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Research Report

Food deprivation enhances the expression but not acquisition of flavor acceptance conditioning in rats

Yeh-Min Yiin, Karen Ackroff, Anthony Sclafani*

Department of Psychology, Brooklyn College of the City University of New York, 2900 Bedford Avenue, Brooklyn, NY 11210, USA

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Abstract

The postingestive actions of nutrients condition strong flavor preferences in rats and may also enhance flavor acceptance (increase total intake) in some situations. This study determined the impact of food deprivation on flavor preference and acceptance conditioned by intragastric (IG) infusions of glucose. Rats fitted with gastric catheters were trained (20 h/day) to associate a CS+ solution (bitter or sour) with IG 16% glucose and a CS− solution with water infusions. One group (FR) was food-restricted during the training sessions, while a second group (AL) was given food ad libitum. All rats were given 2-h access to food prior to the daily sessions. During one-bottle training, the FR rats consumed substantially more CS+ than CS− whereas AL rats drank only slightly more CS+ than CS−. In additional one-bottle acceptance tests, the FR and AL rats consumed substantially more CS+ than CS− when both groups were food-restricted, but only slightly more CS+ than CS− when both groups had food ad libitum. Throughout the experiment, the FR and AL rats displayed equally strong CS+ preferences in two-bottle choice tests irrespective of their deprivation state during the test. The findings indicate that food restriction stimulates the intake of a CS+ flavor that is (or was previously) paired with IG glucose infusions but does not fundamentally alter the learned association between the CS+ flavor and the post-oral nutrient stimulus.

Keywords: Acceptance; Preference; Glucose; Learning; Hunger; Gastric infusions

Food intake and choice are influenced by learned associations between the orosensory (flavor) and post-oral nutrient properties of foods (Capaldi, 1992; Sclafani, 1999). Flavor-nutrient learning is typically studied in the laboratory using a conditioned flavor preference paradigm in which an arbitrary flavor (the target conditioned stimulus, CS+) is paired with a nutrient (the unconditioned stimulus, US) and a different flavor (the CS−) is paired with a non-nutritive source during one-bottle training sessions. This is most effectively accomplished in animals by pairing the intake of a CS+ flavored solution with intragastric (IG) infusions of the nutrient (Sclafani, 1995). In subsequent two-bottle choice tests, rats typically display a strong preference for the CS+ over the CS− solution which is evidence that they learnt the flavor-nutrient association.

In some experiments, animals also increase their intake of the CS+ solution, relative to the CS− solution, during the course of one-bottle training. This conditioned increase in CS+ *acceptance* is a more variable response than is conditioned CS+ *preference*. In particular, animals that develop a strong preference for a CS+ flavor, as measured in two-bottle tests, may or may not consume more CS+ than CS− in one-bottle sessions (Azzara & Sclafani, 1998; Lucas, Azzara, & Sclafani, 1998). Flavor-nutrient acceptance conditioning has also been investigated using a between-group design in which one group is trained with a flavored solution (referred to here as the CS+) paired with IG nutrient infusions and another group is trained with the same flavored solution (referred to here as the CS−) paired with IG water infusions (Ramirez, 1994). Nutrient-conditioned increases in CS+ acceptance have also been a variable outcome using this design (Ramirez, 1994, 1996, 1997a,b). In the case of a CS+ solution paired with IG infusions of an energy-dense nutrient source, this may occur because the satiating action of the IG infusions limits CS+ intake (Azzara & Sclafani, 1998; Lucas et al., 1998; Ramirez, 1997b). With less energy-dense infusions,

* Corresponding author.
E-mail address: asclafani@gc.cuny.edu (A. Sclafani).

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conditioned increases in CS+ acceptance are reported to vary as a function of the nature of the CS+ flavor. Ramirez (1994, 1996, 1997a,b) observed that IG infusions of dilute (3–6%) carbohydrate solutions produced consistent and substantial increases in the one-bottle intake of saccharin solutions during 24 h/day acceptance tests, but had little or no effect on the intake of various non-sweet solutions (e.g. a bitter sucrose octaacetate solution, a sour cherry Kool Aid solution).

Flavor-nutrient learning has been obtained in both food-restricted and non-restricted animals (Ackroff, Touzani, Peets, & Sclafani, 2001; Elizalde & Sclafani, 1990; Fedorchak & Bolles, 1987). Although food deprivation might be expected to facilitate conditioning (Davidson, 1998), in a recent study we observed that food deprivation state had relatively little effect on flavor-nutrient preference conditioning (Yiin, Ackroff, & Sclafani, 2005). IG infusions of carbohydrate, fat, or protein conditioned flavor preferences in both food-restricted animals and freely fed animals trained in short sessions (0.5–2 h/day). In most cases, the magnitude of the conditioned CS+ preference, indexed by the percentage of CS+ consumed in two-bottle tests, was similar in the food-restricted and unrestricted rats. In contrast, data obtained in other studies indicate that food restriction significantly increases CS+ acceptance. That is, in 24 h/day one-bottle sessions, food-restricted rats consumed more of a nutrient-paired CS+ solution than of water-paired CS− solution relative to ad libitum fed animals (Ramirez, 1996; Sclafani, Azzara, Touzani, Grigson, & Norgren, 2001). These results, however, do not establish whether food restriction during original learning enhances flavor acceptance conditioning. That is, flavor acceptance conditioning has not been compared in separate groups of animals that were food-restricted and freely fed during initial CS training sessions. In view of our recent findings (Yiin et al., 2005) that food restriction during training had minimal effects on CS+ preference learning, the present study examined the impact of food restriction state on flavor-nutrient acceptance learning. This was of interest because some data suggest that flavor preference and acceptance conditioning involve different learning processes. In particular, acceptance conditioning is more dependent upon the flavor quality of the CS solutions and is more susceptible to extinction than is preference conditioning (Pérez, Lucas, & Sclafani, 1998; Ramirez, 1996).

Two groups of rats were trained 20 h/day with different flavored solutions paired with IG infusions of glucose or water on alternate days. One group had no food available during the 20-h sessions (food-restricted, FR group) whereas the other group had food ad libitum (AL group). Thereafter, both groups were food-restricted and tested for their one-bottle acceptance of the CS+ and CS− solutions. The CS+ solution was paired with IG water rather than glucose during this test to prevent rapid flavor-nutrient acceptance conditioning in the AL group. If food restriction enhances flavor acceptance learning then the FR group would consume more CS+ than the AL group even though both groups were food-restricted at the time of the acceptance test. The CS solutions used were 'bitter' sucrose octaacetate and 'sour' citric acid solutions, which support robust preference but relatively weak acceptance conditioning in ad libitum fed rats (Drucker, Ackroff, & Sclafani, 1993; Myers & Sclafani, 2003; Pérez et al., 1998; Ramirez, 1996; Sclafani et al., 2001). In the final phase of the experiment, CS+ acceptance was measured with both AL and FR groups fed ad libitum to determine if the initial training experience of the FR rats caused them to over-consume the CS+, relative to the AL rats, when they were no longer food-restricted. In addition to measuring total CS intakes during the 20-h sessions, drinking patterns were analyzed to reveal deprivation state effects on CS bout size and frequency. Two-bottle CS+ vs. CS− preference tests were also conducted to compare how food availability affects flavor preference and acceptance. Additional choice tests were conducted with the CS+ vs. water and CS− vs. water to provide another measure of the magnitude of the conditioned changes in CS+ preference. Prior findings indicate that whereas untrained rats prefer plain water to the bitter and sour CS solutions used here, rats trained with the CS+ paired with IG carbohydrate infusions subsequently prefer the CS+ to plain water as well as to the CS− solution (Pérez et al., 1998; Sclafani, 1991).

Method

Subjects

Adult female Sprague–Dawley rats ($n=21$) bred from stock obtained from Charles River Laboratories (Wilmington, MA) were used. The animals weighed 250–310 g and were housed in individual wire-mesh cages in a room maintained at 21 °C and under a 12:12 h light–dark cycle (lights on at 08:00). Powdered chow (No. 5001, PMI Nutrition International, Brentwood, MO) and tap water were available as noted.

Surgery

The rats were anesthetized with a mixture of ketamine (63 mg/kg) and xylazine (9.4 mg/kg) and a stainless steel intragastric cannula was surgically implanted at the fundus of the stomach as previously described (Elizalde & Sclafani, 1990). After the surgery, animals were given 2 weeks to recover in their home cages. During this time their gastric cannulas were closed with stainless steel screws.

Apparatus

The intragastric (IG) infusion was accomplished using an 'electronic esophagus' apparatus described by Elizalde and Sclafani (1990). In brief, the rats were housed in stainless