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Suboptimal nutrient balancing despite dietary choice in glucose-averse German cockroaches, *Blattella germanica*

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ABSTRACT

Insects have evolved fine-tuned gustatory and post-ingestive physiological mechanisms that enable them to self-select an optimal composition of macronutrients. Their ability to forage optimally among multiple food sources and maximize fitness parameters depends on their ability not only to taste and perceive the nutritional value of potential foods but also to avoid deleterious components; the strength of such avoidance should reflect the severity of the perceived hazard. In German cockroaches (*Blattella germanica*), glucose aversion has evolved in some populations in response to anthropogenic selection with glucose-containing insecticidal baits. In four feeding treatments, we gave newly eclosed glucose-averse female cockroaches free choice to feed from two artificial, nutritionally complementary foods varying in protein and carbohydrate composition, with glucose or fructose as the sole carbohydrate source in either food. After 6 days of feeding, we measured diet consumption and the length of basal oocytes as an estimate of sexual maturation. The females did not compromise on their aversion to glucose in order to balance their protein and carbohydrate intake, and experienced lower sexual maturation rates as a consequence. Nutrient specific hunger via feedback mechanisms, and adjustments to gustatory sensitivity thus do not override the deterrence of glucose, likely due to strong selection against ingesting even small amounts of toxin associated with glucose in baits. In the absence of baits, glucose aversion would be expected to incur a fitness cost compared to wild-type individuals due to lower overall food availability but also to larger difficulty in attaining a nutritionally balanced diet.

1. Introduction

A wide range of animals, spanning from herbivores to omnivores and predators, have been shown to forage optimally for specific nutrients when allowed to compose a balanced diet from nutritionally complementary food sources (Behmer, 2009; Fanson et al., 2009; Lee et al., 2008; Jensen et al., 2012, 2013; Raubenheimer and Jones, 2006; Simpson et al., 2004, 2015). In particular, omnivores that scavenge in nutritionally heterogeneous environments are faced with the challenge of composing an overall balanced diet from food sources that may vary widely in nutritional composition, quality and availability. In order to do so, animals have evolved highly sensitive chemosensory and internal feedback mechanisms that enable them to sense what specific nutrients they need, and to be stimulated to consume them (Simpson and Raubenheimer, 1993, 1996; Simpson et al., 1991). However, little is known about how the evolution of optimal nutrient balancing mechanisms interacts with mechanisms of deterrence and toxin avoidance, which function to prevent the ingestion of lethal or deleterious compounds.

The effects of food deterrents on nutrient balancing are relatively well studied in herbivorous insects (Behmer et al., 2002; Bernays and Raubenheimer, 1991; Hägele and Rowell-Rahier, 1999; Raubenheimer, 1992; Simpson and Raubenheimer, 2001; Singer et al., 2002), but nutrient balancing and performance in relation to potentially lethal toxins has only been investigated very recently (Archer et al., 2014; Schmehl et al., 2014; Shik et al., 2014). Whereas deterrents typically stop the forager from eating before detrimental toxin doses are ingested (Chapman, 2013; Ozaki et al., 2003), toxins that are coupled with phagostimulating nutrients to disguise their taste and facilitate consumption would require highly sensitive gustatory reception capabilities.

The German cockroach (*Blattella germanica* Linnaeus) is an extreme generalist omnivore and a widespread pest in human establishments, where it is apparently able to compose a nutritionally balanced overall diet from various food objects within a household (Jones and Raubenheimer, 2001; Schal, 2011; Schal et al., 1984). German cockroach nymphs have been shown to grow slower when restricted to nutritionally deficient or imbalanced

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diets, and to have a high capacity to rebalance overall nutrient intake and resume growth trajectories when a complementary food appears (Raubenheimer and Jones, 2006). One of the most successful measures of cockroach population control is the use of toxic baits, in which toxins are coupled with phagostimulants to facilitate the ingestion of lethal toxin doses (Appel, 1990; Schal and Hamilton, 1990). However, selection with baits has led to evolution of physiological resistance to the toxins used in baits (Gondhalekar and Scharf, 2012; Schal, 1992; Wang et al., 2004, 2006), as well as behavioral resistance to bait components (Ross, 1997; Strong et al., 1993; Wang et al., 2004, 2006).

An intriguing adaptation that has evolved rapidly in multiple German cockroach populations in response to insecticidal baits containing glucose as phagostimulant is a strong aversion to ingesting glucose (Silverman and Bieman, 1993; Silverman and Ross, 1994). This is in particular interesting because glucose is a ubiquitous and metabolically important nutrient in nature. The adaptation appears to be controlled by a mutation in a single major gene (Silverman and Bieman, 1993), and was recently discovered to be due to a change in the response of taste neurons in the chemosensory appendages (Wada-Katsumata et al., 2011): glucose, which normally stimulates sweet gustatory receptor neurons, in addition strongly stimulates bitter receptor neurons in glucose-averse cockroaches (Wada-Katsumata et al., 2013). In contrast, fructose and other sugars are readily ingested (Silverman and Bieman, 1993; Wada-Katsumata et al., 2011, 2013). If no other food is available over a longer term, however, glucose-averse cockroaches will ingest small amounts of glucose-containing food (Shik et al., 2014; Silverman, 1995; Silverman and Selbach, 1998).

Whereas glucose aversion is highly advantageous in the presence of glucose-containing baits because it prevents intoxication, this adaptation would be expected to be maladaptive in the absence of toxic bait, in particular if glucose-containing foods are a major energy source in the foraging environment. Even in an environment containing glucose-free foods, glucose aversion might be deleterious because individuals might reject nutritionally complementary foods containing glucose which would otherwise enable them to reach their intake target. When given access to nutritionally complementary diets with either glucose or fructose as the sole carbohydrate source in both foods, wild-type German cockroaches reached their protein vs. carbohydrate intake target and maximized oocyte development independently of sugar type (Jensen et al., 2015). In contrast, glucose-averse cockroaches only maximized oocyte development when given nutritionally complementary fructose-containing foods, while consumption and oocyte development were highly suppressed if the foods contained glucose (Jensen et al., 2015). If the presence of glucose in nutritionally complementary foods prevents consumption, glucose-averse individuals foraging in the built environment might thus not be able to reach their nutritional intake target where performance is maximized even though complementary food is available (Jensen et al., 2015; Raubenheimer and Jones, 2006; Simpson et al., 2015). In the situation where glucose-averse individuals have to ingest a glucose-containing food to eat a balanced diet, feedback mechanisms of nutrient specific hunger would have to override the bitter signals from the chemosensory neurons (Simpson and Raubenheimer, 1996; Simpson et al., 1991; Wada-Katsumata et al., 2013), or the cockroaches would have to forage on a diet that is nutritionally imbalanced with the associated costs to performance (Jensen et al., 2015; Raubenheimer and Jones, 2006).

We applied the *Geometric Framework for Nutrition* (Simpson and Raubenheimer, 2012) to analyze the nutrient balancing behavior of glucose-averse female German cockroaches during sexual maturation, and linked specific nutrient intake to the rate of oocyte development. We produced four nutritionally specific, semi-synthetic foods varying in protein-to-carbohydrate ratio with either glucose or fructose as the sole carbohydrate component. We then gave newly eclosed glucose-averse female German cockroaches free choice to compose their diet from single combinations of two nutritionally complementary foods. After 6 days of feeding, we measured cumulative consumption and specific nutrient intake and measured oocyte length as an estimate of sexual maturation. Our results show that females did not balance their intake of protein and carbohydrate from the two foods if one of the foods contained glucose. Failing to ingest a balanced diet incurred a performance cost by slowing the rate of sexual maturation.

2. Materials and methods

2.1. Animals and housing

The glucose-averse German cockroaches used in the experiment originated from the T164 strain which was collected in Gainesville, Florida in 1989 (Silverman and Bieman, 1993), and have been maintained as a laboratory culture on ad libitum water and rat chow (Purina 5001 Rodent Diet, PMI Nutrition International, St. Louis, MO, USA) in transparent plastic containers (45.7 cm × 22.8 cm × 30.4 cm). Before the start of experiments, 400 nymphs were collected from the culture and distributed equally across four plastic containers (18.5 cm × 13.0 cm × 10.0 cm) with ad libitum water and rat chow until eclosion. Adults were collected daily, and newly emerged females were set up in the experiment within 24 h of eclosion. Experiments were performed in a room at 28–29 °C, 25–35% RH, and a 12:12 h L:D regime. Experimental cockroaches were held individually in transparent Petri dishes (100 mm diam × 16 mm), and food and water were provided ad libitum in separate inverted vial caps (foods: 15 mm × 10 mm; water: 25 mm × 12 mm).

2.2. Experimental diets and measuring consumption

We produced four artificial foods differing in protein-to-carbohydrate (P:C) ratio (2:1 or 1:4) and in carbohydrate component (either glucose or fructose), modified from Dussutour and Simpson (2008). Full ingredient lists and compositions of the foods are presented in Table 1. Upon eclosion, experimental animals ($N = 80$) were weighed to the nearest mg and distributed evenly across four dietary treatments, each consisting of free choice to feed on one of the 2:1 foods and one of the 1:4 foods. All foods were dried at 38 °C for 4 days and weighed to the nearest 10 µg before feeding. After 6 days of feeding, the foods were re-collected, re-dried, and re-weighed, and consumption was calculated as the difference in food dry mass before and after feeding. Protein and carbohydrate consumption were calculated respectively by multiplying the proportion of each nutrient group in the foods with the dry mass consumption of the corresponding

Table 1
Ingredient compositions of the four semi-synthetic foods used in the experiment. The numbers in brackets represent the quantity of protein in the respective ingredients. The recipes are modified from Dussutour and Simpson (2008).

P:C ratio	Glucose foods		Fructose foods	
	2:1	1:4	2:1	1:4
Whey protein concentrate (g)	19.3 (16.1)	2.5 (2.1)	19.3 (16.1)	2.5 (2.1)
Calcium caseinate (g)	17.7 (16.1)	2.3 (2.1)	17.7 (16.1)	2.3 (2.1)
Whole egg powder (g)	16.0 (7.8)	16.0 (7.8)	16.0 (7.8)	16.0 (7.8)
Glucose (g)	20.0	48.0	0.0	0.0
Fructose (g)	0.0	0.0	20.0	48.0
Agar (g)	4.0	4.0	4.0	4.0
Vanderzant vitamin mixture (g)	2.0	2.0	2.0	2.0
Methyl 4-hydroxobenzoate (g)	1.0	1.0	1.0	1.0

food, and combining the consumption of either nutrient group from both foods.

2.3. Measuring sexual maturation

To estimate the level of sexual maturation in response to the available combination of complementary foods, all females were dissected immediately after the six-day feeding period and the lengths of five basal oocytes were measured with an eyepiece micrometer in a dissecting microscope (Schal et al., 1993; Uzsák and Schal, 2013). Another group of females ($N = 20$) was dissected within 24 h of eclosion and again the lengths of five oocytes were measured. For all females, the average length of the five oocytes was used as the individual measure for analysis. Basal oocytes of the German cockroach mature synchronously, so there was little variation among the five oocytes within a female.

2.4. Statistical analyses

To ensure that the body mass of experimental females did not differ across treatments, the initial body mass was compared across treatments using analysis of variance (ANOVA). Differences in preference of the 2:1 food versus the 1:4 food were analyzed across dietary treatments using a multivariate ANOVA (MANOVA) test. To test whether one food was preferred over the other within each dietary treatment, consumption from each food in the food pair was compared with a paired *t*-test, and consumption from each food was compared between all dietary treatment groups using a Tukey HSD test. Differences in protein and carbohydrate intake across dietary treatments were analyzed using a MANOVA test followed by separate Tukey HSD tests for protein and carbohydrate intake, respectively. To test whether females provided with at least one glucose-free food regulated protein intake more tightly than carbohydrate intake, we compared the variation in protein versus carbohydrate intake across the three dietary treatments using a Levene's test. Under-ingestion of the nutrient that was deficient in the glucose-free food was compared between the two treatments given only one glucose-free food (either protein- or carbohydrate-rich) by subtracting the average intake of the deficient nutrient from the average intake of the corresponding nutrient by females in the glucose-free treatment. Conversely, over-ingestion of the abundant nutrient in the glucose-free food was compared between the two treatments given only one glucose-free food by subtracting the average intake of the corresponding nutrient by females in the glucose-free treatment from intake of the abundant nutrient by females given either glucose-free food. Under- and over-ingestion of either nutrient were then compared between the two groups given only one glucose-free food using respective *t*-tests. Differences in average oocyte lengths upon dissection were analyzed across all treatments using an ANOVA test, and individual treatments were compared using a Tukey HSD test. All statistical analyses were performed in JMP 7.0 (SAS Institute Inc., Cary, NC, USA).

3. Results

3.1. Animal body mass

Female wet body mass upon eclosion did not differ significantly across treatments (mean ± SE = 67.02 ± 0.53 mg; ANOVA: $F_{4,100} = 0.63$, $P = 0.65$), and no correction for body mass was therefore necessary in further analyses.

3.2. Food and nutrient consumption

The sugar type in the provided foods significantly affected dietary preference (MANOVA: $F_{3,80} = 227.55$, $P < 0.0001$; Fig. 1), which resulted in significantly different protein and carbohydrate intake across dietary treatments in the glucose-averse cockroaches (MANOVA: $F_{3,80} = 13.34$, $P < 0.0001$; Fig. 2). Consumption of glucose-containing foods was very low, and there was no statistical difference in consumption of any glucose-containing food across dietary treatments regardless of food P:C ratio and the sugar type in the complementary food (Fig. 1). Females given only glucose-containing foods therefore consumed very low amounts of both protein and carbohydrate (Fig. 2). In contrast, consumption of fructose-containing foods was highly affected by both the P:C ratio of the food and the sugar type of the complementary food (Fig. 1). In both dietary treatments consisting of complementary foods with different sugar types, consumption of the fructose-containing food was significantly higher than consumption of the same food in the dietary treatment where the females were allowed to select their specific nutrient intake from two fructose-containing foods (Fig. 1). In the dietary treatments consisting of complementary foods with different sugar types, the female cockroaches thus ingested a dietary nutrient composition very close to that of the fructose-containing food while they over-ingested the abundant nutrient (protein or carbohydrate) and under-ingested the deficient nutrient relative to the specific nutrient consumption of the females given complementary fructose-containing foods (Fig. 2). These females consumed more than twice as much of the carbohydrate-rich food than the protein-rich food, and the self-selected nutrient intake was therefore significantly different from what would have resulted if the females had fed at random from the two foods (Fig. 1). Protein intake varied significantly less than carbohydrate intake across dietary treatments with one or two fructose-containing foods (Levene's test: $F_{1,60} = 80.74$, $P < 0.0001$), indicating tighter regulation of protein intake than of carbohydrate intake across the treatments. This resulted in significantly larger under-ingestion of carbohydrate by females given fructose-containing protein-rich

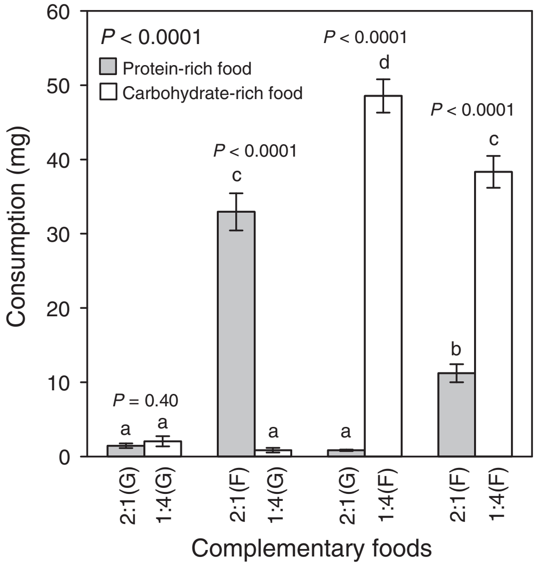

Fig. 1. Consumption (mean ± SE) by glucose-averse female cockroaches from each of the two nutritionally complementary foods within the four dietary choice combinations during foraging for 6 days upon eclosion ($N = 20$ female cockroaches per choice combination). Each choice combination consisted of one food with a protein:carbohydrate (P:C) ratio of 2:1 and one food with a P:C ratio of 1:4, with either glucose (G) or fructose (F) as the sole carbohydrate source in either food. The overall *P*-value is from a MANOVA test which tests for differences in consumption from the 2:1 and the 1:4 food across dietary choice combinations. The *P*-value within each choice combination is from a paired *t*-test testing for differences in consumption from each of the two provided foods. Different letters indicate significant differences (Tukey HSD test, $P < 0.05$).

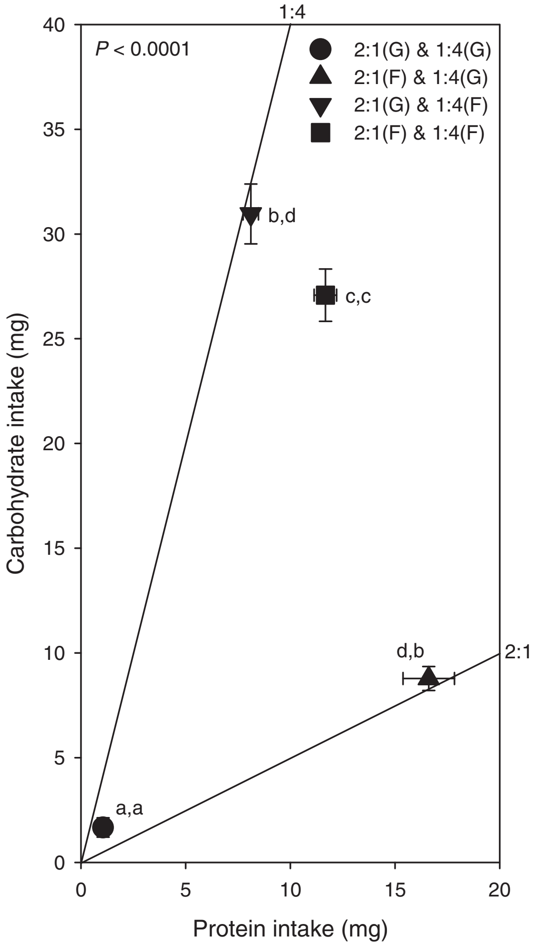

Fig. 2. Protein and carbohydrate consumption (mean ± SE) by glucose-averse female cockroaches within the four dietary choice combinations over the 6 days of foraging after eclosion ($N = 20$ female cockroaches per choice combination). Each choice combination consisted of one food with a protein:carbohydrate (P:C) ratio of 2:1 and one food with a P:C ratio of 1:4, with either glucose (G) or fructose (F) as the sole carbohydrate source in either food. The overall P-value is from a MANOVA test which tests for differences in protein and carbohydrate consumption across dietary choice combinations. Different letters indicate significant differences in protein and carbohydrate consumption, respectively, separated with a comma (Tukey HSD test, $P < 0.05$).

food and glucose-containing carbohydrate-rich food compared to the under-ingestion of protein by cockroaches given fructose-containing carbohydrate-rich food and glucose-containing protein-rich food (t-test: $t_{40} = 474.43$, $P < 0.0001$). However, there was no difference in the amount of over-ingested nutrient (protein or carbohydrate) from the fructose-containing food in the two dietary combinations with different sugar types (t-test: $t_{40} = 0.32$, $P = 0.58$), which indicates that the cockroaches had a similar capacity to over-ingest either nutrient relative to the self-selected intake point.

3.3. Sexual maturation

Females that were given complementary fructose-containing foods contained the largest oocytes by the end of the experiment, although these oocytes were not significantly larger than the oocytes in females given fructose in the carbohydrate-rich (1:4) food and glucose in the protein-rich (2:1) food (Fig. 3). The females given fructose in the protein-rich food and glucose in the carbohydrate-rich food developed significantly smaller oocytes than females given complementary fructose-containing foods, and these oocytes were similarly not significantly different from the intermediate-sized oocytes in the females given fructose in the carbohydrate-rich food and glucose in the protein-rich food (Fig. 3). Females given glucose in both foods failed to mature their oocytes relative to the oocyte size of newly eclosed females (Fig. 3).

4. Discussion

Optimal foraging for specific nutrients has been shown in a diverse array of animals (Behmer, 2009; Fanson et al., 2009; Lee et al., 2008; Jensen et al., 2012, 2013; Raubenheimer and Jones, 2006; Simpson et al., 2004, 2015). However, omnivorous animals foraging opportunistically on a variety of resources must be able to distinguish between nutritious and toxic foods (Scott, 2005), and little is known about how evolution of gustatory perception compromises nutrient balancing with the risk of ingesting deleterious elements and the associated costs. Cockroaches are a particularly interesting group of animals for addressing these questions because they possess unique adaptations for nitrogen storage and recycling in addition to efficient lipid storage (Mullins, 2015). They might therefore be more robust than most other animals as they are able to buffer against the deleterious effects of nutritional imbalance and to quickly recover from nutrient deficiency (Raubenheimer and Jones, 2006). In German cockroach populations, glucose aversion has evolved in response to toxic, glucose-containing baits (Silverman and Bieman, 1993; Silverman and Ross, 1994). We found that glucose-averse female cockroaches did not compromise on their aversion to glucose in order to balance their intake of protein and carbohydrate and thus maximize oocyte development. Instead, these females restricted consumption almost exclusively to the glucose-free, nutritionally imbalanced food and balanced the over- and under-ingestion of the excessive and deficient nutrient in this food (Raubenheimer and Simpson, 1997), at the cost of slower growth of their oocytes (Fig. 3) and therefore delayed reproduction.

In a previous study where migratory locusts (*Locusta migratoria* Linnaeus) had to balance protein and carbohydrate intake from

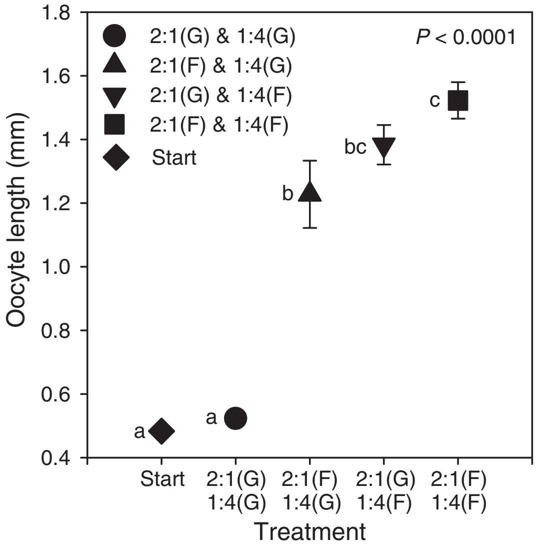

Fig. 3. Average oocyte length (mean ± SE) in the glucose-averse female cockroaches within 24 h after eclosion or after 6 days of feeding on either of the four choice combinations ($N = 20$ female cockroaches per treatment). Each choice combination consisted of one food with a protein:carbohydrate (P:C) ratio of 2:1 and one food with a P:C ratio of 1:4, with either glucose (G) or fructose (F) as the sole carbohydrate source in either food. The overall P-value is from an ANOVA test on differences in average oocyte length across treatments. Different letters indicate significant differences (Tukey HSD test, $P < 0.05$).

complementary foods where tannic acid was present or absent in either, tannic acid affected specific nutrient consumption by compromising nutrient balancing with the intake of tannic acid (Behmer et al., 2002). In contrast, the cockroaches in our experiment did not compromise nutrient balancing with glucose intake and almost exclusively rejected glucose-containing food from their diet despite the cost to sexual maturation. The interplay between nutrient-specific post-ingestive feedback mechanisms and taste stimuli from taste receptors (Simpson and Raubenheimer, 1996) is thus shortcut in these cockroaches when glucose is involved. The much stronger deterrence reaction by glucose-averse cockroaches to glucose than by locusts to tannic acid probably reflects stronger selection against the ingestion of glucose because it is coupled with potent toxins that kill at low intake doses, whereas tannic acid is much less toxic (Behmer et al., 2002; Raubenheimer, 1992; Simpson and Raubenheimer, 2001). Furthermore, the potential of cockroaches to buffer against imbalances due to their specific nutrient storage adaptations and to subsequently rebalance nutrients and quickly recover from imbalances (Mullins, 2015; Raubenheimer and Jones, 2006) would theoretically make cockroaches more nutritionally flexible than other animals and put less of an immediate pressure on compromising taste aversions to deterrents. Another difference between the present study and the study by Behmer et al. (2002) is that even though low amounts of deterrent (glucose) were ingested by the cockroaches in our study, no toxin was ingested; glucose injection into the hemocoel is not toxic to glucose-averse cockroaches (Wada-Katsumata et al., 2011). Thus, performance differences in our experiment were entirely due to differences in nutrient intake with no additional effect of a toxin.

When cockroaches limited their feeding to the protein-rich, fructose containing food, their hunger for non-protein energy should increase, and so their perception of sugar sweetness, including glucose, should increase (Simpson and Raubenheimer, 1996). In fruit flies (*Drosophila melanogaster* Meigen), sugar sensitivity is found to increasingly override bitter sensitivity during starvation (Inagaki et al., 2014). When energetically deprived or when the hemolymph contains high amounts of amino acids relative to sugar, the signaling balance between sweet and bitter sensing neurons could be expected to change towards stronger signals from the sweet sensing neurons. Indeed, *D. melanogaster* fruit flies can sense the caloric value of foods after ingestion by physiological feedback mechanisms independently of the sweetness of the sugar in the food, and are after a short period of phenotypic adaptation able to select the most energetically valuable foods due to modifications in the neuronal mechanisms of taste perception (Fujita and Tanimura, 2011; Stafford et al., 2012). However, these mechanisms seem to play a very limited role with regards to consumption of glucose-containing foods in glucose-averse German cockroaches. Instead, the neuronal input from sweet sensing neurons in glucose-averse German cockroaches is consistently overruled by the signals from bitter sensing neurons when given glucose-containing foods (Wada-Katsumata et al., 2013), probably due to strong selection for a hard-wired aversive response to sweet, glucose-containing baits with well-disguised but highly potent toxins (Silverman and Ross, 1994).

Although the consumption of glucose-containing food was very limited in the present study, glucose-averse cockroaches that are restricted to glucose over a long time frame will start eating at some point, indicating that there is a stage where the requirement for energy overrides deterrence (Silverman and Selbach, 1998). When restricted to glucose-containing foods, glucose-averse females will furthermore feed to an extent that enables full ootheca development and offspring production, although at a much lower intake rate and twice the time from mating to offspring hatch (Silverman, 1995). Whether glucose-averse cockroaches will start ingesting nutritionally complementary glucose-containing foods if nutritional imbalance occurs over a prolonged period, or if the available glucose-free foods were even more nutritionally imbalanced than in the present study, remains to be investigated.

The aversion to glucose restrained the glucose-averse females to single, nutritionally imbalanced foods, and the females instead balanced the over- and under-ingestion of protein and carbohydrate intake from this food. The finding that the female cockroaches regulated protein intake significantly tighter than carbohydrate intake is consistent with earlier findings in German cockroaches and other omnivores (Jensen et al., 2013, 2015; Raubenheimer and Jones, 2006; Sørensen et al., 2008), and in particular cockroaches appear to have low tolerance for excessive protein intake because of efficient nitrogen retention and limited capacity to excrete nitrogen excesses (Cochran, 1985; Mullins, 2015). However, the cockroaches did not over-ingest fructose to a larger extent than protein relative to the intake target, suggesting limited capacity to over-ingest both protein and carbohydrate, although large over-consumption of carbohydrate relative to the intake target has been recorded in wild-type nymphs (Raubenheimer and Jones, 2006). This limited over-ingestion of fructose might be caused by a lower tendency of glucose-averse cockroaches to ingest high amounts of sugars, perhaps in particular monosaccharides, compared to wild-type cockroaches (Jensen et al., 2015), possibly due to lower phagostimulatory power of sugars in general (Shik et al., 2014).

In a previous study, consumption by glucose-averse female cockroaches restricted to single foods was low compared to consumption when given two complementary fructose-containing, glucose-free foods (Jensen et al., 2015). In the present study, however, cockroaches that restricted consumption to a single fructose-containing food did not have much lower consumption than cockroaches given complementary fructose-containing foods. In addition to providing a dietary choice in all treatments and running the experiment at a higher temperature, we also used different compositions of experimental foods including whole egg powder as a base ingredient in the present experiment (Table 1). Whereas the diets used by Jensen et al. (2015) were almost lipid-free, whole egg powder contains substantial amounts of lipids (Dussutour and Simpson, 2008). Similar to the present study, sexually maturing glucose-averse females provided with lipid-containing rat chow (5%, Shik et al., 2014) did not consume significantly less food than wild-type females (Jensen et al., 2015), and glucose-averse nymphs consumed similar amounts of food as wild-type nymphs when given whole egg powder based diets (Shik et al., 2014). While consumption is found to be lower in glucose-averse than in wild-type cockroaches not only when given glucose- but also fructose-containing foods (Jensen et al., 2015), the presence of lipids in the food might counteract this effect. In *D. melanogaster*, fatty acids were shown to serve as an appetitive cue already at low to medium concentrations, detected exclusively through the same subset of neurons that sense sweet substances including most sugars, and induced a robust feeding response (Masek and Keene, 2013). Whether the presence of lipids is more important to stimulate consumption in glucose-averse cockroaches as a response to lower phagostimulatory power of sugars compared to wild-type cockroaches remains to be investigated.

In conclusion, our study shows that glucose aversion prevents glucose-averse German cockroaches from consuming a balanced diet if nutritionally complementary foods contain glucose, and that performance, here measured as sexual maturation, is significantly affected. Glucose-averse cockroaches would thus be expected to have lower fitness than wild-type cockroaches under natural conditions, not only because less food is available overall, but also because glucose aversion is more likely to prevent the ingestion of a balanced diet from the number of food sources that are available. Maintaining the glucose-averse trait might therefore keep wild populations at a significantly lower growth potential than under conditions where wild-type individuals can persist due to lower food availability and lower chances to obtain a nutritionally balanced diet.

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References

Appel, A.G., 1990. Laboratory and field performance of consumer bait products for German cockroach (Dictyoptera: Blattellidae) control. J. Econ. Entomol. 83, 153–159.

Archer, C.R., Pirk, C.W.W., Wright, G.A., Nicolson, S.W., 2014. Nutrition affects survival in African honeybees exposed to interacting stressors. Funct. Ecol. 28, 913–923.

Behmer, S.T., 2009. Insect herbivore nutrient regulation. Annu. Rev. Entomol. 54, 165–187.

Behmer, S.T., Simpson, S.J., Raubenheimer, D., 2002. Herbivore foraging in chemically heterogeneous environments: nutrients and secondary metabolites. Ecology 83, 2489–2501.

Bernays, E.A., Raubenheimer, D., 1991. Dietary mixing in grasshoppers: changes in acceptability of different plant secondary compounds associated with low levels of dietary protein (Orthoptera: Acrididae). J. Insect Behav. 4, 545–556.

Chapman, R.F., 2013. The Insects: Structure and Function. Cambridge University Press, New York.

Cochran, D.G., 1985. Nitrogen excretion in cockroaches. Annu. Rev. Entomol. 30, 29–49.

Dussutour, A., Simpson, S.J., 2008. Description of a simple synthetic diet for studying nutritional responses in ants. Insect. Soc. 55, 329–333.

Fanson, B.G., Weldon, C.W., Peréz-Staples, D., Simpson, S.J., Taylor, P.W., 2009. Nutrients, not caloric restriction, extend lifespan in Queensland fruit flies (*Bactrocera tryoni*). Aging Cell 8, 514–523.

Fujita, M., Tanimura, T., 2011. *Drosophila* evaluates and learns the nutritional value of sugars. Curr. Biol. 21, 751–755.

Gondhalekar, A.D., Scharf, M.E., 2012. Mechanisms underlying fipronil resistance in a multiresistant field strain of the German cockroach (Blattodea: Blattellidae). J. Med. Entomol. 49, 122–131.

Hägele, B.F., Rowell-Rahier, M., 1999. Dietary mixing in three generalist herbivores: nutrient complementation or toxin dilution? Oecologia 119, 521–533.

Inagaki, H.K., Panse, K.M., Anderson, D.J., 2014. Independent, reciprocal neuromodulatory control of sweet and bitter taste sensitivity during starvation in *Drosophila*. Neuron 84, 806–820.

Jensen, K., Engelke, S., Simpson, S.J., Mayntz, D., Hunt, J., 2013. Balancing of specific nutrients and subsequent growth and body composition in the slug *Arion lusitanicus*. Physiol. Behav. 122, 84–92.

Jensen, K., Mayntz, D., Toft, S., Clissold, F.J., Hunt, J., Raubenheimer, D., Simpson, S.J., 2012. Optimal foraging for specific nutrients in predatory beetles. Proc. R. Soc. Lond. B Biol. Sci. 279, 2212–2218.

Jensen, K., Schal, C., Silverman, J., 2015. Adaptive contraction of diet breadth affects sexual maturation and specific nutrient consumption in an extreme generalist omnivore. J. Evol. Biol. 28, 906–916.

Jones, S.A., Raubenheimer, D., 2001. Nutritional regulation in nymphs of the German cockroach, *Blattella germanica*. J. Insect Physiol. 47, 1169–1180.

Lee, K.P., Simpson, S.J., Clissold, F.J., Brooks, R., Ballard, J.W.O., Taylor, P.W., Soran, N., Raubenheimer, D., 2008. Lifespan and reproduction in *Drosophila*: new insights from nutritional geometry. Proc. Natl. Acad. Sci. U.S.A. 105, 2498–2503.

Masek, P., Keene, A.C., 2013. *Drosophila* fatty acid taste signals through the PLC pathway in sugar-sensing neurons. PLoS Genet. 9, e1003710.

Mullins, D.E., 2015. Physiology of environmental adaptations and resource acquisition in cockroaches. Annu. Rev. Entomol. 60, 473–492.

Ozaki, M., Takahara, T., Kawahara, Y., Wada-Katsumata, A., Seno, K., Amakawa, T., Yamaoka, R., Nakamura, T., 2003. Perception of noxious compounds by contact chemoreceptors of the blowfly, *Phormia regina*: putative role of an odorant-binding protein. Chem. Senses 28, 349–359.

Raubenheimer, D., 1992. Tannic acid, protein, and digestible carbohydrate: dietary imbalance and nutritional compensation in locusts. Ecology 73, 1012–1027.

Raubenheimer, D., Jones, S.A., 2006. Nutritional imbalance in an extreme generalist omnivore: tolerance and recovery through complementary food selection. Anim. Behav. 71, 1253–1262.

Raubenheimer, D., Simpson, S.J., 1997. Integrative models of nutrient balancing: application to insects and vertebrates. Nutr. Res. Rev. 10, 151–179.

Ross, M.H., 1997. Evolution of behavioral resistance in German cockroaches (Dictyoptera: Blattellidae) selected with a toxic bait. J. Econ. Entomol. 90, 1482–1485.

Schal, C., 1992. Sulfluramid resistance and vapor toxicity in field-collected German cockroaches (Dictyoptera: Blattellidae). J. Med. Entomol. 29, 207–215.

Schal, C., 2011. Cockroaches. In: Hedges, S., Moreland, D. (Eds.), Handbook of Pest Control, tenth ed. GIE Media, pp. 150–291.

Schal, C., Chiang, A.-S., Burns, E.L., Gadot, M., Cooper, R.A., 1993. Role of the brain in juvenile hormone synthesis and oöcyte development: effects of dietary protein in the cockroach *Blattella germanica* (L.). J. Insect Physiol. 39, 303–313.

Schal, C., Gautier, A.-S., Bell, W.J., 1984. Behavioural ecology of cockroaches. Biol. Rev. 59, 209–254.

Schal, C., Hamilton, R.L., 1990. Integrated suppression of synanthropic cockroaches. Annu. Rev. Entomol. 35, 521–551.

Schmehl, D.R., Teal, P.E.A., Frazier, J.L., Grozinger, C.M., 2014. Genomic analysis of the interaction between pesticide exposure and nutrition in honey bees (*Apis mellifera*). J. Insect Physiol. 71, 177–190.

Scott, K., 2005. Taste recognition: food for thought. Neuron 48, 455–464.

Shik, J.Z., Schal, C., Silverman, J., 2014. Diet specialization in an extreme omnivore: nutritional regulation in glucose-averse German cockroaches. J. Evol. Biol. 27, 2096–2105.

Silverman, J., 1995. Effects of glucose-supplemented diets on food intake, nymphal development, and fecundity of glucose-averse, non-glucose-averse, and heterozygous strains of the German cockroach, *Blattella germanica*. Entomol. Exp. Appl. 76, 7–14.

Silverman, J., Bieman, D.N., 1993. Glucose aversion in the German cockroach, *Blattella germanica*. J. Insect Physiol. 39, 925–933.

Silverman, J., Ross, M.H., 1994. Behavioral resistance of field-collected German cockroaches (Blattodea: Blattellidae) to baits containing glucose. Environ. Entomol. 23, 425–430.

Silverman, J., Selbach, H., 1998. Feeding behavior and survival of glucose-averse *Blattella germanica* (Orthoptera: Blattoidea: Blattellidae) provided glucose as a sole food source. J. Insect Behav. 11, 93–102.

Simpson, S.J., Clissold, F.J., Lihoreau, M., Ponton, F., Wilder, S.M., Raubenheimer, D., 2015. Recent advances in the integrative nutrition of arthropods. Annu. Rev. Entomol. 60, 293–311.

Simpson, S.J., James, S., Simmonds, M.S.J., Blaney, W.M., 1991. Variation in chemosensitivity and the control of dietary selection behaviour in the locust. Appetite 17, 141–154.

Simpson, S.J., Raubenheimer, D., 1993. The central role of the haemolymph in the regulation of nutrient intake in insects. Physiol. Entomol. 18, 395–403.

Simpson, S.J., Raubenheimer, D., 1996. Feeding behaviour, sensory physiology and nutrient feedback: a unifying model. Entomol. Exp. Appl. 80, 55–64.

Simpson, S.J., Raubenheimer, D., 2001. The geometric analysis of nutrient–allelochemical interactions: a case study using locusts. Ecology 82, 422–439.

Simpson, S.J., Raubenheimer, D., 2012. The Nature of Nutrition: A Unifying Framework from Animal Adaptation to Human Obesity. Princeton University Press, Princeton, NJ.

Simpson, S.J., Sibly, R.M., Lee, K.P., Behmer, S.T., Raubenheimer, D., 2004. Optimal foraging when regulating intake of multiple nutrients. Anim. Behav. 68, 1299–1311.

Singer, M.S., Bernays, E.A., Carriere, Y., 2002. The interplay between nutrient balancing and toxin dilution in foraging by a generalist insect herbivore. Anim. Behav. 64, 629–643.

Sørensen, A., Mayntz, D., Raubenheimer, D., Simpson, S.J., 2008. Protein-leverage in mice: the geometry of macronutrient balancing and consequences for fat deposition. Obesity 16, 566–571.

Stafford, J.W., Lynd, K.M., Jung, A.Y., Gordon, M.D., 2012. Integration of taste and calorie sensing in *Drosophila*. J. Neurosci. 32, 14767–14774.

Strong, C.A., Koehler, P.G., Patterson, R.S., 1993. Oral toxicity and repellency of borates to German cockroaches (Dictyoptera: Blattellidae). J. Econ. Entomol. 86, 1458–1463.

Uzsák, A., Schal, C., 2013. Social interaction facilitates reproduction in male German cockroaches, *Blattella germanica*. Anim. Behav. 85, 1501–1509.

Wada-Katsumata, A., Silverman, J., Schal, C., 2011. Differential inputs from chemosensory appendages mediate feeding responses to glucose in wild-type and glucose-averse German cockroaches, *Blattella germanica*. Chem. Senses 36, 589–600.

Wada-Katsumata, A., Silverman, J., Schal, C., 2013. Changes in taste neurons support the emergence of an adaptive behavior in cockroaches. Science 340, 972–975.

Wang, C., Scharf, M.E., Bennett, G.W., 2004. Behavioral and physiological resistance of the German cockroach to gel baits (Blattodea: Blattellidae). J. Econ. Entomol. 97, 2067–2072.

Wang, C., Scharf, M.E., Bennett, G.W., 2006. Genetic basis for resistance to gel baits, fipronil, and sugar-based attractants in German cockroaches (Dictyoptera: Blattellidae). J. Econ. Entomol. 99, 1761–1767.