

Infant and Toddler Responses to Bitter-Tasting Novel Vegetables: Findings from the Good Tastes Study

Susan L Johnson,¹ Kameron J Moding,² Kevin J Grimm,³ Abigail E Flesher,¹ Alyssa J Bakke,^{4,5} and John E Hayes⁴

¹Department of Pediatrics University of Colorado Anschutz Medical Campus, Aurora, CO, USA; ²Department of Human Development & Family Studies, Purdue University, West Lafayette, IN, USA; ³Department of Psychology, Arizona State University, Tempe, AZ, USA; ⁴Sensory Evaluation Center, The Pennsylvania State University, University Park, PA, USA; and ⁵Department of Food Science, The Pennsylvania State University, PA, USA;

ABSTRACT

Background: Infants are born with the biological predisposition to reject bitterness. Dark green vegetables contain essential nutrients but also bitter compounds, making them more difficult to like.

Objective: The Good Tastes Study was designed to determine whether reducing bitterness by adding small amounts of sugar or salt would alter infant acceptance of kale purées.

Methods: Caregivers (n = 106, 94% mothers, 82% Non-Hispanic White) and children (53% male, aged 6–24 mo) participated in a videorecorded laboratory visit during which infants were offered 4 versions of puréed kale: plain, 1.2% or 1.8% added sugar, or 0.2% added salt. Caregivers rated their children's liking for each kale version. Videos were coded for the number of tastes accepted and for children's behaviors and acceptance of each kale version. A multilevel ordered logistic model was fit for the number of accepted tastes and caregiver ratings of child liking of kale versions with age, breastfeeding history, order effects, and kale version as predictors.

Results: Infants 6 to <12 mo accepted more tastes (b = 2.911, P < 0.001) and were rated by caregivers as liking the kale more than older toddlers (\geq 18 mo; b = 1.874, P = 0.014). The plain kale was more likely to be accepted (P < 0.001); also, the first version offered was more likely to be rejected (b = -0.586, P < 0.007). Older infants (\geq 18 mo) exhibited more avoidant behaviors (b = 1.279, P < 0.001), more playing (b = 2.918, P < 0.001), and more self-feeding (b = 1.786, P = 0.005) than younger infants (6 to <12 mo). Children who were reported to have been breastfed more in the last 7 d were more likely to self-feed (b = 0.246, P < 0.001) and play with food (b = 0.207, P < 0.005).

Conclusions: Our findings support that there may be a sensitive period, during the early phase of complementary feeding, to improve success of introducing a novel, bitter, more difficult-to-like food. When low levels of sugar or salt were added, no advantage of bitterness reduction was observed. This study has been registered with ClinicalTrials.gov as NCT04549233. *J Nutr* 2021;151:3240–3252.

Keywords: infancy, eating behavior, complementary feeding, vegetables, bitterness

Introduction

The first 1000 d of life, from conception to 24 mo of age, encompasses a rapid trajectory of developmental changes; critically, these changes include development of food preferences and eating skills (1, 2). Taste and flavor preferences evolve during this period, with the infant and young child learning not only how to eat but also what is edible and preferable (3). These early life experiences matter for nutrition status and outcomes for the young child but are also suggested to be important for the programming of later-in-life food preferences, eating habits, and health outcomes (4). Thus, better understanding of the development of eating patterns during this early window, including the development of flavor preferences, may be critical for promoting the adoption of healthy lifelong eating habits.

It is manifestly clear that some foods are harder to like than others, and these differential preferences are rooted in biology: sweetness is innately liked, whereas bitterness is reflexively disliked initially, although can be overcome by experience and repeated exposure (5–7). Caregivers often want children to consume dark green vegetables, both because of their nutrient density and because of the positive properties they confer for overall health (8, 9). However, such desires are tempered by low innate liking for dark green vegetables, which can be attributed to some combination of their low endogenous

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sweetness, enhanced bitterness, and/or astringency, as well as novel aromas and textures (10, 11).

According to data collected in the United States by the Feeding Infants and Toddlers Study, young children's diets fail to meet recommendations (12). Specifically, although many young infants initially consume a variety of fruits and vegetables $(\sim 73\%$ of 6- to 12-mo-olds) when complementary foods are first being introduced, levels of vegetable consumption decline during toddlerhood (60% of 12- to 24-mo-olds), with only 12% of US toddlers consuming a dark green vegetable within a given day during this developmental period (13, 14). Children's dietary quality continues to decline across childhood, and this is particularly concerning, given that dietary habits are thought to persist into adulthood (15). It would seem, then, that later in the complementary feeding period (~ 12 -24 mo of age), when children are being introduced to foods of the family table and consuming more of what families are consuming, a transition period occurs in which children's eating is challenging for caregivers, warranting further study of this specific developmental window.

Certain foods, like those with simple sugars (e.g., fruits or foods with added sugars), are easily accepted, as sweet taste is highly liked and accepted by most children and adults (4, 7, 16, 17). Conversely, foods lacking sweet aspects to their flavor profiles are often met with less favorable reactions (3, 18). One group of foods that often can be difficult to encourage children to like and eat is dark green vegetables. These foods have chemical compounds (e.g., glucosinolates, polyphenols, isothiocyanates) that confer bitter properties which many individuals, both children and adults, find aversive or difficult to learn to like (19, 20). Negative reactions to bitterness are innate and appear early in infancy via characteristic facial reflexes like grimacing and gaping; signs interpreted as dislike and rejection (4, 6, 21). Although some literature exists that explores maternal decision making regarding offering novel foods (8, 9), comprehensive understanding of caregivers' decisional processes for persisting in offering foods that receive fewer positive reactions from infants and toddlers is lacking.

Some studies in infants and young children have investigated associations of early food experiences and liking and consumption of complementary foods, especially for fruits and vegetables [e.g., (22)]. Infant acceptance during the complementary feeding period positively associates with maternal diet and breastfeeding (23, 24), as well as early introduction to foods (25). Difficultto-like bitter tastes from foods remain understudied across the complementary feeding period. In general, repeated exposure to vegetable flavors—whether through breastmilk (23), flavored formula (26), or infant cereal or purées (26–33)—has resulted in increased liking and intake by children and appears to be at least as, if not more, effective than flavor–flavor learning (e.g., mixing a sweet flavor with a less-liked flavor) or flavor–nutrient learning [e.g., pairing a flavor with increased energy density (28, 34)].

Instead of pairing a novel flavor with energy or a previously liked flavor, or dissipating neophobia via mere exposure, it may also be possible to exploit opponent processes embedded within sensory pathways to improve the palatability of a food. Specifically, bitterness and sweetness exhibit a phenomenon known as mixture suppression (35, 36). When mixture suppression occurs in the central nervous system, the perceived intensities of stimuli presented in a mixture are less than the intensity each component would have been if the stimulus had been presented alone. That is, the sweetness of sugar suppresses the bitterness of quinine, and vice versa (35). Notably, this suppression is asymmetrical-an equimolar mixture of 0.1 mM quinine and 0.32 M sucrose cuts perceived bitterness in half, whereas perceived sweetness is only reduced by $\sim 14\%$ (37). This phenomenon is one reason (of several) that the relation between added sugars and perceived sweetness is not as straight forward as may be commonly assumed (38).

Suppression of bitterness by sweetness is routinely observed in real foods, as should be intuitive to adults who add sugar to coffee. Nonetheless, before embarking on the current study in infants and young children, our team conducted multiple studies in adults to confirm that adding small amounts of sugar would meaningfully reduce bitterness in blended kale (11). We found adding just 1.2% sugar would cut the bitterness of puréed kale by ~47%, and critically for future learning, it does so without altering other sensations and flavors. We also found a similarly low amount of sugar (1%) can significantly decrease the amount of disliking, at least in adults (11). Or to paraphrase the fictional Mary Poppins, "A spoonful of sugar helps the vegetables go down."

A second and separate quirk in taste perception can also be exploited to potentially improve vegetable palatability. Sodium ions block bitterness, and they do so primarily in the periphery, unlike mixture suppression which occurs centrally (39). Critically, this reduction is due to the sodium ions themselves, and not the salty taste, per se, as other salts that are less perceptibly salty (e.g., sodium gluconate) still effectively block bitterness (40). This phenomenon is also seen in foods such as low sodium cheddar cheese, which is both less salty and more bitter than regular cheddar, with expected consequences on liking (11, 41). In the same study in adults described above, we also confirmed that small amounts of added salt can partially block the bitterness of kale, with levels of bitterness reduction similar to those seen for sugar (11).

Collectively, prior work in adults indicates both salt and sugar can effectively reduce the bitterness of kale purée, with downstream effects on liking; whether similar patterns persist in infants and young children when salt and sugar

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Address correspondence to SLJ (email: Susan.Johnson@cuanschutz.edu). Abbreviations used: CEL, Children's Eating Laboratory.

are added to kale is an empirically open question. Data on mixture suppression in early childhood are generally lacking, given the inability to directly assess perceived intensity in this age group; that said, limited data from 12- to 41-mo-olds suggests it may occur [see data in (42), as discussed in (37)].

We conducted an exploratory study to investigate caregiverchild interactions during feeding of 4 versions of a bitter dark green puréed vegetable, 1 plain version, 2 versions in which bitterness was reduced with 2 levels of added sugar, and 1 version in which bitterness was reduced with added salt, in alignment with our prior research in adults. We hypothesized that children would respond more positively to purées in which vegetable bitterness was reduced relative to purées in the plain control condition. Children's observed responses included positive and negative behaviors, the number of tastes (i.e., bites) accepted, and the avidity or rate of the acceptance of each taste. Exploratory aims included determining whether and how children's responses differed across the developmental period of 6–24 mo and whether caregiver perceptions of children's liking varied according to condition.

Methods

Overview

The Good Tastes Study utilized a cross-sectional observational design to assess infant and toddler acceptance of a novel bitter green vegetable purée and to determine whether modification of the inherent taste properties of such foods [i.e., decreased bitterness and increased sweetness; see (11)] could alter acceptance by children. During a laboratory visit, caregivers offered their child tastes of 4 different kale purées [control (plain kale slurry), 1.2% sugar added, 1.8% sugar added, and 0.2% salt added]. Caregivers subsequently rated their perceptions of the child's liking of each version. Caregivers and researchers present in the visit were blinded to the order of introduction. Videos of feeding interactions were recorded digitally and coded by trained research assistants (again, blinded to food condition) to determine acceptance of the food (number of successful tastes, rate of acceptance, and positive and negative behaviors). Caregivers also completed questionnaires to assess family demographic information and infant breastfeeding history. Anthropometric measures were completed at the end of the visit for caregivers and for children. All study procedures were approved by the Colorado Multiple Institutional Review Board (COMIRB #15-2437) and participants were paid for their participation in the study. The study has been registered with ClinicalTrials.gov as NCT04549233.

Participants

Caregivers with infants/toddlers were recruited to participate in this study via a university listserv, flyers posted both on campus and in the community, posts on social media (e.g., Nextdoor, Facebook), and participant referrals between July 2017 and January 2018. Caregivers were eligible to participate if they were between 18 and 51 y of age, could read and speak English, and lived within 75 miles of the Children's Eating Laboratory (CEL) in Aurora, CO. Infants were eligible to participate if they were between 6 and 24 mo of age, were born at term (\geq 37 wk gestation), had experienced at least 1 complementary food (e.g., rice cereal), and did not have food allergies or genetic or metabolic disorders that could affect food intake. After participating in a screening interview by phone, eligible participants were scheduled for a visit at the CEL (see Figure 1). Upon arrival, caregivers provided written consent for their own and their child's participation in the study.

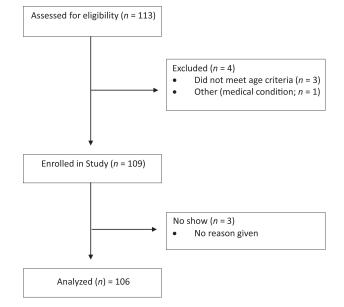


FIGURE 1 Good Tastes Study CONSORT Diagram.

Procedures

Food preparation

Preliminary work to identify appropriate substrates.

We asked mothers to introduce dark green vegetable purées to their infants and toddlers, as these vegetables tend to have bitter taste profiles and are reported to be difficult to engage children to eat (14). Purées were chosen such that the food could be ingested safely by both infants and toddlers. Prior to the current study, a series of studies in adults using both trained panelists and naïve consumers was conducted to identify a dark green vegetable that would be most appropriate for this study, based on the ability to reduce perceived bitterness by the addition of sugar and/or salt (11). Briefly, 3 bitter green vegetables (kale, spinach, and broccoli) were puréed with different levels of sugar (0%, 0.6%, 1.2%, 1.8%) or salt (0.2%) and tasted by trained and naïve adults. Perceived sweetness, saltiness, bitterness, aroma, and texture were assessed for each version of the vegetable purées. Adding small amounts of sugar or salt reduced the perceived bitterness of all 3 green vegetables without altering the texture or other flavors of the purée (e.g., green leafy, grassy, earthy/musty, etc.). That said, perceived sweetness and saltiness also increased, depending on whether sugar or salt was added. Also, as expected, the addition of small amounts of sugar (1.0% sugar and 1.8% sugar) and reduction in bitterness meaningfully increased liking ratings of each vegetable in adults. Ultimately, kale was selected as the target vegetable for the present study because the endogenous bitterness of kale at baseline was greater than for the other 2 vegetables tested, yet this bitterness could still be partially reduced by the addition of sugar or salt.

Sample preparation.

Frozen chopped kale was ordered from a local retailer (Wegmans Food Markets); the purchase was made in bulk to ensure all packages came from a single lot to minimize potential variation that can occur with a natural product grown across seasons or locations. Individual packages were shipped by research staff using containers able to ensure a cold chain; containers were unpacked immediately on receipt and were

stored frozen for the entirety of the study. To ensure consistency of the texture, kale purées were prepared in small batches (20-24 oz or 567-680 g of kale purée) with a standardized protocol developed in pilot testing by staff at the Penn State Sensory Evaluation Center. Briefly, frozen chopped kale was cooked in large glass bowls in a microwave according to package instructions and subsequently cooled in a refrigerator for ~ 1 h. Next, the kale slurries were made according to the protocol of Bakke et al. (11). Ingredients for each version of the kale purées were blended (Nutribullet[®], Capital Brands) for 1 min and 15 s. The puréed kale was frozen in 1-oz cubes (\sim 29 g) in silicone trays, which were color coded to indicate the kale version. The day prior to a study visit, 2 cubes of each version were placed in separate clearly labeled 4-oz (~118 ml) clear plastic soufflé cups with lids and stored in the refrigerator to thaw overnight. On the day of the study visit, plastic cups with kale purées were set out on the counter 30 min before the visit to allow the purée to warm to room temperature prior to serving.

Laboratory visit protocol.

Caregivers and infants/toddlers attended a single laboratory visit lasting ~ 90 min. Caregivers were asked to refrain from feeding their infants/toddlers for 1.5–2 h prior to the visit. Upon arrival at the CEL, caregivers entered a room equipped with 4 dome cameras, 1 in each corner of the room, and 1 omnidirectional microphone in the ceiling. Live outputs from the cameras were viewed on a computer in an adjacent room. Cameras and microphones were turned on to record portions of the visit that would be used for video coding (e.g., during feeding) and turned off during other portions to ensure privacy (e.g., informed consent, anthropometrics).

Laboratory visits were conducted by 2 experimenters: a lead experimenter who directed the visit and a research assistant who assisted with transitions between tasks, managed the cameras, and helped entertain the child when the caregiver was being interviewed. The visit began with the informed consent process, and caregivers gave written consent for their own and their child's participation in the study. Next, caregivers answered questions about their child on the day of the visit (e.g., time the child last ate) and features of typical feeding interactions (e.g., frequency of caregiver feeding versus self-feeding, preference for a highchair or booster seat). After the interview was complete, the lead experimenter instructed the caregiver to play with their child as they normally would at home; a basket of ageappropriate toys was provided. This task lasted ~ 3 min and allowed the child to warm up to the new environment by spending time alone with their caregiver in the laboratory setting.

Feeding interaction.

After the warm-up task, caregivers were asked to place their child in either a highchair or a booster seat next to a small table; whichever best resembled the child's typical feeding environment. The child's chair was placed in the middle of the room facing 2 cameras, 1 in each corner of the room. Caregivers sat directly across from the child if they were in the highchair and next to the child if they were in the booster seat. When the caregiver faced the child, their image was captured by 2 additional cameras in the opposite corners of the room; otherwise, they were videoed using the same cameras that faced the child.

The foods were set up for the caregiver, away from the child's reach, on a foldable tray with a tin containing the 4 kale purées. Clear number labels (1–4, which allowed for blinding

the version to caregivers) indicated the order the purées were to be offered to the child. The order in which the different versions of kale purée were offered followed a modified Williams Latin Square design (43) for the control (plain) kale and kale purées with 1.2% or 1.8% added sugar. The added salt version was always offered last (i.e., a fixed position in the presentation order); this choice was explicitly made to control for the effects of a very different taste exposure (sweet compared with salt) and to minimize any carryover effects of sodium ions on taste perception.

Caregivers of 6- to <12-mo-olds were instructed to spoon feed their child, whereas caregivers of aged children >12 mo were given a choice of spoon feeding or letting their children self-feed, if they wished. The experimenter cued the caregiver when it was time to offer each spoonful of kale. When the child refused an offer, the experimenter allowed more time to elapse (i.e., >30 s) before prompting the caregiver to offer the next spoonful of kale. When the child was upset or the caregiver thought the child needed a break, the procedure was paused and resumed when the child was calm. The overall goal of the task was for the child to taste each kale version 2 times; however, in cases when the child became overly upset, the experimenter and/or caregiver was allowed to terminate the kale version or task early. After 2 tastes or 3 rejections, caregivers were asked to rate their perception of the child's liking for the kale version (see Measures below). Caregivers were then prompted to give their child a sip of water, using a water bottle they brought from home or a small paper cup, to rinse the child's mouth. The experimenter then prompted the caregiver to offer the next kale version using a new spoon. This protocol was repeated until all kale versions had been offered or the task was terminated.

Coding procedures

Trained research assistants coded infant acceptance of each kale version using video recordings of the feeding interaction. Coders were divided into 2 separate teams; 1 team coded the child's rate of acceptance and the other coded child positive and negative behaviors. Both teams were trained by 2 experienced supervisors using video examples of behaviors, group discussions, and practice assignments which were completed independently by the coders. During training, disagreements from the practice assignments were discussed as a group and additional practice videos (3 or 4 per assignment) were assigned to coders. Once coders disagreed infrequently on each code across several assignments, they independently coded 3 gold standard videos. These videos were also coded by the supervisors (gold standard coders) and were selected to represent the range of child ages present in the study sample and a range of codes from the scheme. After coders successfully coded the gold standard videos with acceptable reliability (see Measures below), the study videos (blinded to the coders) were divided between the 2 coders on each team. To assess drift reliability, 20% of the study videos were double coded by coders. Questions and disagreements were resolved by the gold standard coders and the final data set included the resolved codes.

Measures

Demographics and breastfeeding history.

Prior to the laboratory visit, caregivers were mailed and asked to complete a demographic questionnaire to provide their child's and their own age, sex, race, and ethnicity, as well as their marital status, education, and family income. Caregivers also reported information about their child's feeding history, including whether the child had ever been breastfed, if the infant was continuing to receive breastmilk, and the age the child completely stopped receiving breastmilk, if applicable (44). This information was used to create 2 breastfeeding variables that were analyzed in the current study: recent frequency of exposure to breastmilk (number of times the child had been breastfed in the last 7 d, with higher scores indicating more frequent exposure) and time (in months) since last exposure to breastmilk (higher scores indicate more time since the child had been offered breastmilk).

Children's food acceptance

Infant and toddler acceptance of each kale version was assessed using 4 measures: subjective maternal perceptions of their child's liking, observed number of successful tastes, coded rate of acceptance, and behavioral coding of observed positive and negative behaviors displayed by the infants and toddlers during the feeding interaction. Each measure is described in detail below.

Maternal perceptions of liking.

Consistent with previous research (26, 31), caregivers were asked to rate their child's liking for each kale version using a 9-point hedonic scale. Higher scores indicated greater perceived liking (1 = dislikes extremely to 9 = likes extremely). Caregivers were asked to rate their perceptions of liking when their child tasted at least 1 offer of the kale version.

Successful tastes.

We coded each food offer as "successful" if the offer resulted in the child tasting the kale. A taste was defined as the child's tongue contacting the food and offers were categorized as successful (scored as 1) or unsuccessful (scored as 0) based on this definition. The lead experimenter watched for successful tastes during the feeding protocol. When it was unclear whether the child had tasted the food, the experimenter listened and watched for context clues from the child (e.g., vocalizations, facial movements) and also asked the caregiver for a second opinion. Videos were subsequently reviewed to confirm the original decisions for number of tastes. Per the feeding protocol, the number of successful tastes for each kale version ranged from 0 to 2. In cases where the child tasted a kale version more than 2 times (e.g., due to the child self-feeding or the caregiver offering the food without waiting for a prompt from the caregiver), the number of bites consumed by the child still received a score of 2.

Rate of acceptance.

The video coding scheme to assess infant and toddler rate of acceptance of the food was based on the Feeding Infants: Behaviour and Facial Expression Coding System [FIBFECS; (45, 46)]. The coding scheme consists of a 4-point rating scale (0 = refused; 1 = enforced; 2 = accepted; 3 = anticipated).Each of the 4 codes have objective definitions and criteria (see Table 1) to capture infants' rate of acceptance in response to each spoonful of food offered by the caregiver or child. Since this coding scheme was originally validated for use with infants during weaning $[\sim 4-6 \mod 6 \pmod{46}]$, it was adapted here for use with older infants and toddlers. Prior codes and definitions were modified to incorporate cofeeding (both infant and caregiver had their hand on the spoon) and self-feeding (infant/toddlers fed themselves), 2 behaviors that are demonstrated more frequently by older infants and toddlers compared with newly-weaning infants. Additionally, spatial terms (i.e., "a spoon's distance") were incorporated into definitions for acceptance and anticipation codes, similar to the original coding scheme by Mennella and Ventura (47).

Intraclass correlation coefficients (ICCs) were calculated to compare coder reliability using absolute agreement during the training phase and drift reliability coding of the study videos. Both coders demonstrated excellent reliability compared with

TABLE 1 Definitions of codes for infants' and toddlers' acceptance and behaviors

Acceptance codes ¹	Definition	Reliability
Refused (0)	A score of zero (refusal) is given when the child does not open his/her mouth for an offer of food	
Enforced (1)	A score of 1 (enforced) is given when the child opens his/her mouth for a bite, but only after the food/spoon touches the child's lips	
Accepted (2)	A score of 2 (acceptance) is given when the food is close to the child's mouth (less than a spoon's length away) when the child opens his/her mouth to accept the bite	
Anticipated (3)	A score of 3 (anticipation) is given when the child opens his/her mouth for a bite when the spoon is at a distance (a spoon's length or more) from the mouth	
Reliability (ICC) ¹		0.93
Child behavior	Definition	Reliability (κ)
Avoidance ²	A purposeful movement in which a child attempts to move away from or dodge the spoon actively coming towards them (e.g., turning head laterally)	0.38
Prevention ³	Purposeful movement or action to prevent the actively approaching spoon from entering the mouth (e.g., pushing or moving the spoon away)	0.43
Crying/fussing	A distinct, negative vocalization or sound that is sustained for 1 s or longer in response to the food or feeding in general	0.61
Expelling	Ejection or intentional removal of the food or taste from the child's mouth in a non-exploratory way (i.e., not just looking at it) (e.g., spitting the food out of the mouth)	0.62
Play/other	Active exploration, playing, or engagement with food or spoon in hand with visual attention (e.g., finger panting with the food)	0.63
Feeding	Child takes the food or spoon (with food on it) and puts or attempts to put it in his/her mouth on their own or with the help of the parent. This code was selected only for intervals when the child put or attempted to put the spoon in their mouth (e.g., self-feeding)	0.79

¹ICC = intraclass correlation Coefficient reported for entire scale.

²Adapted from the Feeding Infants: Behaviour and Facial Expression Coding System [FIBFECS; (45, 46)].

³Avoidance and prevention were combined into 1 code labeled "avoidant behaviors."

the gold standard coders during training (ICCs > 0.95) and excellent reliability with each other during drift reliability coding of the study videos (ICC = 0.93).

Positive and negative behaviors.

Infant and toddler positive and negative behaviors observed during the offering of each kale version were coded using Noldus The Observer XT coding software (Version 12.5). Based on prior research (46, 48) and a review of the study videos (~20%), the following behaviors were selected to be coded: avoidance, prevention, crying/fussing, expelling, leaning, reaching, feeding, and play/other; leaning and reaching were ultimately removed from the coding scheme due to low reliabilities (described below). Brief descriptions of each behavior used in the final analysis are included in Table 1. The videos of each kale version were segmented into offers that began when the spoon (whether held by the caregiver or child) started approaching the child's mouth and ended at the beginning of the next offer. Since offers varied in length, they were segmented into 5-s intervals to ensure that all infant/toddler behaviors could be more easily coded. Trained pairs of coders rated the presence or absence of each behavior during each 5-s interval and could select multiple codes per interval. Breaks during or between kale versions were not coded.

Both coders demonstrated excellent reliability on the majority of codes during training [$\kappa > 0.75$ for crying/fussing, feeding, leaning, play/other, prevention, and reaching; see Bakeman and Quera, 2011 for a discussion on interpreting κ (49)]. One coder also demonstrated excellent reliability on the avoidance code $(\kappa = 0.77)$, whereas the other coder showed fair reliability ($\kappa = 0.57$). The reliabilities were poor for both coders for the expelling code ($\kappa = 0$ and 0.50), partially due to the low prevalence of expelling in the selected gold standard videos. The decision was made to retain all codes and re-evaluate the inclusion of each code in analysis based on drift reliability statistics calculated for the primary coding of study videos. The drift reliabilities for expelling were good, and so this code was retained for analysis. However, the reliabilities were poor for leaning and reaching, so both codes were dropped from analysis. Drift reliabilities for each code retained for analysis are presented in Table 1.

Variables for analysis.

The original coding scheme was designed to assess positive behaviors (leaning, reaching, feeding) and negative behaviors (avoidance, prevention, crying/fussing, expelling), as well as exploratory behaviors indicative of the child's interest in the materials involved in the feeding, such as the spoon or cup, or the food itself (play/other). Since leaning and reaching were dropped, feeding was left as the single positive behavior examined in the present analysis. The reliabilities for avoidance and prevention were also weak, but these 2 behaviors were correlated in this sample (r = 0.40, P < 0.001). Based on this preliminary analysis and prior research indicating that behaviors indicating avoidant or refusal behaviors tend to be correlated (46, 48), avoidance and prevention codes were collapsed into 1 "avoidant behaviors" code. Intervals that were coded as avoidance, prevention, or both were given a score of "1" or "present." Therefore, the final list of coded behaviors analyzed in the present study are as follows: avoidant behaviors, crying/fussing, expelling, feeding, and play/other. Each of these behaviors were analyzed in separate models at the interval level (1 = present; 0 = absent) in order to capture variability in these codes within and across food offers.

Anthropometric Measures *Caregiver height and weight.*

Height and weight were measured in triplicate by a trained research staff member. Height was measured to the nearest 0.5 cm using a wall-mounted stadiometer (Holtain, Ltd; Stadiometer) and weight to the nearest 0.1 kg using a calibrated bariatric scale (Seca 634 bariatric scale). BMI (in kg/m²) was calculated.

Infant length and weight.

Supine length and weight were measured in triplicate by a trained research staff member. Length was measured to the nearest 0.5 cm using infant length boards (Ellard Instrumentation Ltd) and weight was measured to the nearest 0.1 kg using a calibrated electronic scale (Seca 334 Infant Scale). The WHO Growth Standards were used to calculate weight-for-age, length-for-age, weight-for-length, and BMI *z* scores (50).

Data Analysis

Descriptive statistics were computed (frequencies and means) for the total group and by age groups, and distributions were examined. Given the ordinal nature of the outcomes and the repeated assessments, a multilevel ordered (binary when outcome had only 2 levels) logistic model was estimated for each outcome using Mplus (51). Within-subject predictors included the dummy-coded variables for the type of kale (1.2% or 1.8% added sugar or 0.2% added salt) with the plain kale as the comparison level. Between-subject predictors included age, kale order, and breastfeeding history. Age was dummy coded to draw comparisons between the youngest age group (6 to <12 mo) and the older age groups (12 to <18 mo and \geq 18 to 24 mo). The kale order variable drew comparisons between the first kale version administered and the remaining 3 kale versions, and the breastfeeding history was treated as a quantitative variable.

The multilevel ordered logistic model was fit for the sum of the number of accepted tastes of kale, caregiver ratings of child liking of kale versions, and child behaviors.

The level 1 equation for the multilevel ordered logistic model is written as follows:

$$level - 1$$

$$ln\left(\frac{P(y_{ti} \ge k)}{P(y_{ti} < k)}\right) = b_{0i} + b_1 \cdot sug1.2_{ti} + b_2 \cdot sug1.8_{ti} + b_3 \cdot salt_{ti}$$
(1)

where the log-odds of the outcome (e.g., number of tastes) at trial t for person i is on the left-hand side of the equation, b_{0i} is the random intercept, $sug_{1.2ti}$, $sug_{1.8ti}$, and $salt_{ti}$ are the dummy-coded variables to draw comparisons between the different types of kale (i.e., 1.2% added sugar, 1.8% added sugar, and 0.2% added salt), and τ_k is the threshold for level k. The level 2 equation for the multilevel ordered logistic model is written as follows:

level - 2

$$b_{0i} = \gamma_1 \cdot yng_i + \gamma_2 \cdot old_i + \gamma_3 \cdot kale_{1i} + \gamma_3 \cdot bf_i + u_i - \tau_k$$
(2)

where yng_i and old_i are the 2 dummy-coded variables to draw age comparisons, $kale1_i$ is the dummy-coded variable to indicate whether this is the first kale version, bf_i is the breastfeeding variable, and u_i is the random effect, which is assumed to be normally distributed with a mean of 0 and an estimated variance σ_0^2 .

TABLE 2	Characteristics of caregivers and children who
participated	d in the Good Tastes Study ¹

Demographic characteristic	Value
Child participants	
Group	
Infants (6 to <12 mo)	46 (43%)
Young toddlers (12 to <18 mo)	40 (38%)
Older toddlers (\geq 18 to 24 mo)	20 (19%)
Sex, n(%)	
Female	49 (46%)
Male	57 (53%)
BMI z score, mean \pm SD	0.12 ± 0.97
Underweight	3 (2.9%)
Normal weight	95 (90.5%)
Overweight/obese	7 (6.7%)
Adult participants	
Sex	
Female	100 (94%)
Male	6 (6%)
Age, y	
18–29	25 (24%)
30–49	79 (74%)
Not reported	2 (2%)
Race	
White	87 (82%)
Other	19 (18%)
Ethnicity	
Hispanic/Latino	9 (8%)
Non-Hispanic/Latino	97 (92%)
Education	
<high school<="" td=""><td>3 (3%)</td></high>	3 (3%)
Some college/trade school	7 (7%)
College graduate/postgrad	96 (90%)
Income	
<\$31,950	15 (14%)
\$31,951—\$60,570	29 (27%)
>\$60,570	54 (51%)
Not reported	8 (8%)
BMI	25.7 ± 6.3

¹Values are presented as number of participants (%) unless otherwise indicated.

Results

Participant characteristics

Demographic characteristics of parents and children (n = 106; 53% male children; mean age 13.4 mo) who participated in the Good Tastes Study are described in Table 2. Briefly, parents (94% mothers) were predominantly non-Hispanic white, well educated, and the majority were from middle to upper income families.

Number of successful tastes of kale

The likelihood of accepting tastes was greater for the control version, compared with all other versions (1.2% added sugar b = -1.315, P < 0.001, 1.8% added sugar b = -1.397, P < 0.001, 0.2% added salt b = -2.047, P < 0.001) and there was a lower likelihood of accepting the first version presented (b = -0.586, P = 0.007) compared with other versions (see Figure 2). The likelihood of accepting tastes of kale was greatest for youngest infants (reference group 6 to <12 mo) compared with both other groups of older infants (b = 1.726, P < 0.001 for middle age group; b = 2.911, P < 0.001 for oldest age

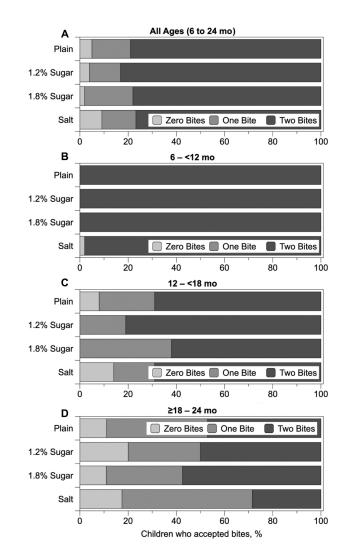


FIGURE 2 Percentage of children who accepted bites (range from 0 to 2 for each version) for the 4 kale versions, by age groups and for the total group. A = percentage of bites accepted for each version and for all children; B = percentage of bites accepted for each version for 6- to <12-mo-old children; C = percentage of bites accepted for each version for 12- to <18-mo-old children; D = percentage of bites accepted for each version for 18- to 24-mo-old children. Some children refused to complete tasting all kale versions (n = 28; infants 6 to <12 mo, n = 1; young toddlers 12 to <18 mo, n = 16; oldest toddlers 18–24 mo, n = 11).

group). Breastfeeding history was not significantly associated with the likelihood of accepting tastes.

Caregiver ratings of child liking of kale versions

There were no differences in maternal ratings of children's liking by kale version, or by order of introduction (see Figure 3). The oldest toddlers were more likely to be rated lower in liking (b = -1.874, P = 0.014), irrespective of version, than the youngest reference group. There was no significant association with breastfeeding history.

Children's rate of accepting kale tastes

For rate of acceptance, children were less likely to easily accept tastes of salty (b = -1.437, P < 0.001) or sweetened kale (b = -0.856, P = 0.015 for 1.2% sugar; b = -0.798, P = 0.001 for 1.8% sugar) compared with the control (plain) version. There was a trend for the first version to be

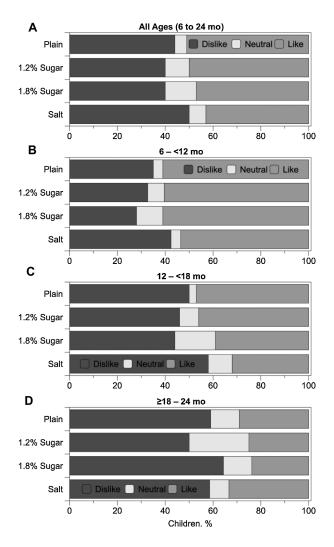


FIGURE 3 Mothers' perceptions of child liking for the 4 kale versions (control, with 1.2% and 1.8% added sugar, and 0.2% salt) by age groups and for the total group. A = percentage of bites accepted for each version and for all children; B = percentage of bites accepted for each version for 6- to <12-mo-old children; C = percentage of bites accepted for each version for 12- to <18-mo-old children; D = percentage of bites accepted for each version for 12- to <18-mo-old children; (n = 28; infants 6 to <12 mo old, n = 1; young toddlers 12 to <18 mo old, n = 16; oldest toddlers \geq 18 to 24 mo old, n = 11).

less likely to be easily accepted compared with subsequent versions (b = 0.451, P = 0.073). With respect to breastfeeding history and age effects, the results for acceptance varied by breastfeeding variable: no effect of breastfeeding in the last 7 d was noted but when the variable capturing how recently the child had experienced consuming breastmilk (i.e., how long since having been breastfed) was included in the model, older children were less likely to easily accept bites compared with the youngest referent group of infants (b = -0.993, P = 0.043).

Children's observed behaviors in response to kale versions

Child behaviors differed by age and, in some cases, by version of kale purée (see Figure 4). The oldest children were more likely to display avoidant behaviors (b = 1.279, P < 0.001), more playing with food (b = 2.918, P < 0.001), and self-feeding

(b = 1.786, P = 0.005) behaviors than the referent group of youngest infants. Children in the middle age group displayed a greater likelihood of crying (b = 1.272, P = 0.011), of playing (b = 2.192, P < 0.001), and of self- or cofeeding (b = 1.525, P = 0.014) than the youngest infants.

Differences in behaviors tended to be more evident in response to the salty version of the kale purées. A significantly higher likelihood of crying (b = 0.553, P = 0.040) and a trend for a lower likelihood of self-feeding (b = -0.425, P = 0.077) occurred in response to the salty version compared with the plain control kale. Children cried less in response to the first version of kale purée (b = -0.432, P = 0.025) that was offered to them. No version or order effects were noted for avoiding, expelling, or playing.

Significant effects for child behaviors were noted for breastfeeding over the previous 7 d such that higher likelihoods of self-feeding (b = 0.246, P = 0.001) and playing with food (b = 0.207, P = 0.005) were found when the child had received more breastmilk feedings in the last 7 d.

In summary, age was the biggest factor related to children's acceptance and behavior in response to tasting the kale purées: younger children were more likely to consume the offered bite and more likely to be rated higher on perceived liking than the oldest age group. Older toddlers were more likely to show avoidant behaviors, to play with the food, and to selffeed than the youngest referent group. Children in the middle age group were more likely to cry, play with the food, and self-feed.

The plain (control) kale purée, without added sugar or salt, was more likely to be accepted and the salty version elicited more crying (although we cannot rule out that results for the salty version may have arisen due to sample position, as discussed below). Also, order effects were noted in that the first version offered to children was less likely to be a successfully accepted taste and crying was less likely to occur on the first taste.

Discussion

The purpose of the Good Tastes Study was to investigate whether adding small amounts of sugar and salt-which should reduce the bitterness of a dark green, difficult-to-like vegetable, kale-would influence infants' and toddlers' acceptance of this vegetable. In contrast to expectations (and prior data from adults), the plain kale purée control was better accepted than all other versions and very few significant effects of taste modification were noted and none were noted for adding sweetness (in the form of small amounts of sucrose). Notably, there was some indication that the first version of kale offered, irrespective of whether it was sweetened or not, was less likely to be accepted than others and this may reflect children's response to novelty, in general, rather than a specific response to the taste, per se (48). Children did not cry more for the first version but did exhibit wariness; particularly older children. Anecdotally, caregivers were observed verbally reasoning with children about the different versions; making statements like "this one is different" or "just try it, it's not the same as the other one," with some success in persuading their child to try a bite.

Effects of added sugar on kale acceptance

Evidence that sweetness did not improve children's acceptance of the kale purées was an unexpected result as, generally

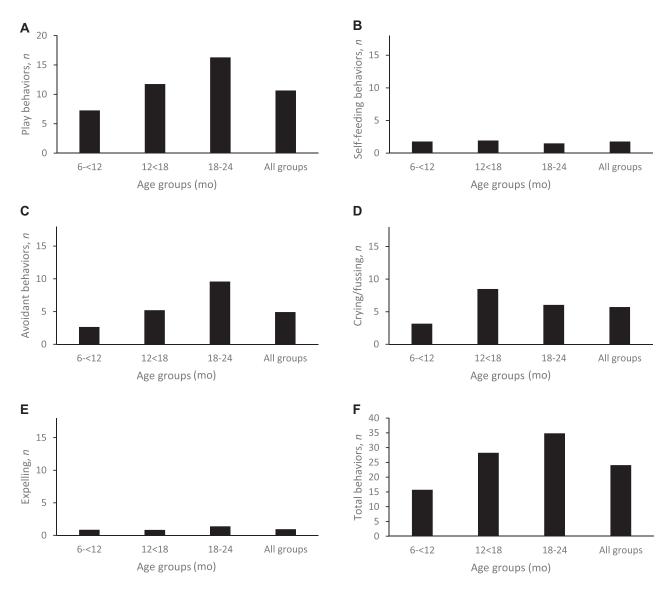


FIGURE 4 Frequency of child behaviors in response to kale by age groups and for the total group. A = frequency of play behaviors; B = frequency of self-feeding behaviors; C = frequency of avoidant behaviors; D = frequency of crying and fussing behaviors; E = frequency of expelling; F = frequency of all behaviors exhibited. Some children refused to complete tasting all kale versions (n = 28; infants 6 to <12 mo old, n = 1; young toddlers 12 to <18 mo old, n = 16; oldest toddlers ≥ 18 to 24 mo old, n = 11).

speaking, children like and accept foods that are sweet (52). The amounts of added sugars used here in purées were purposefully quite low (1.2 or 1.8 g/100 g puree or \sim 1/4 to 1/2 teaspoon per cup), as they were intended to mask bitterness rather than result in high levels of perceived sweetness. The formulations for the amounts of added sugar used here were based upon adult responses, using both naïve consumers and trained descriptive analysis panelists; those data indicated that in adults, the highest level of bitterness reduction was achieved at 1.2 and 1.8% (wt/vol) sucrose and that the purées were perceived to be simultaneously less bitter and somewhat sweet (11). Young children have preferences for higher intensities of sweetness than adolescents or adults (16, 53), and thus it is possible that the addition of higher levels of sweetness could have led to different acceptance and behavioral responses for infants and toddlers in our study. That said, our goal here was mixture suppression (i.e., reduced bitterness), rather than increased affective responses from elevated levels of sweetness. Evidence for mixture suppression in young children is very limited, but prior data support the existence of this phenomenon in 12– 41-mo-olds (37, 42); accordingly, the failure to see an increase in acceptance here is presumably not due to age specificity of mixture suppression.

Notably, young infants' responses to sweetened substrates (e.g., sweetened water) have been reported to be associated with dietary experience such that infants who have previous experience with consuming sugared water preferred higher levels of sucrose; preferences that were sustained at least until 6 mo of age (54). Here, we did not collect information specific to infants' and toddlers' dietary exposure to sweetened foods, so we cannot determine whether such dietary experiences (e.g., puréed vegetables mixed with fruit or fruit juice concentrates) could have influenced their acceptance of the different kale purée versions. Recently, we reported that blending vegetables with fruit increases sweetness but also covers up the other flavors of the vegetables (55).

Age-related effects of kale acceptance

Child age was the most consistent factor associated with children's acceptance of the kale purées and their behaviors exhibited during feeding. The youngest infants, who are least experienced with complementary feeding, accepted almost all bites of kale, irrespective of version, compared with children \geq 12 mo of age. As demonstrated by number of tastes as well as the rate of accepting tastes, children in the oldest age group (18–24 mo of age) consumed kale with less enthusiasm (i.e., more refusals), which could be an indicator of emerging neophobia. In addition, the duration since having been fed breastmilk was longer for older children, and it is possible that the reduction in exposure to similar flavors via breastmilk could have contributed to the decrease in rate of acceptance of kale. Whether learning and experience at an early age (<12 mo) translates to later acceptance is less well known.

These findings are consistent with those of Mennella and colleagues (2017) who reported that young infants (\sim 8 mo of age) readily accepted a novel carrot-flavored cereal after exposure to carrot flavor through their mother's breastmilk (23). They concluded that the early period of complementary feeding "may be an optimum time for [the] infant ... to learn to like the taste of healthy foods." Our findings, though from a cross-sectional study design, align with those of Mennella and others (23, 26, 27, 56) and together indicate that introduction to new foods in general, and in our study, a bitter green vegetable, may be more easily achieved early on in the complementary feeding process. More research could help to determine whether critical periods exist for development of food acceptance patterns and whether early acceptance is associated with later intake (3).

More negative behaviors were elicited from older infants in response to the salted version of kale. In general, although some literature addresses young children's liking for salt (57, 58), less information exists in the literature with respect to infants' (>6 mo of age) and toddlers' (12–24 mo of age) responses to salt taste. A few studies suggest that young infants (2-6 mo of age) prefer water with salt over plain water (59-61). However, this preference does not generalize to foods that are not previously experienced as salty [i.e., breastmilk (62)]. It has been noted that older infants and toddlers are likely to have had more exposure to salt in foods as the number and amount of complementary foods ingested increases. In 1 study with 2- to 3-y-olds, Bouhlal and colleagues (2014) reported increases in intake and caregiver ratings of perceived liking of a root vegetable (salsify) after repeated exposure when 0.2% salt was added to the vegetable (63). Here, toddlers' responses to lightly salted kale in a purée was less positive. This may reflect children's dislike for the perceived level of saltiness in the kale purées or, more likely, it may reflect an order effect, as children always received the salty kale last (see Methods for rationale). It seems likely that by the time children were asked to try the fourth version, they were simply tired of the protocol (or the kale) and showed more avoidant behaviors and cried more in response to the final sample.

In our data, older children were more likely to show lower rates of acceptance of kale. Toddlers (i.e., the oldest age group, from ≥ 18 to 24 mo of age) also were more likely to exhibit behaviors like playing with the food and attempts to self-feed, particularly when having been breastfed more often in the last 7 d. One interpretation of these findings might be that neophobia is beginning to emerge. Alternatively, it may be that mothers who continue to breastfeed also allow or support responsive feeding practices like sensory exploration of food and learning to self-feed. Displaying interest in (playing) and engaging in autonomy demands (self-feeding) are developmentally appropriate and can be considered as important for encouraging food acceptance (46, 64). However, playfulness with food has been interpreted by others to indicate *disinterest* in food and led to food rejection, although these findings were reported with respect to infants 4–6 mo of age (45, 65). Those authors also cautioned that application of their findings should be replicated with older participants.

Caregivers' perceptions of children's acceptance of kale

Caregivers' perceptions of children's liking for kale did not differ by version or by order of introduction. Caregivers did perceive that older children were less likely to like the kale than the youngest children and this is consistent with our findings that older children were less likely to accept tastes and demonstrated more avoidant behaviors, irrespective of taste modification. Most studies that have gathered data about infant exposure to novel foods and infant behaviors in response to foods have focused primarily on infants 6–12 mo of age (33). The results from repeated exposure studies are mixed with respect to maternal ratings of children's liking, with some studies finding evidence of a relationship between maternal liking ratings and children's facial expressions (31) and intake (23, 29), whereas others reported no relation to these acceptance variables (28, 32).

Of note, in a study of infants exposed to a variety of vegetables, mothers did not report immediate postintervention differences in liking for the vegetables to which infants were exposed (at 6 mo of age) but in subsequent follow up at 12 mo of age, mothers in the intervention group reported higher infant liking for the vegetables to which children had been exposed (26). Our findings suggest that some periods are more sensitive than others for introduction of novel foods and that, in accordance with maternal ratings, toddlers liked the kale purées less.

Placing the findings within a greater context of children, families, culture, and child feeding

While important information is provided by our results, these data supply a first look at young children's responses to a novel bitter-tasting vegetable. The convenience sample studied here was homogeneous with respect to many sociodemographic factors: the sample was mostly White, and caregivers were predominantly well-educated with reported family incomes in the middle to upper income range. Some of children's reactions and responses to novel foods is biologically driven: bitterness evokes negative facial reactions across species (59, 66) and response to novelty has been associated with child temperament, which is core to the individual child (31, 48). Addition of a control familiar food to a future protocol would help to disentangle the emergence of neophobia from children's responses to a novel food. However, young children's nutrition status, specifically with respect to iron and zinc, also plays a role in taste acuity and willingness to interact with novelty and these variables are influenced by education, food security, culture, and food beliefs, as well as child feeding practices (67-69). Furthermore, important insights could be gleaned by incorporating information related to maternal liking and consumption of the target food (in this case kale), particularly for mothers who are breastfeeding. Thus, sociocultural influences could lead to different findings when more diverse families and cultures are included in

future research. For example, Keller and colleagues found an interaction between taste genetics, food choices, and the built environment in children aged 4-6 y (70). Furthermore, although our sample included a relatively large sample of young infants and toddlers, the eating and feeding development in this age range accelerates rapidly and would require larger samples to fully elucidate interactions among factors related to development, feeding history, maternal perceptions, child age, and temperament.

This study employed a cross-sectional design to begin to gather data regarding child and maternal factors and, while making new contributions to the evidence base, these relationships would best be studied via longitudinal designs that employ dyadic and sequential coding and analyses to better understand the development of children's acceptance of bitter tasting foods. As an initial study, it may not have been fully powered to detect age and developmental differences. Future studies could use estimates derived from our data to try to ensure that additional studies are adequately powered. Moreover, the study took place in a laboratory under carefully controlled conditions and may not reflect children's and mothers' behaviors and responses in other feeding environments.

Despite these limitations, several strengths of this study should also be highlighted. First, the developmental focus on taste response and the concomitant examination of other child behaviors and maternal perceptions provides a rich picture of young children's responses to a novel food. Our findings also begin to shed light on children's responses to a bitter-tasting food, which have been studied less frequently in exposure studies with infants and toddlers. Further, we conducted wellcontrolled preliminary studies in adults to choose and refine the target foods used here. Last, we utilized observed measures in addition to maternal perceptions, which provide insights into how maternal perceptions align with investigator observations.

Conclusions

Considered together, our findings add important information to the literature focusing on children's acceptance of novel foods and include data from both maternal and child aspects of the dyadic feeding relationship. By combining observed child responses of behavior and acceptance with subjective maternal perceptions of children's liking for the food, the focus shifts from the properties of the food to an emphasis on child age at the time of offering that food. Our findings support the notion that a sensitive period exists—as has been suggested by others—in which it is easier to introduce foods to children. Here, younger infants below the age of 12 mo appeared to more easily accept a dark green vegetable purée. Also, our results support the widespread conjecture that rejection of a first taste does not preclude offering additional tastes to see whether children may change and accept subsequent bites within the same feeding (3).

In contrast to expectations (and prior adult data), reduction of kale bitterness with small amounts of added sugar and salt did *not* result in better acceptance. That is, adding a sweetener did not automatically result in improvements in food acceptance in this group of infants. One interpretation is that the level of sweetness was insufficient to suppress the bitterness of kale. Prior data from trained adult panelists and naïve consumers (11) suggested that bitterness was at least partially reduced at the levels employed by the present study, but it may not have reached the level of masking necessary to influence acceptance in the age group tested here (53). There are other aspects of children's development which may play a role in food acceptance, like the opportunity to explore or play with foods and the extent to which mothers engage in cofeeding or support the older infant to self-feed. Selffeeding during the complementary feeding period has begun to be explored in the context of infant led weaning and has been noted to be associated with lower food fussiness and higher food enjoyment (71). Further research is necessary to determine whether opportunities to engage in these behaviors facilitate or hamper children's food acceptance.

In view of the findings that young infants aged between 6 and 12 mo accepted virtually every taste of kale, irrespective of added sweetness or saltiness, further investigation into whether early exposure facilitates later acceptance (i.e., during the difficult toddler period or when food neophobia begins to emerge) is strongly warranted. Such studies would require longitudinal designs to fully elucidate these relationships.

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The authors' responsibilities were as follows—SLJ, KJM, AJB, and JEH: conceived of the study design; KJG and KJM: led the data analysis; SLJ, KJM, and JEH: obtained funding and supervised the project; AEF: participated in data collection, data interpretation, and contributed to the writing of the manuscript; all authors participated in data collection and data interpretation; SLJ and KJM: led the writing of the manuscript; all authors: contributed to and approved of the writing of the submitted manuscript; and all authors read and approved the final manuscript.

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