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FEEDING, ENERGY PROCESSING RATES AND EGG PRODUCTION IN PAINTED LADY BUTTERFLIES

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Summary

Volume and energy ingestion rates, meal sizes (intakes to satiation) and meal frequencies were measured for previously unfed adult painted lady butterflies (*Vanessa cardui* L.) fed sucrose solutions or nectar from *Lantana camera* flowers in the laboratory. Volume and energy rates of crop emptying, assimilation efficiencies and mature egg production over 1 week were measured for *V. cardui* fed on sucrose solutions to assess mechanisms for and consequences of maximizing net meal energy. Viscosity reduced volume ingestion rates as sugar concentration increased, and 35–52.5 % (w/v) sucrose produced a maximum rate of energy gain from sucrose solutions. Ingestion rates were lower from *Lantana* flowers. Increasing *Lantana* nectar concentration from 33 to 70 % sucrose would produce about the same rate of energy gain for a meal. Virtually all ingested sugars were assimilated. Energy processing rates of 30 μl meals did not vary with sex, varied little with concentration and were 12–30 times the rate of energy use for maintenance. For females this may be due to the linear dependence of mature egg production on the amount of sugar ingested. Average meal timing compensated for variations in food concentration. Meals may be initiated before complete crop emptying, and this would increase the overall rates of energy processing, particularly for small meals. If *Vanessa* are not time-constrained while foraging, selecting concentrated nectars would decrease foraging frequency and increase the number of mature eggs produced after a meal.

Introduction

Some nectar feeders are exceptions to the optimality rule of maximizing long-term rate of net energy gain while foraging (e.g. Stephens and Krebs, 1986). Given a choice, they prefer concentrated sugars consumed at low energy gain rates (Dethier and Rhoades, 1954; Hainsworth and Wolf, 1976; Montgomerie *et al.* 1984; Hainsworth, 1989), but the choice of more concentrated sugar solution yields greater net gains for the amount consumed (Hainsworth, 1989, 1990). It has been suggested that they select foods to maximize net meal energy, or ($cM-fh$), where c is food energy concentration, M is meal volume, f is rate of energy expenditure

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while foraging (including searching through to ingestion) and h is foraging time for a meal (Hainsworth, 1990). Although nutrient requirements can modify choices with respect to net energy gains (Belovsky, 1978), nectar is the primary energy source for most nectarivores.

In some cases, maximizing rate of net energy gain for a meal would also maximize net meal energy. For example, if c, M and f are not variable, maximizing $(cM-fh)h^{-1}$ would maximize $(cM-fh)$ by minimizing fh. This suggests that maximizing net meal energy could be a general criterion for many animals, foods and situations, but how it is achieved may vary depending on foods and situations.

It thus is important to study meals, particularly for nectar feeders. Also, dynamic theories of foraging emphasize integrating short-term behaviour with variable long-term consequences (Mangel and Clark, 1988). A study of how meal energy is used could expand the time scale for feeding to include a variety of dynamics and allow us to assess how and to what extent short-term foraging mechanisms affect fitness.

A way to expand the time scale is to measure meal timing relative to rates of energy use. Based on meal net energy gains and rates of energy use:

$$T = \frac{[(cM - fh) - a]}{m + s}, \tag{1}$$

where T is time between meals, c, M, f and h are defined above, a is digestive energy loss, m is rate of energy use for short-term maintenance and s is rate of energy use for long-term storage, growth or reproduction (Hainsworth, 1990).

Two methods have been used to measure T. One method measures inter-meal intervals relative to net energy consumed (LeMagnen and Devos, 1970; Wolf and Hainsworth, 1977; Hainsworth, 1980; Hainsworth *et al.* 1981; Simpson, 1983; Simpson *et al.* 1989). Long-term rates of energy storage are estimated from net gains over the time to a subsequent meal. The other method measures processing of consumed energy (Gelperin, 1966; Hainsworth, 1974; McHugh and Moran, 1979; Jobling, 1986; Karasov *et al.* 1986; McCann and Stricker, 1986; Simpson *et al.* 1989; Hainsworth *et al.* 1990). Rates of crop or stomach net energy emptying are compared to rates of energy use for maintenance, and excess rates give a measure of long-term energy storage rates. The two methods are related when stomach or crop emptying is controlled relative to rates of energy use and meal initiation is related to emptying (McHugh and Moran, 1979; Hunt, 1980; Simpson and Ludlow, 1986; Simpson *et al.* 1989).

Both approaches should account for long-term energy storage rates (s in equation 1). Measurements of this are limited to a few species and indicate that the assumption of energy additivity across foods of different qualities (Stephens and Krebs, 1986) is not always correct. Long-term energy storage rates increase with food concentration for blowflies (Hainsworth *et al.* 1990) and hummingbirds (Hainsworth, 1990), so energy-rich foods have a potential use beyond that reflected by additive ranking based on net energy gains per unit of foraging time.

It would be desirable to relate s to fitness (Clark, 1989). For many animals this is

difficult because complex nutritional factors influence fitness and food choice for energy, or because consequences must be measured over long periods, or both. The experiments reported here concern sugar feeding and energy processing by adult painted lady butterflies (*Vanessa cardui* L.). Long-term consequences are measured as egg production, which depends simply on sugar intake over a brief period (days).

Materials and methods

Animals

Test animals were raised in the laboratory from animals obtained from a commercial source. Adults were kept 7–9 to a cage (0.03 m^3) containing sources of 30 % sucrose (w/v). Hollyhock plants were placed in the breeding cage 4–5 days after butterfly emergence until eggs were laid. Adults used in experiments were placed in 3.75 l individual cages with access to water *via* a cotton wick. Day of emergence was designated day 1. Photoperiod was 15 h light:9 h dark, and ambient temperature was 22±2 °C.

Meal sizes, intake rates and egg production

A meal is usually considered to be the amount eaten when feeding ceases, even though food is still available (LeMagnen and Devos, 1970; Dethier, 1976; Wolf and Hainsworth, 1977; Simpson and Ludlow, 1986). Unfed day 2 butterflies were weighed and held by their wings with their feet in either a 35 % (6 females, 9 males) or 70 % (33 females, 31 males) sugar solution. Feeding was allowed to continue until the proboscis was retracted for 1 min despite continued tarsal stimulation. When satiated, butterflies were reweighed, and the volume ingested was calculated from weight gain divided by fluid specific gravity. The duration of proboscis contact with the sugar solution was measured with a stopwatch to calculate the volume and energy rates of intake. Measurements obtained using the same method with 80 % sucrose (Hainsworth, 1989) and with 17.5 and 52.5 % sucrose were included in the analysis of ingestion rates.

To measure meal sizes under more natural conditions, 10 unfed butterflies were allowed to feed from 10 *Lantana camera* (L.) inflorescences. Each inflorescence contained 25–45 flowers with a modal nectar volume of 0.4 μl per flower [mean=0.5±0.3 μl (S.D.), N=155] and a modal nectar concentration of 33 % sucrose (mean=37.4±9.6 % sucrose, N=35) measured with a temperature-compensated refractometer (Hainsworth, 1974). Inflorescence stems were placed through holes in the floor of a plastic chamber (32 cm×16 cm×12 cm high) in three rows of three, four and three, spaced 8 cm apart within rows and staggered between rows so that nearest neighbor centers were 5.6 cm apart. The chamber was elevated 6 cm and placed in an aquarium containing 3 cm of water so that the stems extended into the water. A butterfly was weighed and allowed to feed until the proboscis was coiled and 10 min had passed with no further feeding. This criterion was used because none of six butterflies videotaped to measure intervals

to the next meal (see below) probed flowers in the interval between 10 and 64 min following proboscis coiling. Also, by 10 min all butterflies had assumed a characteristic non-feeding posture with wings clasped above them: this contrasts with their feeding posture with the wings held down. Butterflies were reweighed and intake was calculated from modal nectar specific gravity. Day 5 butterflies were used to ensure that they fed.

Foraging was videotaped for six butterflies to measure flower probing times with a stopwatch from proboscis insertion to withdrawal. It was assumed that the short probe times for flowers revisited three or more times represented time to probe a flower with no substantial nectar intake. The average of these times was subtracted from the time to probe flowers on the first and second visits to calculate ingestion time. Volume ingested was divided by total ingestion time to calculate nectar volume rate of ingestion.

Mature eggs were counted on day 8 to relate sugar consumption to egg production. Butterflies that had been fed a varying amount of sucrose in 1–6 meals over 1–3 days were returned to their cages, and three groups of 4–7 butterflies were not fed but were placed in individual breeding cages with access to water. The butterflies were frozen on day 8 and weighed. The abdomen was opened and the ovaries were removed. Mature eggs were easily distinguished by their size and by 12–14 longitudinal ridges on the outer membrane.

Meal frequencies

Six day 5 butterflies (three females, three males) that consumed a meal of *Lantana* nectar were videotaped at 30 s intervals following feeding to measure times to the next meal to the nearest minute. A similar method was used to examine the effect of sugar concentration on time to the next meal. Unfed day 5 butterflies were fed 17.5 % (11 males, 8 females), 35 % (9 males, 14 females) or 70 % (8 males, 9 females) sucrose by tarsal stimulation to satiation after they had demonstrated that they would approach and probe a flower on a *Lantana* inflorescence. They were then placed in a 14.5 cm × 11.5 cm × 10.5 cm high plastic chamber with a single *Lantana* inflorescence and videotaped to determine the time elapsed before feeding. Butterflies that visited the *Lantana* inflorescence within 15 min were removed and fed additional sucrose.

Crop emptying

Energy ingested, corrected for assimilation (see below) over the time to empty the crop and minus the rate of energy use for short-term maintenance, provides a measure of the rate of energy allocation for long-term functions (LeMagnen and Devos, 1970; McHugh and Moran, 1979; Hainsworth, 1980, 1990; Hainsworth *et al.* 1981, 1990; McCann and Stricker, 1986). Unfed butterflies were fed 30 μl of 17.5, 35, 52.5 or 70 % sucrose on day 2, 15 μl of 35 % sucrose on day 2 or 30 μl of 35 % sucrose on day 5 from calibrated capillary tubes. They were held by their wings, and the proboscis was uncoiled and placed inside the tube. This was sufficient to induce feeding. Sugar solutions were tinted with red food coloring to

aid subsequent visualization of crop contents. After feeding, butterflies that were returned to their cages generally remained inactive.

The volume in the crop was measured five (lower concentrations) or six (higher concentrations) times after feeding, to provide a good measure of emptying time at each concentration and volume (N=6–11 for each sample time). The butterflies were anesthetized by cooling and then quick-frozen in an alcohol and dry-ice bath. The crop was dissected and an opening made above the air space (empty crops were filled with air). Crop contents were drained into 10 μl calibrated capillary tubes. After most of the contents had been removed, the crop was lifted from the abdomen to remove any remaining fluid. The refractive index of crop fluid was measured with a temperature-compensated refractometer as a check for contamination with other fluids. Data were not used unless the refractive index was the same as that of the fluid fed to the butterflies.

Analysis of crop emptying

We compared six regression models that have been used in studies on other species or physical models of elastic emptying structures (Hopkins, 1966; Stubbs, 1977; McHugh and Moran, 1979; Hunt, 1980; Smith *et al.* 1984; Jobling, 1986; McCann and Stricker, 1986; Hainsworth *et al.* 1990): (1) linear, (2) square root, (3) cube root, (4) hyperbolic, (5) inverse cube root and (6) exponential changes in volume with time. Data were transformed and analyzed by linear, least-squares regressions. Models were compared using squared residuals for untransformed data (Smith *et al.* 1984; Jobling, 1986) in an analysis of variance (ANOVA) with multiple comparison (Scheffe's test) (Hainsworth *et al.* 1990). Values are presented as means±S.D. unless stated otherwise.

Digestive assimilation

The refractive index of excreted fluid of butterflies fed 17.5, 35 or 52.5 % sucrose was measured with a temperature-compensated refractometer. Butterflies were fed three meals per day of a sucrose solution on days 2 and 3. Between feedings they were kept in 3.75 l plastic cages with floors of aluminum foil. Freshly produced excreta were collected with capillary tubes. The first excretion was often red (food contained no dye) with subsequent fluids pink and eventually colorless. The red fluid was assumed to be fluid in the digestive tract prior to feeding, so only the refractive indices of colorless fluids were used to calculate assimilation efficiencies.

Results

Intake rates from sucrose solutions

Average volume intake rate decreased with increasing concentration, as in other butterfly species where fluid intake rate depends on viscosity (Boggs, 1988; Kingsolver and Daniel, 1979; May, 1985; Pivnick and McNeil, 1985) (Fig. 1). There were no differences between the sexes in average volume rate of intake for

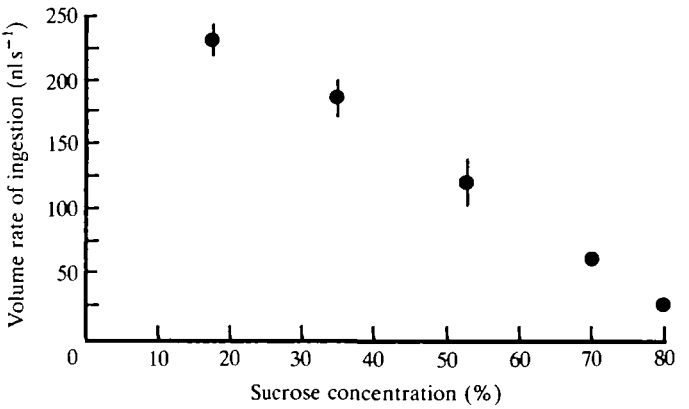

Fig. 1. Average±S.D. [N=7 (17.5%), 15 (35%), 8 (52.5%), 64 (70%), 10 (80%)] volume rate of ingestion by *Vanessa cardui* on day 2 as a function of sucrose concentration for individuals fed by tarsal stimulation.

butterflies fed 70% (males 62.1±14.0 nl s^{-1}, females 63.0±13.7 nl s^{-1}; $t_{62}=-0.26$, $P=0.8$) or 35% sucrose (males 195.4±37.8 nl s^{-1}, females 174.8±66.0 nl s^{-1}; $t_{13}=0.773$, $P=0.453$).

For the large number of butterflies fed 70% sucrose (masses 109–318 mg, mean=192.2±40.3 mg) there was no difference in average mass between sexes (males 191.4±39.8 mg, females 192.9±48.2 mg; $t_{62}=-0.152$, $P=0.9$) and volume rates of intake ranged from 32 to 107 nl s^{-1}. Volume rate of intake was positively correlated with mass ($r=0.365$, volume rate=37.8+0.13W, where W is body mass, $F_{1,63}=9.532$, $P=0.003$). Based on Poiseuille's equation, flow through the proboscis should depend on the pressure difference across its length divided by resistance [$8\mu l(\pi R^4)^{-1}$, where μ is viscosity, l is length and R is radius]. The cibarial pump or proboscis dimensions may vary with mass, although change in mass explained only 13% of the total variation in volume intake rate.

When volume rates of intake were converted to energy rates of intake there was a maximum at 35–52.5% sucrose (Fig. 2). This has been predicted and measured for other species of butterflies and is a consequence of the relationship between flow through the proboscis (influenced mainly by viscosity) and fluid energy content (May, 1985; Pivnick and McNeil, 1985).

Meal sizes

From tarsal stimulation

Fig. 3 is a frequency distribution of meal sizes for the 64 butterflies fed 70% sucrose on day 2. There was no difference between the sexes in average meal size (mean=36.2±9.9 µl, males 35.2±9.7 µl, females 37.0±10.6 µl; $t_{62}=-0.74$, $P=0.5$). Fifteen butterflies fed 35% sucrose on day 2 had an average meal size of 40.6±10.6 µl. Meal size was significantly greater with 35% sucrose (Mann–Whitney U-test, $P=0.049$), but the average energy content of a meal was significantly

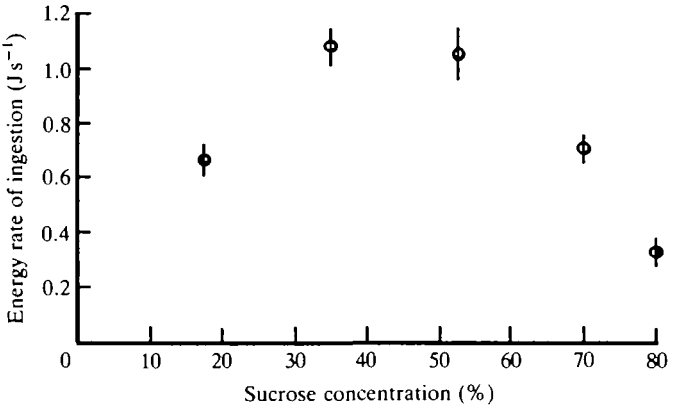

Fig. 2. Average±S.D. [N=7 (17.5%), 15 (35%), 8 (52.5%), 64 (70%), 10 (80%)] energy rate of ingestion by *Vanessa cardui* on day 2 as a function of sucrose concentration for individuals fed by tarsal stimulation. Energy was calculated from the heat of combustion of sucrose.

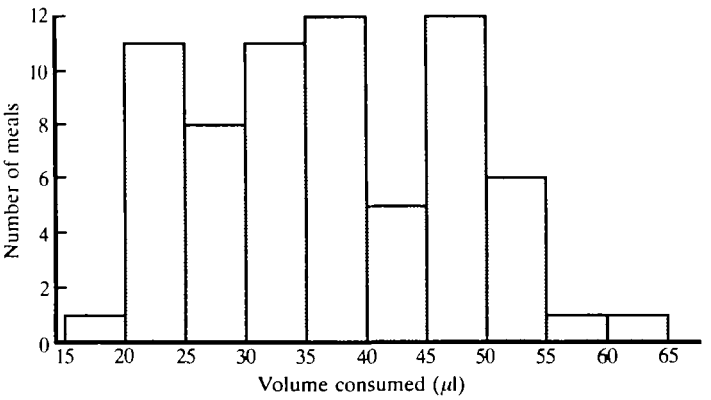

Fig. 3. Frequency distribution of volumes consumed to satiation for 64 *Vanessa cardui* fed 70% sucrose by tarsal stimulation on day 2.

greater for the higher concentration (417.6±114.2J for 70% *vs* 234.2±61.1J for 35% sucrose; Mann–Whitney U-test, $P<0.01$).

For butterflies fed 70% sucrose there was a positive correlation between meal size (μl) and body mass (W in mg) (r=0.33, meal size=20.2+0.08W, $F_{1,63}$=7.571, P=0.008). Crop volume may increase with body size, but variation in mass explained only 11% of the variation in meal size.

From Lantana *nectar*

304±28 flowers were available, and an average of 145±56 flower probes (some multiple probes of the same flower) occurred in 48.5±24 min before foraging

ceased. Meal size averaged 28.0±9.3 μl, significantly less than the 40.6 μl of 35 % sucrose ingested after tarsal stimulation on day 2 (Mann-Whitney U-test, $P<0.01$). Based on calculated ingestion times (21.1±9.7 min), the average volume rate of ingestion was 19.3±10.4 nl s^{-1}, considerably less than the 185±49.8 nl s^{-1} for 35 % sucrose ingested after tarsal stimulation (Fig. 1).

Meal frequencies

Because of variation in meal volumes, and for comparison with crop emptying (see below), meal frequencies were expressed as microliters consumed divided by hours to the next meal. There was no difference in average meal timing between butterflies that fed from *Lantana* nectar [12.4±7.5 μl h^{-1} (95 % confidence interval)] and those fed 35 % sucrose (12.8±2.3 μl h^{-1}). Average meal timing with 17.5 % (19.9±5.6 μl h^{-1}) and 70 % (6.8±2.4 μl h^{-1}) sucrose indicated relatively precise compensation for variation in concentration. A two-way ANOVA showed a significant effect of concentration ($F_{2,60}=15.18$, $P=0.0001$) and no significant effect of sex (mean for 33 males 12.84±7.94 μl h^{-1}, for 33 females 13.00±9.77 μl h^{-1}, $F_{1,60}=0.139$, $P=0.711$) or interaction ($F_{2,60}=0.396$, $P=0.396$). The variation in timing within concentrations resulted in no significant correlations between hours to the next meal and meal size (μl) for 17.5 % ($r=0.13$, $P=0.60$) and 35 % ($r=0.33$, $P=0.13$) sucrose, but a significant negative correlation for 70 % sucrose ($r=-0.53$, $P=0.03$).

Crop emptying

Day 2

Crop volumes immediately after feeding were not statistically different for butterflies fed 30 μl of different sucrose concentrations [mean values 17.5 %=26.1±1.4 μl ($N=6$), 35 %=24.8±1.6 μl ($N=7$), 52.5 %=25.7±1.8 μl ($N=7$), 70 %=25.3±1.1 μl ($N=6$), $F_{3,22}=0.815$, $P=0.5$]. The average of 25.5±1.5 μl indicates that 4–5 μl bypassed the crop for each concentration. About the same volume also bypassed the crop for butterflies fed 15 μl of 35 % sucrose, because adding 15 μl did not make the volumes statistically different from the average values immediately after ingesting 30 μl ($t_{11}=-1.964$, $P=0.08$).

Multiple comparisons of squared residuals for butterflies fed 30 μl showed that the inverse cube root (17.5 % and 35 % sucrose) and hyperbolic regressions (35 %, 52.5 % and 70 % sucrose) produced significantly higher variation (P values <0.05, Scheffe's F values ≥ 2.40). No discrimination between other regressions could be made (P values >0.05, Scheffe's F values ≤ 2.19) (Table 1). Similar results were obtained from analysis of crop emptying for blowflies (*Phormia regina*) fed different concentrations of sucrose or fructose (Hainsworth *et al.* 1990). Multiple comparisons of squared residuals for butterflies fed 15 μl of 35 % sucrose showed that only the hyperbolic regression produced significantly higher variation (P values <0.05, Scheffe's F values ≥ 2.51).

Table 1. *Scheffe's* F *values for comparisons of untransformed residual squared variation from regressions for* Vanessa cardui *fed 30 μl of different concentrations of sucrose on day 2*

	$\sqrt{V}$	$\sqrt[3]{V}$	$\frac{1}{\sqrt[3]{V}}$	$\frac{1}{V}$	ln$V$
17.5 %					
V	7.5×10^{-12}	7.0×10^{-12}	4.1*	3.3×10^{-7}	1.9×10^{-10}
$\sqrt{V}$		6.0×10^{-15}	4.1*	3.3×10^{-7}	2.7×10^{-5}
$\sqrt[3]{V}$			4.1*	3.3×10^{-7}	2.7×10^{-10}
$\frac{1}{\sqrt[3]{V}}$				4.1*	4.1*
$\frac{1}{V}$					3.2×10^{-7}
35 %					
V	5.3×10^{-5}	1.1×10^{-7}	3.8*	4.0*	0.009
$\sqrt{V}$		5.7×10^{-5}	3.9*	4.0*	0.01
$\sqrt[3]{V}$			3.8*	4.0*	0.009
$\frac{1}{\sqrt[3]{V}}$				0.002	3.5*
$\frac{1}{V}$					3.7*
52.5 %					
V	6.3×10^{-5}	1.0×10^{-5}	0.041	6.7*	6.3×10^{-5}
$\sqrt{V}$		1.7×10^{-7}	0.043	6.8*	1.3×10^{-4}
$\sqrt[3]{V}$			0.043	6.8*	1.2×10^{-4}
$\frac{1}{\sqrt[3]{V}}$				5.7*	0.038
$\frac{1}{V}$					6.7*
70 %					
V	5.5×10^{-6}	3.4×10^{-4}	0.453	12.4*	0.005
$\sqrt{V}$		2.5×10^{-4}	0.45	12.4*	0.005
$\sqrt[3]{V}$			0.429	12.3*	0.003
$\frac{1}{\sqrt[3]{V}}$				8.1*	0.359
$\frac{1}{V}$					11.9*

Crop volume (V) is measured in microlitres.
* $P<0.05$.

Day 5

Crop volumes from butterflies fed on day 5 were used to test for differences between sexes. A two-way ANOVA showed no significant effect of sex (mean values: 31 males=17.12±9.23 μl, 34 females=15.61±9.25 μl, $F_{1,55}=1.492$, $P=0.23$), a significant effect of time ($F_{4,55}=245.5$, $P=0.0001$) and no significant

Table 2. *Intercepts, slopes and ± their 95 % confidence limits for linear regressions of crop volumes (μl)* vs *time (h) for* Vanessa cardui *fed sucrose solutions of 30 μl or 15 μl on day 2*

% Sucrose	Intercept	Slope
Fed 30 μl		
17.5	23.1±1.9	−12.5±1.3
35	24.2±1.3	−5.9±0.4
52.5	23.6±1.4	−3.5±0.3
70	25.5±1.2	−2.8±0.2
Fed 15 μl		
35	10.6±0.5	−4.3±0.6

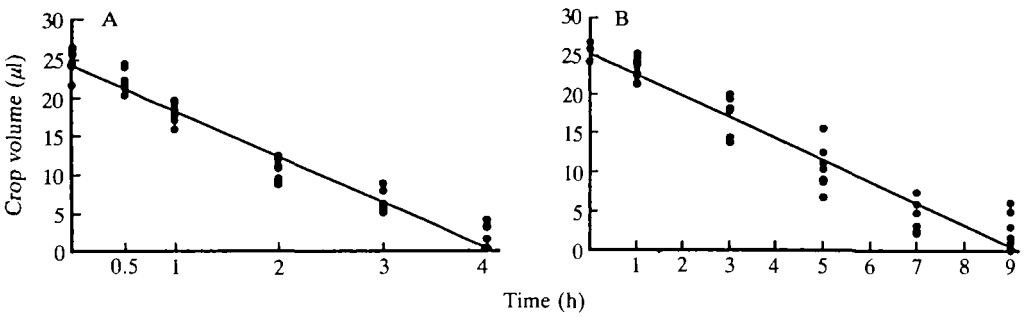

Fig. 4. Individual data and linear regression equations for crop volume *vs* time for *Vanessa cardui* fed 30 μl of 35 % (A) or 70 % (B) sucrose on day 2.

interaction ($F_{4,55}$=0.139, P=0.97). Multiple comparisons of squared residuals for combined data showed that only the hyperbolic regression produced significantly higher variation (P values <0.05, Scheffe's F values ≥13.61).

Comparisons of linear regressions

Linear regressions described crop emptying with relatively low residual variation (Table 1) and were considered most parsimonious for comparisons (Table 2). Fig. 4 shows linear regressions with individual measurements of crop volumes for butterflies fed 35 and 70 % sucrose on day 2. Volume rates of emptying decreased significantly with each increase in concentration for butterflies fed 30 μl on day 2 (Table 2). Butterflies fed 15 μl of 35 % sucrose on day 2 had a significantly lower volume rate of emptying than butterflies fed 30 μl (Table 2).

Combined data for male and female butterflies fed 30 μl of 35 % sucrose on day 5 gave the linear regression V=25.9–8.07t (N=65, r^2=0.91), where t is time in hours and V is crop volume in microliters. Therefore, about the same volume bypassed the crop, but the crop emptied significantly faster compared with butterflies fed the same food on day 2 (Table 2). From the linear regression, the crop would reach zero volume in 3.2 h, so the 30 μl fed would be processed at an overall rate of

9.3 μl h^{-1}. This is significantly less than the rate calculated from meal intervals for butterflies fed 35 % sucrose on day 5 (12.8 μl h^{-1}, $P<0.05$). The difference could be due to variations in meal size or to initiation of meals when about 7 μl remained in the crop. The latter might well occur, because experiments show that Australian sheep blowflies (*Lucilia cuprina*) feeding *ad libitum* feed again before complete crop emptying (Simpson *et al.* 1989).

Digestive assimilation

The average refractive index of excreted fluid was the same for butterflies fed 17.5 and 35 % sucrose (1.3338±0.0004, 40 samples from three butterflies fed 17.5 % and 31 samples from four butterflies fed 35 % sucrose). If all solutes in the fluid were sugars, assimilation efficiencies would be 96 % (17.5 % sucrose) and 98 % (35 % sucrose). The average refractive index of excreted fluid was significantly higher for butterflies fed 52.5 % sucrose (1.3345±0.0011, 48 samples from five butterflies, Scheffe's $F>9.01$), but assimilation efficiency remained high (98 %). Based on these results, *Vanessa* are considered to assimilate essentially all ingested sugars regardless of concentration.

Overall energy processing rates

Energy processing rates were calculated from linear regressions for the time to reach a crop volume of 0.5, 1.0, 3.0 or 7.0 μl. Several volumes were used to assess the consequences of initiating meals when the crop had emptied to different volumes (see above; Simpson *et al.* 1989; Hainsworth *et al.* 1990). Because we wanted to know the rates for processing amounts fed, including what may have bypassed the crop, calculated times for each regression were divided into volumes fed minus final crop volume. Volume rates of processing were converted to energy rates using the heat of combustion of sucrose (16.48 J mg^{-1}).

There was little change in processing rate as concentrations increased within a final crop volume for butterflies fed 30 μl on day 2 (Table 3). A larger volume remaining in the crop at feeding would increase overall energy processing rates because the amount bypassing the crop becomes an increasing fraction of the total amount processed. Butterflies fed 15 μl of 35 % sucrose on day 2 had lower overall processing rates for emptying the crop to 0.5–3.0 μl, as expected from the lower slope for volume rate of emptying, but emptying the crop to 7.0 μl would raise overall processing rate so that it exceeds rates for butterflies fed 30 μl (Table 3). Butterflies fed 30 μl of 35 % sucrose on day 5 had higher overall processing rates (Table 3), as expected from their higher rate of crop emptying.

Mature egg production

The butterflies fell into two groups: (i) those producing mature eggs without sugar intake and those with a steep slope for egg production *vs* sugar intake (Fig. 5, closed symbols), and (ii) those requiring sugar to produce eggs and with a lower slope (Fig. 5, open symbols). The former were 22 % of all females and they occurred in one generation. The difference may have reflected the nutritional

Table 3. *Overall energy processing rates in Jh^{-1} for times for* Vanessa cardui *to reach variable crop volumes (times calculated from linear regression equations)*

	Final crop volume (μl)			
% Sucrose	0.5	1.0	3.0	7.0
Fed 30 μl on day 2				
17.5	47.0	47.3	48.4	51.5
35	42.3	42.6	43.4	45.5
52.5	38.7	38.8	39.7	42.0
70	38.1	38.2	38.7	40.2
Fed 15 μl on day 2				
35	35.7	36.2	39.1	55.1
Fed 30 μl on day 5				
35	54.1	54.2	54.9	56.6

Energy is standardized for the volume fed, i.e. all energy is accounted for, not just energy emptied from the crop.

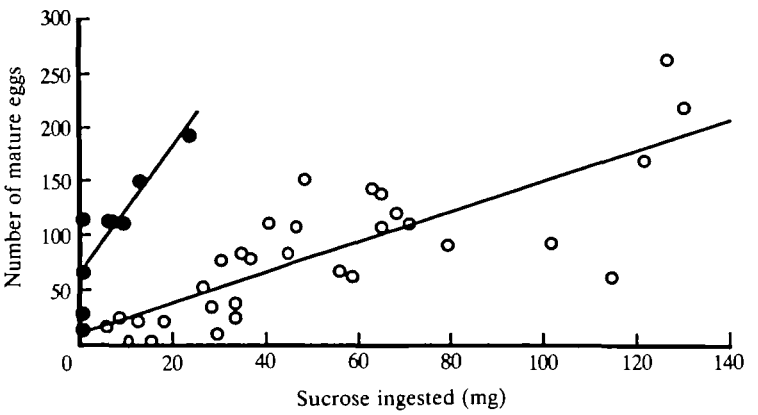

Fig. 5. Number of mature eggs produced by day 8 as a function of the mass of sucrose ingested by virgin *Vanessa cardui*. See text for description and equations for data represented by filled and open symbols.

condition of newly emerged adults. For both groups there was a significant linear correlation between the number of eggs produced and the mass of sucrose ingested (r=0.80, $F_{1,30}$=53.862, P=0.0001, y=6.8+1.44x for open symbols, r=0.86, $F_{1,7}$=19.394, P=0.0031, y=60.2+6.09x for closed symbols).

Discussion

Laboratory studies show the importance of adult nutrition for lepidopteran fecundity and longevity (Murphy *et al.* 1983; Leather, 1984; Hill and Pierce, 1989;

Hill, 1989). In general, sugar intake increases longevity and fecundity. Although the effects of sugars may vary (Hill and Pierce, 1989), newly emerged adults could benefit considerably by maximizing net gains of energy per meal from nectar during foraging.

How butterflies maximize net meal energy [$(cM-fh)$ in equation 1] under natural conditions should depend on factors influencing net energy ingestion from floral nectars. May (1988) studied nectar foraging by *Agraulis vanillae* and *Phoebis sennae* visiting several plant species in central Florida. Energy per flower was correlated with nectar volume per flower, and nectar volume per flower was correlated with corolla length. There was no correlation between energy per flower and nectar concentration (May, 1988). It is not clear, however, whether butterflies should always select plants to visit based on energy per flower, because a greater net meal energy could be obtained by visiting flowers with higher concentrations but lower total energy. Whether this occurs can depend on the time required to consume a meal from different nectars. If meal time is constrained, then a greater net meal energy could be obtained from nectars with higher volumes and lower concentrations (Hainsworth, 1989). An average meal of 28 μl of *Lantana* nectar took 48.5 min followed by an average of 2.2 h to the next meal, so 27 % of time was spent feeding. This should vary with nectar availability, but the calculations suggest there can be situations where foraging time is not constrained and net meal energy could be maximized by visiting flowers with high nectar concentrations.

Butterflies spend time moving between and probing flowers, so continuous ingestion from unlimited volumes does not reflect their usual time use (May, 1985, 1988). *Vanessa* foraging from *Lantana* flowers took an average of 48.5 min to consume an average meal of 28.0 μl, so the average rate of energy gain for a meal (0.05 J s^{-1}) was considerably less than with continuous ingestion of 35 % sucrose (1.08 J s^{-1}, Fig. 2). The difference partly involves the time for travel between flowers and the time taken to probe empty flowers (an average of 27.4 min or 56 % of meal time) but, even after accounting for these, the average volume rate of ingestion was less from *Lantana* nectar than from a pool of 35 % sucrose. Emptying flowers of small volumes may require more time per microliter, or rates may increase with experience (Laverty and Plowright, 1988).

What would be the consequence of a higher *Lantana* nectar concentration? If it is assumed that increasing the concentration from 33 to 70 % sucrose only influences ingestion time by the factor shown in Fig. 1 (3.04 times), average ingestion time would increase from 21.1 to 64.1 min. This would increase the total average time for a meal from 48.5 to 48.5+(64.1–21.1)=91.5 min, and the average *rate* of energy gain for a meal would be slightly higher for 70 % sucrose. With an average meal size of 28.0 μl, 33 % sucrose would give 188.4 J h^{-1} and 70 % sucrose 211.8 J h^{-1}. The rate of net energy gain involves expenditures that would be higher for a longer meal, but this should involve only a few joules per hour. This emphasizes the importance of considering meal energy gains for efficient foraging, because nectar with 70 % sucrose yields more than twice the energy, even though

the rate of gain may be little different from that of a meal of *Lantana* nectar containing 33 % sucrose.

In laboratory experiments (with no time constraint) *Vanessa* preferred concentrated sucrose, even though it produced a lower rate of net energy gain during ingestion (Hainsworth, 1989). Newly emerged females that maximized net meal energy by selecting concentrated food would maximize the number of mature eggs produced from a meal of that food (Fig. 5). The production of eggs could eventually plateau, and this may depend on how rapidly egg production increases with sugar ingestion, but a plateau was not apparent over the time and range of sugar intakes used in these experiments. Thus, at least for newly emerged female *V. cardui*, there is a rapid and direct impact of ingested sugars on potential reproductive performance. The lack of difference between sexes in meal sizes and subsequent processing from the crop suggests that newly emerged male *Vanessa* may be influenced in a similar way to females in reproductive performance, perhaps associated with mate location or spermatophore provisioning (Boggs and Watt, 1981). Depending on the sugars obtained within the first few days of foraging, it is likely that males and females would shift time investment towards mating and oviposition, respectively, and that minimizing the time for this should have an important impact on fitness.

The use of sugars for reproduction (s in equation 1) is reflected in food processing rates. Studies of stomach or crop emptying have related rate of emptying to rates of use of assimilated energy in several species (Treherne, 1957; McHugh and Moran, 1979; Hunt, 1980; Hainsworth *et al.* 1981, 1990; Simpson, 1983; McCann and Stricker, 1986; Wolf and Hainsworth, 1977). A striking feature for *Vanessa* is the high rate of energy processing relative to the maintenance rate of energy use. From measurements of oxygen consumption rates for insects (Zebe, 1954; Bartholomew and Casey, 1978; Gromysz-Kalkowska and Hubicka, 1988), and assuming a respiratory quotient of 1.0, a 198 mg *V. cardui* would use 1.6–2.4 $J\,h^{-1}$ for maintenance at rest. This is 12–30 times less than the overall energy processing rates calculated with linear regressions for *Vanessa* fed 30 μl on day 2 (Table 3). The uniform and high energy processing rate for *Vanessa* is probably due to the direct impact of ingested sugars on reproduction.

Although energy processing rates were high relative to maintenance rates of energy use on day 2, the rates were even higher following 4 days of food deprivation (Table 3). This suggests a feedback mechanism between energy stores and digestive processing that may compensate for low net energy gains from feeding just after emergence.

Vanessa were fed a meal after specified deprivation times but, with continuous access to food, their meal sizes and feeding patterns could be very different. With *ad libitum* feeding, the sheep blowfly consumes larger meals of a dilute sugar solution, but with 24 h of deprivation they consume larger meals of a more concentrated solution (Simpson *et al.* 1989). An interplay between tarsal thresholds (influenced by previous meals) and available food could produce variation in meal sizes and timing, as could changing nutritional condition from

prior feeding. Even for the second meal consumed by *Vanessa* it is likely that feeding involves stimuli other than just an empty crop, because their crops would not have been empty at the time they initiated a second meal from *Lantana*.

Feeding before the crop empties increases overall energy processing rates (Table 3; Hainsworth *et al.* 1990). It is particularly pronounced with small meals because the food bypassing the crop is a relatively large fraction of the amount processed before the next meal. Although smaller meals are emptied more slowly from the crop, meal initiation before the crop is empty could make overall energy processing rates comparable to those for larger meals. Additional experiments are needed to measure crop volumes at feeding following small meals to determine the extent to which the mechanism represents an adaptation for maximizing energy processing rates.

The dynamics of food processing indicate energy additivity in long-term energy storage rates across concentrations, and meal frequencies suggest a simple hypothesis for how newly emerged *Vanessa* can maximize fitness through feeding behaviour. There is variation in nectar concentration within and among plants (Heyneman, 1983; Pivnick and McNeil, 1985; May, 1988), so not all butterflies will consume meals of concentrated nectar. However, because energy processing rates are similar across concentrations, a butterfly should be able to achieve high egg production despite lower nectar concentration by increasing meal frequency. This may carry a cost if predation rate is related to feeding frequency. The variation of T with c (equation 1) shows the appropriate adjustment for *Vanessa*. In blowflies (Hainsworth *et al.* 1990) and hummingbirds (Hainsworth, 1990) energy storage rates between meals increase with sugar concentration, so foods of high concentration have a greater long-term use per unit of food energy.

Most nectar concentrations are intermediate in value (an average of 25% sucrose for 85 species of butterfly-pollinated plants, Heyneman, 1983), and it has been suggested that the evolution of nectar concentration reflects factors producing a maximal rate of net energy gain during foraging (Baker, 1975; Heyneman, 1983; Kingsolver and Daniel, 1979; May, 1985, 1988; Pivnick and McNeil, 1985). *Vanessa* would benefit from higher concentrations unless this constrained the time for a meal (Hainsworth, 1989). The time required for a meal from *Lantana* suggests that this does not have to be the case, so the evolution of nectar concentrations may involve factors necessary and sufficient for visitation, with appropriate adjustment for costs to the plants, but without design for what is 'best' for pollinators.

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