

High-fat diet alters weight, caloric intake, and haloperidol sensitivity in the context of effort-based responding

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High-fat (HF) diets result in weight gain, hyperphagia, and reduced dopamine D_2 signaling; however, these findings have been obtained only under free-feeding conditions. This study tested the extent to which HF diet affects effort-dependent food procurement and the extent to which dopamine signaling is involved. Male Sprague-Dawley rats consumed either a HF ($n = 20$) or a standard-chow ($n = 20$) diet. We assessed the sensitivity to effort-based reinforcement in 10 rats from each group by measuring consumption across a series of fixed-ratio schedules (FR 5–FR 300) under a closed economy and quantified performance using the exponential-demand equation. For each FR, acute injections of 0 or 0.1 mg/kg of haloperidol, a D_2 antagonist, were administered to assess dopamine-related changes in consumption. Rats fed a HF diet consumed more calories and weighed significantly more than rats fed standard-chow. Food consumption decreased in both groups in an effort-dependent manner, but there

were no group differences. Haloperidol reduced responding in an FR-dependent manner for both groups. Animals exposed to a HF diet showed an altered sensitivity to haloperidol relative to rats fed a standard diet, suggesting that HF diet alters sensitivity to DA signaling underlying effort-based food procurement. *Behavioural Pharmacology* 00:000–000 Copyright © 2017 Wolters Kluwer Health, Inc. All rights reserved.

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Introduction

Diet-induced obesity (DIO) is a laboratory model of obesity in which nonhuman animals – usually rodents – are provided chronic access to a high-fat (HF) diet, which results in hyperphagia, excess body weight, and changes in neurobiology related to food intake, relative to controls fed standard-chow diets (Rolls *et al.*, 1980; Geiger *et al.*, 2009; Johnson and Kenny, 2010; Vucetic *et al.*, 2012). DIO-related hyperphagia is traditionally tested in environments that use unlimited access to food, in which large amounts of food are readily available with little response-cost required (Rolls *et al.*, 1980). Therefore, these economic conditions likely favor heightened food consumption (see Rasmussen *et al.*, 2016).

Studies from the field of behavioral economics (Hursh, 1980, 1984) show that effort plays an integral part of consumption of food, drugs, and other reinforcers relevant to human behavior (see Hursh, 2000; Bickel *et al.*, 2014). Demand refers to the extent to which behavior for a commodity changes as a function of its price. In non-human animal studies, price is manipulated by fixed-ratio (FR) schedules of reinforcement, where a set number of responses result in a reinforcer (e.g. a single food pellet). To assess demand, the number of reinforcers earned at a range of response requirements is plotted as a function of FR (log scaled) to produce a demand curve (Hursh and Silberberg, 2008). Demand curves typically show that consumption decreases as the FR increases in a positively

accelerating fashion, with little change in consumption at lower prices (inelasticity) and higher reductions in consumption at higher prices (elasticity; Hursh and Winger, 1995).

One way to quantify sensitivity to effort is the exponential-demand equation, which quantifies the essential value of a reinforcer (Hursh and Silberberg, 2008):

$$\log Q = \log Q_0 + k(e^{-\alpha Q_0 P} - 1), \quad (1)$$

where Q_0 is the number of reinforcers consumed at the lowest price (e.g. akin to a free-feeding environment), k is the range of reinforcers earned expressed in log units, and α (i.e. essential value) is a free parameter that estimates changes in consumption as a function of price (P ; Hursh and Silberberg, 2008). Lower values of α indicate lower elasticity (i.e. consumption is relatively insensitive to price increases) and are therefore associated with a commodity with higher reinforcer efficacy, whereas higher values of α indicate higher elasticity and lower reinforcer efficacy. Behavioral-economic analyses offer a fuller characterization of the reinforcing efficacy of food by measuring consumption across a wide range of prices (i.e. response requirements) rather than simply observing consumption at one price, such as in free-feeding environments.

Previous studies have utilized demand procedures to characterize effort-related food consumption in the genetically obese Zucker rat (*fal/fa*) – an animal model of obesity that has repeatedly been shown to engage in hyperphagia (Rasmussen *et al.*, 2010, 2012). In these studies, genetically obese Zucker rats consumed significantly more sucrose pellets than lean Zucker rats, but only when food was delivered contingent on lower response requirements. When higher response requirements were tested, there were no group differences. Obese Zucker rats also had higher Q_0 estimates compared with lean Zucker rats, supporting that the differences were related to low effort. No group differences in elasticity (α values) were evident. It is important to note that the Rasmussen *et al.* (2010, 2012) studies assessed demand in an open economy, in which additional food was available outside of the experimental session. Research suggests that demand in open economies is more elastic than demand in closed economies, in which all daily food is earned (Hursh, 1980, 1984; Collier *et al.*, 1992). Therefore, because the studies by Rasmussen *et al.* (2010, 2012) were carried out in open economies, this may have limited the ability for differences in elasticity to be observed between lean and obese Zucker rats. Nonetheless, these studies support the finding that differences between lean and obese rats observed in previous studies using animal models of obesity (i.e. Rolls *et al.*, 1980) may be inflated by ready access to food.

AQ3

One likely neurobiological mechanism underlying difference in food-reinforced behavior in models of obesity is disturbed dopamine (DA) neurotransmission. DIO reduces DA D_2 signaling in brain areas that underlie choice and motivation, which may lead to alterations in sensitivity to reinforcement (Geiger *et al.*, 2009; Johnson and Kenny, 2010; Vucetic *et al.*, 2012). For example, Johnson and Kenny (2010) showed that rats exposed to 40 days of a HF, cafeteria-style diet showed a 75% reduction in striatal D_2 receptor availability that was associated with hyperphagia. Whether hyperphagia following DIO was maintained across a variety of response requirements is unknown as food consumption was measured in a free-feed (low effort) environment.

AQ4

Research suggests that DA D_2 signaling underlies effort-based food procurement. Salamone *et al.* (1991, 1994) have shown that depleting DA in the nucleus accumbens by neurotoxic lesions reduces responding for food under FR schedules of reinforcement, and does so in a ratio-dependent manner, that is, higher FRs reduce food consumption more strongly than lower FRs. However, these same DA-depleting lesions do not reduce the amount of food consumed in a free-feeding situation (Cousins and Salamone, 1994; Cousins *et al.*, 1996; Salamone *et al.*, 2001). A similar pattern of results is observed when haloperidol, a D_2 antagonist, rather than neurotoxic lesions, is used to reduce DA activity, such that following a 0.1 mg/kg injection of haloperidol, rats

tend to consume more food that is less preferred, but freely available in the experimental chamber relative to a more preferred food that is available contingent on a FR 5 schedule (Salamone *et al.*, 1991). Therefore, accumbens-based D_2 appears to be involved in effort-based food procurement.

Another study by Soto *et al.* (2011) supports D_2 involvement in effort-based food consumption. Here, genetic variations in D_2 receptor densities affected demand for food in three different mouse strains: a D_2 knockout lean mouse (KO), which lacked D_2 receptors, a heterozygous (HET) mouse, and a wild-type (WT) mouse: the latter two had typical levels of D_2 receptors and served as controls. Across a range of FRs from 5 to 160, HET and WT mice showed similar rates of demand, reaching elasticity around 80 lever presses. However, demand curves for D_2 receptor KO mice were much steeper, reaching elasticity around FR 20. The estimated α values for D_2 receptor KO mice were higher than α values for WT and HET mice, indicating that the food had a lower essential value for the lean D_2 receptor KO mice. Thus, DA activity – especially D_2 signaling – is implicated in effort-based food procurement.

Given that reduced D_2 signaling has been shown to reduce the amount of effort that rodents will produce to gain access to food, it is possible that DIO-induced DA reductions also modulate effort-based food responding. Behavioral pharmacological procedures offer one alternative to test the role of DA in effort-based feeding by comparing behavior of HF diet exposed rats to controls following an injection of a DA antagonist and measuring the extent to which demand changes as a function of diet condition (i.e. HF vs. standard) and drug administration. Haloperidol has been shown to unmask differences in delay discounting between rats fed a HF diet and rats fed standard-chow (Boomhower and Rasmussen, 2014), although it is not clear whether effort-based responding is also relevant. Acute administration of haloperidol may allow general diet-induced changes in DA levels, which may be too subtle to detect under baseline conditions, to be unmasked.

The current study had two primary aims. First, we examined the extent to which the essential value of food changed as a result of exposure to a HF diet in rats using a demand procedure in a closed economy. Rats responded under a variety of FR schedules and the number of reinforcers earned and responses made were analyzed using the exponential-demand equation. Second, we examined the extent to which HF diet would result in dopaminergic alterations that may manifest in sensitivity to haloperidol. As such, behavior following an acute injection of haloperidol was compared between animals fed a standard diet versus a HF diet.

Methods

Subjects and diets

Male Sprague-Dawley rats ($n = 40$) were obtained from a commercial breeder (Harlan, Livermore, California, USA) at 3 weeks of age and individually housed in plexiglass, shoebox cages with unlimited access to water and food. Rats were housed in a temperature-controlled and humidity-controlled colony room and maintained on a 12-h light/dark cycle (lights on at 07:00 h). Dietary exposure was based on that described in Huang *et al.* (2006). Upon arrival, rats were randomly assigned to a HF diet ($n = 20$; 5.49 kcal/g, 36% from fat; Bioserv, Frenchtown, New Jersey, USA) or a standard-chow diet ($n = 20$; 3.0 kcal/g, 17% from fat), and remained on these diets for 15 weeks. Following this dietary exposure period, the 10 rats with the highest body mass (g) from the HF diet group and the 10 rats with the lowest body mass (g) from the standard-chow diet group were selected for operant testing (Huang *et al.*, 2006; Boomhower and Rasmussen, 2014). This standard practice (e.g. Levin and Keesy, 1998; Huang *et al.*, 2003, 2006; Johnson and Kenny, 2010; Boomhower and Rasmussen, 2014) is followed to maximize the differences in weights and D_2 in the striatum between groups (Wang *et al.*, 2001; Johnson and Kenny, 2010) and to ensure that the animals were a model of obesity, and not simply an evaluation of the effects of a HF diet, *per se*. It is common to capitalize on individual differences in this manner to establish or begin to establish animal models of various phenomena, including obesity and alcohol use disorders (see Bell *et al.*, 2006; Rasmussen *et al.*, 2016). During operant testing, rats earned all of their food during the experimental session (i.e. closed economy). The Idaho State University Institutional Animal Care and Use Committee approved all procedures.

Apparatus

Seven Coulbourn Habitest (Coulbourn Instruments, Whitehall, Pennsylvania, USA) standard experimental chambers for rats were used for data collection. Each chamber was equipped with two levers and stimulus lights situated on the right side of the chamber and 5 cm above the grid floor. Only the right stimulus light was illuminated and the right lever was active during each session. When a response contingency was fulfilled, a 45-mg grain-based Precision pellet (3.35 kcal/g; Bioserv) was dropped into a food receptacle situated between the two response levers. White noise was generated through a speaker situated on the top-right corner of the left wall. The chamber was ventilated through a 5 cm \times 5 cm fan on the top-left corner of the left wall. Each chamber was placed in a sound-attenuating cubicle. Experimental events were controlled and data were collected with a 0.01 s resolution using GraphicState software (Coulbourn Instruments) on a Windows-based computer.

Behavioral testing

Training

After the 15-week diet exposure concluded, lever-press training commenced. Lever-press training consisted of placing each rat in an operant chamber in which an FR 1 schedule of reinforcement was in effect for the right lever. After a single lever press, the house light was illuminated for 3 s and a 45 mg pellet was delivered. Once 70 responses were made, lever pressing was considered trained. Rats that did not fulfill the criterion in 3 days were trained to lever press by hand shaping (i.e. reinforcing successive approximations) until 70 responses were made independently.

Behavioral testing

Once rats were trained to lever press, demand curves were determined. In this phase, rats earned all of their daily food during 6 h experimental sessions (i.e. closed economies) that were performed 5 days/week (Sundays, Mondays, Tuesdays, Thursdays, and Fridays). On 2 days/week (Wednesdays and Saturdays), rats were allowed access to their originally assigned diet condition (standard-chow vs. HF) for 10 h. Water was freely available during the behavioral testing session. During experimental sessions, rats earned food pellets under FR schedules that increased systematically. For the first 3 weeks, an FR 5 was implemented, such that for every five lever presses, a single food pellet was delivered. In subsequent 3-week periods, the FR values increased to 15, 30, 50, and 90. Only 1 week of testing was used for the FR 150 and FR 300 contingencies as body masses decreased markedly. Free-feeding body weights were calculated on the basis of weights at the end of the 15-week diet exposure. Rats that showed weight losses to below 85% of their free-feeding weights were not exposed to subsequently higher FRs as this compromised the health of the rats. This practice was also in compliance with institutional welfare guidelines.

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Drug

Haloperidol (Sigma-Aldrich, USA) was dissolved in a 1:1:18 vehicle solution of lactic acid (Sigma-Aldrich), buffering agent, and saline (1 ml/kg). Both drug and vehicle solutions (which included the lactic acid, buffering agent, and saline) were maintained at a pH of 7. Acute intraperitoneal injections were delivered 30 min before the experimental session to ensure that the drug was behaviorally active. Rats received one dose of 0.1 mg/kg, as well as one injection of vehicle for each FR schedule tested as this dose has been shown to be behaviorally active, although not toxic, using high-effort and low-effort operant conditions (e.g. Salamone *et al.*, 1991; Rasmussen and Newland, 2001). Seven days elapsed between drug injections to minimize potential carry-over effects.

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Statistical analysis

Statistical analyses were carried out using IBM SPSS for Macintosh, version 23.0 (IBM Corp., Armonk, New York, USA). Weight gain during diet exposure was analyzed using nonlinear regression. For baseline data, reinforcers and responses for each rat under each FR were determined by averaging data from the last three sessions of each FR and compared using a two-way analysis of variance (ANOVA) with repeated measures, with FR as a within-subjects variable and dietary exposure as a between-subjects variable. Aspects of demand were quantified by fitting the exponential-demand equation to the reinforcer data (expressed as number of food pellets earned and number of kcal consumed). Differences in free-feeding food consumption (g) and estimated parameters from the demand equation were compared using independent *t*-tests. Drug effects between groups were compared using a three-way repeated-measures ANOVA, with dose and FR as within-subject variables and diet as a between-subject variable. In some instances (e.g. higher FRs), when rats' behavior was placed under higher FR schedules, but no reinforcers were earned, a 1 was added to the values (including 0 s) in that rat's data (e.g. if the rat earned 15 reinforcers under vehicle and 0 under haloperidol, we recorded it as 16 reinforcers under vehicle and 1 under haloperidol). This allowed us to not only carry out the analyses, as 0 s in the denominator yields undefined values, but to retain the relative values in the data set consistent. This method has been used with other studies that use demand procedures (Hursh, Association for Behavior Analysis International 2016 Convention).

Results

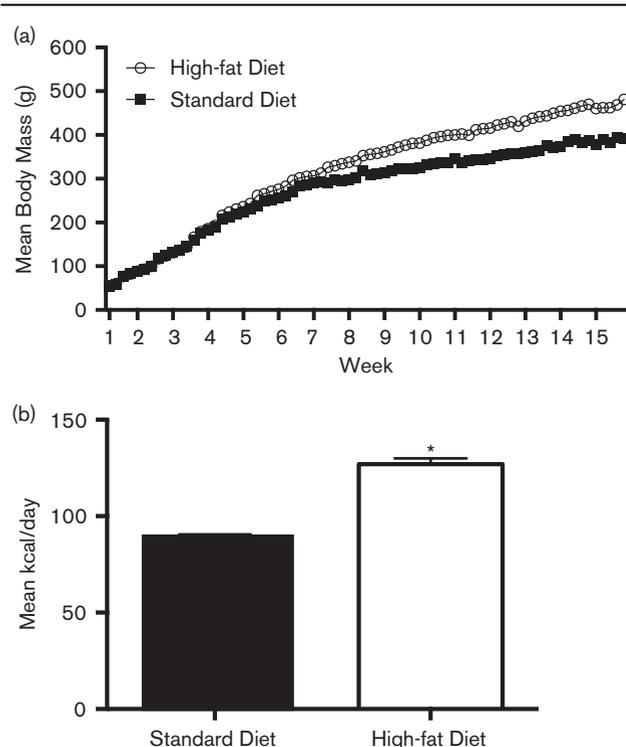
Weight gain and food intake

Weight gain was analyzed for the 20 rats that moved on to behavioral testing ($n = 10$, standard diet; $n = 10$ HF diet). Weight increased with age for both diet groups [HF diet: $F(3, 70) = 8557.00$, $P < 0.001$, $R^2 = 0.99$; standard-chow diet: $F(3, 70) = 5415.31$, $P < 0.001$, $R^2 = 0.99$] across the 15-week exposure period (Fig. 1a). By the final day of exposure, rats exposed to the HF diet showed significantly higher weights than those in the standard diet conditions [$t(38) = -2.69$, $P < 0.01$, $d = 0.87$]. The rats used in the operant portion of the study that were exposed to the HF diet also consumed significantly more calories than the rats exposed to the standard diet [$t(9.978) = -3.85$, $P < 0.01$, $d = -1.72$] (Fig. 1b).

Baseline demand

Demand curves (reinforcer data as a function of FR) are summarized as means for each group in Fig. 2. Graphs showing three representative individual animals from each group are also included. Generally, as FR increased, the number of reinforcers decreased [$F(2.10, 37.88) = 106.36$, $P < 0.001$, $\eta_p^2 = 0.85$]; however,

Fig. 1



Weight gain for rats that continued to behavioral testing ($n = 20$; a) and their caloric consumption ($n = 20$; b) for rats administered standard-chow versus a high-fat diet. (b) The mean caloric intake during the last 3 days of exposure as a function of diet. Error bars represent 1 SEM.

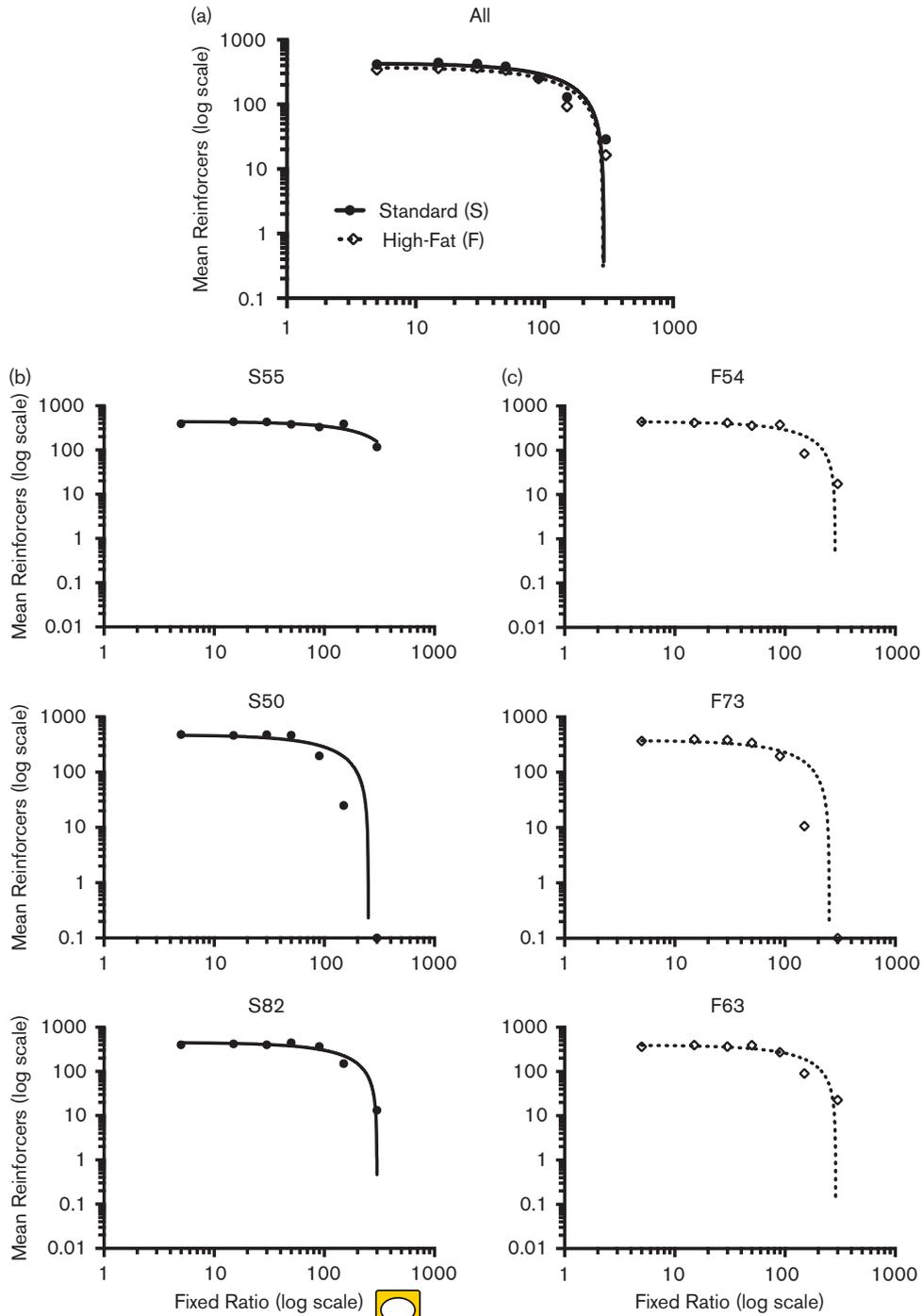
there was no significant main effect of diet ($P = 0.69$) or interaction. Data from individual animals show similar patterns. Note that S55 showed a shallow demand curve with little elasticity. Free parameter values were estimated using the exponential-demand analysis (Table 1). A significant between-groups difference in R^2 was found, but there were no differences for other estimated parameters. An exponential-demand analysis was also carried out on the reinforcer data, expressed as the number of kcal consumed at each FR (Table 1); there were no significant group differences in these data.

Response output curves are shown as a function of FR and summarized as means for each group; data from representative animals from each group are also shown (Fig. 3). There was a significant main effect of FR [$F(1.72, 30.98) = 22.97$, $P < 0.001$, $\eta_p^2 = 0.56$], as responses increased and then decreased as FR increased, but no significant main effect of diet or interaction (P 's > 0.76).

Acute drug challenge

Figure 4a and b shows the mean reinforcers earned across all FRs for standard and HF groups for both vehicle (Fig. 4a and c) and 0.1 mg/kg (Fig. 4b and d) haloperidol conditions. The dotted horizontal line refers to the baseline half- Q_0 value or the value at which half of the

Fig. 2



(a) The mean number of reinforcers earned as a function of diet. Error bars represent 1 SEM. (b, c) Representative data for individual demand curves of rats exposed to standard-chow (b) or a high-fat diet (c).

maximal reinforcers were earned [this is a value based on the mean 0.5 (Q_0) values across group in Table 1]. This line assists in observing deviations from baseline. For reinforcers, there was a significant main effect of FR [$F(6, 96) = 51.04, P < 0.001, \eta_p^2 = 0.76$] such that reinforcers decreased as FR increased, but there was no

significant main effects of drug or group. However, there was a significant FR \times diet interaction [$F(6, 96) = 2.45, P < 0.05, \eta_p^2 = 0.13$] such that the lowest number of reinforcers was earned by rats exposed to the HF diet at the highest FRs. There was also a significant

Table 1 Mean (SEM) free parameters of the exponential-demand equation fit to reinforcer and kcal earned during operant testing

	Standard	High-fat	<i>t</i> (<i>d.f.</i>)	<i>P</i> -value
Reinforcers				
O_0	526.06 (8.15)	474.52 (8.97)	-1.34 (8)	NS
α	8.50×10^{-6} (6.57×10^{-7})	5.02×10^{-6} (1.99×10^{-7})	-1.60 (10.63)	NS
k	2.74	2.74		
R^2	0.75 (0.01)	0.89 (0.005)	3.57 (18)	0.002
n	10	10		
kcal				
O_0	82.90 (1.13)	83.88 (1.95)	0.137 (18)	NS
α	6.72×10^{-5} (5.51×10^{-6})	5.15×10^{-5} (3.21×10^{-6})	-0.78 (18)	NS
k	1.95	1.95		
R^2	0.72 (0.02)	0.84 (0.01)	1.79 (14.8)	NS
n	10	10		

FR \times diet \times drug interaction [$F(6, 96) = 2.46$, $P < 0.05$, $\eta_p^2 = 0.13$] reflecting the observation that the lowest number of reinforcers was earned at the highest FRs following an injection of haloperidol in rats exposed to a HF diet. The interaction is best observed by comparing the number of bars above and below the half- O_0 line for vehicle (Fig. 4a and e) versus 0.1 mg/kg (Fig. 4b and d) haloperidol and also attending to the differences between the standard and HF diet conditions across FRs.

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Figure 4c and d shows responses. There was a significant main effect of FR [$F(6, 96) = 11.19$, $P < 0.001$, $\eta_p^2 = 0.41$]. Although there were no significant main effects of group or dose, there were marginally significant FR \times diet [$F(3.42, 54.73) = 2.30$, $P < 0.08$] and FR \times dose \times diet interactions [$F(3.42, 54.73) = 2.36$, $P < 0.07$] such that responses were lowest under the lowest and highest FRs under haloperidol and in animals exposed to the HF diet. These effects can best be observed by comparing the number of bars above and below the half- O_{\max} lines and also examining differences between the standard and HF diet conditions across FRs. O_{\max} values were calculated using the linear-elasticity demand equation (Hursh and Winger, 1995; Hursh, 2000) and represent the maximum responding during baseline for rats fed a HF diet ($M = 21842.02$, $SEM = 575.30$) and rats fed a standard diet ($M = 26068.17$, $SEM = 1793.41$). Because it is difficult to assess between-group differences in drug sensitivity from the data shown in Fig. 4, the data were converted into percent change from vehicle. These data are shown in Fig. 5. A note about analysis is warranted. Given that we used repeated-measures ANOVAs to analyze these data, only animals that completed all FR schedules under haloperidol (FR 5–FR 90) can be included in the analysis. Data from FR 150 to FR 300 are shown in Fig. 5b and d and were excluded from the repeated-measures analysis, but are addressed independently.

As Fig 5a shows, data below the 100% line indicate a haloperidol-induced reduction in reinforcers and any above the line represent an increase. Rats in the standard

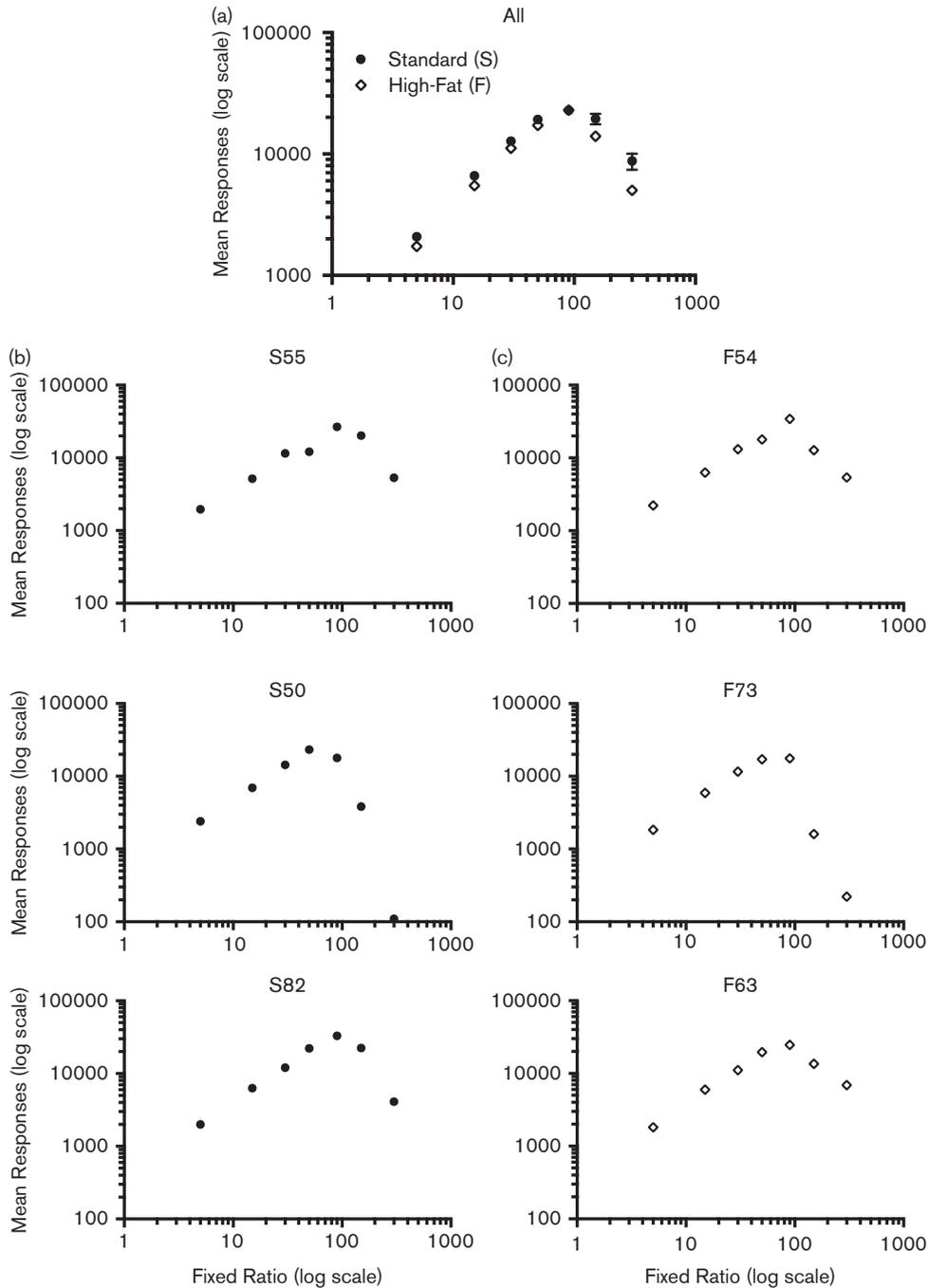
diet condition showed haloperidol-induced reductions in reinforcement across all FRs. Rats exposed to a HF diet showed lower haloperidol-induced suppression than rats in the standard diet group under FR 5–FR 50. In some cases, a haloperidol-induced increase from vehicle was observed in the HF diet rats. In other words, for the HF diet group, sensitivity and behavioral change to haloperidol depended highly on FR. A repeated-measures analysis supported a significant FR \times diet interaction [$F(4, 36) = 2.53$, $P = 0.057$, $\eta_p^2 = 0.22$]. There was no significant main effect of FR or diet.

Figure 5c shows percent change from vehicle for responses. Consistent with the reinforcer analysis, only FR 5–FR 90 were included. A repeated-measures ANOVA showed a significant main effect of FR [$F(4, 40) = 2.60$, $P = 0.053$, $\eta_p^2 = 0.22$]. In addition, rats exposed to the HF diet showed lower haloperidol-induced suppression than standard diet rats, but it depended on FR. A FR \times diet interaction trended toward significance [$F(4, 40) = 2.36$, $P = 0.07$, $\eta_p^2 = 0.21$].

Figure 5b and d also shows individual data (each datum = one rat) for FR 150 and FR 300, the schedules under which all rats did not complete the FRs. As mentioned previously, these schedule values were assessed separately because of the number of animals that were could not complete FR 150 and FR 300. For FR 150, five standard diet rats and nine of the HF diet rats completed the schedule. A χ^2 -analysis confirmed a significant group difference [$\chi^2(1) = 3.81$, $P < 0.05$]. In addition, the medians and range of the data within each group indicate robust differences in the distributions of the groups. The majority of the data (eight of nine rats) from the HF diet group showed increases in both reinforcers and responses as a result of haloperidol, whereas four of five rats in the standard diet group showed reductions in reinforcers and responses (one rat showed a 500-fold increase). Indeed, the median score for the HF diet group is 10-fold higher than the mean of the standard diet group. A statistical analysis (i.e. *t*-test) on these data would be difficult to interpret, however, given the unequal sample sizes and large differences in variance; thus, none are reported here. For FR 300, only three and seven standard diet and HF diet rats completed the schedule, respectively, and there was a trend toward a significant difference [$\chi^2(1) = 3.20$, $P = 0.074$]. The medians were similar, but as with the FR 150 data, a statistical analysis was not useful because of unequal *n*'s and unequal variances.

Because the haloperidol-induced reductions for reinforcers and responses were scaled more linearly (i.e. not logarithmically), an exponential-demand analysis is not reported.

Fig. 3



(a) The mean number of responses emitted for each fixed-ratio for rats exposed to standard-chow (closed circles) or a high-fat diet (diamonds). Error bars represent 1 SEM. (b, c) Representative data for individual animals exposed to standard-chow (b) or a high-fat diet (c).

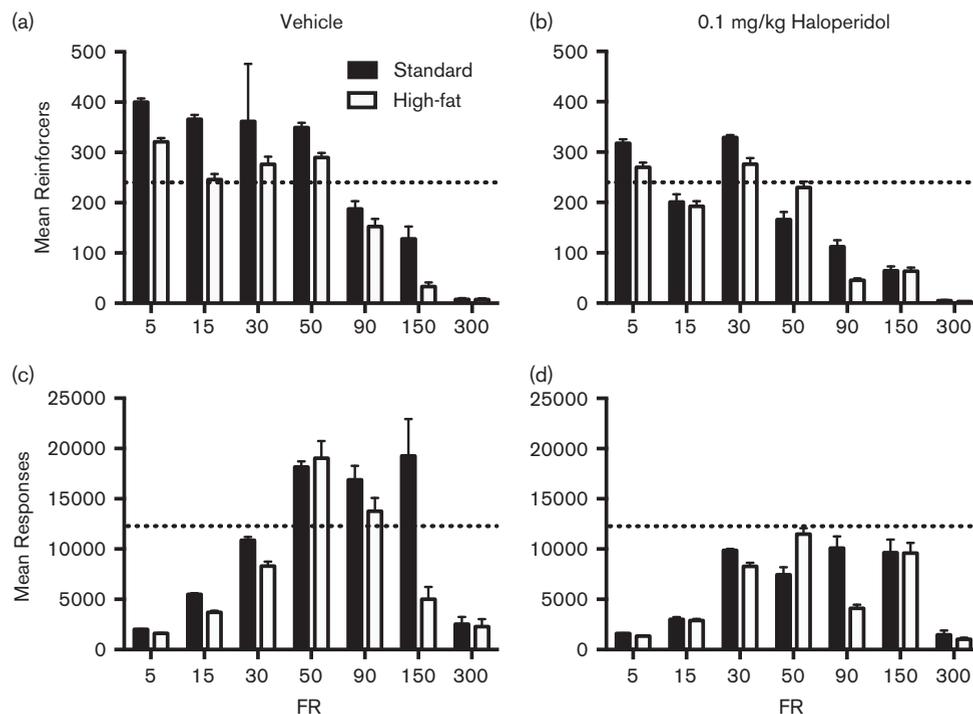
Discussion

Chronic exposure to a high-fat diet resulted in greater caloric intake and body mass than exposure to a standard diet

Rats allowed free access to a chronic HF diet versus standard-chow showed differential patterns of food consumption and weight gain. Rats administered the HF

diet consumed significantly more calories and showed significantly higher body mass than rats exposed to a standard diet. This is consistent with previous literature (e.g. Rolls *et al.*, 1980; Johnson and Kenny, 2010).

Fig. 4



(a, b) Reinforcers as a function of FR for each group. Dotted horizontal lines show baseline half- Q_0 values (241) to allow for between-dose comparisons. (c, d) Responses; the dotted horizontal line represents half- Q_{max} baseline values. The vehicle is represented in (a) and (c) and the 0.1 mg/kg dose of haloperidol in (b) and (d). Note: means in the FR 150 condition include only seven and nine rats from standard-chow and high-fat diet, respectively; the means in FR 300 include three and five rats, respectively. Error bars = 1 SEM. FR, fixed-ratio.

Baseline food consumption decreased as a function of effort requirement, although there were no diet-related differences

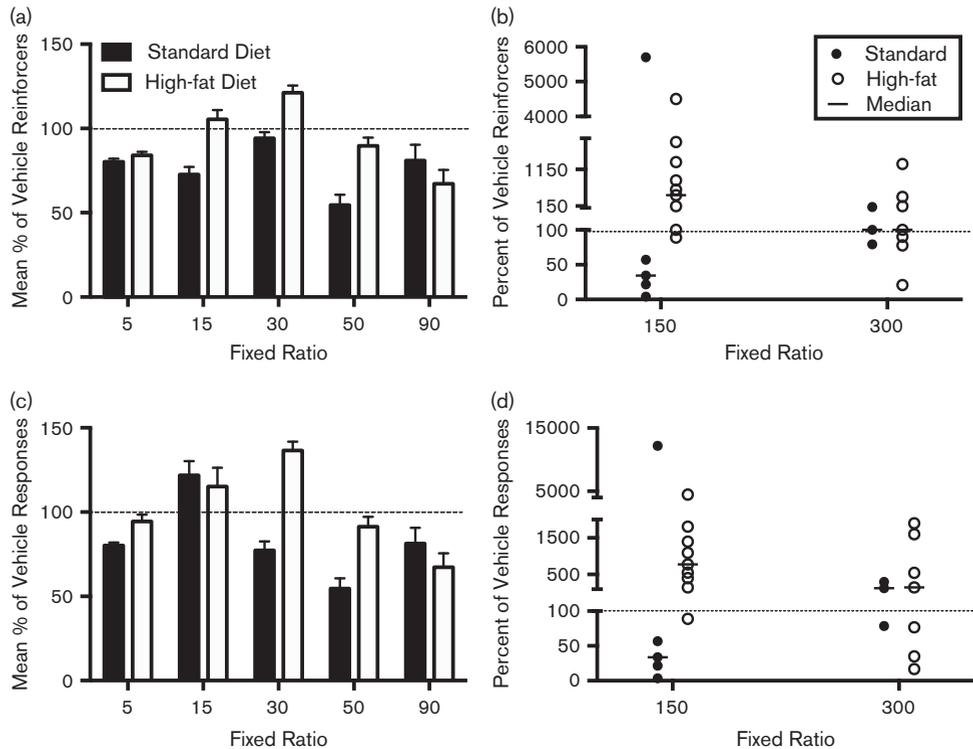
We examined the extent to which chronic exposure to a HF diet altered demand for food in a closed economy across FR values ranging from 5 to 300. At all FR values, rats in both groups emitted a similar number of responses and earned a similar number of food reinforcers (i.e. there were no group differences). The exponential-demand analysis did not yield significant group differences, but rats exposed to a HF diet showed a significantly higher R^2 than rats fed a standard diet. This finding is consistent with Rasmussen *et al.* (2010), who found that obese Zucker rats showed a significantly better fit of the exponential-demand equation than controls. Irrespective of the differences in R^2 , the fits of the exponential-demand model generally described the pattern of data for both groups well for both exponential-demand analyses (average $R^2 = 0.82$ for reinforcer analysis and $R^2 = 0.86$ for kcal analysis), but did not show any differences in behavior.

Previous literature suggests that rats will engage in hyperphagia when exposed to a chronic HF diet (Johnson and Kenny, 2010). The current study showed that when a response requirement is added to food

consumption, rats with different dietary histories show similar sensitivities to effort-based reinforcement. It is important to point out that under free-feeding conditions, rats in the HF diet group consumed 40% more calories daily than the standard-chow group (130 vs. 90 kcal, respectively). However, under the closed economy in which all food was response dependent, both groups consumed a maximum of about 90 kcal/day. One interpretation of this observation is that 90 kcal/day is an average number of kcal that a rat will defend. This interpretation is further supported by the exponential-demand analysis applied to the reinforcer data expressed as kcal earned, in that the Q_0 values are 83.26 and 87.08 for the rats fed the standard diet and the HF diet, respectively (Table 1). Other conditions that make food more readily available (i.e. the free-feed environment) or palatable (i.e. HF diet) may increase the amount consumed and promote excessive weight gain. Higher response requirements (e.g. >90 lever presses) may challenge this threshold, resulting in reductions in food consumption and weight loss.

These findings replicate aspects of Rasmussen *et al.* (2010, 2012), which examined demand for sucrose in genetically lean and obese Zucker rats. In these studies, obese Zuckers earned more reinforcers at lower FRs than

Fig. 5



(a) The mean number of reinforcers earned expressed as percent of vehicle (haloperidol/vehicle) for FR 5–90. (b) The individual data for the number of reinforcers earned expressed as percent vehicle (haloperidol/vehicle) for FR 150–FR 300. (c, d) Mean number of responses emitted expressed as percent vehicle. The dotted line shows the point at which responding under vehicle and haloperidol was equal. Bars that fall below the line indicate reductions in responding as a function of haloperidol administration and bars that fall above the line show increases as a function of haloperidol administration. FR, fixed-ratio.

lean Zucker rats defended consumption at similar rates. The finding that the Zucker and DIO models of obesity show similar patterns of demand suggests that effort required for food procurement may be an important factor in obesity. The current study also expands these papers by using a closed economy (see Hursh, 1980; Collier *et al.*, 1992) and grain-based pellets (instead of sucrose). Further, these findings support human correlational and experimental studies that show that proximity to food results in greater consumption and a higher likelihood of obesity (Wansink *et al.*, 2006; Epstein *et al.*, 2007; Hanks *et al.*, 2012).

Chronic exposure to a high-fat diet interacts with haloperidol-induced reductions in effort

An established behaviorally active, but nontoxic dose of haloperidol was used to assess the extent to which diet-induced alterations in D_2 manifested as changes to effort-based food consumption. Figure 4 shows a number of findings. First, the 0.1 mg/kg dose of haloperidol appeared to generally decrease the number of reinforcers and responses in an FR-dependent manner. For vehicle, the vast majority of reinforcers in the lower FRs (1–50)

were higher than the half- Q_0 values. Under the 0.1 mg/kg dose, however, there were fewer means above this line. Also, across the higher FRs in which the vehicle values are below the half- Q_0 line, the values for the 0.1 mg/kg dose are even lower. These haloperidol findings replicated Salamone *et al.* (1991) and also extends this work by including FRs greater than those in the original report (FR ranged from 1 to 5) as the current study included FR values up to 300.

The most notable findings, however, were found in the interactions of dietary history, response requirement, and haloperidol. Specifically, the lowest number of reinforcers appeared to be earned at the highest FRs in rats exposed to the HF diet condition. In addition, diet interacted with haloperidol and FR, such that the lowest number of reinforcers earned tended to be in the highest-effort conditions following an injection of haloperidol in rats with a history of a HF diet. Haloperidol also influenced responses emitted. In the vehicle condition, the peak responses were in the medium FR range (50–90), which replicates other behavioral-economic studies (Rasmussen *et al.*, 2010; Soto *et al.*, 2011; Rasmussen *et al.*, 2012). For most FRs, especially those in that range,

an injection of haloperidol reduced the number of responses emitted. Responses did not exceed half- O_{max} values in the 0.1 mg/kg conditions for any FR. However, these haloperidol-induced reductions resulted in only marginally statistically significant interactions, possibly because lower responses emitted at very low (e.g. FR 5) and very high (e.g. FR 300) response requirements created a floor effect.

Chronic exposure to a high-fat diet changed sensitivity to effort following an injection of haloperidol compared with rats fed a standard diet

Although the raw data suggest that haloperidol reduced reinforcers in a manner that was FR and diet dependent, it is difficult to determine how diet influenced sensitivity to haloperidol by an examination of Fig. 4. As such, drug data were analyzed as percent of vehicle (Fig. 5). Assessment of these effects statistically with a repeated-measures analysis was difficult because the number of rats in each group completing higher FRs (150 and 300) under haloperidol was reduced, making a repeated-measures analysis across all FRs challenging. We approached this problem by analyzing the data for FR 5–FR 90 using a repeated-measures analysis and examining behavior under FR 150 and FR 300 separately. The analyses showed a number of findings. First, consistent with Fig. 4, the 0.1 mg/kg dose of haloperidol generally decreased the number of reinforcers and responses for rats fed a standard diet. The single exception to this finding is that rats fed a standard diet showed a higher level of responding, but not reinforcers, at FR 15 under haloperidol. These haloperidol findings from the standard-chow group replicate previous literature that has documented decreases in DA D_2 by genetic (Soto *et al.*, 2011), pharmacological (Salamone *et al.*, 1991), and neurotoxic lesion (Cousins and Salamone, 1994; Cousins *et al.*, 1996; Salamone *et al.*, 2001) in standard rat models resulting in a reduction in effort-dependent food consumption.

Rats exposed to a HF diet, however, showed an altered sensitivity to haloperidol compared with the rats fed standard-chow. At most FRs (FR 5–FR 50, but not FR 90), rats exposed to the HF diet showed a lower haloperidol-induced reduction in reinforcers than rats fed a standard diet and in a number of cases haloperidol actually increased the number of reinforcers earned in this group relative to vehicle. This pattern also suggests the possibility that the effects of haloperidol on effortful responding documented in other studies (i.e. Salamone *et al.*, 1991) depend highly on diet and FR value.

Data from the haloperidol sessions from FR 150 and FR 300 schedules add to these findings. First, significantly more rats from the HF diet condition completed the FR 150 schedule than the rats fed a standard diet and this trend was observed for FR 300. Second, for four of the five rats that completed FR 150, haloperidol decreased

responding and reinforcers earned at these schedule values for rats fed a standard diet; however, haloperidol tended to increase responding for eight of the nine rats fed a HF diet. These differences support the repeated-measures analysis and offer some additional evidence that rats exposed to HF diets show a lowered sensitivity to haloperidol (in terms of its reducing effects) that interact with effort.

The altered sensitivity to haloperidol in rats exposed to a HF diet (compared with controls) extends the literature that chronic exposure to a HF diet leads to a reduction in D_2 and changes in food consumption in free-food intake, often interpreted as a ‘blunting’ of food reward processes (Geiger *et al.*, 2009; Johnson and Kenny, 2010; Vucetic *et al.*, 2012). This interpretation would support the conclusion that HF diet alters the D_2 receptor in a manner that changes the rewarding properties of food; however, the findings in this study suggest that a reduction of D_2 activity may enhance the reinforcing properties of food under some effort-based conditions and reduce them in others, although they are reduced to a lesser extent than controls.

Despite the strengths of this study, there were also some limitations. First, a closed economy with the same food type was necessary to examine a range of possible differences in elasticity. The inherent limitation with this methodological setup is that the earlier diet exposure effects could be ‘washed out’ during operant testing. To accommodate this problem, we continued some exposure to diet by allowing rats to come into contact with their respective diets twice per week. Although it would be ideal to allow the animals to earn their daily diets in operant sessions across the entire study, the differences in macronutrient content and palatability of these pellets would confound interpretation of the results. Future research could investigate this question by manipulating dietary exposure duration between demand curve sessions or by using open economies, in which rats are allowed outside access daily to their respective diets. This is complicated, however, because caloric intake must be controlled and the use of an open economy reduces elasticity for food. A second limitation is that we used only a single dose of haloperidol to test differences in sensitivity. Although this dose of haloperidol has been used as an effective dose in a number of studies (Salamone *et al.*, 1991, 1994; Randall *et al.*, 2012) and allowed a diet-related interaction to be observed, a fuller characterization using smaller and larger doses would provide more dose–response information and may improve the ability to detect altered sensitivity to dopaminergic function.

Conclusion

Despite these limitations, the data in the present study have implications for public health. Food environments often have HF foods, such as fast food or highly

processed food, available at low-effort and monetary costs (Drewnowski and Darmon, 2005). These data suggest that rats exposed to a HF diet will defend consumption of food at similar rates as rats exposed to standard-chow, but that HF diet can alter sensitivity to food reward when unmasked with a D₂ compound at response requirements that vary. Nonetheless, one way to reduce overeating and promote healthy food choices may be to make HF foods less convenient and more expensive than healthier alternatives to promote healthy food choices (Wansink *et al.*, 2006; Epstein *et al.*, 2007; Hanks *et al.*, 2012).

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Conflicts of interest

There are no conflicts of interest.

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