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Nucleus accumbens shell moderates preference bias during voluntary choice behavior

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Abstract

The nucleus accumbens (NAc) shell lies anatomically at a critical intersection within the brain's reward system circuitry, however, its role in voluntary choice behavior remains unclear. Rats with electrolytic lesions in the NAc shell were tested in a novel foraging paradigm. Over a continuous two-week period they freely chose among four nutritionally identical but differently flavored food pellets by pressing corresponding levers. We examined the lesion's effects on three behavioral dynamics components: motivation (when to eat), preference bias (what to choose) and persistence (how long to repeat the same choice). The lesion led to a marked increase in the preference bias: i.e., increased selection of the most-preferred choice option, and decreased selection of the others. We found no effects on any other behavioral measures, suggesting no effect on motivation or choice persistence. The results implicate the NAc shell in moderating the instrumental valuation process by inhibiting excessive bias toward preferred choice options.

Key words: mesolimbic reward pathway; foraging decision-making behavior; motivation; neurobiology of reward valuation; inhibitory control

Introduction

Reward is a critical driving force of behavior, and many psychological and neural disorders stem from dysfunction in reward circuitry. A pivotal pathway implicated in reward processes is the mesolimbic reward pathway, which includes a major dopaminergic neuronal projection from the ventral tegmental area (VTA) of the midbrain to the nucleus accumbens (NAc) in the ventral striatum of the basal ganglia. The NAc consists of a *core* and surrounding *shell* region, each with different afferent and efferent connections as well as distinct functions (Groenewegen *et al*., 2004 Dalley *et al*., 2011). The NAc shell in particular appears to be ideally situated anatomically to play a key role in reward-related processes, with principal afferents from VTA, thalamus, amygdala, hippocampus and prefrontal (i.e., infralimbic) cortex, and principal efferents to VTA, lateral hypothalamus and ventral pallidum (that in turn projects back to prefrontal cortex via thalamus) (Groenewegen *et al*., 1999; Kelley, 2004; Dalley *et al*., 2011).

Functionally, the NAc shell has been implicated in Pavlovian and instrumental behavior (Berridge *et al*., 2010; Berridge and Kringelbach, 2013). For the Pavlovian system, multiple studies have shown that the NAc shell mediates reward prediction, incentive

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salience of cues associated with reward (leading to increased behavioral output in the presence of these cues), and hedonic impact of rewards (Johnson *et al.*, 1996; Zhang *et al.*, 1998; Basso and Kelley, 1999; Zhang and Kelley, 2002; Zhang *et al.*, 2003; Pecina and Berridge, 2005; Woolley *et al.*, 2006; Wassum *et al.*, 2009; Berridge *et al.*, 2010; Smith *et al.*, 2011; Berridge and Kringelbach, 2013). For hedonic impact, studies have shown that the NAc shell mediates likeability, including a specific effect on taste (Basso and Kelley, 1999; Zhang and Kelley, 2002; Zhang *et al.*, 2003; Woolley *et al.*, 2006).

Less is known about the relationship of the NAc shell to instrumental behavior. There is evidence for NAc shell influence on instrumental behavior mediated through Pavlovian incentive salience, termed Pavlovian-Instrumental Transfer (PIT) (Wyvell and Berridge, 2000; Corbit *et al.*, 2001; Corbit and Balleine, 2011; Pecina and Berridge, 2013). Moreover, as opposed to a general PIT effect in the NAc core region—in which previous stimulus-reward pairings with *any* reward type can enable a stimulus to enhance instrumental responding for a given reward—the PIT effect in the NAc shell appears to be specific—requiring the previous stimulus-reward pairing to include the same reward type as the current one (Corbit and Balleine, 2011). Thus, the NAc shell appears to track specific stimulus-reward pairings, which can in turn influence instrumental behavior.

There is some evidence for involvement in the valuation process that drives instrumental behavior, such as for magnitude processing (Stopper and Floresco, 2011). However, the NAc shell appears to be more involved in such processing in risk-based scenarios in which the outcome varies across trials between receiving a large reward or nothing at all. In these cases, the NAc shell promotes a 'risky' preference for the variable option with a larger reward over a certain option with a smaller reward when the former entails the larger overall value (Stopper and Floresco, 2011; Sugam *et al.*, 2014). Closer analysis suggests that this risk preference may derive from NAc shell promotion of a win-stay, lose-shift behavioral strategy (propelled by the impact of the larger payoff on the win-stay component) (Stopper and Floresco, 2011).

This evidence for NAc shell involvement in instrumental valuation and behavior leaves multiple questions unanswered. For example, to what extent does the NAc shell promote stay-versus-switch behavioral strategies (i.e., beyond risk-based scenarios)? To what extent is the NAc shell involved in the valuation of basic reward parameters that drive instrumental behavior? Moreover, if involved, how exactly would the valuation influence instrumental behavior?

The NAc shell's causal role in instrumental valuation and behavior also remains unclear: i.e., whether being directly involved in the valuation process or having secondary control over it, and if the latter, whether as a promoter or inhibitor. Although most findings for Pavlovian effects appear to suggest enhancement of reward efficacy (Berridge *et al.*, 2010; Berridge and Kringelbach, 2013), actual NAc shell neuronal activity is often unknown; and some evidence has shown an inhibitory relationship with, for example, lower-level feeding circuits (Stratford and Kelley, 1997; Taha and Fields, 2006).

To address these questions regarding the potential relationship of the NAc shell to the instrumental behavioral system, we targeted a shell 'hotspot' for reward processing in mammals (Smith *et al.*, 2011; Berridge and Kringelbach, 2013). We induced electrolytic lesions in the NAc shell of rats to test for causality and to produce a clear direction of effect—a loss of NAc shell neural activity. We tested the rats in a novel naturalistic foraging task, in which they were housed in an operant chamber for a continuous 2-week period, i.e., a 'closed economy' (Atalayer and Rowland, 2009), and allowed to make choices freely among four nutritionally identical but differently flavored food pellets by pressing a corresponding lever for each flavor. We then examined the temporal and sequential dynamics of the foraging behavior that reflect two key behavioral components: when and what to eat (Jung *et al.*, 2014). We found that the NAc shell lesion selectively changed the bias across the distribution of choices, with the favorite item being selected more and the others less, while all other behavioral components measured remained unchanged. To better characterize the biasing effect, we developed two novel indices that disentangled choice option preferences and sequential choice dependencies, which provided further evidence that the NAc shell lesion affected actual choice preferences.

Materials and methods

Subjects

Fifteen 7-week-old male Sprague-Dawley rats (Koatec, Pyeongtaek, South Korea) were used for the study. All animal care and experimental procedures were performed according to the KAIST guidelines for the care and use of laboratory animals and approved by the KAIST Institutional Animal Care and Use Committee.

Surgery

Because the NAc shell is closely located to other subcortical structures, especially the NAc core, we used the electrolytic lesion, which enabled a precise focal lesion, with the caveat that any structure within this lesion would be damaged, including potential passing fibers. Histological analysis showed that focal lesions within the NAc shell were indeed made (described below). Rats were pseudo-randomly grouped into the electrolytic lesion ($n = 8$) and sham-operated groups ($n = 7$). The electrodes consisted of twisted stainless steel wires and were implanted bilaterally into the shell of the NAc (AP: +1.4 and AP: +2.0, for each electrode in a hemisphere; ML: ± 0.8; DV: −6.5, measured from the brain surface), for a total of four. For the lesion group, the implanted electrode was connected to a stimulus isolator (World Precision Instruments), and a 250 μA current was applied for 10 s to damage neurons in the shell of the NAc. The electrodes were removed a few minutes later. For the sham group, the same surgical procedure (see Supplementary Information) was performed as the lesion group, except no current was applied.

Behavioral apparatus, procedure and task

After one week of recovery from the surgery, the rats were individually housed in an experimental operant chamber and maintained on a 12-h light/dark cycle with *ad libitum* access to water for 2 weeks (see Supplementary Information) (Figure 1A). The rats freely chose and ate among four nutritionally identical but differently flavored 45 mg pellets: chocolate, banana, coffee, and cinnamon (Bio-Serv, Frenchtown, NJ, USA). No other food was available in the chamber, requiring the subjects to obtain all food via the closed economy, i.e., from their efforts. A trial began by breaking the photo-beam with a nose-poke and four levers were extended into the chamber on the opposite wall. Once the rats pressed a lever, a flavored pellet assigned to the chosen lever was delivered and then all levers were retracted. The levers corresponding to specific flavors were pseudo-randomly determined across rats. Thus, the task is considered as an unforced task, in that the rats learned to obtain food spontaneously, and all rats used in the experiment learned to obtain pellets within 3.5 days on average. This paradigm produced minimal intervention by the experimenter, it allowed us to

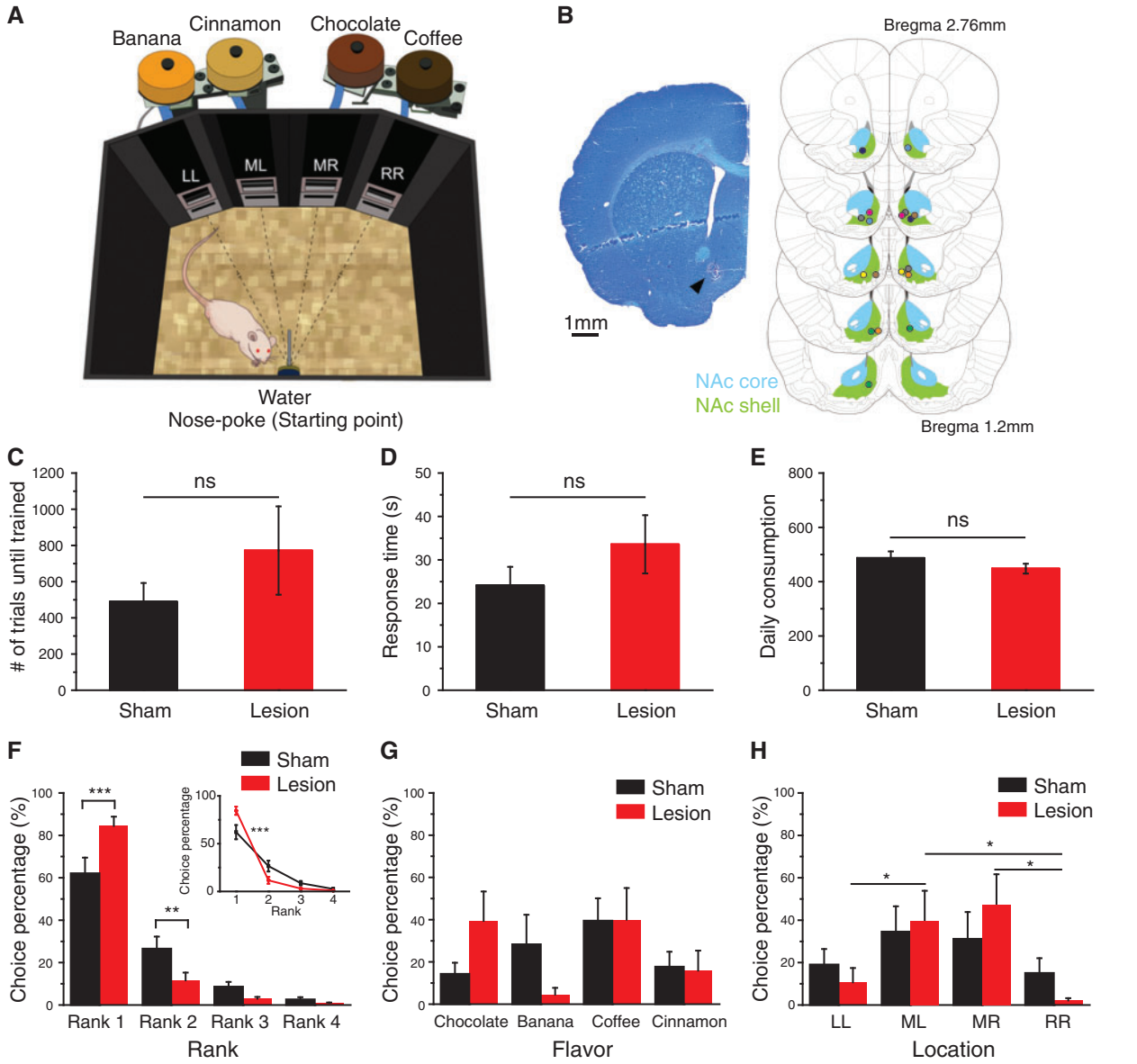

Fig. 1. Preference bias is enhanced in the NAc lesion group. (A) Schematic of the experimental setup. The custom-made operant chamber was constructed with four levers (retracted in the diagram) connected to four different pellet dispensers for the four flavors, as well as a nose-poke opening and sensor on the opposite wall to monitor consummatory behavior for two weeks. The levers were equidistant from the nose-poke sensor. A water container (spout shown) was mounted above the nose sensor, and beta chip bedding covered the floor. (B) Histological results of the NAc shell lesions. Photograph showing a coronal section of a representative shell lesion of the NAc (left). The triangle indicates the lesion. Locations of lesion from all rats in the NAc lesion group are schematically represented with circles color-coded by subject (right). The NAc core and shell are highlighted in blue and green, respectively. (C–E) Comparison of general performance between the lesion and sham groups. Trials to learning criterion (C), response time (D), and daily consumption (E) of the NAc lesion and sham groups. (F–H) Comparison of the preference bias between the NAc lesion and sham groups. Choice percentages with respect to rank (F), flavor (G), and location (H) are shown. (F inset) Interaction between rank and choice percentage is also shown. The values are mean ± SEM. ns: not significant; *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.

observe naturally emerging choice patterns, and importantly, it mimicked everyday, uninterrupted foraging behavior. After the experiment was completed, histology was conducted as described in the Supplementary Information.

Data analysis

Pressing a lever within 5 s after a nose poke was considered a successful trial. We used the 5 s response time requirement to differentiate intentional choices from unintentional lever presses. Rats were considered to have learned the task once the successful trial rate in the last 20 trials exceeded 90%. All animals met the task-learning criterion in the first 6 days. We analyzed the choice behavior data of the last 8 days of the 2-week task period, which is the period where all animals met the learning criterion. We first examined whether the lesion impaired general learning and task performance by comparing multiple factors such as daily consumption between sham and lesion groups using the Mann-Whitney Test.

For the sequential dynamics, we focused on two key features: (1) the choice distribution, measured as the *preference bias*, i.e., the degree of preference toward the most preferred option; and (2) the tendency to repeat a previous selection, measured with *choice persistence*. For preference bias, we examined rank, flavor and location,

using two-way ANOVAs with group (lesion or sham) as one factor with subsequent post-hoc tests. We defined *rank* as the order of the choice options to capture the rats' individual preferences for different flavors from most to least (i.e., rank 1 was the most consumed flavor, rank 2 the next most, etc.). For the second sequential component, choice persistence, to capture the nature of 'stay' and 'switch' behavior quantitatively, we first defined a *run* as a series of consecutive identical choices. We then assessed the sequential dependency by comparing the empirical run distributions with (a) randomly shuffled and (b) entirely sorted ones (e.g., comparing ACACABADBB to AAAABBBCCD) using Monte Carlo hypothesis testing (see Supplementary Information). We then examined each rank separately comparing the groups using the Mann-Whitney test.

For the temporal dynamics, we examined three features: (1) variation in the eating pattern during a day, by comparing the groups with *t*-tests and a two-way repeated measures ANOVA (across the day), (2) period of the eating pattern, by comparing the periods of the autocorrelograms of both groups using a *t*-test, and (3) burstiness of eating actions. For burstiness, we first calculated the inter-choice intervals (ICIs, i.e., the time latency between consecutive choices) for the lesion and sham groups, and then compared the slopes of the cumulative ICI distributions in a log-log scale using a *t*-test.

Finally, because a given choice distribution could reflect actual choice option preferences or sequential choice dependencies we developed two novel indices to isolate the effects: the *B-index* and *P-index*, respectively (see Supplementary Information). To further assess the ability of the *B-* and *P-indices* to discriminate the lesion and sham groups, we applied receiver operating characteristic (ROC) analysis.

Results

The NAc shell lesion led to an increase in the preference bias

After the animals recovered from surgery, they were individually housed in a custom-designed operant chamber and their foraging behavior was monitored for 2 weeks (Figure 1A). To obtain food pellets, the rats needed to learn to nose-poke in the nose-poke well when lit and then press one of the four levers on the corresponding wall within 5 s. All rats spontaneously learned the task, meeting the task-learning criterion within 6 days. Upon completion of the experiment we confirmed bilateral focal lesions in the NAc shell with microscopic histological examination (lesion size $= 0.213 \pm 0.019\,mm^2$ for 8 rats, Figure 1B). We then tested whether the lesion impaired learning and task performance by comparing the number of trials to reach the learning criterion, response time (i.e., time interval between nose pose and lever press), and daily consumption between the lesion and sham groups (Figure 1C–E). We found no significant differences between the lesion and sham groups (Mann–Whitney Test, learning rate, $P = 0.563$; daily consumption, $P = 0.165$; response time, $P = 0.298$), indicating that the NAc shell lesion did not significantly impair operant learning, performance and daily food consumption.

To assess the sequential dynamics, we first examined how strongly choices were biased with respect to rank, flavor and location (Figure 1F–H). There was a significant main effect of rank ($F_{3,52} = 141.648$, $P < 0.001$, Figure 1F) and a significant interaction between group and rank ($F_{3,52} = 8.218$, $P < 0.001$, Figure 1F inset). For both groups, the choice percentage of rank 1 was significantly different from the choice percentage of the other ranks (Post-hoc tests, $P < 0.001$), indicating a strong bias towards the favorite flavor. However, the lesion group exhibited a significantly higher bias toward the favorite flavor compared with the sham group, with a higher choice percentage of rank 1 ($F_{1,52} = 16.288$, $P < 0.001$) and lower choice percentage of all other ranks (rank 2: $F_{1,52} = 7.203$, $P < 0.01$; rank 3: $F_{1,52} = 1.060$, $P = 0.31$; rank 4: $F_{1,52} = 0.104$, $P = 0.75$, Figure 1F).

For the specific flavors (regardless of rank), we found no main effect on the choice percentage ($F_{3,52} = 1.910$, $P = 0.139$) and no interaction between group and flavor (two-way ANOVA, $F_{3,52} = 1.584$, $P = 0.204$, Figure 1G). For location, there was no interaction between group and location ($F_{3,52} = 0.744$, $P = 0.531$), but there was a main effect of the four locations of left (LL), middle left (ML), middle right (MR) and right (RR) on choice percentage (two-way ANOVA, $F_{3,52} = 4.070$, $P < 0.05$), and post-hoc pairwise comparisons revealed a significant difference in the choice percentage between center locations and side locations (LL-MR, MR-RR, ML-RR, $P < 0.05$) only in the lesion group ($F_{3,52} = 4.363$, $P < 0.01$). However, there was no difference in the choice percentage for each location between the lesion and sham groups (*t*-test, $P = 0.4171$ for LL, $P = 0.7941$ for ML, $P = 0.4138$ for MR, and $P = 0.071$ for RR, Figure 1H). To address whether the location effect might have generated the rank order effect, we compared the choice percentages of the highest rank and the most frequently chosen location. We found significantly different intakes for rank 1 and the MR location (*t*-test, $P < 0.05$). This result suggests that the preference bias toward an individual's favorite flavor did not appear to be due to a preference for the center locations.

We next examined the second main sequential dynamics component: choice persistence. Animals with the lesion appeared to exhibit longer runs than the sham animals (Figure 2A). However, longer runs could be due to greater choice persistence or simply the higher preference bias found in the lesion group. To distinguish these possibilities, we calculated the cumulative distribution of runs, defined as the probability of runs longer than a given run length (i.e., the survival function). Both the lesion and the sham groups exhibited heavy-tail distributions in a log-log scale, indicating that the choice pattern consisted of a large number of short runs and a few very long runs (Figure 2B and C). We next compared the empirical choice sequences to randomly shuffled ones (and thus sequential dependencies removed) and found that the cumulative run distribution of the empirical data significantly deviated from the randomly shuffled ones for all subjects in both the lesion and sham groups (Monte Carlo hypothesis testing, $P < 0.001$) (Figure 2B and C). Thus, the choice behavior of the rats was highly dependent on the previous choice histories regardless of the NAc shell lesion (Lau and Glimcher, 2005).

To assess the degree of choice persistence, we next compared the empirical sequences to fully sorted sequences (e.g., ACACABADBB to AAAABBBCCD). The cumulative run distributions of the empirical data were significantly different from that of the sorted choice sequences for all subjects in both the lesion and sham groups (Monte Carlo hypothesis testing, $P < 0.001$) (Figure 2B and C). Thus, both the lesion and sham groups showed a moderate degree of persistence in a range between completely random and completely perseverant.

Next, to assess each rank separately, we compared the cumulative run distributions for each rank with those of a randomly shuffled choice sequence (Figure 2D and E). In the lesion group, for all ranks except the lowest rank (rank 4), the cumulative run distribution deviated from the randomly shuffled one. The deviation for the lowest rank was not clear in the lesion group due to the low choice frequency for rank 4 (average 29.75

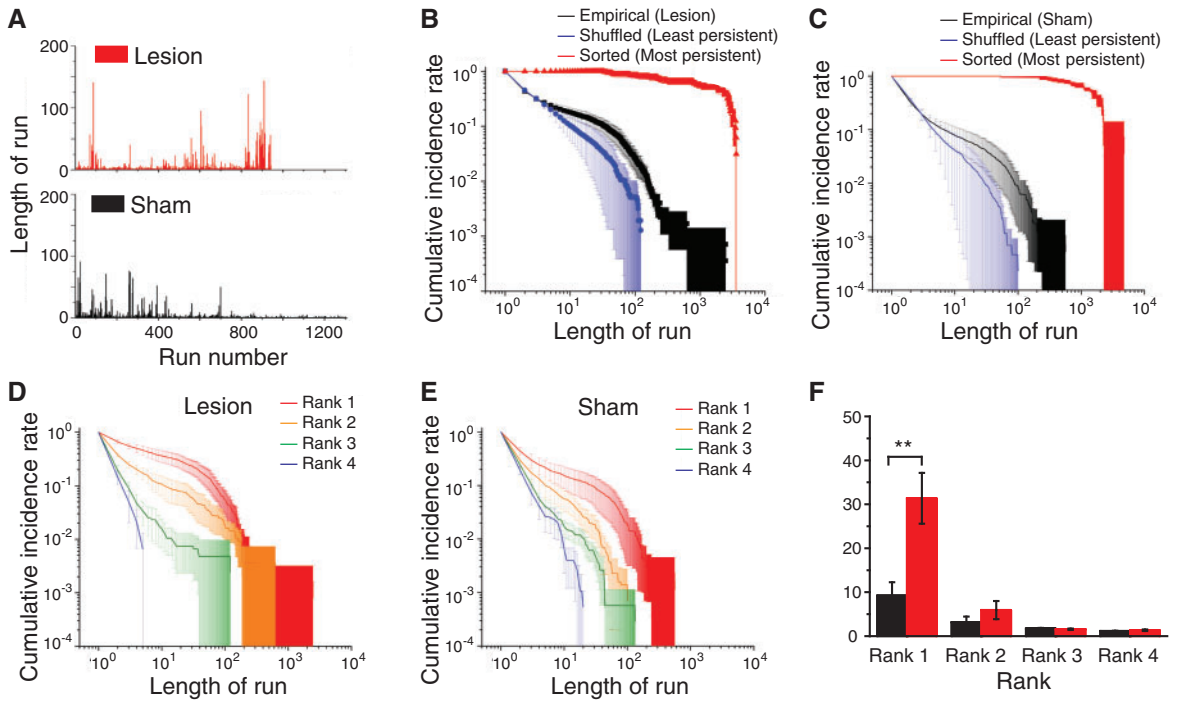

Fig. 2. Comparison of the sequential features of choice behavior between the NAc and sham groups. (A) The length of runs (i.e., a sequence of consecutive identical choices) for representative animals in the lesion (top) and sham (bottom) groups. In both groups, short runs were abundant while a few long runs were intermittently observed in a periodic manner. (B, C) The cumulative run distributions in a log-log scale for the entire choice sequence for the lesion (B) and sham (C) groups. Black, blue, and red lines represent the empirical, randomly shuffled (least persistent possible), and sorted choice (most persistent possible) sequences, respectively. In both groups, the distributions of empirical choice sequences were similarly located between shuffled and sorted sequences. (D, E) Log-log representation of the cumulative distributions of runs for each rank for the lesion (D) and sham (E) groups. Red denotes runs from rank 1; orange from rank 2; green from rank 3; and blue from rank 4. (F) Comparison of mean run lengths with respect to rank order between the lesion (red) and sham (black) groups. $^{**}P < 0.01$.

choices during experiment, less than 1% of total number of choices), i.e., a possible floor effect. In the sham group, for all ranks, the cumulative run distributions deviated from the randomly shuffled one. This deviation from the randomly shuffled sequences indicates that the animals in both groups were likely to repeat their previous action regardless of rank (except rank 4 in the lesion group), leading to the generation of long runs.

Although the cumulative run distributions for all ranks (except rank 4 in the lesion group) also showed a heavy tail and deviated from randomly shuffled ones in both groups, the thickness of the tails for each rank appeared to decrease in accordance with rank order. Indeed, the mean length of runs decreased across rank (Figure 2F). Moreover, the lesion group showed significantly longer runs for rank 1 than the sham group (Mann–Whitney test, $P < 0.05$). However, the relationship between rank order and run lengths suggests that preference bias in choice behavior may significantly contribute to run length, which we examine further below.

The NAc shell lesion did not affect the temporal dynamics

We assessed the temporal dynamics via three key features: (1) daily eating pattern variation, (2) period of the eating pattern, and (3) burstiness of eating actions. For the first, we found no significant differences in food consumption between the groups during the light and dark cycles (t-test, $P = 0.19$ for both comparisons, Figure 3A). Both groups showed a significantly greater intake during the dark cycle (12 p.m. to 12 a.m.) than during the light cycle (12 a.m. to 12 p.m.) (paired t-test, $P < 0.01$ lesion group, $P < 0.001$ sham group). Food consumption across the entire day did not differ between the two groups (Figure 3B, two-way repeated measures ANOVA, $F_{Group} = 0$, $P = 1$). For the eating pattern period, we extracted the pitch of the average autocorrelogram (Figure 3C). Both the lesion and sham groups exhibited a time interval between peaks approximately equal to 24 hours, consistent with their circadian rhythm (t-test, $P = 0.241$, Figure 3D). Thus, the lesion did not appear to impair the general circadian rhythmic foraging patterns of the animals. For burstiness, we compared the inter-choice interval (ICI, i.e., the time latency between consecutive choices) distributions between the lesion and sham groups. The time interval between actions has been suggested as an indicator of motivational state and of a Pavlovian salience effect on behavior (Robbins and Everitt, 1996; Salamone *et al.*, 2003; Niv *et al.*, 2006; Zanutto and Staddon, 2007; Jung *et al.*, 2014). In both groups, the majority of ICIs were short, but extremely long ICIs also sporadically occurred, indicating that there were bursts of activity separated by relatively long inactive periods (Figure 3E). The cumulative ICI distributions of both groups were highly right-skewed, exhibiting similar degrees of heavy tails in a log-log scale (slope: -1.97 ± 0.04 for the lesion group, -1.91 ± 0.03 for the lesion group; t-test, $P = 0.27$; Figure 3F). The close agreements between the lesion and sham groups in both the periodicity of foraging behavior and the heavy-tailed ICI distributions indicate that the lesion did not significantly alter the temporal dynamics of the foraging behavior.

Preference bias and choice persistence indices show that the NAc shell lesion affected the preference bias only

The lesion group showed a stronger preference bias and longer runs for the preferred option (rank 1), however, we cannot as

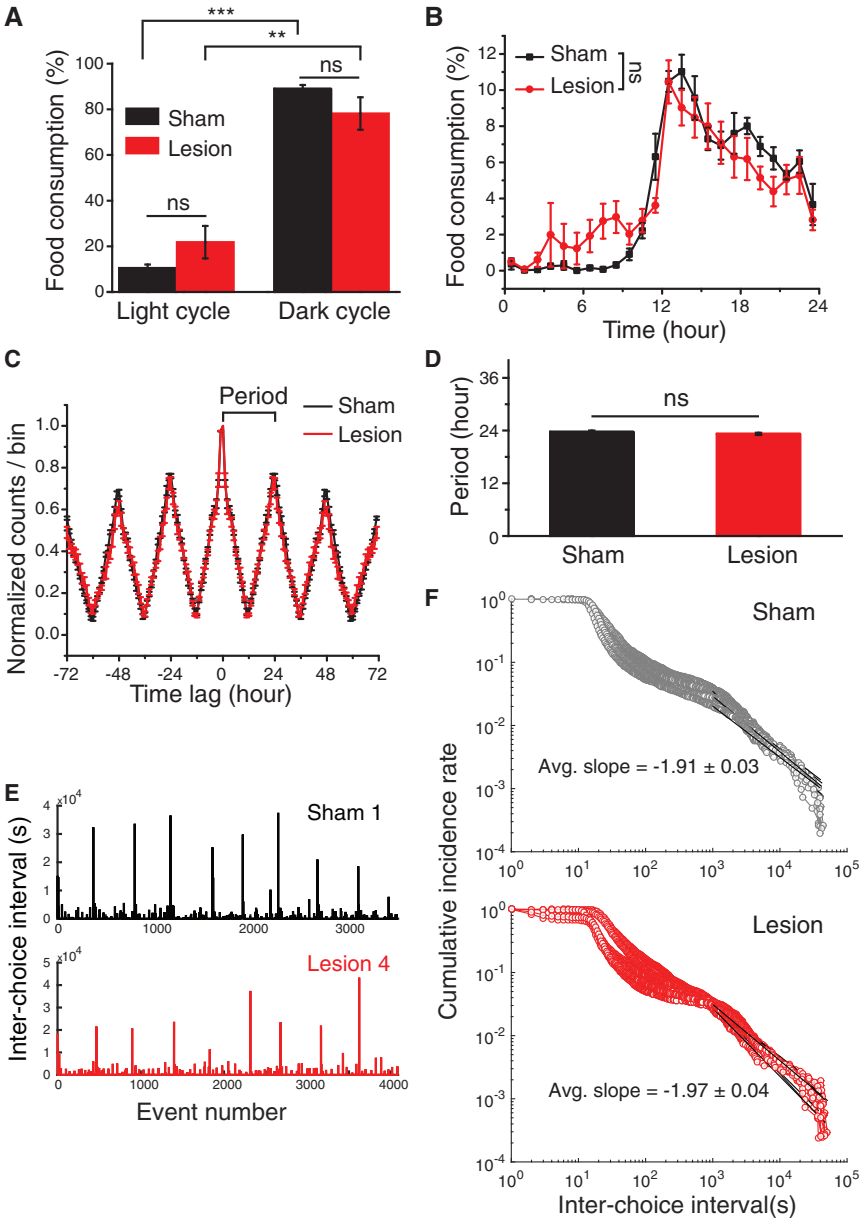

Fig. 3. Comparison of the temporal features of feeding behavior between the NAc lesion and sham groups. (A) Food consumption during light and dark cycles. (B) The hourly variation of normalized food consumption across the day. (C) Autocorrelogram of feeding behavior for the lesion and sham groups. (D) Period of feeding behavior. (E) Example inter-choice interval (ICI) patterns of lesion (top) and sham (bottom) groups. In both groups, short ICIs were abundant while a few long ICIs were intermittently observed in a periodic manner. (F) Cumulative ICI distributions of all rats in the lesion and sham groups in a log-log scale. The black solid lines denote the slope of each distribution. For all figures, error bars are standard errors of the mean (SEM). ns: not significant; $^{**}P < 0.01$; $^{***}P < 0.001$.

yet rule out the possibility that the longer runs in the lesion group resulted from the more biased choice behavior. To clarify the lesion effects on the preference bias versus the choice persistence, we developed the *B-* and *P-indices* (Figure 4A). Corroborating our previous findings, we found a significant difference in the *B-index* between the lesion and sham groups (Mann–Whitney *U* test, $Z = -2.662$, $P < 0.01$). Thus, rats with NAc lesions were indeed more likely to choose their favorite option. For the *P-index*, however, there was no significant difference between the lesion and sham groups (Mann–Whitney *U* test, $Z = -0.926$, $P = 0.397$). Thus, the lesion did not appear to significantly influence choice persistence.

To further assess the ability of the *B-* and *P-indices* to distinguish the lesion and sham groups, we applied receiver operating characteristic (ROC) analysis. Discriminability was determined by computing the area under the ROC curve, with 1.0 being perfect separation of the lesion and sham groups, and 0.5 being no separation of the groups. The area for the *B-index* was 0.911 and for the *P-index* 0.643, indicating that the *B-index* discriminated the lesion and sham groups to a high degree, whereas the *P-index* performance was poor (Figure 4B). Finally, to provide a further diagnostic measure, we obtained the optimal threshold value of the *B-index* for discriminating between the groups from the ROC analysis, which was equal to 0.35839. This optimal value remained highly effective at classifying 12 normal rats used in our previous study under the same experimental paradigm (green dots in Figure 4A) (Jung *et al.*, 2014).

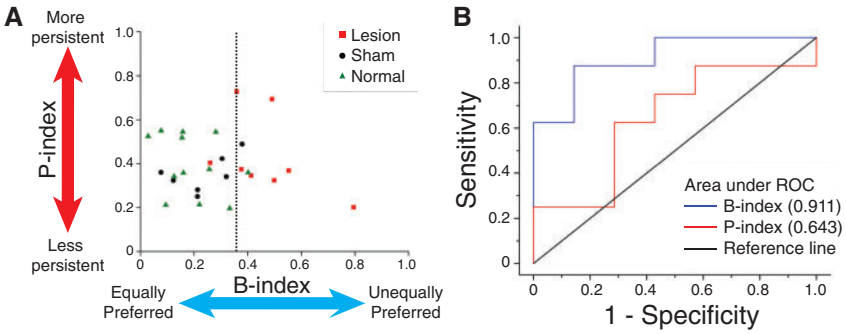

Fig. 4. Quantitative comparison of the preference bias and the choice persistence based on the *B* and *P indices* between the lesion and sham groups. (A) A plot of the *B-index* (*x*-axis) and *P-index* (*y*-axis) of the lesion (red), sham (black), and normal control (green) groups, representing the degree of preference bias and choice persistence, respectively. (B) ROC curves for the *B* and *P indices*, showing the proportion of correct classifications (sensitivity) of lesion (blue) and sham (red) group rats at different false-positive rates (*1 - Specificity*). The numbers inside the legend indicate the area under the ROC curve for the corresponding index, which reflects classification ability, with 1.0 being perfect separation of the two groups, and 0.5 being no separation of the two groups (high for *B-index*; low for *P-index*). The dashed vertical line in (A) is the optimal threshold value for the *B-index* obtained from the ROC analysis.

Discussion

The mesolimbic pathway, including one of its key structures, the NAc shell, is critical for the processing of multiple reward features and the control of behavior (Smith *et al.*, 2003; Johnson *et al.*, 1996; Zhang *et al.*, 1998; Basso and Kelley, 1999; Zhang and Kelley, 2002; Pecina and Berridge, 2005; Woolley *et al.*, 2006; Wassum *et al.*, 2009; Berridge *et al.*, 2010; Smith *et al.*, 2011; Berridge and Kringelbach, 2013). However, the role of the NAc shell in instrumental valuation and behavior is less clear. Here we investigated whether a NAc shell lesion influences choice behavior in a novel 'closed economy' foraging paradigm that mimicked natural foraging by providing the freedom to choose when, what and how long to obtain food. We first discuss possible effects of lesion on learning and execution, then on the sequential and temporal dynamics.

The lesion and sham groups both spontaneously learned the operant task reaching the learning criterion at similar times. Thus, task learning was not significantly affected by the lesion, which is consistent with previous studies that the NAc core, but not the shell, is required for instrumental learning (Corbit *et al.*, 2001; de Borchgrave *et al.*, 2002; Balleine and O'Doherty, 2010). Regarding performance, similar response times between the two groups indicates that general motor execution remained sufficiently intact with the lesion. In addition, the lesion did not significantly affect food intake: either pellets consumed over the entire experiment or consumed per day. This suggests that the general motivation for food was not affected by the NAc shell lesion, discussed below.

For the sequential dynamics, i.e., what to select and whether to continue selecting it, the lesion group exhibited a heightened preference bias compared to the shams. And although both groups showed strong repetitive choice behavior, reflected in heavier tails in the cumulative run distributions compared to randomly shuffled choice sequences, the lesion group exhibited longer runs with the most preferred flavor than the sham group. However, the repeated choices could result from either a biased preference or perseverative behavior. Using the *B* and *P* indices developed to tease apart these factors, we found that the NAc shell lesion enhanced preference bias and not choice persistence; and thus, the longer run lengths in the lesion group resulted from choosing a favorite option with high frequency rather than perseveration. Thus, we did not find evidence for involvement of this NAc shell region in a switch-or-stay behavioral strategy. Future research will need to determine why the NAc shell appeared to affect the win-stay strategy in the Stopper and Floresco (2011) study but did not appear to do so here. Differences between the studies include the specific NAc shell site targeted and the testing paradigms used (e.g., risk-based versus certain reward outcomes).

For the temporal dynamics, in addition to food intake, the time interval between actions is an indicator of motivational state (Robbins and Everitt, 1996; Salamone *et al.*, 2003; Niv *et al.*, 2006; Zanutto and Staddon, 2007; Jung *et al.*, 2014). However, the lesion and sham groups in our study exhibited similar circadian rhythm eating patterns and heavy-tailed distributions in the temporal choice patterns (i.e., inter-choice intervals). Thus, the NAc shell lesion did not significantly alter processes underlying foraging timing events, further suggesting that the lesion had no apparent effect on general motivation.

Thus, overall, the NAc shell lesion led to a singular effect: an increase in the preference bias. Our results are particularly similar to another study that, although targeting a different region in the NAc (more dorsal to ours), provided a choice to rats between two foods that were nutritionally identical but differing in flavor: chocolate *vs* banana pellets (Woolley *et al.*, 2006). When a mu-opioid agonist was infused in their targeted NAc region, they also obtained an increase in consumption of the most-preferred flavor (although with no effect on the other flavor). The authors suggested that the opioid infusion heightened the preferred flavor's palatability. In our study, although we also found heightened selection of the most-preferred flavor, we did not observe an increase in overall consumption, with instead consumption of the other options decreasing, resulting in a change in the preference bias. Such a change is consistent with an underlying change in the *relative* palatability, i.e., relative value. Nonetheless, why there was no evident change in motivation as typically seen in other studies remains unclear. One possibility derives from the novel closed-economy paradigm that we developed, which had no temporal constraints such as on session length. This freedom may promote more stable and consistent temporal dynamics. A second possibility is the lack of obvious external Pavlovian cues to provoke increased wanting and behavior. Another possibility may be due to other instrumental behavioral components of the paradigm. For example, the heightened effort of the response may promote an overall constant behavioral output, as is highlighted in Herrnstein's law of effect (Herrnstein, 1961; 1.970). Finally, we cannot rule out the possibility that the electrolytic lesion may

have led to a broader effect that balanced out differential effects seen via other means, such as pharmacological manipulations, leading to no overall change in consumption. Thus, further studies are needed to continue to delineate the specific mechanisms underlying the behavioral dynamics. In any case, the lesion produced a clear effect on the underlying preference structure and the resulting behavioral preference bias.

The specific causal role played by the NAc shell has also remained unclear. Multiple studies may suggest involvement in the enhancement of reward or associated cue efficacy (Johnson *et al.*, 1996; Zhang *et al.*, 1998; Basso and Kelley, 1999; Zhang and Kelley, 2002; Zhang *et al.*, 2003; Pecina and Berridge, 2005; Woolley *et al.*, 2006; Wassum *et al.*, 2009; Berridge *et al.*, 2010; Smith *et al.*, 2011; Berridge and Kringelbach, 2013); yet the actual effect on NAc shell neural activity is often unknown. At the same time, GABA agonist inactivation within the NAc shell leads to increased food consumption via the releasing of feeding mechanisms mediated by the lateral hypothalamus (Maldonado-Irizarry *et al.*, 1995; Stratford and Kelley, 1997; Basso and Kelley, 1999; Zhang *et al.*, 2003; Taha and Fields, 2006; Stratford and Wirtshafter, 2012). It has thus been suggested that the NAc shell may participate in the regulation of more instinctive, homeostatically driven motivation and feeding mechanisms while promoting more sophisticated, hedonically driven ones (Kelley, 2004; Berridge, 2009). Yet, there is also evidence for a mechanism of extinction that includes NAc-shell inhibition of Pavlovian-cue efficacy (Floresco *et al.*, 2008).

In our study, the lesion's effect on biasing preference was evident. Because neuron loss due to the lesion did not lead to a preference decrement, and in fact led to the opposite, i.e., a heightened preference bias, the region appears to provide an inhibitory control signal to moderate preferences. A preference bias decrease in the normal state suggests excitatory input from an upstream source that activates NAc shell inhibition of a downstream target as part of a larger control circuitry. However, since there are multiple inputs to the NAc shell, as noted in the introduction, the key input to the control circuitry has yet to be identified (Groenewegen *et al.*, 1999; Kelley, 2004 Dalley *et al.*, 2011). One leading candidate may be the VTA, given that, for example, denervation of dopaminergic afferents from VTA to the NAc shell by 6-hydroxydopamine (6-OHDA) lesion abolished conditioned place preference (Sellings *et al.*, 2008). A second leading candidate may be infralimbic prefrontal cortex, given evidence for its role in inhibitory control (Dalley *et al.*, 2011). A determination of the NAc shell inhibitory target(s) also requires further study, given that preference processing could in fact include any of the NAc shell's principal targets, for example, VTA, lateral hypothalamus or ventral pallidum (Heimer *et al.*, 1991; Groenewegen *et al.*, 1999; Kelley, 2004; Trigo *et al.*, 2010; Smith *et al.*, 2011).

In sum, our findings provide evidence for NAc shell control over valuation processes used for decision-making. Indeed, if lower-level reward processes tend to have strong biases toward the most-preferred options (Taha and Fields, 2005; Woolley *et al.*, 2006; Roesch *et al.*, 2009), such inhibitory dampening may be necessary to broaden this narrow-mindedness, and may constitute an important mechanism to tip the balance toward higher-level control of behavior.

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Supplementary data

Supplementary data are available at *SCAN* online.

Conflict of interest. None declared.

References

Atalayer, D., Rowland, N.E. (2009). Meal patterns of mice under systematically varying approach and unit costs for food in a closed economy. *Physiology & Behavior*, **98**, 85–93.

Balleine, B.W., O'Doherty, J.P. (2010). Human and rodent homologies in action control: corticostriatal determinants of goal-directed and habitual action. *Neuropharmacology*, **35**, 48–69.

Basso, A.M., Kelley, A.E. (1999). Feeding induced by GABA receptor stimulation within the nucleus accumbens shell: Regional mapping and characterization of macronutrient and taste preference. *Behavioral Neuroscience*, **113**, 324–36.

Berridge, K.C. (2009). 'Liking' and 'wanting' food rewards: brain substrates and roles in eating disorders. *Physiology & Behavior*, **97**(5), 537–50.

Berridge, K.C., Kringelbach, M.L. (2013). Neuroscience of affect: brain mechanisms of pleasure and displeasure. *Current Opinion in Neurobiology*, **23**, 294–303.

Berridge, K.C., Ho, C.-Y., Richard, J.M., DiFeliceantonio, A.G. (2010). The tempted brain eats: Pleasure and desire circuits in obesity and eating disorders. *Brain Research*, **1350**, 43–64.

Corbit, L.H., Balleine, B.W. (2011). The general and outcome-specific forms of Pavlovian-instrumental transfer are differentially mediated by the nucleus accumbens core and shell. *Journal of Neuroscience*, **31**, 11786–94.

Corbit, L.H., Muir, J.L., Balleine, B.W. (2001). The role of the nucleus accumbens in instrumental conditioning: evidence of a functional dissociation between accumbens core and shell. *Journal of Neuroscience*, **21**, 3251–60.

Dalley, J.W., Everitt, B.J., Robbins, T.W. (2011). Impulsivity, compulsivity, and top-down cognitive control. *Neuron*, **69**, 680–94.

de Borchgrave, R., Rawlins, J.N.P., Dickinson, A., Balleine, B.W. (2002). Effects of cytotoxic nucleus accumbens lesions on instrumental conditioning in rats. *Experimental Brain Research*, **144**, 50–68.

Floresco, S.B., McLaughlin, R.J., Haluk, D.M. (2008). Opposing roles for the nucleus accumbens core and shell in cue-induced reinstatement of food-seeking behavior. *Neuroscience*, **154**, 877–84.

Groenewegen, H.J., Wright, C.I., Beijer, A.V., Voorn, P. (1999). Convergence and segregation of ventral striatal inputs and outputs. *Annals of the New York Academy of Sciences*, **877**, 49–63.

Heimer, L., Zahm, D.S., Churchill, L., Kalivas, P.W., Wohltmann, C. (1991). Specificity in the projection patterns of accumbal core and shell in the rat. *Neuroscience*, **41**, 89–125.

Herrnstein, R.J. (1961). Relative and absolute strength of response as a function of frequence of reinforcement. *Journal of the Experimental Analysis of Behavior*, **4**, 267–72.

Herrnstein, R.J. (1970). On the law of effect. *Journal of the Experimental Analysis of Behavior*, **13**, 243–66.

Johnson, P.I., Parente, M.A., Stellar, J.R. (1996). NMDA-induced lesions of the nucleus accumbens or the ventral pallidum increase the rewarding efficacy of food to deprived rats. *Brain Research*, **722**, 109–17.

Jung, K., Jang, H., Kralik, J.D., Jeong, J. (2014). Bursts and heavy tails in temporal and sequential dynamics of foraging decisions. *Plos Computational Biology*, **10**, e1003759.

Kelley, A.E. (2004). Ventral striatal control of appetitive motivation: role in ingestive behavior and reward-related learning. *Neuroscience & Biobehavioral Reviews*, **27**, 765–76.

Lau, B., Glimcher, P.W. (2005). Dynamic response-by-response models of matching behavior in rhesus monkeys. *Journal of the Experimental Analysis of Behavior*, **84**, 555–79.

Maldonado-Irizarry, C.S., Swanson, C.J., Kelley, A.E. (1995). Glutamate receptors in the nucleus accumbens shell control feeding behavior via the lateral hypothalamus. *Journal of Neuroscience*, **15**, 6779–88.

Niv, Y., Joel, D., Dayan, P. (2006). A normative perspective on motivation. *Trends in Cognitive Sciences*, **10**, 375–81.

Pecina, S., Berridge, K.C. (2005). Hedonic hot spot in nucleus accumbens shell: Where do mu-opioids cause increased hedonic impact of sweetness? *Journal of Neuroscience*, **25**, 11777–86.

Pecina, S., Berridge, K.C. (2013). Dopamine or opioid stimulation of nucleus accumbens similarly amplify cue-triggered "wanting" for reward: entire core and medial shell mapped as substrates for PIT enhancement. *European Journal of Neuroscience*, **37**, 1529–40.

Robbins, T.W., Everitt, B.J. (1996). Neurobehavioural mechanisms of reward and motivation. *Current Opinion in Neurobiology*, **6**, 228–36.

Roesch, M.R., Singh, T., Brown, P.L., Mullins, S.E., Schoenbaum, G. (2009). Ventral striatal neurons encode the value of the chosen action in rats deciding between differently delayed or sized rewards. *The Journal of Neuroscience*, **29**, 13365–76.

Salamone, J.D., Correa, M., Mingote, S., Weber, S.M. (2003). Nucleus accumbens dopamine and the regulation of effort in food-seeking behavior: implications for studies of natural motivation, psychiatry, and drug abuse. *The Journal of Pharmacology and Experimental Therapeutics*, **305**, 1–8.

Sellings, L.H.L., Baharnouri, G., McQuade, L.E., Clarke, P.B.S. (2008). Rewarding and aversive effects of nicotine are segregated within the nucleus accumbens. *The European Journal of Neuroscience*, **28**, 342–52.

Smith, K.S., Berridge, K.C., Aldridge, J.W. (2011). Disentangling pleasure from incentive salience and learning signals in brain reward circuitry. *Proceedings of the National Academy of Sciences of the United States of America*, **108**, E255–64.

Stopper, C.M., Floresco, S.B. (2011). Contributions of the nucleus accumbens and its subregions to different aspects of risk-based decision making. *Cognitive, Affective & Behavioral Neuroscience*, **11**, 97–112.

Stratford, T.R., Kelley, A.E. (1997). GABA in the nucleus accumbens shell participates in the central regulation of feeding behavior. *Journal of Neuroscience*, **17**, 4434–40.

Stratford, T.R., Wirtshafter, D. (2012). Evidence that the nucleus accumbens shell, ventral pallidum, and lateral hypothalamus are components of a lateralized feeding circuit. *Behavioural Brain Research*, **226**, 548–54.

Sugam, J.A., Saddoris, M.P., Carelli, R.M. (2014). Nucleus accumbens neurons track behavioral preferences and reward outcomes during risky decision making. *Biological Psychiatry*, **75**, 807–16.

Taha, S.A., Fields, H.L. (2005). Encoding of palatability and appetitive behaviors by distinct neuronal populations in the nucleus accumbens. *Journal of Neuroscience*, **25**, 1193–202.

Taha, S.A., Fields, H.L. (2006). Inhibitions of nucleus accumbens neurons encode a gating signal for reward-directed behavior. *Journal of Neuroscience*, **26**, 217–22.

Trigo, J.M., Martin-Garcia, E., Berrendero, F., Robledo, P., Maldonado, R. (2010). The endogenous opioid system: A common substrate in drug addiction. *Drug and Alcohol Dependence*, **108**, 183–94.

Wassum, K.M., Ostlund, S.B., Maidment, N.T., Balleine, B.W. (2009). Distinct opioid circuits determine the palatability and the desirability of rewarding events. *Proceedings of the National Academy of Sciences of the United States of America*, **106**, 12512–7.

Woolley, J.D., Lee, B.S., Fields, H.L. (2006). Nucleus accumbens opioids regulate flavor-based preferences in food consumption. *Neuroscience*, **143**, 309–17.

Wyvell, C.L., Berridge, K.C. (2000). Intra-accumbens amphetamine increases the conditioned incentive salience of sucrose reward: Enhancement of reward "wanting" without enhanced "liking" or response reinforcement. *Journal of Neuroscience*, **20**, 8122–30.

Zanutto, B.S., Staddon, J.E.R. (2007). Bang-bang control of feeding: role of hypothalamic and satiety signals. *Plos Computational Biology*, **3**, e97.

Zhang, M., Kelley, A.E. (2002). Intake of saccharin, salt, and ethanol solutions is increased by infusion of a mu opioid agonist into the nucleus accumbens. *Psychopharmacology*, **159**, 415–23.

Zhang, M., Balmadrid, C., Kelley, A.E. (2003). Nucleus accumbens opioid, GABAergic, and dopaminergic modulation of palatable food motivation: contrasting effects revealed by a progressive ratio study in the rat. *Behavioral Neuroscience*, **117**, 202–11.

Zhang, M., Gosnell, B.A., Kelley, A.E. (1998). Intake of high-fat food is selectively enhanced by muopioid receptor stimulation within the nucleus accumbens. *Journal of Pharmacology and Experimental Therapeutics*, **285**, 908–14.