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Does cribbing behavior in horses vary with dietary taste or direct gastric stimuli? $\!\!\!^{\bigstar}$

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ABSTRACT

Concentrated feed diets have been shown to drastically increase the rate of the cribbing, an oral stereotypy in horses, but the specific component causing the rise has not been identified. Furthermore, the mechanism through which feed affects cribbing has not been explored. In the first experiment of this study, we quantified the latency to crib and number of cribs in 15 min after the horses tasted various grain, sugar, and artificial sweetener solutions. Undiluted grain stimulated the most cribs (P < 0.01) compared with all other solutions, and shortest latency to crib, although this was significantly higher only when compared with diluted grain (P=0.03). In Experiment 2, latency to crib and number of cribs in 15 min after the grain and sugar solutions were administered via nasograstric tube were also evaluated. There were no statistical differences among cribbing responses to grain, fructose, and water administered directly to the stomach although grain stimulated cribbing behavior more quickly than 10% fructose (P=0.03) and 100% tap water (P=0.04). These results confirm that highly palatable diets, possibly mediated through the opioid and dopaminergic systems, are one of the most potent inducers of cribbing behavior. The highly palatable taste remains the probable "cribogenic" factor of concentrated diet, although gastric and post-gastric effects cannot be excluded.

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1. Introduction

Stereotypies are abnormal repetitive, invariant, and seemingly functionless behaviors, and these behaviors are associated with suboptimal environmental conditions, although the exact causal factors remain unknown (Mason, 1991). Cribbing, or crib-biting, is an equine oral stereotypy characterized by a horse placing its upper incisors on a horizontal surface and drawing air into the esophagus while flexing the ventral neck muscles (McGreevy et al., 1995). The prevalence of the behavior has been estimated to be around 2–10%, with some breeds demonstrating a predisposition for cribbing (Albright et al., 2010; Bachmann et al., 2003; Luescher et al., 1998; Vecchiotti and Galanti, 1986). Management practices related to social contact, pasture time, and high-concentrate diet have also been linked to the performance of cribbing (reviewed by Wickens and Heleski, 2010).

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http://dx.doi.org/10.1016/j.applanim.2017.01.015 0168-1591/© 2017 Elsevier B.V. All rights reserved. Provision of concentrated feed to young horses at weaning is a particularly strong risk-factor for the development of cribbing (Waters et al., 2002). Sweet feed is also known to increase the rate of cribbing immediately post-ingestion in established cribbing horses (Kusunose, 1992). Consuming roughage (Gillham et al., 1994) and plain oats (Whisher et al., 2011) does not have the same crib-inducing effect. Some hypothesize that highly palatable diets induce the release of endogenous opioids and, in turn, cribbing through a complex interplay of the opioid, dopaminergic, and glutaminergic neural systems (Dodman et al., 1987; Gillham et al., 1994).

The goal of this study was to characterize further the relationship between sweet-tasting substances and cribbing. The first objective was to compare the effects of sugars, artificial sweetener, and sweet feed ingested by mouth on cribbing behavior. We measured the number of cribs and latency to crib as indicators of the strength of motivation to crib. We hypothesized that the taste of food influenced cribbing rate and predicted the commercial sweet feed would be the strongest stimulator of cribbing, followed by sucrose and fructose solutions. The second objective was to compare cribbing after delivery of a single-sugar or grain solution directly to the stomach via nasogastric tube. We hypothesized that some aspect of taste, such as neurophysiologic mechanisms associ-

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ated with the activation of sweet taste receptors and reward neural circuitry, is the critical factor in palatable diet-initiated stereotypies; therefore, we predicted that bypassing these receptors would not result in a difference in the post-delivery cribbing rate and latency to crib among the various solutions.

2. Materials and methods

2.1. Animals and husbandry

All procedures were approved by the Cornell University Institutional Care and Use Committee. Six client-owned horses (two mares, four geldings; four thoroughbreds, one warmblood, and one horse of undetermined breed), ranging in ages between 4 and 25 years old, were lent to Cornell University for this project. The horses were all otherwise healthy established cribbers, although the exact age at onset of cribbing behavior was unknown. The horses were housed in 3.3×3.3 m stalls with a combination of solid and slatted wood walls. The horses had visual and tactile access to adjacent animals. The horses were fed hay ad libitum and approximately 1 kg sweet feed (Respond Original Beet Pulp Formula, Agway, Syracuse, NY, USA) at 07:00 h and 16:00 h. The horses were allowed at least 1 h daily of grassless paddock time in compatible social groups. Horses were returned to the owners at the conclusion of the study.

2.2. Experimental procedures

The study took place between March and July of the same year and consisted of two parts conducted sequentially (Experiment 1: March–May; Experiment 2: May–July). All testing occurred between 10:00–12:00.

2.2.1. Oral solutions-Experiment 1

Once daily, 3-5 times per week, six horses were provided with a shallow bucket containing one of several sweet feed mixtures (Respond; 12% crude protein and 10% crude fiber). The ingredients of the sweet feed, in order of weight, were beet pulp, alfalfa meal, cracked corn, molasses, wheat middlings, soy bean meal, and distiller's grain. Mixtures included sweet feed diluted with water or solutions of with either sucrose, (fructose [D-(-)-fructose], Sigma Aldrich, St. Louis, MO, USA) or saccharin artificial sweetener (Sigma Aldrich) in a 250–1000 mL water solution except for the 100% grain, which contained no water (500 mL, Respond). Solutions (sugar or sweetener solid dissolved in tap water) of 5% sucrose, 10% sucrose, 10% fructose, 0.1% saccharin, 0.2% saccharin, 12% grain, 25% grain, 100% grain, and 100% tap water were presented to the horses in a randomized order and a minimum of two times. Sucrose concentrations were chosen based on previous findings of the strongest preference in foals for 10% sucrose solutions compared to tap water (Randall et al., 1978). Fructose is included in many commercial horse feeds, but to the authors' knowledge, no published data of fructose taste preference is available. We elected to test only a 10% fructose solution based on our preliminary findings that this concentration was the most preferred for fructose as well as sucrose. Grain buckets and hay from regular feedings were removed at least 1 h prior to the experimental trial. Once the horses were provided with the test bucket, the time (s) from latency to cribbing and the number of individual cribbing events in 15 min were recorded. The trial was included if the horse put its muzzle in the bucket and the tongue and lips moved, indicating the horse tasted the solution. The buckets were removed after the 15-min period.

2.2.2. Nasogastric administration-Experiment 2

The same six horses were used in this experiment, which involved instilling several different solutions directly into the stomach using a nasogastric tube $(1.9 \text{ cm} \times 3 \text{ m})$. To decrease

subject distress and improve safety, a low dose (0.6 mg/kg) alpha-2-adrenergic agonist xylazine (Rompun, BayerDVM, Shawnee Misson, Kansas, USA) was administered intravascularly prior to applying a nose twitch and passing the nasogastric tube. A twitch with a cotton rope nose loop was used. The horses were intubated no more than once every 48 h, and to decrease the number of procedures, only three solutions were administered in a randomized order: 12% grain, 10% fructose, and 100% tap water, all at a 1L total volume. The grain suspension was made by grinding feed into mash using a food processor and mixing with water before pumping through the nasogastric tube. The highest concentration grain slurry that would move through the nasogastric tube was 12%. Fructose was chosen because it triggered the most cribbing when ingested orally compared to the other sugar or sweetener solutions (Section 3.1). Immediately after the nasogastric tube and twitch were removed and any solution was wiped from the nasal and oral area to prevent tasting external remnants, the time from latency to crib and the number of cribs in 15 min were recorded. Latency was recorded as 900s for horses that did not crib within the 15 min. Additional trials were conducted to test for effects of the restraint techniques by evaluating twitch restraint with and without sedation. The latency to crib and number of cribs were recorded during the same 15-min period after the twitch alone was applied for 3 min, or when chemical sedation plus the twitch were used for 3 min but no nasogastric tube passed.

2.3. Statistical analysis

The data were analyzed using a repeated measures multivariate analysis of variance (MANOVA) with the number of cribs and latency to crib as the response variables, and the various solutions consumed or delivered via nasogastric tube as the independent variables to investigate the effect of taste or nasogastric delivery of various solutions on the number of cribs and latency. Post hoc analysis for multiple comparisons was conducted with Tukey's adjustment to compare all pairwise differences between mean effects of oral or nasogastric delivery of various solutions. Statistical significance was identified at the level of P < 0.05. Rank transformation was conducted on response variables because the normality assumption had been violated. All analysis was conducted using PROC GLM in the SAS system for Windows, version 9.4 (SAS Institute Inc., Cary, NC, USA).

3. Results

3.1. Effect of consuming various oral solutions on the number of cribs and latency to crib

There was no significant difference in the number of cribs or latency to crib between the two concentrations of diluted grain (12% and 25%) (P=0.94 number of cribs; P=0.99 latency), saccharin (0.1% and 0.2%; P=0.28, 0.99), or sucrose (5% and 10%; P=0.99, 1.00); therefore, the results were combined into one variable per sugar, sweetener, or diluted grain for this experiment. Horses cribbed a significantly greater number of times after consuming 100% grain compared with the same period after consuming diluted grain, saccharin, sucrose, fructose, or tap water-only solutions (P<0.01 for all comparisons; Table 1). No significant difference in crib number was found among the various diluted grain, sugar, saccharin, or water solutions, although there was a trend for saccharin to trigger less cribbing especially when compared to other sweet solutions.

The undiluted grain mixture triggered horses to crib quickly after ingestion, although this was significant only when compared with diluted grain (P=0.03; Table 1). Latency to crib time was also

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