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Effect of Corncob Bedding on Feed Conversion Efficiency in a High-Fat Diet-Induced Prediabetic Model in C57Bl/6J Mice

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Laboratory facilities use many varieties of contact bedding, including wood chips, paper products, and corncob, each with its own advantages and disadvantages. Corncob bedding, for example, is often used because of its high absorbency, ability to minimize detectable ammonia, and low cost. However, observations that mice eat the corncob lead to concerns that its use can interfere with dietary studies. We evaluated the effect of corncob bedding on feed conversion (change in body weight relative to the apparent number of kcal consumed over 7 d) in mice. Four groups of mice (6 to 12 per group) were housed in an individually ventilated caging system: (1) low-fat diet housed on recycled paper bedding, (2) low-fat diet housed on corncob bedding, (3) high-fat diet housed on recycled paper bedding, and (4) high-fat diet housed on corncob bedding. After 4 wk of the high-fat diet, feed conversion and percentage body weight change both were lower in corncob-bedded mice compared with paper-bedded mice. Low-fat–fed mice on corncob bedding versus paper bedding did not show statistically significant differences in feed conversion or change in percentage body weight. Average apparent daily feed consumption did not differ among the 4 groups. In conclusion, these data suggest that corncob bedding reduces the efficiency of feed conversion in mice fed a high-fat diet and that other bedding choices should be favored in these models.

Abbreviations: CC, corncob bedding; HF, high-fat diet; LF, low-fat diet; P, paper bedding.

The *Guide to the Care and Use of Laboratory Animals* (the *Guide*) is used as a reference for the standard of care for many laboratory animals. Under rodent housing, the *Guide* suggests the use of contact bedding on solid-bottom caging for all rodents for both enrichment of environment and to absorb urine and minimize ammonia buildup.[2] Many beddings that have been tested for absorbency, comfort, cost, biodegradability, toxicity, and effects on research are available for rodent housing.[9,11] Corncob bedding is a common choice for rodent bedding because of its biodegradability, ability to decrease the amount of detectable ammonia,[7] high absorbency per unit volume,[1] and cost.

An active area of animal research involves modeling the weight gain that is seen in the average human diet in Western societies and that induces insulin resistance and prediabetes. Unintentional effects of housing conditions, including the potential effects of corncob bedding on metabolic parameters, need to be avoided in these models. In addition to uncontrolled dietary intake, corncob bedding has been associated with decreased time spent in slow-wave sleep, presumably because of decreased comfort[4] in rats. These findings prompt the concern that corncob bedding might also alter activity status, a parameter affects intended weight gain.

Here, we conducted a retrospective analysis of weight gain, apparent feed consumption, and feed efficiency of mice housed on corncob bedding compared with recycled paper bedding and fed either low-fat or high-fat diet. This study resulted from a noticeable reduction in expected weight gain in an established high-fat–fed mouse model of obesity and prediabetes after animals were moved from one building within a facility that routinely uses recycled paper bedding to another building that uses corncob bedding. This decrease in weight gain continued for weeks past the expected acclimation to transportation and the new facility. We hypothesized that this suppression in weight gain was associated with the use of corncob bedding.

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Materials and Methods

Male C57Bl/6J mice ($n = 36$; Jackson Laboratory, Bar Harbor, ME) were used. All mice were 4 wk of age on arrival and housed singly in IVC (Lab Products, Seaford, DE, or Alternative Design, Siloam Springs, AR) on autoclaved recycled pelleted paper (P; catalog no. 7084, Harlan Teklad, Madison, WI) or 1/4-in. corncob bedding (CC; catalog no. 7097, Harlan Teklad). The study for which these mice were obtained initially involved a forced treadmill exercise model that was initiated after the data for the current study were collected. The mice were housed individually to standardize the effects of exercise within the home cage between groups. All mice were provided with Enviro-dri (Shepherd Specialty Papers, Cincinnati, OH) and one nesting square (Nestlets, Ancare, Bellmore, NY) for environmental enrichment. Animals were maintained on a 12:12-h light cycle, with an ambient temperature of 70 to 72 °F (21.1 to 22.2 °C) and a target humidity of 30% to 70%. Pelleted food was provided in excess weekly, placed in a culture dish on the cage floor. Because of anecdotal reports that high-fat feed can become rancid and moldy rapidly, requiring changing more frequently than weekly, we closely monitored the condition of the feed daily, but we did not need to change the feed more often. Water was provided ad libitium throughout the study. Cages were changed every other week, with spot changes as needed. Health screening was accomplished through contact bedding sentinels, which were found to be free of mouse rotavirus, mouse hepatitis virus, *Mycoplasma pulmonis*, mouse parvovirus, minute virus of mice, pneumonia virus of mice, reovirus 3, Sendai virus, Theiler mouse encephalomyelitis virus, *Clostridium piliformis*, ectromelia virus, lymphocytic choriomeningitis virus, Hantaan virus, mouse adenovirus, mouse cytomegalovirus, and ectoparasites. This colony was housed in a room containing a sentinel confirmed to be positive for *Aspicularis tetraptera*, but our animals were not treated during the study. In addition, the mice in this study were not tested for the presence of pinworms at euthanasia, but all mice in all groups were housed in the same room, with the same probability of pathogen exposure. The animal care and use program was in compliance with federal regulations and was accredited by AAALAC. All procedures were approved by the institutional IACUC prior to initiation of the studies.

Mice were assigned randomly to 4 experimental groups, 6 mice per group for the low-fat diet (groups 1 and 2) and 12 mice per group for the high-fat diet (groups 3 and 4). The low-fat purified, pelleted diet (D01030107, Research Diets, New Brunswick, NJ) contained 10% kcal from palmitate fat and 3.85 kcal/g and the high-fat purified, pelleted diet (D01030108, Research Diets) had 45% kcal from palmitate fat source and 4.73 kcal/g. On arrival, all mice received a standard extruded, natural-ingredient rodent diet (Teklad Global Diets 2018SX, Harlan Teklad) and housed on pelleted paper bedding. After 2 wk, all mice were given the low-fat diet, to acclimate them to the palmitate fat source. At 3 wk after arrival, the mice were housed on either paper or corncob bedding. At 5 wk after arrival, feeding of the high-fat diet was initiated as appropriate. Mice, the amount of feed provided, and the amount remaining were weighed weekly. Care was taken to recover as much of the feed as possible from within the bedding, but we presume that some of the feed provided was neither consumed nor recovered for weighing.

For statistical analysis, raw data was collected for each mouse weekly. These data are presented as a mean for each treatment group (n = 6 or 12). One-way ANOVA with a Newman–Keuls Multiple Comparison test was performed by using Prism for Windows (version 5.03, GraphPad Software, San Diego, CA).

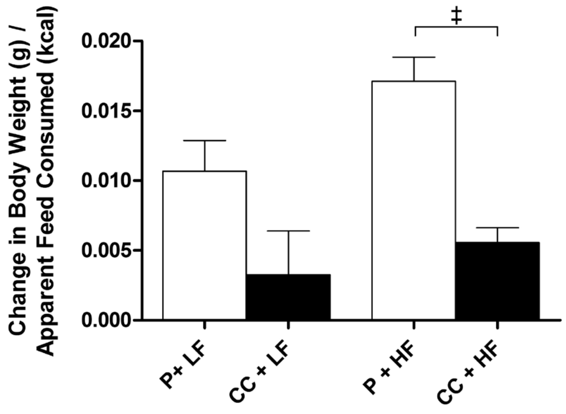

Figure 1. Feed conversion of mice housed on recycled paper bedding and fed a low-fat diet (P + LF) and those housed on corncob bedding and fed a low-fat diet (CC + LF) did not differ (P = 0.081). However, feed conversion of mice housed on recycled paper bedding and fed a high-fat diet (P + HF) differed significantly (‡, $P < 0.0001$) from that of mice housed on corncob bedding and fed a high-fat diet (CC + HF).

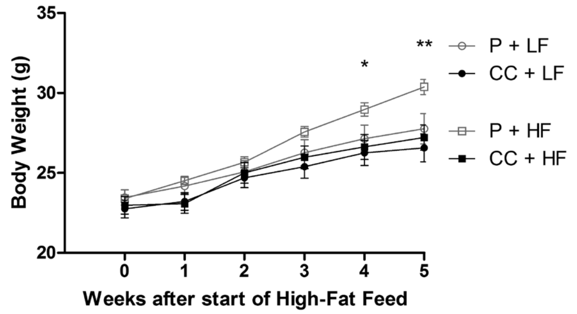

Figure 2. Mean body weights of mice housed on recycled paper bedding and fed a high-fat diet (P + HF) and those housed on corncob bedding and fed a high-fat diet (CC + HF) differed significantly at 4 wk (†, P = 0.0148) and 5 wk (‡, P = 0.0023) after the start of the diet. In contrast, body weight did not differ between mice housed on recycled paper bedding and fed a low-fat diet (P + LF) and those housed on corncob bedding and fed a low-fat diet (CC+LF) at either 4 wk (P = 0.454) or 5 wk (P = 0.376).

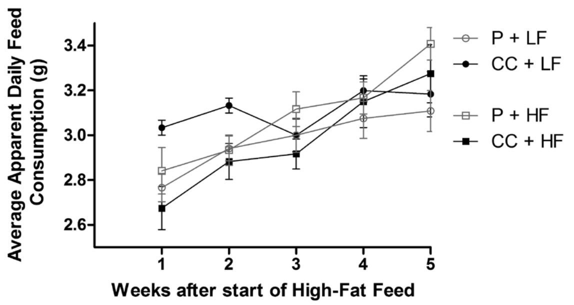

Figure 3. Average apparent daily feed consumption for the 5-wk period after initiating high-fat feed did not differ between either mice housed on recycled paper bedding and fed a low-fat diet (P + LF) and those housed on corncob bedding and fed a low-fat diet (CC + LF; P = 0.088) or between mice housed on recycled paper bedding and fed a high-fat diet (P + HF) and those housed on corncob bedding and fed a high-fat diet (CC + HF; P = 0.379).

Results

After 4 wk on the high-fat diet, feed conversion (Figure 1) and body weight gain (Figure 2) were significantly decreased in the mice housed on corncob bedding compared with those on high-fat feed housed on paper bedding ($P < 0.0001$ and P = 0.0148, respectively). After 5 wk of high-fat feeding, the significance between the body weights of the mice housed on corncob bedding compared with paper bedding increased (P = 0.0023; Figure 2). Feed conversion did not differ between low-fat–fed mice housed on corncob compared with paper bedding (P = 0.081), nor did body weights at 4 or 5 wk of mice fed a low-fat feed differ between those housed on corncob or paper bedding (P = 0.4535 and P = 0.3758, respectively). Average apparent daily feed consumption (Figure 3) was similar between mice on low-fat feed housed on corncob compared with paper bedding (P = 0.088) and between mice on high-fat feed housed on corncob compared with paper bedding (P = 0.379).

Discussion

This study was conceived when, for logistical and equipment availability reasons, a group of mice in a diet-induced prediabetic study were moved from one facility that routinely used recycled paper in their cages to another facility that used corncob bedding. A high-fat diet model is often used in diabetes research, and the weight gain of rodents on a high-fat diet is well described.[3,8] At our facility, the mice housed on corncob bedding were not reaching the expected weight gain for the model, thus prompting the retrospective statistical analysis of data already collected from these mice compared with a later group of mice housed solely in the second facility on recycled paper bedding. Because these mice were not part of a specific study design for the effects of corncob bedding on diet-induced weight gain, other variables, including the changes in the macro- and microenvironment of the mice when they switched facilities, need to be considered during data analysis. The mice housed on corncob bedding started in the first facility in an IVC unit that ran at least 50 air changes per hour, on recycled paper bedding, and with a staff dedicated to that facility. When these mice were moved to the subsequent facility, they were housed in an IVC unit with 4 air changes per hour, on corncob bedding, and a different dedicated staff. The mice on recycled paper bedding were in the second facility throughout the study, in IVC racks at 4 air changes hourly. In addition, the mice housed on corncob bedding experienced the stress of moving from one facility to the other at 4 wk after arrival at the institution and 1 wk prior to starting the high-fat diet. However, the changes in weight gain and feed efficiency were calculated by using data collected 4 and 5 wk after this move. During the time of collection, the staff, caging, and environmental enrichment were the same for all groups. The stress of changing environments might be expected to cause a decrease in apparent feed consumption and therefore a decrease in weight gain in mice, but this effect likely would be negligible after a 4-wk acclimation period. Importantly, this study shows no significant difference in apparent feed consumption between the 4 groups.

Another factor to consider is a possible change in intestinal microbiota due to bedding change or the physical move from one building to another. Dysbiosis of the microbiota could cause a change in the digestion and absorption of feed. However, according to work done in another laboratory, bedding changes have a negligible effect on microbiota.[5] In addition, after 5 d of acclimation, microbiota returned to its original distribution in animals moved to a different facility.[5] The current work supports the belief that any changes seen in feed conversion between weeks 4 and 5 are unaffected by differences in microbiota induced by changes in bedding or a physical move between facilities.

The effects of environmental changes, including bedding, on various aspects of experimental design have been documented in other studies.[4,6,10] The current study supports caution when choosing bedding types for diet-induced models. Although the corncob bedding did not reduce the amount of feed consumed by the mice, it did affect the efficiency with which animals were able to convert the consumed feed into body mass. Additional research should be performed to find the cause of this corncob-associated decrease in feed conversion.

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