group. To limit environmental influences, we did not allow the animals to have access to a running wheel. This allowed us to observe how their preselected genes would affect their ability to handle an 8-wk WD intervention with respect to body composition, energy intake, lipid profiles, and glucose homeostasis. We addressed the following questions: *1*) do HVR animals display resistance to commonly measured negative metabolic effects of a WD if they are not permitted to have voluntary running; and 2) are there any advantageous effects of having preselected genes for high exercise behavior in HVR rats compared with LVR rats selected for low running behavior, or compared with nonselected controls? We tested the hypothesis that, in the absence of voluntary running, inherent genes for high levels of voluntary running by selective breeding would have co-selected functions to protect against metabolic dysfunction occurring while consuming a WD.

Methods

Animal welfare and experimental design

All animal procedures were approved by the Institutional Animal Care and Use Committee at the University of Missouri, Columbia. Rats were killed by carbon dioxide asphyxiation. The experiment was performed in the summer of 2015.

Male WT (outbred Wistar rats) were purchased from Charles River Laboratories. Male HVR and LVR rats from the 12th and 13th generations of an artificial selection process for high and low voluntary wheel running behavior were bred in the animal quarters of the College of Veterinary Medicine, University of Missouri, Columbia. The selective breeding process used to generate HVR and LVR rats has been described previously (14). Briefly, the founding population consisted of outbred Wistar rats (Charles River). Thirteen families were bred for each HVR and LVR line, and each generation was provided access to running wheels from 28 to 34 d of age for selection of subsequent breeding pairs. Within each HVR and LVR family, the highest and lowest, respectively, running male and female are then chosen for the next breeding cycle based upon wheel distance run during nights 5 and 6 of the selection period.

Rats were housed in temperature-controlled facilities on a 12 h:12 h light:dark cycle and consumed ad libitum water and normal diet (ND), which comprised LabDiet Certified Rodent 14% protein rodent diet (5CR4) containing the following profile: 3.5 kcal/g, 16.1% protein (expressed as % kcal), 69.3% carbohydrate (expressed as % kcal), and 14.6% fat (expressed as % kcal) (16). At 33–35 days of age, all rats were moved to single housing to undergo a 1-wk acclimatization period. At 40–43 d of age (and 1 wk older for WT), rats either continued on a ND or switched to a Western Diet (WD; TD.88137; Harlan Laboratories) (17) that contained the following macronutrient profile: 4.5 kcal/g, 15.2% protein (expressed as % kcal), 42.7% carbohydrate (expressed as % kcal), and 42.0% fat (expressed as % kcal) to generate the following experimental groups: LVR-ND ($n = 7$), LVR-WD ($n = 7$), WT-ND ($n = 6$), WT-WD ($n = 7$), HVR-ND ($n = 8$), and HVR-WD ($n = 8$), for a total of 43 rats after the death of 1 rat. The WD contained high sucrose (34% by food weight), whereas the ND contained low sucrose (2.6% by food weight) (16). Due to the limited amount of cage space, rats were double caged when possible during the 8-wk experiment. No running wheels were provided during the experimental period.

Food intake and feed efficiency

During the 8-wk diet intervention, food intake was recorded each week. Data were analyzed for the first 6 wk to accommodate the glucose tolerance testing in week 7. After measuring food weights at the end of each week, a new total amount of food was recorded and added to each cage. Cumulative caloric intake was done by summing individual weekly intake of calories for each animal. Feed efficiency was calculated by dividing caloric consumption by the grams of body weight gained over the same period (18).

Body weight and composition

During the 8-wk diet intervention, body weights were recorded weekly. Body composition was measured before and after the 8-wk diet intervention in the LVR and HVR animals (40–43 d of age during premeasurement) and after 8 wks of diet intervention in the WT under isoflurane anesthesia by dual-energy X-ray absorptiometry with the use of a Hologic QDR-1000/w machine calibrated for rats.

Blood lipid and hepatic triglyceride measurements

Blood lipids and hepatic triglycerides (TGs) were measured after the dietary treatment periods in overnight-fasted rats. Directly after carbon dioxide asphyxiation, ~1 mL of blood was collected via heart stick with the use of a 22G needle connected to a 3-mL syringe. The blood was placed in 200 μL of chilled 0.1 M EDTA and kept on ice until it was centrifuged at 7000 × *g* for 10 min at 4°C. The top layer of plasma was carefully extracted and stored at −80°C. Plasma was sent to the laboratory of Charles E Wiedmeyer (Comparative Clinical Pathology Services, University of Missouri, Columbia, MO) for measurements of total cholesterol, high-density lipoprotein (HDL) cholesterol, low-density lipoprotein (LDL) cholesterol, TGs, and nonesterified fatty acids (NEFAs).

To measure hepatic TGs, the median lobe of the liver was extracted from each rat, placed into liquid nitrogen immediately, and stored at −80°C. A 30-mg liver sample was homogenized with a mixture of

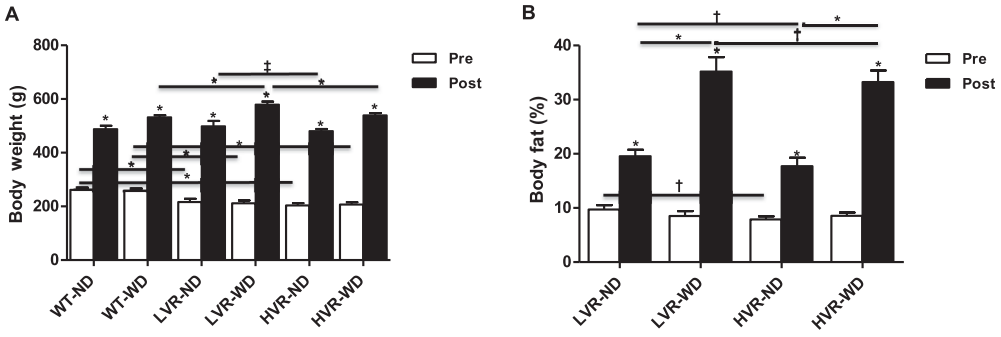

FIGURE 1 Body weight and body fat percentage. (A) Body weights and (B) percentage body fat of LVR, WT, and HVR rats at the beginning (pre, white bars) and end (post, black bars) of an 8-wk diet intervention; normal diet (ND), Western diet (WD). Percentage body fat was determined at the end of the intervention. Data are reported as means with error bars indicating SEMs. Significant differences are noted as follows: $^{\dagger}P < 0.05$; $^{\ddagger}P < 0.01$; $^{*}P < 0.001$. HVR, high-voluntary running; LVR, low-voluntary running; WT, wild-type.

2:1 chloroform:methanol with TissueLyser LT (Qiagen) for 2 × 2 min at 20 Hz. The test tubes were put in a cold room on a rotating wheel overnight. The following day, 1 mL of MgCl (4 mM) was mixed in each tube, followed by centrifuging for 1 h at 1000 × *g* at 4°C. The organic solvent phase was left to evaporate overnight. Subsequently, the dried lipids were mixed with butanol:Triton-X114 (3:2 vol:vol) to dissolve the dried lipids. Finally, 3 μL of standards and samples were added into each well of a 96-well plate loaded with 300 μL of mixed glycerol and TAG reagent. The plate was then incubated for 5 min at 37°C (or 15 min at room temperature) followed by a reading at 540 nm.

Intravenous glucose tolerance testing

The protocol for the intravenous glucose tolerance testing (IVGTT) was based on the methods of Vieira-Potter et al. (19). Testing occurred during week 7 of the 8-wk diet intervention. The rats were fasted overnight and subjected to IVGTT the following morning. A baseline blood sample was collected with 25G × ¾″ winged needles (Termo) followed by injection of 50% dextrose (Vetone). The amount of 50% dextrose injected was calculated based on body weight (4 μL of 50% dextrose/g body weight). Blood samples were taken at time points 15, 30, 45, 60, and 120 min after the injection of dextrose. For each time point, ~100–200 μL of blood was taken (19).

Glucose concentrations (mg/dL) were determine with an Alpha Trak 2 blood glucose monitoring system with test strips (Abbott). For insulin, the blood was then centrifuged at 7000 × *g* for 10 min at 4°C and the top layer of plasma was carefully extracted via a pipette and stored at −80°C. Plasma was sent to the laboratory of Charles E Wiedmeyer (Comparative Clinical Pathology Services, University of Missouri, Columbia, MO), where insulin concentrations were determined by ELISA (ng/mL). AUC estimates for glucose ($AUC_{glucose}$) and insulin ($AUC_{insulin}$) were calculated. Select individual animal time points for insulin were not obtainable due to insufficient blood volume. These rats were excluded from the $AUC_{insulin}$ calculation. To estimate insulin sensitivity, the product of $AUC_{glucose} \times AUC_{insulin}$ was calculated (20) and multiplied by 10^{-7} to obtain numbers ranging from 1 to 30.

Data analysis

Analysis of data was performed with Prism statistical software and Microsoft Excel. Food intake, feed efficiency, IVGTT measurements, and lipids were analyzed through the use of a 1-factor ANOVA, with a Neuman-Keuls multiple comparisons analysis. Body weight and body composition were analyzed through the use of a 2-factor ANOVA with Bonferroni post-hoc analyses. Data are reported as means ± SEMs with a significance set at $P < 0.05$.

Results

Body weight and fat mass

At the start of dietary treatments, WT groups weighed significantly more than either LVR or HVR selectively bred lines (all $P < 0.001$), with no difference between LVR and HVR (**Figure 1**A). Seven weeks into the diet intervention, body weights were measured before IVGTT testing and all 3 rat lines consuming the WD weighed significantly more than their respective ND groups ($P < 0.001$). Additionally, the LVR-WD animals weighed more than both the WT-WD and HVR-WD after the WD ($P < 0.001$). Furthermore, after the ND, LVR-ND animals weighed more than HVR-ND ($P < 0.001$).

Before starting diets, baseline percentage body fat was higher in the LVR-ND than in the HVR-ND groups ($P < 0.05$); however, there were no differences between the LVR-WD and HVR-WD groups (Figure 1B). WT rats did not undergo body composition testing prior to diets. After the diet treatments, the WT-ND body fat was 19.1 ± 1.5% and the WT-WD was 28.4 ± 2.1%. After the diets, body fat percentage increased in all groups ($P < 0.001$), but groups that were fed WD had a significantly higher body fat percentage than those fed ND (both $P < 0.001$), and the LVR-ND and LVR-WD groups had greater increases in body fat percentage than HVR groups on the same diet (both $P < 0.05$). The body fat percentage of LVR-WD animals was 1.7% less than that of HVR-WD animals.

Total energy intake and feed efficiency

The LVR, WT, and HVR groups had similar total energy intakes (**Figure 2**A). However, when on WD, all groups exhibited greater energy intake than groups on ND. Interestingly, both LVR-WD and HVR-WD rats had higher energy intakes than WT-WD rats. To provide some insight into the impact of the diets on weight gain, the summed, caloric intake was divided by the grams of body weight gained to assess feed

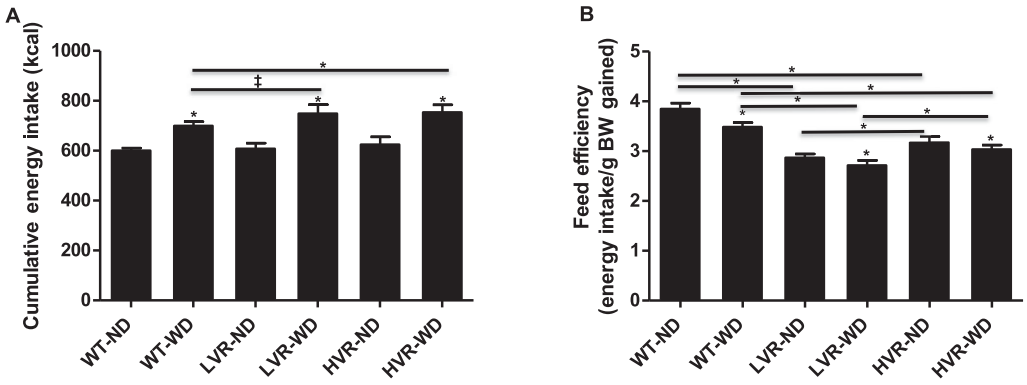

FIGURE 2 Energy intake and feed efficiency. (A) Cumulative energy intake from the summed weekly energy intakes from weeks 1–6. (B) Feed efficiency of energy intake divided by body weight gain during weeks 1–6. Data are reported as means with error bars indicating SEMs. Significant differences are noted as follows: $^{‡}P < 0.01$; $^{*}P < 0.001$. HVR, high-voluntary running; LVR, low-voluntary running; ND, normal diet; WD, western diet; WT, wild-type.

efficiency (Figure 2B). This analysis revealed significantly more caloric intake per gram of weight gained in the ND groups than in the WD groups ($P < 0.001$). All WD rats had a higher feed efficiency—less energy per gram of body weight gained—than ND rats within their respective groups ($P < 0.001$). Both LVR groups were also more efficient than the corresponding HVR groups ($P < 0.001$).

Plasma lipids and liver TG concentrations

Plasma total cholesterol was not significantly different when comparing all 3 rat lines on an ND (**Figure 3**A). Additionally, both the WT and HVR rats consuming the WD did not differ in total cholesterol concentration from their ND counterparts; however, the WD led to significantly higher total cholesterol in the LVR rats ($P < 0.001$). There were significant increases in total cholesterol in both HVR-WD ($P < 0.05$) and LVR-WD ($P < 0.001$) compared with WT-WD, as well as in LVR-WD compared with HVR-WD ($P < 0.01$). Similarly, each line's LDL cholesterol concentrations on a WD did not differ compared with their ND counterparts. However, LDL cholesterol was significantly higher in the HVR-WD than in the WT-WD rats ($P < 0.05$) (Figure 3B). Furthermore, plasma HDL cholesterol (Figure 3C) was significantly higher in LVR-WD than in HVR-WD ($P < 0.01$) and WT-WD ($P < 0.05$). Analysis of blood TG concentrations showed no statistical difference between rats consuming ND (Figure 3D), whereas WD caused an elevation in blood TGs in both HVR and LVR rats compared with their ND counterparts ($P < 0.001$). LVR-WD rats had greater blood TG concentrations than both HVR-WD and WT-WD rats ($P < 0.001$). Additionally, HVR-WD rats had significantly greater plasma TG concentrations than WT-WD rats ($P < 0.01$). NEFAs concentrations were elevated in both HVR-WD ($P < 0.001$) and LVR-WD ($P < 0.01$) compared with their ND counterparts; however, there was no difference between WT-WD and WT-ND (Figure 3E). Both LVR-WD ($P < 0.001$) and HVR-WD ($P < 0.001$) had significantly higher NEFA concentrations than found in WT-WD rats.

Within diets, only the LVR-ND had a significantly higher concentration than the WT-ND ($P < 0.05$). Liver TG content (Figure 3F) was also assessed, and similar to plasma TGs, there were no differences in groups consuming ND or between strains consuming WD. However, all groups consuming WD exhibited similar elevations in hepatic TGs compared with their respective ND counterparts ($P < 0.01$).

Glucose tolerance testing

For the IVGTT, we analyzed $AUC_{glucose}$, $AUC_{insulin}$, and the product of $AUC_{glucose} \times AUC_{insulin}$. First, analysis of the $AUC_{glucose}$ for revealed that WD groups had a greater $AUC_{glucose}$ than ND groups for all rat lines ($P < 0.001$) (**Figure 4**A). The $AUC_{glucose}$ values for HVR on both diets were higher than for both other groups on the respective diets (all $P < 0.001$); additionally, the values for the WT-WD rats were also significantly higher than for the LVR-WD rats ($P < 0.01$). For the $AUC_{insulin}$, WD rat lines had significantly higher $AUC_{insulin}$ than their ND counterparts in the WT and LVR groups (WT, $P < 0.05$; LVR, $P < 0.001$) (Figure 4B), but the HVR-WD was not significantly higher than the HVR-ND. The LVR-WD rats had significantly higher $AUC_{insulin}$ than both the HVR-WD and WT-WD rats ($P < 0.001$). Interestingly, there was no significant difference between any of the rat lines consuming the ND. When analyzing $AUC_{glucose} \times AUC_{insulin}$ as a surrogate for insulin sensitivity, the WD rats exhibited significant increases compared with all rat lines consuming ND (WT, $P < 0.01$; LVR, $P < 0.001$; HVR, $P < 0.001$) (Figure 4C). In addition, the LVR-WD exhibited a higher $AUC_{glucose} \times AUC_{insulin}$ than both the HVR-WD ($P < 0.001$) and WT-WD ($P < 0.001$). However, there was no significant difference between HVR-WD and WT-WD rats.

Discussion

Over the past several years, we have investigated various physiologic effects in our selective breeding model for high and low voluntary running distances (14). Recently, we investigated the impact of a WD in this model and noted increased energy intake in HVR compared with LVR rats (21). Further, we have reported that when HVR rats were fed a WD, increased voluntary running appeared to offset the increased energy consumption noted in these animals, negating any potential greater metabolic dysfunction from energy excess. This result has been observed in other short-term feeding studies looking at rats with increased aerobic capacity consuming a short-term, high-fat diet (22). Metabolic syndrome induced by a high-fat diet in Sprague-Dawley rats is largely reversible with exercise training (23). To follow-up the earlier work, the present study was performed to investigate whether HVR rats are genetically protected from 8 wk of WD-induced metabolic

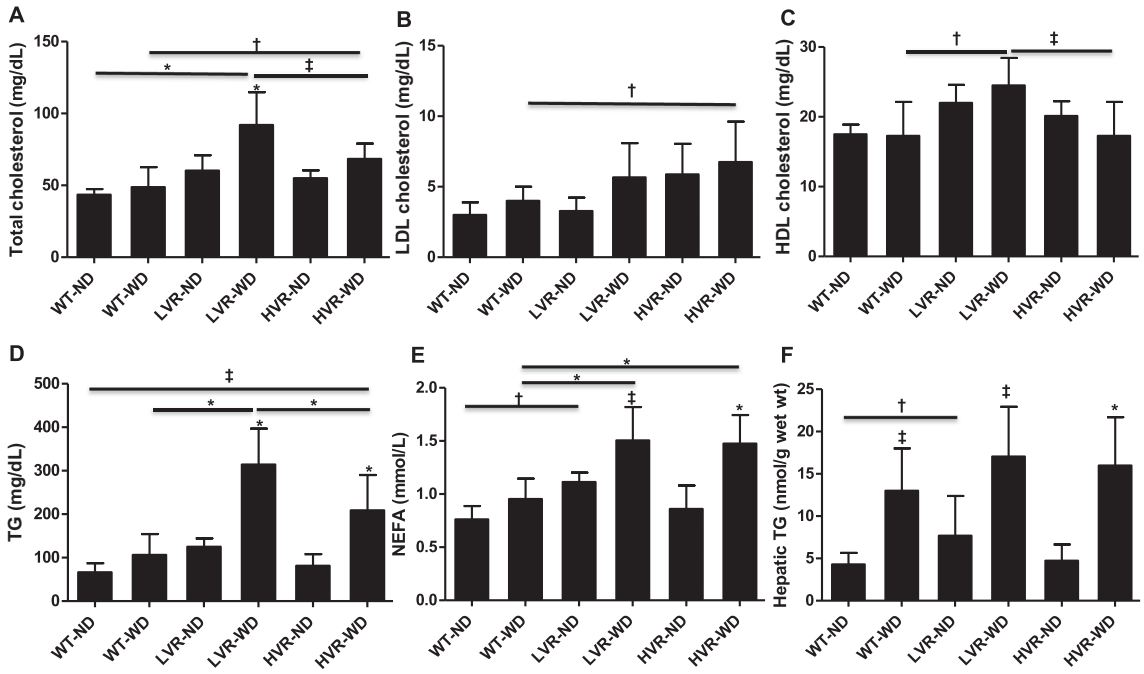

FIGURE 3 Blood lipids, hepatic TG, and NEFA. Data are levels of (A) total cholesterol, (B) LDL cholesterol, (C) HDL cholesterol, (D) TG, (E) NEFA, and (F) hepatic TG after 8 wk of diet. Data are reported as means with error bars indicating SEMs. Significant differences are noted as follows: $^{\dagger}P < 0.05$; $^{\ddagger}P < 0.01$; $^{*}P < 0.001$. HDL, high-density lipoprotein; HVR, high-voluntary running; LDL, low-density lipoprotein; LVR, low-voluntary running; ND, normal diet; NEFA, nonesterified fatty acid; TG, triglyceride; WD, western diet; WT, wild-type.

dysfunction compared with LVR rats when they are sedentary, i.e., not permitted to engage in voluntary running. One of our findings herein, contrary to our hypothesis, is that having the preselected HVR genetic background in the absence of accessibility to voluntary wheel running was not protective against metabolic dysfunction under the experimental conditions employed.

WD consumption for 8 wk led to significant increases in body weight in sedentary LVR, WT, and HVR rats compared with ND-fed cohorts. LVR-WD rats weighed significantly more than HVR-WD rats, although the differences in body weight between the LVR and HVR groups were modest. Similar findings were noted for adiposity, as measured by body fat percentage, reflecting increased susceptibility for dietary calories to lead to increased fat gain, i.e., a higher feed efficiency in LVR rats when fed a WD. Again, although the LVR rats exhibit higher adiposity as a result of WD consumption, both the LVR and HVR groups demonstrated similar patterns of adiposity gain in response to WD. Although not determined in our study, it is possible that the greater increases in weight and fat gain in the LVR than in the HVR animals may be also be due, in part, to the lower spontaneous cage locomotor activity in LVR rats, as we have previously reported (24). Others have reported the importance of caloric expenditure in a different running model. Rats were selected for the highest capacity to run (HCR) the longest distances by avoiding mild electric shock during forced, endurance running on a motor-driven treadmill. HCR rats had higher body temperature and a higher metabolic rate, which in turn slowed weight gain and the associated negative health outcomes found in rats born with a low capacity for running, which exhibited higher chronic disease risk (25). HCR rats were more physically active and lost more weight during calorie restriction than the low-capacity runners (26). Interestingly, HCR rats also displayed increased voluntary wheel running compared with low-capacity rats (27). Thus, although inherently greater aerobic capacity and improved metabolic fitness were associated with increased voluntary physical activity, increased drive for physical activity does not innately improve metabolic health during WD consumption. Likewise, human cardiorespiratory fitness, rather than physical activity levels, is more strongly associated with all-cause mortality (28).

For blood lipid parameters, our results offer a unique perspective. A relatively consistent theme emerged in which WD promoted changes in the lipid profiles of both LVR and HVR rats, but surprisingly not in WT rats. Within the selectively bred rat groups, the LVR-WD animals showed a significant increase in total cholesterol, TGs, and HDL cholesterol compared with the HVR-WD animals. However, the HVR-WD animals exhibited a significant increase in LDL cholesterol compared with the WT-WD animals, although not different from the LVR-WD rats. Higher HDL cholesterol and lower LDL cholesterol are correlated to lower the risk of cardiovascular disease (29). Thus, the lower HDL cholesterol and lack of difference in LDL cholesterol in the HVR-WD compared with the LVR-WD suggests the HVR line was not protected from the dyslipidemic effects of the WD. However, the HVR-WD rats did exhibit less of rise in TGs when consuming the WD than found in the LVR rats. Overall, the metabolic profiles of the LVR-WD and HVR-WD groups showed a worsened metabolic profile compared to the WT-WD rats. We speculate that this may be due, in part, to the WT-WD rats having a lower total energy intake compared with the WD selectively bred rat groups, and HVR rats being sedentary.

To establish a better idea of the metabolic effects of WD in LVR and HVR rats, insulin and glucose were measured in response to an

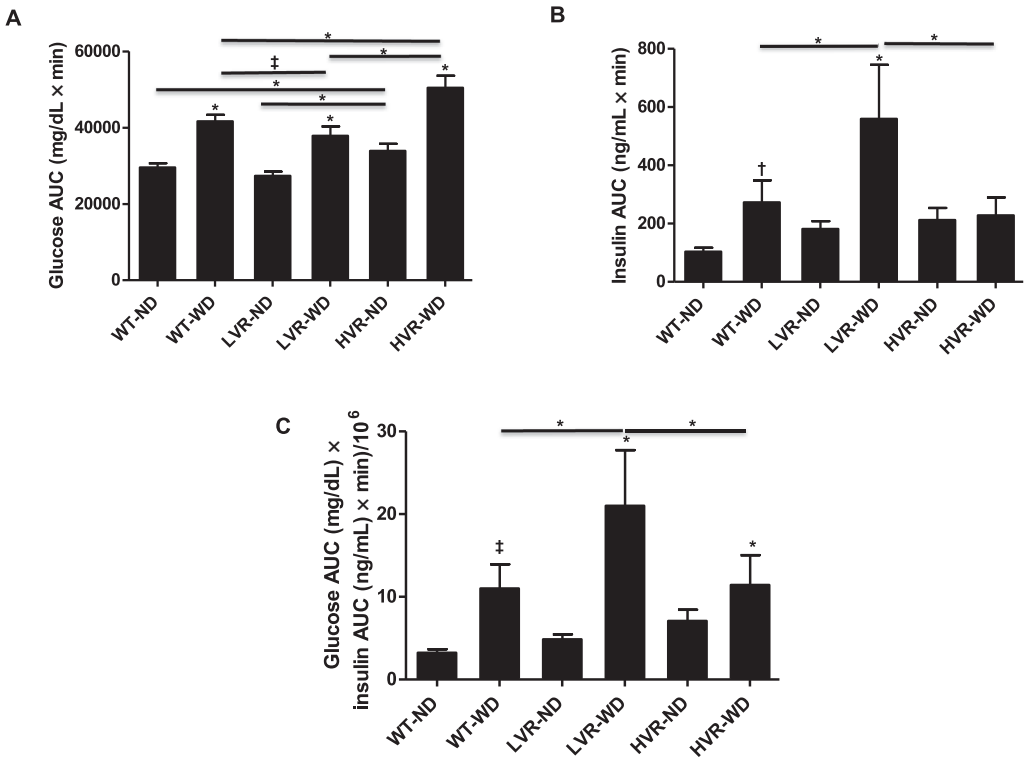

FIGURE 4 Insulin dynamics. (A) $AUC_{glucose}$, (B) $AUC_{insulin}$, and (C) $AUC_{glucose} \times AUC_{insulin}$ estimated from IVGTTs performed during week 7. Data are reported as means with error bars indicating SEMs. Significant differences are noted as follows: $^{\dagger}P < 0.05$; $^{\ddagger}P < 0.01$; $^{*}P < 0.001$. $AUC_{glucose}$, area under curve, glucose; $AUC_{insulin}$, area under curve, insulin; HVR, high-voluntary running; IVGTT, intravenous glucose tolerance test; LVR, low-voluntary running; ND, normal diet; WD, western diet; WT, wild-type.

intravenous glucose load. High-sugar diets have been repeatedly shown to cause hyperinsulinemia (30–32) and high-fat diets can also induce insulin resistance (33). We found that all 3 groups showed a significant increase in $AUC_{glucose}$ and $AUC_{glucose} \times AUC_{insulin}$ when comparing ND with WD. HVR-WD was the only group that did not see an increase in $AUC_{insulin}$ from consuming WD, although this may have been affected by a few of the rats in this group being excluded due to a lack of blood at selected insulin time points during the IVGTT. Regardless, on a WD, group differences existed in an intravenous glucose load test. WD produced greater hyperglycemia in HVR rats but showing greater hyperinsulinemia in LVR rats. As the product of the AUCs was greater in LVR-WD than in HVR-WD rats, we speculate that insulin resistance was primarily driven by the $AUC_{insulin}$ in the LVR-WD group, whereas it was driven, to a lesser extent, by the $AUC_{glucose}$ in the HVR-WD group. Others have reported metabolic dysfunction in WT rats. For example, feeding male Sprague-Dawley rats a WD resulted in higher serum glucose, insulin, triglycerides, total cholesterol, and liver TGs (34). After 4 wk of a high-fat/high-sugar diet, Wistar rats developed hyperglycemia, hyperinsulinemia, glucose intolerance, and a diminished insulin response to a glucose load (31). Similarly in male Sprague-Dawley rats, 10 wk of WD consumption led to significantly higher fasting insulin and the development of insulin resistance (35). Here, more metabolic dysfunction was seen in the selectively bred rats when they were not given access to voluntary running wheels. It is possible that the HVR line has co-selected genes that require a high amount of inherent activity to maintain a healthy lipid profile and glucose homeostasis. This could also account for the irregular blood lipid markers observed in the HVR-WD group. We speculate that the HVR line does not receive metabolic benefits, nor is it inherently protected from the negative effects of a WD by simply being bred for high running behavior.

One limitation of the current study is that blood lipids and IVGTT were not measured prior to the diets. A second limitation, alluded to earlier, was that cage locomotor activity was not measured, which could have helped to explain some of the metabolic differences between the LVR and HVR groups, such as body weight, body fat, and total energy consumed. We have, however, previously demonstrated that HVR rats are inherently more active than LVR rats in an open field environment (24). A third limitation is that the rats used to establish disease protection were generations 12–13, and it is possible that this may not have been sufficient to establish disease protection. The fourth limitation of our data is that only a single gender was employed, and therefore we cannot extrapolate our findings to female rats.

Overall, the interaction of 2 diets (ND and WD) with 3 genetically different populations (WT, HVR, and LVR) combined with a sedentary lifestyle was examined to mimic the US adult population. Importantly, the value of physical activity is evidenced by our observations that an absence of voluntary physical activity in rats fed an ND did not alter selected metabolic measures in rats genetically bred for HVR performance. However, metabolic function was disrupted for some variables in the same HVR genetic pool exposed to the dietary challenge of a high-sugar/high-fat diet that has previously been noted to induce

a variety of cardiometabolic abnormalities (36–40). Thus, selectively bred HVR animals, when not allowed voluntary wheel running, are not protected from some of the deleterious metabolic effects of a WD. If sustained in the long term in the absence of voluntary physical activity, a WD may increase the risk of future cardiometabolic disease, even in the absence of a predisposition for low physical activity levels.

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