Do sunbirds use taste to decide how much to drink?

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Summary

1. Nectarivorous birds typically consume smaller meals of more concentrated than of less concentrated sugar solutions. It is not clear, however, whether they use taste to decide how much to consume or if they base this decision on post-ingestive feedback.

2. Taste, a cue to nectar concentration, is available to nectarivores during ingestion whereas post-ingestive information about resource quality becomes available only after a meal. When conditions are variable, we would expect nectarivorous birds to base their decisions on how much to consume on taste, as post-ingestive feedback from previous meals would not be a reliable cue to current resource quality.

3. Here we tested whether white-bellied sunbirds (\textit{Cinnyris talatala}), foraging from an array of artificial flowers, use taste to decide how much to consume per meal when nectar concentration is highly variable: they did not. Instead, how much they chose to consume per meal appeared to
depend on the energy intake at the previous meal, i.e. how hungry they were.

4. Our birds did, however, appear to use taste to decide how much to consume per flower visited within a meal. Unexpectedly, some individuals preferred to consume more from flowers with lower concentration rewards and some preferred to do the opposite.

5. We draw attention to the fact that many studies perhaps misleadingly claim that birds use sweet taste to inform their foraging decisions, as they analyse mean data for multiple meals over which post-ingestive feedback will have become available rather than data for individual meals when only sensory information is available.

6. We discuss how conflicting foraging rules could explain why sunbirds do not use sweet taste to inform their meal size decisions.

**Key-words:** foraging choices, hummingbirds, nectar concentration, nectarivores, post-ingestive feedback, sunbirds, taste

**Introduction**

Natural food resources vary both spatially and temporally in their quality and quantity, and animals encountering rewards of different value must decide whether and how fully to exploit them (Whelan & Brown 2005). Such decisions are expected to be important for ‘optimising’ feeding efficiency. One way animals can maximise feeding efficiency is to maximise their instantaneous rate of energy intake (DeBenedictis *et al.* 1978; Schmid-Hempel, Kacelnik & Houston 1985; Chen & Jiang 2006; Kim, Gilet & Bush 2011). For example, specialist nectarivores
typically prefer higher concentrations of sucrose over weaker ones as stronger solutions allow a higher rate of instantaneous energy intake, up to the point where viscosity imposes physical constraints on consuming (Hainsworth & Wolf 1976; Roberts 1995; Roberts 1996; Blem et al. 2000; Leseigneur & Nicolson 2009; Köhler et al. 2010; Kim, Gilet & Bush 2011).

In addition, because of the energetic costs of carrying a meal around and how often it is necessary to travel to feed, it is more economical for non-social nectarivores such as hummingbirds to consume slightly smaller meals of stronger than of weaker solutions (DeBenedictis et al. 1978). A meal can be thought of as the total nectar consumed from multiple flowers from when a bird first starts feeding until it leaves a food source. Like hummingbirds, sunbirds also typically consume less per meal when sucrose concentrations are higher than when they are lower (Köhler, Verburgt & Nicolson 2008). Volumetric intake of higher concentrations may also be lower than intake of lower concentrations: more concentrated sucrose solutions take longer to pass through the gut, limiting capacity for further food intake (Downs 1997; Savory 1999).

To demonstrate a preference for higher sucrose concentrations birds must be able to distinguish them from lower concentrations. It is not clear, however, what types of information sunbirds will use to do this or to inform their meal size decisions: current information about the resource, such as taste, or post-ingestive feedback from previous meals, such as satiation (Bacon et al. 2011).

Sensory cues of meal quality may be available prior to ingestion (e.g. sight and smell) or during ingestion (e.g. taste) via receptors in the mouth (Berkhoudt 1985; Yearsley et al. 2006; Skelhorn, Griksaitis & Rowe 2008; Baldwin 2014;
Zhao, Li & Zhang 2015). In addition, some taste information may continue to be available after ingestion via receptors in the gut (Yearsley 2006; Depoortere 2014). Post-ingestive feedback is available only when the decision about how much to consume of a resource has already been taken (Provenza 1995; Yearsley et al. 2006). This past experience of the resource can then be used to predict its value at future visits (Lima 1983; Duncan & Young 2002; Yearsley et al. 2006; Gil, De Marco & Menzel 2007).

It is possible to examine which of these types of information an animal is using in its decision-making by investigating how quickly it responds to changes in resource quality. An immediate response indicates that information about the current state of the resource such as taste is being used, whereas slower responses (e.g. behaviour does not change until subsequent meals) indicate that animals are likely using post-ingestive feedback. How long it takes for post-ingestive feedback to become available will, however, depend on gut transit times. Sunbirds have high metabolic rates and therefore to survive need to forage efficiently and assimilate dietary sugar very rapidly (Nicolson & Fleming 2014). Gut transit times in sunbirds increase with increasing food concentration; in the Palestine sunbird *Cinnyris oseus* from as little as 26 min for 10% sucrose solutions to around 47 min for 50% solutions and transit times are 23 min for 20% solutions in the white-bellied sunbird *C. talatala* (Lerch-Henning and Nicolson 2015; Roxburgh & Pinshow 2002). Food passage rates are similarly rapid for hummingbirds (Karasov et al. 1986; Hainsworth 1989; Tiebout 1989). Post-ingestive feedback about a meal will become available before the entire meal has been processed and we would therefore expect nectarivorous birds to have access to some post-ingestive feedback within a few minutes of ingestion.
Evidence from previous studies suggests that where resources are stable over several hours, hummingbirds (a group of birds known to be able to taste sweet: Baldwin et al. 2014), preferentially use post-ingestive feedback from previous meals rather than the sweetness of the current meal to decide how much to consume: they do not change how much they consume per meal the first time they encounter a different concentration, when they should be able to taste the difference (Bacon, Hurly & Healy 2011). Likewise sunbirds do not immediately change their sucrose intake rate following changes in nectar concentration (Köhler et al. 2008). Such stable resources mean that birds can learn (1) what the energetic value of a resource is, and (2) that it is unchanging, allowing them consume a fairly set volume per meal according to post-ingestive feedback and habit after the first few encounters with the resource. However, feeding for some time on one concentration and then feeding on another concentration, again for a number of sequential meals, seems an unlikely scenario in nature. Given the variation in nectar concentration and volume found in flowers (Corbet et al. 1979; Corbet 2003; Keasar, Sadeh & Shmida 2008), birds may rarely experience two consecutive meals of the same nectar concentration and may rarely consume a single nectar concentration per meal. Thus where resources are variable sunbirds must make two types of decisions per meal about what volume to consume: (1) What volume to consume from each flower visited and (2) What volume to consume in total at that meal. In such variable conditions birds might adjust their meal volumes using sensory information such as taste, as sufficient post-ingestive feedback about the sucrose concentration of the current flower/meal in question will not be available in time to be informative. Multiple decision points also increase the risk of error and could
conceivably reduce feeding efficiency compared to feeding on a constant resource.

Here we provided white-bellied sunbirds *Cinnyris talatala* with a diet that was either constant or varied in concentration between meals (temporal variation) or within meals (spatial variation) to see if they adjusted how much they consumed per meal based on the concentration of sucrose they encountered in any of these contexts. We predicted that (see table 1 for a summary):

**Table 1:** A summary of our main predictions concerning how much sunbirds would be expected to consume per meal or flower visited depending on what type of information they base their decisions on, taste or post-ingestive feedback.

<table>
<thead>
<tr>
<th>Information type used in decision</th>
<th>Scenario</th>
<th>Decision: how much to consume per Meal/ Flower</th>
<th>Expected behaviour</th>
<th>Prediction</th>
</tr>
</thead>
<tbody>
<tr>
<td>Taste</td>
<td>Stronger sucrose solution</td>
<td>Meal</td>
<td>Consume less</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>Flower</td>
<td>Consume more</td>
<td>2</td>
<td></td>
</tr>
<tr>
<td>Post-ingestive feedback</td>
<td>Stronger sucrose solution</td>
<td>Meal</td>
<td>Consume less at next meal</td>
<td>4a</td>
</tr>
<tr>
<td></td>
<td>Flower</td>
<td>Consume the same volume from all flowers</td>
<td>4b</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Longer time since last meal</td>
<td>Meal</td>
<td>Consume more</td>
<td>4a</td>
</tr>
<tr>
<td></td>
<td>Stronger sucrose solution over five consecutive meals</td>
<td>5 meals</td>
<td>Consume less in total across those 5 meals</td>
<td>5</td>
</tr>
</tbody>
</table>
1. **Use of taste in meal size decisions:** When sucrose concentration varied between meals sunbirds, in order to optimise flight costs (frequency of feeding flights and weight of the meal DeBenedictis et al. 1978), would use taste to consume less per meal when the sucrose concentration consumed was stronger than when it was weaker.

2. **Use of taste in individual reward exploitation:** When sucrose concentration varied within meals sunbirds, in order to obtain as high a concentration meal as possible, would use taste to consume less per flower visited within that meal when the sucrose concentration in that flower was weaker than when it was stronger.

3. **Sucrose and energy intake:** When sunbirds foraged on a constant resource compared to one with spatial or temporal variability their meal sizes would be more consistent and they would take in fewer calories, as they would be able to feed more efficiently.

4. **Use of post-ingestive feedback:** If sunbirds used post-ingestive feedback rather than taste in meal size decisions they would (a) consume less when energy intake at the previous meal was relatively high and more when the time since the last meal was relatively long (proxies for relative satiation), (b) consume similar amounts from all flowers visited within a meal regardless of the sucrose concentration they contained as insufficient time elapses during consumption of nectar in individual flowers for post-ingestive feedback to be available.
5. **Importance of measurement period**: Even if there was no relationship between sucrose concentration and meal volumes, post-ingestive feedback accumulated over five consecutive meals would result in birds consuming more during those meals when mean meal concentration was relatively low than when it was relatively high. This would illustrate how measuring intake over a long enough timescale for post-ingestive feedback to become available (5 meals) results in data that could be misinterpreted as evidence for taste.

**Materials and methods**

*Study subjects and housing*

We used one female and four male wild caught white-bellied sunbirds, *Cinnyris talatala*, that were housed in a large outdoor aviary. Sunbirds were mist-netted at Jan Celliers Park, Pretoria, South Africa during the nonbreeding season of 2012. Birds were released at the place of capture after experiments were completed in February 2014. All birds were adult at the time of testing.

The aviary (9.0 m long, 5.5 m wide, 1.8 m high) was constructed of wire mesh (1 × 2.5 cm) over a steel frame. The entire aviary was covered in netting to provide shade for the birds and to exclude bees from their feeders. The aviary was divided into five equal compartments (1.8 m wide) across its length by wire mesh walls with doors (0.68 m wide, 1.8m high) between them of similar construction. The fifth compartment with the external door was used for storing equipment so birds had access only to the first four compartments. The internal doors were all adjacent to the front wall of the aviary and except during testing
were left open so the birds had access to the whole area.

During testing the focal bird was confined to one compartment but could both see and hear its flock mates who had access to the other three compartments. Birds were separated from their flock mates by gently herding them between compartments, thereby avoiding any need to handle them. The birds were provided with natural branches to perch on at a variety of levels and were maintained on an *ad lib* diet of 20 % w/w sucrose solution with a nutritional supplement (Ensure®, Abbott Laboratories, Johannesburg, South Africa) to provide protein, vitamins and minerals. This food was replaced daily. The maintenance feeders were modified 60 ml or 20 ml syringes with the tips blocked and a feeding hole marked with red drilled in the side. The birds also had access to water baths and water feeders as well as any small insects entering the aviary.

*Training*

The birds were trained to feed from an array of artificial flowers, consisting of a sheet of Perspex (5 mm thick x 30 cm wide x 50 cm high) with nine 250 μl microcentrifuge tubes with the lids removed (henceforth ‘wells’), mounted in holes drilled through it. The wells were filled with sucrose solution and lay horizontally when the Perspex sheet (henceforth ‘array’) was hung on the aviary wall. There was a 10 × 0.4 cm perch 4 cm below each well. The wells were all marked with self-adhesive paper reinforcers (internal diameter 5 mm, external diameter 9 mm). Reinforcers were white so as to avoid confusion with any of the colours planned for use in the experimental treatments and to get the birds used to feeding from resources of a novel colour compared to the red circle on their
feeders. The hole in the reinforcer was lined up with the opening of the well so the paper part stuck to the well rim forming a circle of colour around the well.

As in experimental treatments the wells were arranged in a pattern of equilateral triangles with sides 10 cm: this meant that each well was the same distance from its horizontal as from its vertical neighbours in the row above. The wells were laid out such that there was a row of three horizontally across the centre of the array flanked above and below by a row of two with single wells at the mid point at the top and bottom, to form a diamond shape.

Birds were trained to use the array by placing it close to one of their standard feeders and then moving the feeder closer to and eventually onto the array. If the birds did not investigate the wells on the array once the feeder was in amongst them, the colour surrounding the hole on the feeder was changed from red to white to match the wells on the array and it was gradually made harder for the birds to feed from it by turning the feeding hole towards the back. If this did not work (two birds) then five dots of red were added to the reinforcer around the well on the array closest to the feeder to attract the birds’ attention to it. Once the birds had started to feed from the wells on the array, we removed the feeder and continued to let them feed from the array for at least 10 meals and until they had visited all nine wells at least once. Training was completed at least 48 hours before testing.

**Treatments**

There were three experimental treatments: constant, spatial variability and temporal variability. The volume of rewards was the same in all treatments (60 µl per well). We used three sucrose concentrations (10, 20 and 30% w/w)
selected to span the range of nectar concentrations that sunbirds typically consume in the wild (Nicolson & Fleming 2014). Birds were allowed to visit as many wells on the array as they chose to per meal. A meal was defined as the time from a bird first starting to drink from a well to the time it left the array. There were two exceptions to this. Firstly, if the bird left the array for less than five seconds before returning, for example to catch an insect, the time from it starting to feed from the array again to leaving it more permanently was added to the preceding meal. Secondly, if a bird sat on the array without feeding for over a minute the meal was considered to have ended.

In the constant treatment, rewards were the same concentration (20% w/w sucrose solution) in all nine wells.

In the spatial variability treatment, rewards consisted of three different sucrose concentrations (10, 20 and 30% w/w sucrose solutions equivalent to 0.303, 0.632 and 0.988 mol/l). Each concentration was allocated at random to three of the nine wells in the array and the pattern of reward locations was altered at random after each meal.

In the temporal variability treatment, rewards were one of three different sucrose concentrations (10, 20 or 30%). In this treatment all nine wells were filled with the same concentration for each meal and the concentration was changed between meals. The concentration experienced by birds at each meal was selected pseudo-randomly so that each concentration appeared twice in every six meals.
*Experimental Feeding Array*

The feeding array used in experimental treatments differed slightly from the Perspex training array to reduce the chances of location preferences. It was slightly smaller (5 mm thick x 35 cm wide x 30 cm high) and more crucially the layout of wells differed so that no row contained only a single well. The nine sucrose wells each with a wooden dowel perch below were arranged in three rows of three wells with the middle row being slightly offset. The array was hung vertically from the aviary walls at about 165 cm from the ground. Wells were surrounded by coloured paper reinforcers, all the same colour within treatments (e.g. constant = yellow, spatial = blue, temporal = pink). Each bird was allocated a different colour for each treatment so that no two birds had the same colour for the same treatment. *We avoided using red because of the pre-existing association with the colour of the feeding aperture on the maintenance feeders.*

*Experimental Protocol*

Each bird completed the three treatments consecutively. The order of treatments was pseudo-randomised to ensure that no two birds received them in the same order. Each treatment lasted for 30 meals (2.5 to 4 hours). Observations were taken over eight hours each day between 0900 and 1700 such that each bird experienced one to all three treatments within a day. Two birds completed one treatment on their first day and two treatments on their second day, two birds completed two treatments on their first day and one treatment on their second day and one bird completed all three treatments in a single day. *We did not let birds start a treatment unless we were sure they would finish it within the same day. During the night birds were returned to the flock.*
Meal durations and the time of each meal from the start of the treatment were recorded using a stopwatch. For each meal we recorded which of the nine wells the bird visited, the concentration of sucrose in those wells and, using 75 µl haematocrit tubes, how much sucrose solution birds left in each well they had visited. To ensure that experimenters spent a similar amount of time in the aviary taking measurements with the haematocrit tube after each meal in all three treatments all wells were emptied and refilled between meals.

Experimenters made observations from outside the aviary by looking through the transparent array to see what the bird was doing. They sat at least four m away from the aviary in order to get a clear view without disturbing the birds.

**Data analysis**

Data were analysed in JMP version 7. Models were linear mixed effects models (LMM) with bird included as a random factor. Random slopes were fitted for all main effects. Data were square root transformed for normality where appropriate.

**Overview: Consumption of different concentrations**

We used an LMM to assess whether birds visited a similar number of wells and consumed a similar volume of all concentrations in the temporal and spatial treatments. Data were the total number of wells visited and the total volume consumed of each concentration by each bird. Data for the constant treatment where only one concentration was encountered were excluded.
1. Use of taste in meal size decisions

To see if sunbirds consumed smaller meals of higher than of lower concentrations, we calculated the total volume they consumed per meal and for the spatial treatment where multiple concentrations were consumed per meal, the mean concentration (sweetness) of the meal. Excluding data for the constant treatment where only one concentration was encountered, we used an LMM to assess whether meal volume was related to: meal concentration, treatment or an interaction between the two.

2. Use of taste in individual reward exploitation

We examined data from the spatial variability treatment to see if there was any indication that the sunbirds were using taste within meals to decide how much to consume from individual wells. We used an LMM to test if the volume consumed per well was related to the concentration of sucrose in that well and a t-test to see if birds consumed a higher concentration than the mean concentration available. We used F-tests to determine if, relative to one another, birds consumed more similar amounts per reward of some concentrations than other concentrations. Data were the variance of the mean volume consumed per well by each bird of each concentration.

3. Sucrose and energy intake

To assess whether the volume and energy consumed per meal was more consistent in the constant treatment than in either of the variable treatments, we calculated for each treatment the mean and variance of each bird’s meal volumes and energy content. Data were for each bird and treatment and were analysed
using LMMs. In addition, to test whether the sucrose that sunbirds consumed provided them with a similar amount of energy per unit time for all three treatments we divided the energy they consumed per meal by the time elapsed since their last meal and used an LMM to assess the relationships between their energy intake/time elapsed and treatment. Data were for each bird, treatment and meal.

Energy consumed was calculated as follows:

Energy (j) = (Volume consumed (l) × concentration (mol/l)) × (molecular mass of sucrose: 342g) × (energy content of sucrose per gram: 16800 j).

4. Use of post-ingestive feedback in meal size decisions

To test whether birds consumed a smaller volume when energy intake at the previous meal was relatively high and a larger volume when the time since their last meal was relatively long we calculated their energy intake per meal and used an LMM to assess the relationships between the volume consumed per meal, energy intake at the previous meal and time since the previous meal. Data were for all three treatments.

5. Importance of measurement period

To illustrate how data collected over multiple rather than single meals could be misinterpreted as the use of sweetness to decide how much to consume rather than post-ingestive feedback, we calculated the total volume birds consumed over five consecutive meals (a period averaging 21.59 ± 0.79 min, over which time post-ingestive feedback could have contributed to decision-making) and
using LMM tested whether this volume was related to: the mean concentration of those meals (sweetness), treatment or an interaction between the two. Data for the constant treatment where only one concentration was encountered were excluded to make this analysis directly comparable to analysis 3.

Results

Overview

On average, sunbirds fed from the experimental array once every 5.34 ± 0.14 S.E. min and during each meal they visited 2.75 ± 0.08 wells and consumed a total of 95.55 ± 3.34 μl of sucrose solution.

Consumption of different concentrations: Within the spatial and temporal treatments birds visited a similar number of wells ($R^2 = 0.13, F_{2,8} = 1.31, p=0.321$) and consumed a similar total volume ($R^2 = 0.33, F_{2,8} = 1.75, p=0.233$) of each concentration.

1. Use of taste in meal size decisions

Sunbirds did not adjust the volume of their meals relative to mean meal concentration (Adjusted $R^2 = 0.13, F_{1,4} = 0.17, p=0.697$: Figure 1). Prediction 1 was therefore not met. Meal volume was not affected by treatment ($F_{1,4} = 0.89, p=0.396$), and there was no interaction between meal concentration and treatment ($F_{1,283} = 1.74, p=0.188$).
Figure 1: Volumes consumed during single meals as a function of the mean sucrose concentration of those meals. The five dashed lines are trend-lines for individual birds. N= 300 data points. Data are for the spatial variability and temporal variability treatments (constant treatment was excluded).

2. Use of taste in individual reward exploitation

Overall, there was no difference among the three concentrations in how much birds drank per well when they encountered them (Adjusted $R^2 = 0.47$, $F_{1,4} = 1.22$, $p=0.329$: Figure 2). Only two of the five birds did as predicted and consumed more from wells containing stronger solutions than they did from wells containing weaker solutions (Figure 2). Consequently, the mean concentration consumed by sunbirds in this treatment ($21.56 \pm 0.87\%$) did not differ from 20%, the mean concentration available: t-test: $t_4 = 1.78$, $p=0.14$).

Prediction 2 not met, prediction 4b met.
**Figure 2:** In the spatial variability treatment, the volume birds consumed per well did not vary on average among sucrose concentrations. There was however greater variation among birds in how much they chose to consume from 10% and 20% sucrose wells compared to how much they chose to consume from 30% sucrose wells. Each line represents a different bird. The styles used for each bird are the same as in previous figures. Data are the mean ± SE volume (µl) consumed per well of each sucrose concentration by each bird, across all 30 meals.

We did, however, observe that there was greater variance among birds in how much they chose to consume per well when they encountered 10% or 20% wells than when they encountered 30% wells (F-test two-sample for variance: 10 | 30, $F_{4, 5} = 77.64, p < 0.001$; 20 | 30, $F_{4, 5} = 24.05, p = 0.004$: Figure 2).
3. Sucrose and energy intake

The volume consumed per meal was similar across treatments ($R^2 = 0.69$, $F_{2, 8} = 2.31$, $p=0.161$) as was variance in the volume consumed per meal ($R^2 = 0.71$, $F_{2, 8} = 0.91$, $p=0.439$) and the mean energy consumed per meal ($R^2 = 0.35$, $F_{2, 8} = 0.152$, $p=0.275$). Variance in energy consumed per meal was however lowest in the constant treatment and highest in the temporal treatment ($R^2 = 0.67$, $F_{2, 8} = 5.18$, $p=0.036$; Variance constant = 22.88, spatial =32.77, temporal = 41.42; paired t-test, constant | temporal $t_4 = 3.77$, $p= 0.019$). Prediction 3 partly met.

The energy consumed/time elapsed was similar in all three treatments (Adjusted $R^2= 0.03$, $F_{2, 8} = 1.54$, $p=0.271$).

![Figure 3: The volume birds consumed at each meal depended on their energy intake at their last meal. Data are shown for each meal and each bird. The five dashed lines are trend-lines for each bird (the styles used for each bird are the same as in figures 1). N= 450 data points. Data are for all three treatments.](image)
4. Use of post-ingestive feedback in meal size decisions

Birds consumed more sucrose following meals with a lower energy intake (Adjusted $R^2 = 0.22$, $F_{1, 8} = 13.46$, $p=0.009$: Figure 3) and tended to consume more when it had been longer since they last fed ($F_{1, 1} = 40.62$, $p=0.099$). There was no effect of treatment ($F_{2, 7} = 1.98$, $p=0.200$). Prediction 4a met.

**Figure 4:** Volumes consumed during five consecutive meals as a function of the mean sucrose concentration of those five meals. The data are exactly the same as for Fig. 1 except that the single meals presented in Fig. 1 are grouped into sets of five consecutive meals. The five dashed lines are trend-lines for individual birds (the styles used for each bird are the same as in Fig 1 and 2). N= 60 data points. Data are for the spatial variability and temporal variability treatments (constant treatment was excluded).
5. Importance of measurement period

The total volume consumed over five meals was related to the mean concentration (sweetness) of those meals (Adjusted $R^2 = 0.23$, $F_{1,5} = 10.56$, $p=0.024$; Figure 4), the volume consumed in five meals was not affected by treatment ($F_{1,1} = 1.86$, $p=0.252$), and there was no interaction between meal concentration and treatment ($F_{1,16} = 0.25$, $p=0.619$). Prediction 5 met.

Discussion

Use of taste and post-ingestive feedback in deciding how much to consume

Contrary to our predictions, sunbirds did not appear to use taste to inform their feeding decisions about how much to consume per meal. They did not adjust the volume of their meals depending on the concentration of sucrose they were consuming, either when they experienced different single concentrations among meals or when they experienced a mix of concentrations within meals. Furthermore, when the sunbirds encountered a range of concentrations within a meal, there was on average no difference among concentrations in how much they consumed per well. This is a contrast to what has been observed in experiments with hummingbird where birds encounter a number of different sucrose concentrations per meal and are more likely to fully empty wells containing higher (e.g. 35% compared to 25%) concentrations (Bailey et al unpublished). However, while all the sunbirds consumed a similar amount of 30% sucrose per well encountered, they varied considerably in how much they drank per well of 20% or 10% sucrose, with some birds consuming less and some consuming more of lower than of higher concentrations. This suggests that...
they may have been using taste to decide how much to consume from individual wells.

There are three possible reasons why sunbirds did not adjust their meal sizes to reflect the sucrose concentration consumed: 1). They cannot taste sweetness, 2). They can taste sweet but cannot or are insufficiently motivated use this information alone to make decisions about meal size (Halpin et al. 2012), 3). They can taste sweet and can use taste to make decisions about meal size, but chose not to in this instance as the costs of not consuming ‘optimal’ volumes of different concentrations were outweighed by the benefits of following a fairly fixed feeding routine (Mclinn & Stephens 2005), discussed below.

*Can sunbirds taste sweet?*

The apparent unwillingness or inability of sunbirds to use taste to decide how much to consume leads us to wonder if they do have a sense of sweet taste. Hummingbirds are the only birds known to have a taste receptor sensitive to sweet; it is a repurposed umami receptor (Baldwin et al. 2014). This ability to sense sweet has, however, evolved since hummingbirds diverged from the swifts (Baldwin et al. 2014). Hummingbirds and swifts (Apodiformes) are in a different order from the sunbirds (Passeriformes). Therefore, if sunbirds do have a sense of sweet taste they would have evolved it separately from hummingbirds. Some birds such as chickens do not appear to be able to taste sweet (Roura, Baldwin & Klasing 2013; Baldwin et al. 2014). Experimental evidence however, indicates that others do. For example frugivorous cedar waxwings (*Bombycilla cedrorum*), were able to distinguish between agar cubes with and without sugars before
swallowing the sugar cubes (Martinez del Rio, Karasov & Levey 1989),
suggesting that the ability to taste sweet may have also evolved in some
passerine birds.

There is some evidence that sunbirds at least can taste the difference
between sucrose and hexose solutions, preferring sucrose to a 1:1 mixture of
glucose and fructose in choice tests with small volumes (Nicolson et al. 2015). As
the sweet-taste receptors of hummingbirds respond to a number of different
sugars (Baldwin et al. 2014), it seems likely that the same taste receptors that
allow sunbirds to differentiate between sucrose and hexose would also allow
them to taste differences among different sucrose concentrations. Furthermore,
we observed in the spatial variability treatment that although overall there was
no difference among concentrations in how much birds consumed on average
per well, individual birds responded differently to 10% and 20% wells but all
consumed a similar mean volume from 30% wells. This suggests to us that
individual birds could taste differences among sucrose concentrations but then
made different decisions from one another in response to this information. We
consider it most likely that sunbirds can taste sweet but either cannot, choose
not to or are insufficiently motivated to use this information to inform decisions
about how much to consume per meal. Indeed in another context, whether or not
birds discriminate among mimetic prey, motivation appears to be an important
factor (Halpin et al. 2012). Even if sunbirds cannot taste sweet they may still
have been able to use solution viscosity as a cue to its concentration and post-
ingestive consequences but this information also appears to have been largely
ignored.
Most past experiments fail to distinguish between taste and post-ingestive feedback

As most studies of bird sugar concentration preferences record preferences that develop over multiple meals (1 to <24 hours) rather than within meals, it is unclear for species used in these studies if they can actually taste sweet or are developing preferences via post-ingestive feedback (for example: Duncan 1960; Vince 1963; Brindley 1965; Harriman & Milner 1969; Schuler 1983; Levey 1987; Martinez del Rio et al. 1988; Matson, Millam & Klasing 2000; Matson, Millam & Klasing 2001).

The preference of nectarivorous and frugivorous birds for different sugars has also attracted considerable attention (see Napier et al 2013 for review). Unfortunately, like studies of nectar concentration preferences, almost all experiments have been conducted over multiple meals, making it impossible to disentangle taste and post-ingestive responses (Fleming et al. 2004; Fleming et al. 2008; Wellmann & Downs 2009; Odendaal et al. 2010; Medina-Tapia et al. 2012). The consistent finding of preference for sucrose at high sugar concentrations and preference for hexose sugars at low sugar concentrations can be explained by differences in sucrase activity in the intestine of different species (Napier et al. 2013). This demonstrates a clear role for post-ingestive feedback in nectar choices, but does not rule out a role for taste in detecting sugar concentration at the time scale of single meals – the focus of this study.

Experiments aimed at decoupling taste and viscosity cues to sucrose concentration through the use of thickening agents (e.g. Tylose™) indicate that viscosity is a major determinant of food intake rate, as both licking rate and tongue loads of white-bellied sunbirds are reduced at higher viscosities (Köhler et al. 2010). We would therefore expect birds to be able to detect viscosity both
directly by its texture and indirectly via its effects on intake. Unfortunately, however, studies that manipulate viscosity and look at bird preferences for different solutions report only on intake rates and preferences that have developed over multiple meals rather than within a meal, making it impossible to determine if birds will base their decision on viscosity alone in the absence of post-ingestive information (Stromberg & Johnsen 1990; Köhler et al. 2010). It would be informative therefore to investigate how birds respond to variation in solution viscosity within the timescale of single meals.

The volume our sunbirds chose to consume per meal appears to be simply a result of how hungry they were as meal volume was related to their energy intake at the last meal and the time since that meal. Therefore, if by chance they encountered only low concentrations over five consecutive meals they consumed a greater total volume of sucrose solution over that time than when they encountered a higher mean concentration over five consecutive meals. This relationship between meal size and meal concentration across multiple (post-ingestive feedback available) but not single (post-ingestive feedback not available) meals illustrates how a response to sucrose concentration can arise even when the subject cannot taste sweet and illustrates how misleading it would be to interpret such data as evidence of an ability to taste sweet or use sweetness as cue to sucrose concentration in feeding decisions.

**Implications for feeding behaviour**

Animals are generally expected to use cues to inform behavioural decisions where cue reliability is greater than environmental certainty (Mclinn & Stephens 2005). They are also expected to respond more rapidly to changes in resource
quality and place more weight on current information when environmental conditions are unpredictable than when conditions are stable (Shettleworth et al. 1988; Dunlap & Stephens 2012). Thus in our experiments where sucrose concentration was highly unpredictable and the taste (current information) was a reliable cue to sucrose concentration, we would certainly have expected the birds to use taste to adjust their meal sizes if they could, even though flight distances were probably shorter and food more abundant in the aviary than in many wild situations. That they did not and behaved much as observed in situations where concentration is held stable over much longer periods (Köhler et al. 2008; Bacon, Hurly & Healy 2011), suggests to us that while sweet taste may be useful in other contexts the benefits of using taste to adjust meal size are outweighed by other unquantified factors. Indeed, in general the tendency for animals to favour following a fixed behavioural routine over paying attention to potentially beneficial cues is poorly understood (Mclinn & Stephens 2005).

There is some dichotomy between the two basic rules that govern how much birds are expected to consume of different sugar concentrations. The first rule suggests that, given the choice, birds should consume higher rather than lower sucrose concentrations as this maximises their sugar intake rate, at least between about 10-40% sucrose (Hainsworth & Wolf 1976; Roberts 1995; Leseigneur & Nicolson 2009; Kim, Gilet & Bush 2011). The second rule is that they should consume smaller meals when meal concentration is high than when it is low (DeBenedictis et al. 1978). So on the one hand they should reject low concentrations (when encountered within a meal) and on the other they should consume more of low than of higher concentrations (when they constitute whole meals). If, as is often the case in nature but not in our experiments, the volume of
nectar available is highly variable, birds may frequently have to move to a new flower patch before they are satiated. Under such conditions the definition of where one ‘meal’ ends and another begins may become blurred and it becomes unclear which rule the birds should prioritise. It is then possible that on average it is better for birds to consume as much as is required to satisfy current hunger levels rather than consuming less of higher concentrations and risking an energy deficit if the next flower patch visited contains very little sucrose, or consuming more of low concentrations and risking not being in a position to fully exploit any high concentration flowers encountered in the next few minutes.

Our data from the spatial variability treatment suggest that different individuals may prioritise different rules, with some choosing to consume smaller amounts of higher rather than lower concentrations and some choosing to do the opposite. In the wild this could lead to resource partitioning among individuals. Such resource partitioning may help reduce competition and aggression within small foraging flocks and has potential implications for pollination dynamics: sunbirds would more fully exploit flowers containing nectar of different concentrations than if all individuals rejected lower concentrations.

In conclusion, white-bellied sunbirds do not use taste to decide how much to consume per meal when concentration is highly variable and unpredictable, even though theory suggests that using taste to decide how much to consume should increase their feeding efficiency in these conditions. Further work will be necessary to establish just what the costs and benefits associated with choosing to follow a fixed feeding routine may be, while disregarding cues such as taste that could be used to improve feeding efficiency.
Acknowledgements:

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References


Data accessibility
Supplementary Material 1.
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