Effects of Partial Sugar Deprivation on Lifespan and Carbohydrate Mobilization in the Parasitoid *Macrocentrus grandii* (Hymenoptera: Braconidae)

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ABSTRACT We compared the lifespan of *Macrocentrus grandii* (Goidanich) adults fed a 50% sucrose solution at various intervals throughout their lives. Treatments included starvation, continuous feeding, feeding on the first day of life only, and feeding every second, third, or fourth day of life. Life expectancy for starved males and females was less than 3 d, and providing sugar during the first day of life increased life expectancy by 2 d for males and 4 d for females. Life expectancy was highest when adults were fed continuously (14 d for males and 21 d for females) or every 2 d (17 d for males and 23 d for females). The life expectancy of adults that were fed either every 3 or every 4 d ranged between 9 and 16 d. Together, these results demonstrate that a constant supply of sugars is not necessary to achieve maximum survivorship, and limited sugar availability may suffice to increase substantially the lifespan of *M. grandii* over starvation values. A series of anthrone tests was used to determine levels of gut sugars, simple storage sugars ('body sugars'; primarily trehalose), and glycogen over the first 6 d of life of female and male *M. grandii* that were either fed 50% sucrose continuously, the first day of life only, or not at all. A single day of sugar feeding led to apparently maximum levels of gut sugars, body sugars, and glycogen, and parasitoids fed only on the first day of life maintained high levels of these nutrients for 1 d postfeeding. After this time, glycogen and gut sugars decreased substantially, but body sugar levels remained essentially constant. This pattern suggests a strategy in which gut sugars and glycogen are mobilized to maintain high levels of body sugars in starving parasitoids.

KEY WORDS *Macrocentrus grandii*, anthrone, carbohydrate, glycogen, sugar, parasitoid

THE MAIN ENERGY source for adult parasitoids is sugar, which can be obtained in the field from nectar or honeydew (e.g., Rogers 1985; Hagen 1986; Elliot et al. 1987; Idoine and Ferro 1988; Hagley and Barber 1992; Evans 1993; Jervis et al. 1993, 2002; Patt et al. 1997; Stapel et al. 1997; Tooker and Hanks 2000). Although numerous laboratory studies have shown that the lifespan of adult parasitoids can be increased on the order of 10-fold by sugar feeding (e.g., Leius 1961, Avidov et al. 1970, Syme 1975, 1977, Hagley and Barber 1992, Wäckers and Swaans 1993, Dyer and Landis 1996, Heimpel et al. 1997, McDougall and Mills 1997, Olson et al. 2000), effects of sugar on field longevity are not known (Rivero and Casas 1999). Both the quality and abundance of sugar sources is likely to be more variable in the field than in laboratory assays where high-quality sugar sources are typically presented ad libitum over the parasitoid's lifespan. Sources of variability such as temporal patterns of floral and extrafloral nectar availability, variability in nectar consumption by pollinators, and population fluctuations of honeydew-producing Homoptera and the ants that tend them, make it unlikely that sugar availability will be constant over the life of most adult parasitoids. Furthermore, costs associated with searching for sugar sources (as opposed to hosts) may limit circumstances under which foraging for food is profitable (Lewis and Takasu 1990; Takasu and Lewis 1993, 1995; Wäckers 1994; Sirot and Bernstein 1996; Lewis et al. 1998; Clark and Mangel 2000). The ability to withstand periods of host-deprivation may therefore allow parasitoids to allocate more time to searching for hosts versus food.

Here, we explore temporal sugar requirements of the parasitoid *Macrocentrus grandii* (Goidanich). Our first objectives were to determine (1) the effects of a single day of sugar feeding on *M. grandii* life expectancy, (2) how often *M. grandii* adults need to feed on sugar to attain maximum longevity, and (3) how various levels of temporary sugar deprivation affect life expectancy.

A second set of objectives focused on the physiological response to starvation in *M. grandii*. We were interested primarily in utilization of limiting amounts of sucrose, because sucrose and its component monosaccharides, fructose and glucose, are major components of nectar (van Handel et al. 1972, Harborne 1988). Once ingested, the disaccharide sucrose

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is enzymatically cleaved inside the gut and absorbed into the hemocoel as glucose and fructose (Wyatt 1967, Friedman 1985). In most insects, both glucose and fructose are converted to the disaccharide trehalose and/or glycogen within the fat body surrounding the gut (Wyatt 1967, van Handel 1984, Friedman 1985). Olson et al. (2000) found that meals of 50% sucrose solution produced elevated levels of gut sugars, body sugars (presumably primarily trehalose) and glycogen in *M. grandii*. Various metabolic strategies by which trehalose and glycogen reserves are used during starvation have been documented in insects. These strategies range from rapid depletion of both trehalose and glycogen (e.g., Ziegler 1991) to homeostatic regulation of trehalose levels mediated by the breakdown of glycogen (Chapman 1998). Here, we use a series of biochemical assays based on the anthrone reaction (Mokrasch 1954) to characterize patterns of sugar mobilization during starvation in *M. grandii*.

Materials and Methods

Macrocentrus grandii is a polyembryonic larval parasitoid of the European corn borer, *Ostrinia nubilalis* (Hübner) (Lepidoptera: Crambidae) and was introduced to the United States in the 1920s and 1930s (Parker 1931, Baker et al. 1949, Edwards and Hopper 1998). The maximum longevity of starved adults is 4–5 d and can be greatly increased by feeding on a 50% sucrose solution (Olson et al. 2000) or floral nectar (Orr and Pleasants 1986). *Macrocentrus grandii* females do not host feed, and are synovigenic (i.e., females mature eggs posteclosion) (Olson et al. 2000, Jervis et al. 2001).

Parasitoids used in our assays were reared in third to fifth instar European corn borer larvae as described by Olson et al. (2000). The *M. grandii* colony came from broods collected from diapausing feral larvae of *O. nubilalis* in Minnesota in 1997 and was maintained at 27°C, 75 ± 5% RH, and a photoperiod of 16:8 (L:D) h. European corn borer hosts were obtained from a laboratory colony maintained as described by Leahy and Andow (1994). *Macrocentrus grandii* pupal masses typically contained between 15 and 40 pupae and were held singly in 10-cm-diameter plastic petri dishes until emergence. Wasps in the food treatments were provided sugar ≈3 h after emerging in the dishes, and all wasps were provided water by filling a 0.50 ml microcentrifuge tube with distilled water and threading a cotton string through a hole in the cap of the tube.

Parasitoid Survivorship. The survivorship of adult male and female *M. grandii* was compared under six different feeding regimes in which 'food' was a 50% sucrose solution (by weight), and water was provided in every treatment: (1) Starved (i.e., provided with water only). (2) Fed continuously from emergence. (3) Fed only during the first day of emergence and starved thereafter until death. (4) Fed every 2 d beginning from emergence until death such that there was 1 d of starvation between feeding days. (5) Fed every 3 d starting from emergence until death such that there were 2 d of starvation between feeding days. (6) Fed every 4 d starting from emergence until death such that there were 3 d of starvation between feeding days.

Parasitoids emerging from each brood were placed in groups of two wasps of the same sex in plastic petri dishes (6 cm diameter). The dishes holding the wasps were randomly assigned to all treatments such that wasps from the same brood were nearly equally represented in all treatments. For the five feeding treatments, the sucrose solution was smeared on the inside of the petri dish cover with a sterile cotton-tipped applicator. For treatments 3–6, parasitoids were fed *ad libitum* for 24 h during the day of feeding. On starvation days, sugar-coated petri dish covers were replaced with new (sterile) petri dish covers. Parasitoids that were fed only during the first day of emergence and then starved (treatment 3) were transferred to fresh dishes without sucrose (but with water) after exposure in sucrose-smeared dishes for 24 h. Petri dishes were kept at 27°C, 75 ± 5% RH, and a photoperiod of 16:8 (L:D) h and checked daily for survival. At least 29 parasitoids were used for each treatment/sex combination. Effects of the diet treatment and parasitoid sex on survivorship were evaluated using proportional hazards modeling, a nonparametric analysis designed for testing for effects of multiple factors on cohort survivorship (SAS Institute 1995). In addition, separate proportional hazards analyses were run for all of the possible pairwise comparisons of the treatments, in which an experiment-wide error rate of $\alpha' = \alpha/n = 0.0033$ ($\alpha = 0.05, n = 15$) was used to assign significant differences. This procedure is analogous to using the Bonferroni correction for means comparisons in analysis of variance (ANOVA) and provides a conservative test for individual statistical differences (Neter et al. 1990).

Sugar Metabolism. Daily changes in the amounts of fructose (i.e., gut sugars), glycogen, and 'body sugars' (i.e., nonglycogen sugars not present in the gut; presumably primarily trehalose) were quantified from *M. grandii* that were either starved, fed only during the day of emergence, or fed continuously. Parasitoids were placed in groups of two wasps of the same sex within plastic petri dishes (6 cm diameter). Individuals from the same broods were distributed evenly across the three treatments. Water was provided in each treatment as described above, as was 50% sucrose solution for the two treatments in which food was offered. petri dishes were kept at 27°C, 75 ± 5% RH, and a photoperiod of 16:8 (L:D) h. Wasps from each group were frozen and assayed daily from ages 1–6 d in the two sucrose-fed treatments, and from ages 0–5 d in the starvation treatment. Since starved male parasitoids do not normally live beyond 4 d, nutrient assays were conducted for 0- to 4-d-old male parasitoids in the starvation treatment. The amount of each nutrient obtained from newly emerged unfed (starved, day 0) wasps was regarded as the baseline amount present in male and female wasps at emergence. One forewing of each parasitoid was slide mounted and measured from the outer edge of the anal cell to the outer edge of the

tip of the wing to the nearest 0.02 mm as an estimate of parasitoid size. Nutrient analyses were conducted on at least ten individuals per day for each treatment/sex combination, except in some starvation treatments where less than ten parasitoids were available per treatment due to the difficulty of keeping parasitoids alive without food for longer than 3 d.

The amounts of fructose, body sugars and glycogen in individual wasps were estimated as described by Olson et al. (2000) with the exception that ovaries were not removed from the females. The assays were based on a series of biochemical tests developed by van Handel (1965, 1967, 1985a, 1985b). Individual parasitoids were crushed with a plastic pestle in a 1.5-ml microcentrifuge tube containing 50 μl of 2% sodium sulfate in distilled water and placed on ice. After cooling, 450 μl of chloroform-methanol (1:2) was added and the tubes were vortexed. Tubes were then centrifuged at 14,000 rpm for 2 min and 200 μl of the resulting supernatant was transferred to a glass tube (12 mm diameter by 75 mm) for the sugar assays (the 'sugar tube'). An additional 200 μl of the supernatant was discarded and the precipitate was left in the microcentrifuge tube for the glycogen assay (the 'glycogen tube'). The sugar tube was heated at 90°C until ≈50 μl of solution was left in the tube, and the glycogen tube was heated at the same temperature for 15 min. Analyses were done in batches of between four and eight samples for each of the nutrients described below, and a blank tube was run alongside each sample/nutrient combination so that absorbance values from the blanks could be subtracted from sample absorbances.

The cold anthrone test was used to estimate amounts of fructose and gut sugars from individual parasitoids. To estimate the amount of fructose, 950 μl anthrone reagent (prepared as described in van Handel 1985a) was added to the sugar tube, mixed, and left to react for 1.5 h, and the absorbance measured at 625 nm (cold anthrone reading). Standard curves were generated to transform absorbance values into absolute amounts. Pure fructose (Fisher, Fairlawn, NJ) was tested at 625 nm in amounts ranging from 1 to 50 μg in three replicate runs. The resulting linear regression was highly significant ($F = 883$, df = 21, $P < 0.0001$, $r^2 = 0.98$) and generated the following regression equation: absorbance = 0.053 + 0.035 (μg fructose). The total amount of sugars present in the gut was estimated from this value as follows. First, the cold anthrone reading was multiplied by two because sucrose (which was fed to the parasitoids) is composed of equal parts fructose and glucose, and the glucose does not react with anthrone at room temperature (van Handel 1967; Olson et al. 2000). Second, this amount was multiplied by 2.5, because two-fifths of the original 500 μl of the solution was used for the assay. Thus, the amount of fructose detected by the cold anthrone assay was multiplied by 5 to estimate the amount of sugars present in the gut of *M. grandii*.

The hot anthrone test measures total sugars present in the sample (van Handel 1967; Olson et al. 2000). To estimate the amount of nonglycogen sugars present within parasitoid hemolymph and other tissues (i.e., 'body sugars'), the gut sugars value was subtracted from results of a hot anthrone test done on the supernatant from which glycogen had been precipitated. The hot anthrone test was carried out on the same solution that was used for the cold anthrone test. After the cold anthrone test was completed, the solution was heated at 90°C for 15 min and then cooled on ice. The absorbance was read at 625 nm to give a measurement of total sugars. Standard curves for the hot anthrone test were run in two replicates at a range of 1–50 μg using pure glucose (Dextrose; Fisher), which has virtually the same absorbance as fructose and glucose using the hot anthrone test (Olson et al. 2000). The linear regression was highly significant ($F = 539$, df = 12, $P < 0.0001$, $r^2 = 0.98$) and generated the following regression equation: absorbance = 0.150 + 0.036 (μg glucose). The amount of sugars estimated from the hot anthrone test was multiplied by 2.5 to compensate for the fact that two-fifths of the original solute was used and the gut sugars were subtracted from this amount to arrive at the estimated amount of body sugars.

Glycogen levels were estimated by adding 1 ml of anthrone reagent to the microcentrifuge tube containing the precipitate after centrifugation. The tube was heated at 90°C for 15 min., cooled on ice and the absorbance measured at 625 nm. Because all glycogen in the sample is presumed to precipitate to the bottom of the tube, the absorbance amount is considered representative of the whole insect. Standard curves were generated at 625 nm based on three replicates of between 1 and 100 μg of oyster glycogen (ICN Biomedicals, Aurora, OH). Regression analyses showed that a quadratic fit was superior to linear regression, with both first and second-order polynomial parameters significant ($P < 0.0001$ for each parameter, with $F = 1{,}640$, df = 22, $P < 0.0001$, and $r^2 = 0.993$ for the quadratic fit). The quadratic regression generated the following equation: absorbance = $-0.007 + 0.044$ (μg glycogen) -0.0002 (μg glycogen)^2.

Results from the starvation treatment were excluded from the statistical analyses of nutrient levels to stabilize variances and because nutrient levels in starved individuals were so clearly different from nutrient levels in the other two treatments (see *Results*). We used multiple regression analyses in which the diet treatment was coded as a binary variable and age and winglength were continuous variables. The analyses were run on absorbance values to avoid slight inaccuracies inherent in using standard curves and other forms of estimation. Of particular interest in these analyses was the interaction between effects of diet treatment (i.e., fed for 1 d versus fed continuously) and age on nutrient absorbance levels. For nutrients that were significantly affected by parasitoid winglength in the multiple regression analysis, we did linear regressions of nutrient levels onto winglength separately for each diet treatment.

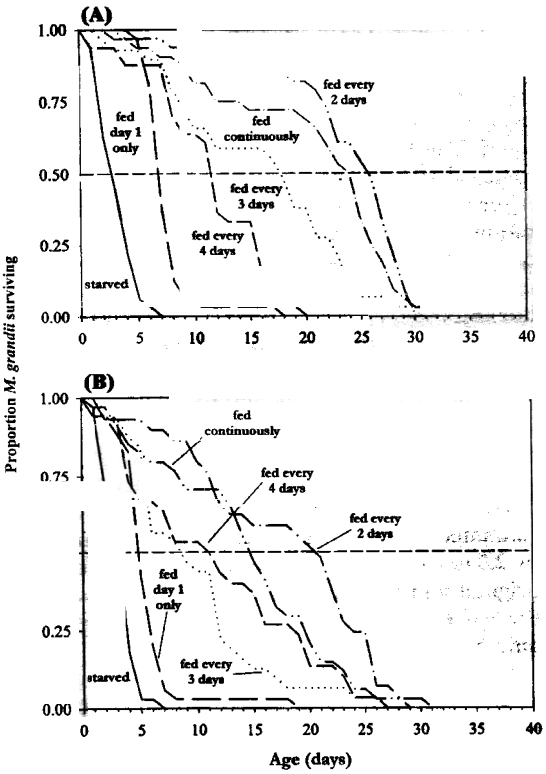

Fig. 1. Survivorship curves for female (A) and male (B) *M. grandii* subjected to each of six diet treatments. The dashed line at 0.5 survivorship indicates median longevity for each treatment.

Results

Parasitoid Survivorship. Female *M. grandii* lived significantly longer than male *M. grandii* and the feeding treatments had significant effects on survivorship as well (Proportional hazards analysis including feeding treatment and gender: feeding treatment, $\chi^2 = 275.3$, df = 5, $P < 0.00001$; gender, $\chi^2 = 13.9$, df = 1, $P < 0.0001$). Survivorship curves for all six treatments for both sexes are shown in Fig. 1, and life expectancies are given in Table 1. Providing *M. grandii* with 50% sucrose for 1 d at the beginning of their lives increased life expectancy by ≈4 d for females and 2 d for males (Table 1). Survivorship was further increased by feeding the parasitoids at least every 4 d (Fig. 1; Table 1). Maximum survivorship for both females and males was observed in the treatment in which parasitoids were fed every 2 d, with slightly lower life expectancies observed when wasps were fed throughout their lives, but this difference was not significant (Fig. 1; Table 1). However, wasps fed every 2 d lived significantly longer that wasps fed every 3 d, whereas there was no significant difference between wasps fed every 3 d and fed continuously (Table 1).

Nutrient Analyses. As expected, gut sugars were absent in starved individuals and present during the complete 6-d observation period in individuals that were fed continuously (Fig. 2). When parasitoids were fed only on the first day of their life, gut sugars were detectable in substantial amounts from females

Table 1. Life expectancy in days for female and male *M. grandii* subjected to six feeding regimes

Feeding regime	Females	Males
		2.9a
		5.1b
		11.4cd
		9.4d
		17.3c
		14.1cd

Life expectancy was calculated from day zero according to Southwood (1978):

$$\sum_{x}^{w}\left(\frac{l_x + l_{x+1}}{2}\right)$$

where l_x is survivorship at day x, and w is the day at which zero parasitoids remain alive. Significant differences among treatments were calculated separately for females and males. Life expectancies associated with different letters were significantly different in proportional hazards survivorship analyses for which the criterion for significant differences was the experiment-wide error rate, $\alpha' = \alpha/n = 0.0033$, with $\alpha = 0.05$ and $n = 15$ pairwise comparisons.

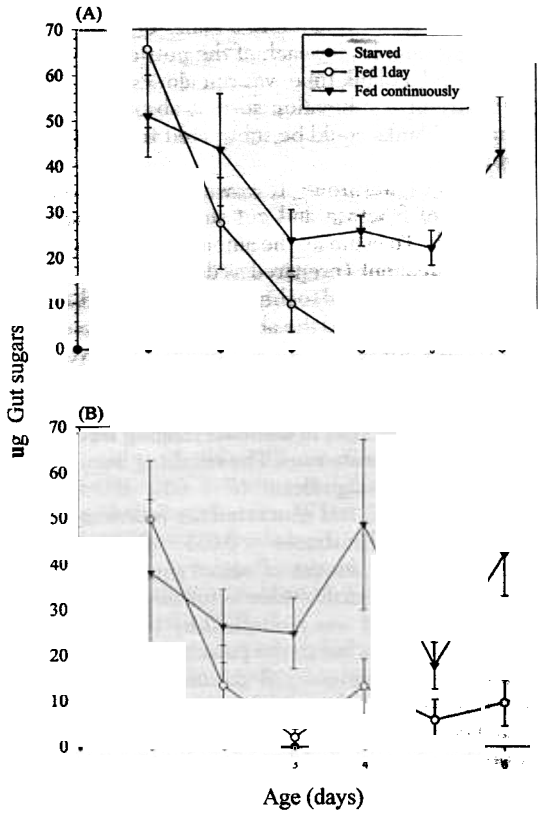

Fig. 2. Micrograms of gut sugars of female (A) and male (B) *M. grandii* that were either starved, fed 50% sucrose for the first day of life only, or fed 50% sucrose over their entire life.

Table 2. Multiple regression analysis testing for effects of diet treatment (fed only on day 1 versus fed continuously), age, the interaction between diet and age, and winglength on absorbance values for gut sugars, body sugars and glycogen

	df	Females			Males		
		MS		P	MS		P
Gut sugars							
Diet		0.04	0.9	0.35	0	0	0.95
Age		1.35	31.1	<0.0001	0.14	2.6	0.10
Diet × Age		0.40	9.1	0.003	0.21	4.0	0.047
Winglength		0.11	2.5	0.11	0.03	0.65	0.42
Body sugars							
Diet		0.03	1.0	0.32	0.01	1.0	0.33
Age		0.06	2.1	0.15	0.00	0.19	0.67
Diet × Age		0.01	0.50	0.47	0.01	0.36	0.55
Winglength		0.38	14.1	0.0003	0.21	11.91	0.0008
Glycogen							
Diet		0.22	1.2	0.28	0	0	0.98
Age		7.52	40.6	<0.0001	1.56	6.88	0.01
Diet × Age		4.26	23.0	<0.0001	1.11	4.9	0.029
Winglength		0.86	4.66	0.033	4.67	20.6	<0.0001

for the first 3 d of life and in males for the first 2 d of life (Fig. 2). Low levels of gut sugars were detected in 4–6 d old males, but no gut sugars could be detected in females within this age range (Fig. 2). The decline in gut sugars over time in parasitoids fed for only 1 d led to a significant interaction between the effects of age and diet treatment on fructose levels in both female and male *M. grandii* (Table 2). We could detect no effect of parasitoid size (as estimated by winglength) on gut sugar levels in either male or female *M. grandii* (Table 2).

Starved female and male *M. grandii* had low levels of body sugars, whereas parasitoids in both of the feeding treatments had relatively high levels of body sugars throughout their lives (Fig. 3). There were no significant effects of the diet treatment, age, or the interaction between these two variables on body sugar levels in female and male *M. grandii* from the two nonstarvation treatments (Table 2). However, a significant effect of winglength on body sugars was found for both males and females (Table 2).

Glycogen levels of starved female and male *M. grandii* were low over the entire observation period, but relatively high for individuals fed sucrose continuously (Fig. 4). The glycogen levels of females fed only on their first day of life dropped to near-zero levels by the end of the observation period (Fig. 4A), leading to a significant interaction of age and treatment on the glycogen levels (Table 2). The decrease in glycogen levels in males fed only 1 d was less pronounced, but still resulted in a significant age x diet interaction (Fig. 4B, Table 2). Winglength had a significant effect on glycogen levels for both female and male *M. grandii* (Table 2).

The average experiment-wide winglength was 3.58 ± 0.23 mm (SD; $N = 167$; range. 2.98–4.02 mm) for female *M. grandii* and 3.40 ± 0.22 (SD; $N = 178$; range: 2.86–3.94 mm) for male *M. grandii* ($t_{343} = 7.7$; $P < 0.0001$). We used linear regression analyses pooled over age for each of the diet treatments to characterize the effect of winglength on body sugar and glycogen levels (similar analyses were not done for gut sugars because there was no significant effect in the overall ANOVAs; Table 2). The relationship between winglength and body sugars was positive and significant for treatments in which sucrose was offered, but not significant for the starvation treatment (Table 3). A positive effect of winglength on glycogen levels was found in *M. grandii* that were starved or fed continuously, but not in individuals that were fed for 1 d (Table 3).

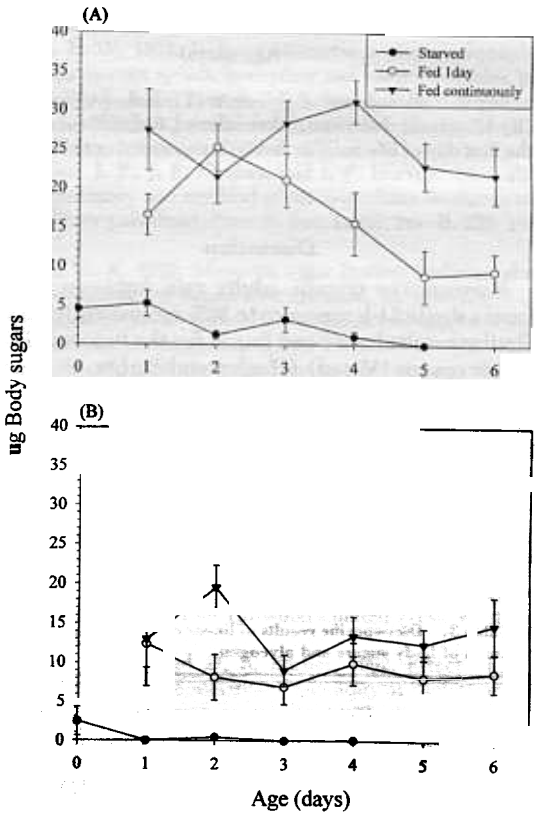

Fig. 3. Micrograms of body sugars of female (A) and male (B) *M. grandii* that were either starved, fed 50% sucrose for the first day of life only, or fed 50% sucrose over their entire life.

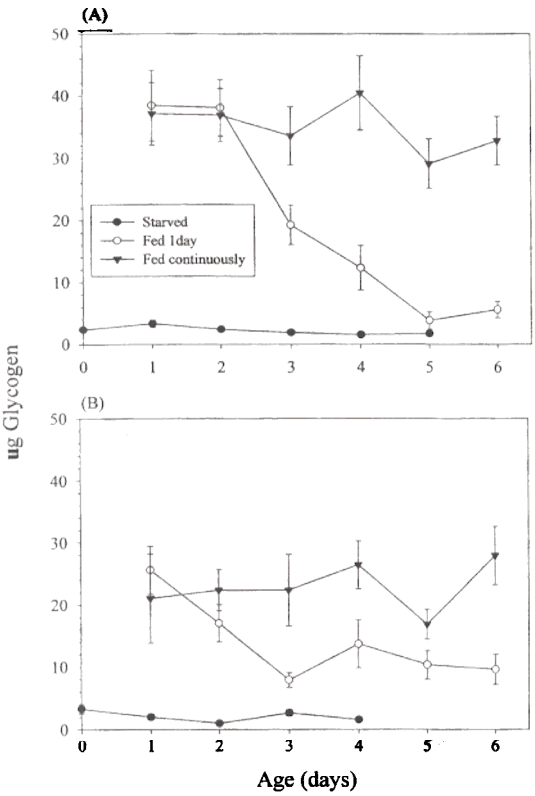

Fig. 4. Micrograms of glycogen of female (A) and male (B) *M. grandii* that were either starved, fed 50% sucrose for the first day of life only, or fed 50% sucrose over their entire life.

Discussion

Macrocentrus grandii adults gain between 2–4 d from a single 24-h exposure to 50% sucrose early in life. Similar results have been found for the braconid *Pholetesor ornigis* (Weed) (Hagley and Barber 1992) and the ichneumonid *Venturia canescens* (Grav.) (Fletcher et al. 1994). Maximum longevity can be attained by feeding on sugar every 2 d, but a feeding rate less than this leads to substantially decreased life expectancy. Still, sugar meals offered every 4 d increased life expectancy by over 8 d for both female and male *M. grandii*. These results suggest that *M. grandii* adults do not need to feed every day in the field to attain maximum longevity, and that even limited sugar availability can substantially increase life expectancy.

In addition to avoiding starvation, the ability to withstand periods of host-deprivation should allow parasitoids to allocate more time to searching for hosts or mates versus food (Lewis et al. 1998). A number of empirical studies have shown that female parasitoids switch from host-searching to food-searching after periods of sugar-deprivation (Lewis and Takasu 1990; Takasu and Lewis 1993, 1995; Wäckers 1994) and optimality models have identified hypothetical threshold levels of nutrient reserves below which females should switch from host- to food-searching (Sirot and Bernstein 1996; Clark and Mangel 2000). Lewis and Takasu (1990) showed that starved 2-d-old female *Microplitis croceipes* (Cresson) oriented toward odors associated with sugar, while 2-d-old females that had been fed a 20% sucrose solution for 5 min at the end of the first day of life oriented toward host-associated odors. Thus, it appears that the deprivation time that it takes to induce sugar- as opposed to host-searching is greater than 24 h for *M. croceipes*. We expect that the same would be true for *Macrocentrus grandii* since we could detect no decrease in survival from 24-h periods of sugar deprivation.

From a single day of feeding, *M. grandii* adults appear to achieve maximum levels of gut sugars, simple storage sugars ('body sugars'), and glycogen. All three of these forms of sugar remain high (i.e., indistinguishable from continuously-fed individuals) for the first day of starvation following the feeding period. This presumably explains the similarity in life expectancy for *M. grandii* fed continuously versus every 2 d. The combination of (1) no effect of parasitoid winglength on gut sugar levels, and (2) a positive effect of parasitoid size on storage sugars, is consistent with a relatively size-invariant gut volume and a higher rate of sugar intake in larger wasps. Alternatively, larger wasps may be more efficient at processing dietary sugar.

While gut sugars and glycogen dropped to relatively low levels over the 6-d observation period in parasitoids fed only on the first day of their life, body sugar levels remained relatively constant in this treatment. This pattern suggests mobilization of gut sugars and glycogen to body sugars. We presume that trehalose is the dominant sugar present in *M. grandii* 'body sugars'

Table 3. Diet-specific results of linear regression analyses testing for the effect of female and male *M. grandii* winglength on estimated amounts of body sugars and glycogen

Nutrient	Diet treatment	Females				Males			
		P	r^2	slope	intercept	*P*	r^2	slope	intercept
Body sugars	Starved	0.22				0.60			
	Fed 1 day	0.001	0.18			0.029	0.08		
	Fed continuously	0.031	0.08			0.01	0.10		
Glycogen	Starved	0.036	0.09			0.0004	0.22		
	Fed 1 day	0.23	—			0.23	—		
	Fed continuously	0.047	0.07			<0.0001	0.24		

Regression parameters are only reported if *P* for the slope parameter is <0.05.

because of the dominance of this sugar in insect hemolymph and other tissues (Wyatt 1967). The relative stability over time of this carbohydrate fraction in our study was probably a reflection of homeostatic regulation of trehalose (Friedman 1978, 1985). In *Manduca sexta* (L.) larvae, the regulation of trehalose levels is hormonally linked to glycogen breakdown: low blood sugar levels trigger the release of adipokinetic hormone (AKH), which activates glycogen phosphorylase, an enzyme that cleaves glucose residues from glycogen in preparation for trehalose synthesis (Gies et al. 1988; Chapman 1998). Patterns of glycogen and trehalose levels in some other larval and pupal Lepidoptera under conditions of starvation support this model (Horie 1960; Saito 1963; Wyatt 1967; Friedman 1978, 1985) as do data from adult boll weevils (Nettles et al. 1971). However, Ziegler (1991) found no evidence for AKH-regulated trehalose regulation in adult *M. sexta*, and he suggested that differences in life-histories between larval and adult *M. sexta* may have led to the evolution of trehalose regulation in larvae but not adults. Data consistent with trehalose regulation has been provided for continually fed adult cockroaches (Bowers and Friedman 1963) and mosquitoes during flight (Nayar and van Handel 1971) however. In starved adult mosquitoes, trehalose and glycogen levels decline together (Nayar and Sauerman 1975; van Handel 1984; Yuval et al. 1994), but appreciable levels of hemolymph sugars (primarily trehalose) can be detected for up to 10 d after feeding (Nayar and Sauerman 1975; van Handel 1984). Also, eclosion levels of hemolymph sugars stayed relatively constant for 48 h in *Aedes aegypti* (L.) (Naksathit et al. 1999a, 1999b). It is not clear from these studies whether trehalose regulation occurs in starving mosquitoes, but Foster (1995) stated that hemolymph trehalose levels of adult mosquitoes remain steady under unspecified conditions.

Our results support regulation of trehalose at the expense of glycogen in adult *M. grandii*, but the mechanisms of regulation remain to be identified. Keeping body sugar levels high at the expense of gut sugars and glycogen is presumably a strategy to maintain sugar resources in the most usable form under conditions of starvation. Although this mobilization strategy potentially contributes to lower mortality rates than would otherwise occur, the fact that mortality increases 2 and 3 d postfeeding despite unchanged levels of body sugars suggests that relatively high levels of body sugars are not sufficient to prevent mortality.

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