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Food Pulses Increase Longevity and Induce Cyclical Egg Production in Mediterranean Fruit Flies

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Abstract

1. Inasmuch as virtually all studies on mortality and reproduction in insects are conducted under conditions in which food availability is constant, little is known about the demographic response of insects to variable food environments. For example, it is not known if and to what extent the life expectancy of insects subjected to shortages of high-quality food will increase and/or whether this increase is associated with major decreases in lifetime reproduction.
2. Therefore cohorts of 100 individual female medflies were subjected to different sets of conditions of protein availability (interspersed with sugar-only diets) including *ad libitum* sugar-only (no protein), *ad libitum* protein and full (protein) diet either every 2nd, 4th, 6th, 11th, or 21st day, as well as two lag-treatments (1 day full diet followed by 30 days sugar-only, followed by one of two cyclical treatments).
3. Both life expectancy and lifetime reproduction were strongly affected by specific treatments. Specifically (i) mortality was inversely related to frequency of protein availability whereas lifetime

reproduction was directly related; (ii) distinct cycles in reproduction began to appear when food pulse cycles were as short as every 4 days. However, egg-laying peaks and troughs were particularly pronounced in the 10- and 20-day food pulse cycles; (iii) the peak and trough levels were inversely related to cycle length; and (iv) the within-cycle height was independent of cycle length, occurring 4 days after protein food was made available to the cohort whether the cycle length was 5, 10 or 20 days.

4. The results shed new light on the within- and between-cycle and lifetime dynamics of reproduction when insects are subjected to variable food environments and indicate that medfly females track food level very closely.

Keywords: food restriction, life span, life table, reproduction, variable environment

Introduction

Protein availability in nature is a fundamental determinant of individual fitness and population growth because amino acids (the basic ingredients of proteins) are required by individuals for both physiological maintenance and offspring production. Although the accessibility of protein-rich diets for most species in nature is highly variable, the vast majority of life-history studies have focused on the birth and death response of individuals that have constant access to *ad libitum* diets. Very few studies have been conducted in which access to dietary protein by individuals is pulsed. For example, it is not known whether the life expectancy of female fruit flies subjected to periodic pulses of protein-rich food increases relative to cohorts with unlimited access to protein-rich food, whether flies rapidly use the dietary protein to synthesize and lay new eggs but at the expense of little or no subsequent egg-laying, or whether their days of reduced egg-laying due to the absence of protein are recovered later when protein again becomes available. The reason answers to these questions are not known is that historically most investigations concerned with the relationship between food and birth and death rates have focused on either caloric restriction and/or food depression (Masoro 1988; Austad 1989; Lynch 1989; Masoro & Austad 1996; Sohal & Weindruch 1996; Weindruch 1996; Kirk 2001), reproductive physiology (Engelmann 1970; Boggs & Ross 1993), biodemography (Carey et al. 1998a,b,c; Sgro & Partridge 1999), cost of reproduction (Bell 1982, 1984; Reznick 1985; Bell & Koufopanou 1986; Partridge 1987; Roitberg 1989), life-history trade-offs (Jacome et al. 1995; Tatar & Carey 1995), and the effects of "egg load" on foraging behavior (Prokopy, Roitbert & Vargas 1994).

We believe that understanding the impact of variability in access to high-quality (high protein-content) food on birth and death rates is important for several reasons. First, conditions in which food availability fluctuates are more consistent with the conditions to which most animals are subjected in nature. Therefore the results of experiments involving variable food levels will provide new insights into the demographic response of animals reared under conditions that are more consistent with what they would experience in nature (Begon 1976; Bouletreau 1978; Webster et al. 1978; Drew, Courtice & Teakle 1983; Courtice & Drew 1984; Hendrichs et al. 1993). In particular, the results may reveal if and to what extent animals subjected to shortages of high-quality food can extend their lives.

Second, food regimes that vary with time will necessarily generate patterns of egg production that vary with age. Therefore analysis of both the local and lifetime dynamics of egg production and mortality in females denied access to protein-rich food sources part of the time will deepen understanding of the reproductive physiology (Engelmann 1970) and mortality dynamics of insects in general and fruit flies in particular. Third, earlier studies demonstrated that both food (Carey et al. 1998b) and host (Carey, Krainacker & Vargas 1986) deprivation in the Mediterranean Fruit Fly (*Ceratitis capitata* Wiedemann) will extend longevity. However, these and similar studies on *Drosophila* (Chippindale et al. 1993; Chippindale et al. 1997) were conducted using constant and not variable conditions. It is currently not known whether longevity is increased further with variable food availability as a result of a caloric restriction effect or whether it will be decreased due to increased stress and/or partial starvation. Fourth, one of the central questions in the host foraging ecology of adult insects involves trade-offs between "egg load" and foraging time (Roitberg 1989; Prokopy et al. 1994; Rosenheim 1996; Rivero-Lynch & Godfray 1997; Rosenheim 1999). Knowledge of the relationship of both egg production and survival to access to protein sources will shed light on this important area of population dynamics. Fifth, understanding the physiological or behavioral mechanisms that organisms use to buffer life histories against environmental variation is critical to understanding range shifts, population dynamics under global change scenarios, and impacts of introduced predators or hosts (Boggs 2002). Sixth, studies on the effects of diet on aging have a long history including healthy living (Willett 1994; Casper 1995) and caloric restriction (Masoro & Austad 1996; Sohal & Weindruch 1996; Weindruch 1996; McAdam & Millar 1999). Thus the results of investigations concerned with the effects on vital rates of variable food availability will complement the literature on the effects of diet on aging, shed light on the underlying mechanisms aging, and provide new insights into the dynamics of cost of reproduction.

The current study builds on previous investigations on the mortality (Carey et al. 1999, 2001) and reproductive (Carey et al. 1998b) response of the medfly subject to changing dietary conditions. We present the results of a study on the Mediterranean Fruit Fly designed to determine the impact of periodically alternating food quality (food pulses) on its reproductive dynamics and longevity. Three general questions are addressed: (1) Is the life expectancy increased in females that have periodic access to a full (protein-rich) diet? (2) What are the general dynamic properties of egg-laying in females subjected to regular pulses of protein-rich food? (3) To what extent does the inability to produce eggs at young ages due to the lack of dietary protein pre-empt and/or enhance egg-laying at older ages?

Methods

A total of 100 medfly females were subjected to one of nine different dietary regimes classified in one of three ways: (1) Cyclical. This group consisted of four treatments in which food was available to flies either 2nd, 4th, 6th, 11th or 21st days. These were coded as 1:1, 1:3, 1:5, 1:10, and 1:20, respectively (i.e., the second number indicates the number of days flies were provided sugar-only diet after the one day of full diet). (2) Cyclical-lag. This group consisted of two treatments in which flies were given 1 day of protein at eclosion followed by 30 days of sugar-only diet and then followed by either a 1:3 or a 1:10 pattern

of food availability. (3) Controls. Flies were monitored in three sets of controls including (i) *ad libitum* sugar-only (Control A); (ii) *ad libitum* full diets (Control B); and (iii) first day after eclosion flies were given full diet but maintained on sugar-only diet thereafter (Control C). Pairs of flies were housed in $6.5 \times 6.5 \times 12$ cm³ clear plastic containers. The full adult diet consisted of yeast hydrolysate and pure sucrose (1:3 ratio by volume) whereas the sugar-only diet consisted of pure sucrose only. The yeast hydrolysate (ICN Biomedicals Inc., Costa Mesa, California, USA) contained 60% protein along with vitamins and minerals (Vargas et al. 1997).

Results

Descriptive Statistics

Summary statistics for the reproductive and longevity responses of female medflies subjected to the seven treatments and three control conditions are presented in tables 1 and 2. The results merit several comments. First, it is clear that dietary pattern has a profound influence on longevity. Life expectancy at eclosion among the treatments differed by 35% or nearly 13 days ranging from 37.0 days in the 1:1 cyclical treatment to nearly 50 days in the 1:30–1:3 lag treatment. Interestingly neither the shortest nor the greatest life expectancy was observed in the controls. This suggests that, with respect to survival, constancy of either protein availability or sugar-only alone is better under some circumstances but worse under others. Second, proportionally reproduction was affected to a much greater degree by dietary pattern than was life expectancy. For example, net reproduction ranged from slightly over 50 eggs/female in the sugar-only controls (Control A) to slightly less than 900 eggs/female in the *ad libitum*, full diet control (Control B). Clearly, reproduction is far more sensitive to fluctuations in the availability of protein-rich food than is life expectancy. Third, flies in the two treatments with the lowest life expectancy (<40 days for full diet Control B and the 1:1 treatment) experienced the highest lifetime reproduction. Gross reproduction was only 30% higher than net reproduction for the *ad libitum* control but 70% higher for the 1:1 treatment. This indicates that many more flies died in the 1:1 treatment that were still capable of producing a substantial number of eggs than those in the *ad libitum* controls. In other words, rate of reproductive senescence was more closely linked to somatic senescence in the *ad libitum* treatment than the cyclical treatment. Fourth, life expectancy was greatest for treatments in which females were subjected to long periods of sugar-only diet and lowest for treatments in which females had access to a short period of sugar-only diet. For example, the longest-lived flies were those subjected to 1 day of protein followed by 30 days of sugar-only diet and then followed by a 1:3 cycle (i.e., the 1:30–1:3 treatment). Generally speaking the demographic summary measures reveal that reproduction increases with increasing access to a full diet, life expectancy increases with decreasing access to a full diet, and the longevity trade-offs with reproduction are complex due to the disparity between the rate of aging of the reproductive system and the rate of aging in the fly as a whole.

Table 1. Life expectancy, e_0 (days), and lifetime reproduction (eggs/female) for medfly cohorts subject to different food cycles. SD denotes standard deviation, and CI (L) and CI (U) denote lower and upper confidence intervals, respectively. Codes indicate ratio of days in which flies were given access to a full diet (sugar + yeast hydrolysate) relative to days with sugar-only (see Methods).

Treatment	Life expectancy		Reproduction		
	e_0	SD	Mean	95% CI (L)	95% CI (U)
Controls					
Control A (sugar)	44.1	26.83	65.0	53.5	76.5
Control B (full)	39.7	13.94	914.4	817.2	1,011.6
Control C (1:0)	46.8	30.64	762.0	664.7	859.3
Cyclical treatments					
1:1	37.0	17.56	532.0	453.7	610.4
1:3	43.6	34.57	508.5	444.9	572.0
1:5	47.5	25.28	317.4	272.9	361.8
1:10	49.6	28.73	182.5	154.5	210.5
1:20	47.9	33.22	332.0	275.7	388.2
Lag treatments					
1:30–1:3	49.9	31.44	220.9	181.9	259.8
1:30–1:10	47.6	31.00	136.7	120.4	152.9

Table 2. Mean and standard error for first egg peak and its location (age)

Treatment	Number eggs		Age	
	Mean	SE	Mean	SE
Controls				
Control A (sugar)	33.8	2.110	6.0	0.785
Control B (full)	41.2	1.369	10.33	0.891
Control C (1:0)	39.8	1.588	11.09	1.136
Cyclical treatments				
1:1	45.4	2.189	8.63	0.908
1:3	49.7	2.415	6.89	0.609
1:5	49.3	2.813	6.46	0.888
1:10	44.1	3.107	5.97	0.842
1:20	50.8	3.000	8.31	0.848
Lag treatments				
1:30–1:3	47.3	3.137	7.80	1.246
1:30–1:10	36.6	2.331	6.35	0.883

Cohort Survival

Patterns of cohort survival (l_x) differed across treatments shown in figure 1a–d. The 4-day difference in life expectancy for females in Control A versus Control B (table 1) was due to age differences in the patterns of survival—the survival rate of young flies maintained on *ad libitum* full diet was high initially but then rapidly declined after 1 month (fig. 1a). In contrast, survival of flies given access to a sugar-only diet was nearly linear through the last fly's death. Differences existed between survival patterns of flies subjected to the different cycle treatments. For example, female survival among treatments where flies were given access to a full diet only on either the 2nd, 4th, or 6th days (treatments 1:1, 1:3, and 1:5, respectively) was similar through day 20 (fig. 1b) but diverged thereafter. Survival after day 20 was inversely related to the frequency flies had access to a full diet—flies with more frequent access to a full diet died more quickly (treatment 1:1) whereas flies with less frequent access to a full diet died off more slowly (treatments 1:3 and 1:5). Differences in female medfly survival for treatments with long full-diet intervals were relatively small as shown in figure 1c for treatments in which flies had access to a full diet either every 11th day (treatment 1:10) or every 21st day (treatment 1:20). Similarly differences in survival among flies subjected to the lag treatments shown in figure 1d were also relatively minor. In general, it appears that survival patterns of medfly subjected to different cycles of protein availability were mediated primarily by qualitative (switch on or off) rather than quantitative (egg production levels) reproductive efforts.

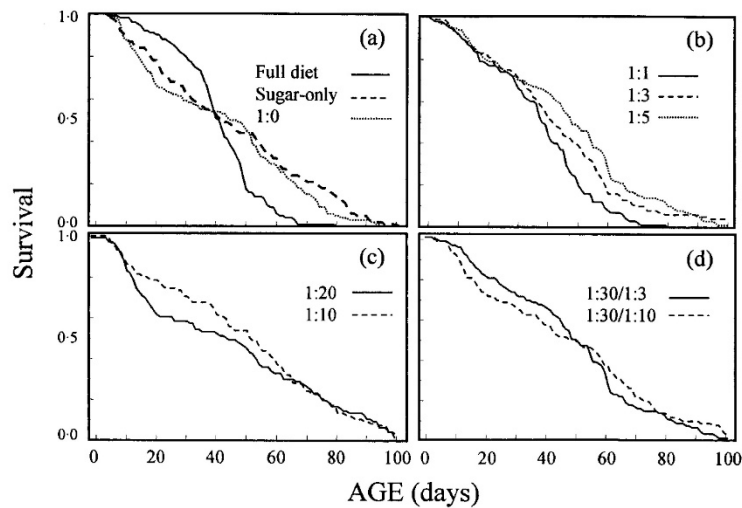


Figure 1. Survival (l_x) schedules for flies in each of the seven treatments and three controls: (a) three control cohorts including *ad libitum* full diet, sugar-only diet, and first day full diet followed by sugar-only diet for remainder of cohort life (1:0); (b) three short-cycle treatments including full diet every 2nd, 4th, and 6th day for treatments 1:1, 1:3, and 1:5, respectively; (c) two long-cycle treatments including full diet every 11th and 21st day for treatments 1:10 and 1:20, respectively; (d) two lag-cycle treatments including full diet on first day followed by 29 days of sugar-only diet followed, in turn by either full diet every 4th or every 11th day.

Age Patterns of Reproduction

The age-specific egg production in the cohort of flies in the *ad libitum* full diet treatment (fig. 2a) was nearly indistinguishable from the daily pattern of egg production in the cohort of flies that had access to full diet only on alternate days, treatment 1:1 (fig. 2b). In other words, despite medfly females having access to a full diet half as many days in the 1:1 treatment relative to the *ad libitum* control, females were still capable of producing eggs at virtually the same rate as flies with access to a full diet 100% of the time. This result shows that medflies are capable of compensating for the absence of food for 24-h periods with very little loss of lifetime productivity. One reason for the nearly identical lifetime egg production between the *ad libitum* control and the 1:1 treatment was that flies in the 1:1 treatment laid slightly fewer eggs at younger ages but compensated for this slight decrease by laying more eggs at older ages.

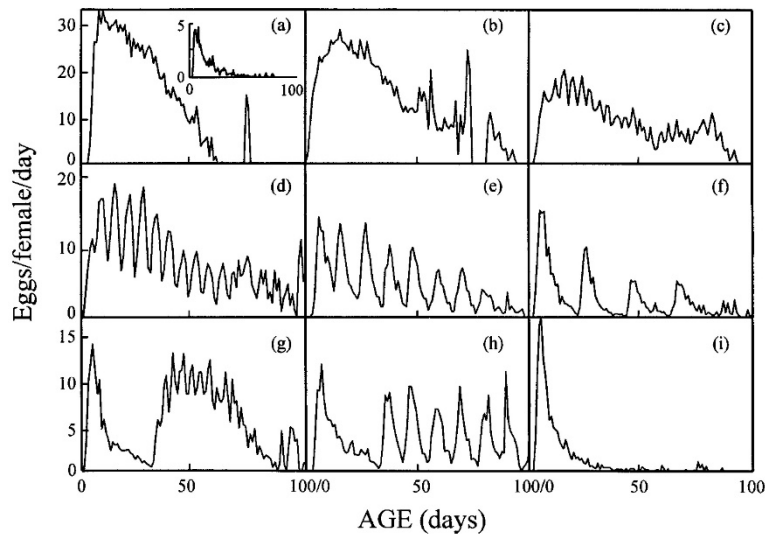


Figure 2. Average number of eggs/female/day in the medfly study consisting of seven treatments and three control cohorts: (a) *ad libitum* full diet (inset—sugar-only diet); (b) full diet every other day (1:1); (c) full diet every 4th day (1:3); (d) full diet every 6th day (1:5); (e) full diet every 11th day (1:10); (f) full diet every 21st day (1:20); (g) full diet on first day followed by sugar-only diet for 29 days, which was then followed by full diet every 4th day; (h) same as g except full diet every 11th day starting on day 30; (i) full diet on first day followed by sugar-only diet.

Distinct peaks and troughs in egg production begin to appear in the overall reproductive schedule when flies are denied access to full diet for 3 or more days with both the peaks and the troughs directly related to the length of the period. This is evident in figure 2c–f which shows the reproductive rates for flies subjected to the 1:3, 1:5, 1:10, and 1:20 treatments, respectively. Note that the peaks and troughs vary by only around 5 eggs per day in the shortest cycle treatment (3 days without full diet) but by up to 15 eggs per day

at young ages in the longest cycle treatment (20 days without full diet). The cycle highs and lows in egg production generally decreased with age. Also cohorts of flies that were denied access to a full diet at younger ages were capable of producing eggs at much older ages than are flies fed *ad libitum* (fig. 1a) or fed a full diet on alternate days (fig. 2b).

The reproductive schedules of females subjected to the lag cycles shown in figure 2g–h reinforce the observation in other treatments that flies denied access to a full diet at younger ages are capable of producing substantial numbers of eggs at older ages relative to *ad libitum* flies. For example, the egg production rates of the female cohorts at ages beyond 50 days shown in figure 2g–h are substantially higher than egg production rates of the females subjected to similar treatment cycles but which were not denied access to a full diet at young ages (fig. 2c,e). The egg production rate did not fall to zero until females were denied access to a full diet for at least 20 days (fig. 2f). This suggests that medfly females have adopted a reproductive strategy in which they retain the ability to produce at least a few eggs over long periods when they do not have access to dietary protein.

The data on daily reproduction for individual medfly females were used to construct event history diagrams shown in figure 3a–i. These diagrams are useful because they provide insights into the within-cohort variation in age patterns of egg-laying for the flies for each of the treatments. The charts reveal that the reproduction of flies denied access to a full diet part of the time was lower than that for flies with unlimited access to a full diet by reducing both the average daily egg output (particularly the high egg-laying days) and by increasing the frequency of zero egg-laying days. For example, females with the highest lifetime egg production were maintained on the *ad libitum* full diet (Control B). These females produced at least some eggs over 72% of the time and laid in excess of 30 eggs per day over 33% of the time. In contrast, the females with substantially lower lifetime egg production were those maintained on the 1:5, 1:10, and 1:20 treatments which produced one or more eggs per day 58, 48, and 33% of the time, respectively, and produced over 30 eggs per day 6.8, 3.4, and 2.5% of the time, respectively. It thus appears that the reproductive strategy for medflies under conditions of food scarcity is to produce a small number of eggs over a sustained period rather than to produce a large number of eggs over an abbreviated period, *ceteris paribus*.

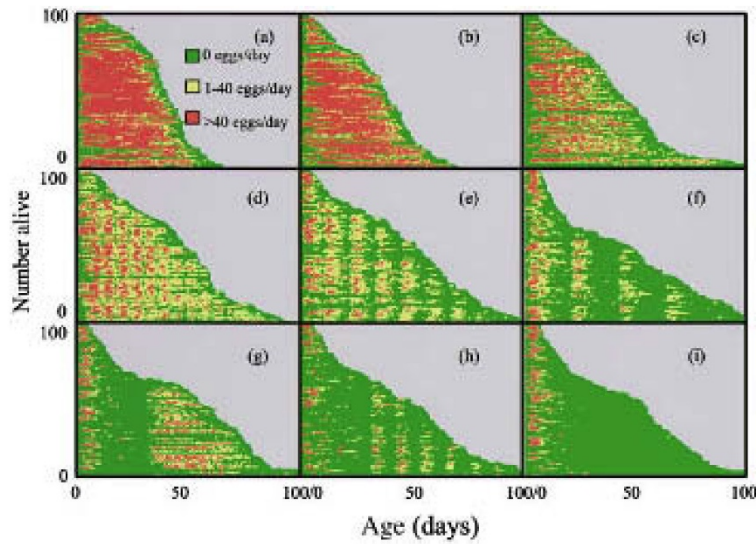


Figure 3. Event history graphs of individual female reproduction in each of the 10 treatments. Figure 3(a)–(i) same as figure 2 labels. Each individual female within a treatment is represented by a horizontal “line” proportional to her life span (Carey et al. 1998c). Each day of a female is color-coded according to whether she laid zero (green), 1–40 eggs (yellow), or greater than 40 eggs (red).

Within-Cycle Reproductive Patterns

Composite within-cycle schedules of egg production in medfly females for selected treatments were constructed by averaging the per capita fecundity for each day of the cycle over all ages. These composite schedules thus represent the daily pattern of egg production of a hypothetical female who lived to the last day of possible life. These schedules are the within-cycle equivalent of the gross reproductive rate in classical demography—the number of eggs a hypothetical female would lay in her lifetime if she lived to the last day of possible life. Three of these hypothetical schedules are shown in figure 4 (for the treatments 1:5, 1:10 and 1:20) and reveal several important patterns regarding the reproductive dynamics of medflies in general and the relationship between protein availability and egg production in particular. First, in all three treatments the maximum number of eggs was laid an average of exactly 4 days after females were given access to a full diet. The same time lag between when females had access to a new source of dietary protein and their maximal egg production flies independent of the cycle length suggests that the rate of production of new eggs is regulated solely by when new protein becomes available. Second, in contrast to the timing of maximal egg production relative to the availability of dietary protein, the egg production level is dependent upon the cycle length. This is evident in the differences in the peak heights between treatments. For example, peak egg production in the 1:5 treatment was greatest at 13.1 eggs/female after 4 days but in the other two treatments peak egg production was less than 11 eggs/female. This suggests that a female’s current blood protein level determines whether she has enough reserves to immediately

synthesize proteinaceous and lipid yolk for egg maturation (Engelmann 1970). Third, the rate of increase in egg production depends on the level of egg production on the day when females are given access to new protein. For example, egg production in the 1:5 treatment increased by only 3.4 eggs per 4 days from food-day to peak whereas egg production in the 1:10 and 1:20 treatments increased by 9.3 and 9.6 eggs per 4 days, respectively. Also note that egg production began increasing immediately after the flies had access to new protein. Fourth, the number of eggs per "food-cycle" differs by only around 20% between the 6-day (1:5) cycle and the 21-day (1:20) cycle suggesting that both short-term and lifetime fecundity is more food rather than time-limited, *ceteris paribus*. That is, females subjected to the 6-day dietary cycle produced 60 eggs per cycle but females subjected to a cycle over 3-fold greater produced only 12 eggs more.

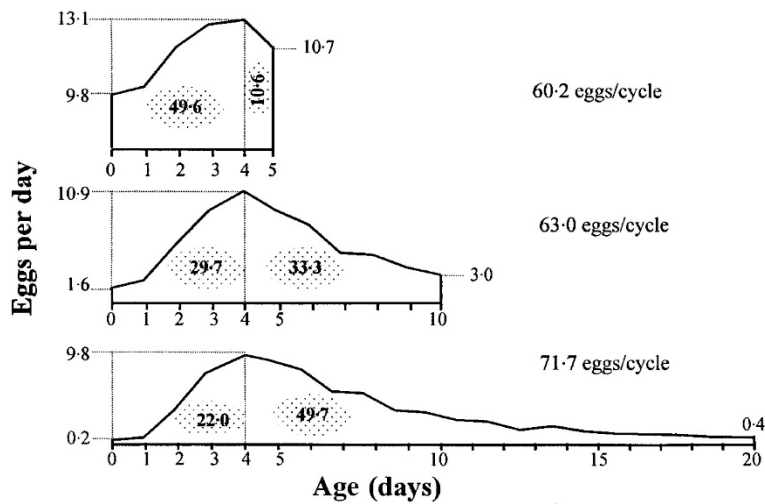


Figure 4. Patterns of egg production for composite egg-laying cycles averaged over the life course of all flies. These composites correspond to a within-cycle, gross reproductive rate—the average number of eggs laid per day (within cycle) for a female who lives to the last day of possible life. Note that the number of eggs at the start of the cycle and its peak are given for each treatment on the left of the schematic and the number of eggs at the end of the cycle are given on the right of the schematic. The average total number of eggs both before and after the peak are contained within each graph and the total eggs per cycle is given to the right of each schematic.

Modeling Relationships and Graphical Analysis

We examined the dynamic relationships between feeding regimen, egg production, peak egg-laying, age at peak egg-laying, and longevity using several statistical regression models, including a linear model fitted by least squares with lifetime number of eggs as response, a Poisson regression model fitted by maximum likelihood with lifetime (age-at-

death) as response, and a variety of non-parametric regression fits to describe the relationships between pairs of variables.

Model 1

This model relates the total number of eggs (EggNo) as response variable with the predictors lifetime (age-at-death), number of eggs at peak egg production (peak-level), age at peak egg-laying (peakage), and eight treatments. The eight treatments considered in model 1 are coded by seven treatment indicator variables, assuming the values 0 or 1, as follows, where the designations for the treatments are the same as those used in table 1: $\text{tr}(1:1) = 1$ for the 1:1 cyclical diet (full diet on alternate days), $\text{tr}(1:3) = 1$ for the 1:3 cyclical diet, $\text{tr}(1:5) = 1$ for the 1:5 cyclical diet, $\text{tr}(1:10) = 1$ for the 1:10 cyclical diet, $\text{tr}(1:20) = 1$ for the 1:20 cyclical diet, $\text{tr}(C) = 1$ for Control C (1-day protein, then all other days sugar), and $\text{tr}(A) = 1$ for Control A (sugar only). Since the egg-laying patterns for the treatments with switching diets, the lag treatments of table 1, were quite different, the two lag treatments were not included in this model. If the fly is under a specific treatment, the respective treatment indicator is = 1 while all other treatment indicators are = 0. The case where all treatment indicators are zero corresponds to the baseline treatment which is Control B (*ad libitum* full diet).

The fitted model involves a log transformation of the response and various transformations of the predictors. As a main criterion for model selection and variable transformations the Akaike information criterion (AIC) was used (McCullagh & Nelder 1986). For variable transformations, we investigated Box-Cox transformations and also checked for interactions between the predictors that were not included in the final model. The residuals of the model were reasonably normal. The equation of the fitted model is given by

$$\begin{aligned} \text{EggNo} = \{ & \exp[1.96 + 0.0076^* \times \text{lifetime} + 0.1537^* \times \\ & \log(\text{peak-level}) - 0.1363^* \times \text{sqrt}(\text{peakage}) + \\ & 0.061 \times \text{tr}(1:1) - 0.1277^* \times \text{tr}(1:3) - \\ & 0.2060^* \times \text{tr}(1:5) - 0.4188^* \times \text{tr}(1:10) - \\ & 0.6418^* \times \text{tr}(1:20) - 0.6884^* \times \text{tr}(C) - \\ & 0.8673^* \times \text{tr}(A)] \}^{8/3}. \end{aligned}$$

eqn 1

(The asterisk (*) indicates that the corresponding predictor has a significant effect on the response, $P < 0.05$.)

This model reveals three important relationships. First, with the exception of the 1:1 cyclical treatment (full diet on alternate days), which is almost indistinguishable from the baseline *ad libitum* diet in terms of the effect on lifetime number of eggs, all of the treatments led to significantly different amounts of total eggs as compared to the baseline control B treatment (*ad libitum* full diet). This is seen as the corresponding treatment indicator variables that are predictors in the model have significant regression coefficients. Second, again ignoring the 1:1 cyclical treatment, there exists a monotonic dose-response relationship in that the number of eggs laid increases in a monotonic fashion with the frequency of protein days. In other words, the more frequent the availability of full diet is, the greater

is the number of total eggs produced. While increased protein in the diet is also related to shortened longevity with an associated shortening of the egg-laying period, this finding implies that this shortening of the egg-laying period is more than compensated for by an increase in the intensity of egg-laying. Third, the significance of the coefficients for lifetime (age-at-death) and for both the peak height and the location (age) of the egg production peak indicate that early and high peaks of egg production and a long female lifetime are positively related to lifetime egg production. Highly productive egg-layers can therefore be identified early in life, in agreement with the findings in Müller et al. (2001).

Model 2

Lifetime (age-at-death) as response was related to treatment and peak characteristics (it is important to note that all of these variables are available at an early age of the fly) as predictors. We found that a Poisson regression model which is a special case of the generalized linear model (McCullagh & Nelder 1986), using the log link function, was adequate for modeling this regression relationship. The predictors are the seven treatment indicators as defined in model 1, plus two indicators for the two lag treatments, $\text{tr}(1:30-1:3) = 1$ if the fly received the lag treatment 1:30–1:3, $\text{tr}(1:30-1:10) = 1$ if the fly received the lag treatment 1:30–1:10 (see table 1). As in model 1, the baseline treatment when all indicators are zero is Control B (*ad libitum* full diet). The predictors for the peak characteristics are again peak-level and peakage, as described in model 1. The equation of the fitted model is

$$\begin{aligned} \text{Lifetime} = & \exp(3.6651 - 0.0764 \times \text{tr}(1:1) + \\ & 0.0534 \times \text{tr}(1:3) + 0.1681 \times \text{tr}(1:5) + \\ & 0.2041 \times \text{tr}(1:10) + 0.1537 \times \text{tr}(1:20) + \\ & 0.1403 \times \text{tr}(\text{C}) - 0.01058 \times \text{tr}(\text{A}) + \\ & 0.2116 \times \text{tr}(1:30 - 1:3) + \\ & 0.1145 \times \text{tr}(1:30 - 1:10) - \\ & 0.00192 \text{ peak-level} + \\ & 0.01837 \times \text{peakage}). \end{aligned}$$

eqn 2

All of the coefficients in this model turn out to be significant, $P < 0.05$. The main inference to be drawn is that longevity is reduced when the peak location of egg-laying is earlier and the peak is higher. This is consistent with the general concept of reproductive tradeoffs with mortality: early egg-laying tends to reduce later egg-laying and to reduce longevity. Also, less frequent protein days tend to enhance longevity, with two notable exceptions: full diet every other day, the 1:1 cyclical diet, and the sugar-only diet Control A are associated with a decrease in longevity. The maximum increase in longevity is obtained for the cyclical 1:10 treatment.

Graphical displays of the egg-laying data plotted against female lifetime (fig. 5a), peak size (fig. 5b), and peak location (fig. 5c) help to visualize the modeling results and reveal other important relationships. The significance of the relationships was established in models 1 and 2. In these graphs non-parametric regression fits are used to assess the shape

of the relationship which in most cases is non-linear. The non-parametric regression estimates allow nearly arbitrary shapes for the regression function and are therefore very useful for modeling non-linear trends which cannot be easily parameterized (Müller 1987).

Several aspects of these graphs merit comment. First, lifetime egg production is positively correlated with female lifetime but only up to a certain age at which point no correlation exists. The initial correlation is the result of limits on daily egg production constraining lifetime production—females must live several weeks before they have time to lay a substantial complement of eggs. The lack of correlation of lifetime egg production and longevity at high female lifetimes indicates that lifetime egg production may ultimately be limited by an endowment concerning the potential eggs a female is capable of producing in her lifetime. Second, the relationship between both the size of the initial egg peak and its location (age) are shown in figure 5b,c, respectively. These graphs reveal that the size of the egg peak is positively associated with lifetime egg production whereas the location is negatively associated with lifetime egg production; in other words, the older the age at which the initial peak occurs, the lower the number of lifetime eggs produced.

Discussion and implications

The current studies were designed to both complement and build on the results of previous medfly demographic research including baseline studies on birth and death rates (Carey 1984; Carey, Yang & Foote 1988, Carey et al. 1998a,c; Vargas & Carey 1989; Vargas et al. 1997), experimental investigations in which conditions of food or host availability are manipulated (Carey et al. 1986, 1998b, 1999; Krainacker, Carey & Vargas 1987), and modeling cost of reproduction (Müller et al. 2001). None of these previous studies provided insights into the life-history response of medfly females to cyclical environmental perturbations at the level of the individual. That is, comparative studies in which daily reproduction of individual females was monitored for a wide variety of treatments over their entire lifetime have not been done previously.

We believe that the life-history response of individual females to these different patterns of environmental manipulations sheds important new light on the reproductive dynamics of medflies in particular and insects in general. First, the current findings shed important light on the dynamics of insect reproduction including the strategic trade-offs between reproduction and survival in the medfly—how females retain amino acids in their system as “common currency.” Specifically the within-cycle patterns of reproduction suggest that the reproductive strategy of medfly females with respect to protein allocation involves three levels: (1) a fraction of amino acids are always held in reserve in the fat bodies and haemolymph rather than used for the immediate production of yolk protein; (2) even though flies may not have access to a new protein source, a fraction of this reserve is allocated daily to vitellogenesis; (3) new protein food stimulates flies to allocate a greater fraction of the amino acids reserve for egg production than would be allocated if no new protein food was available.

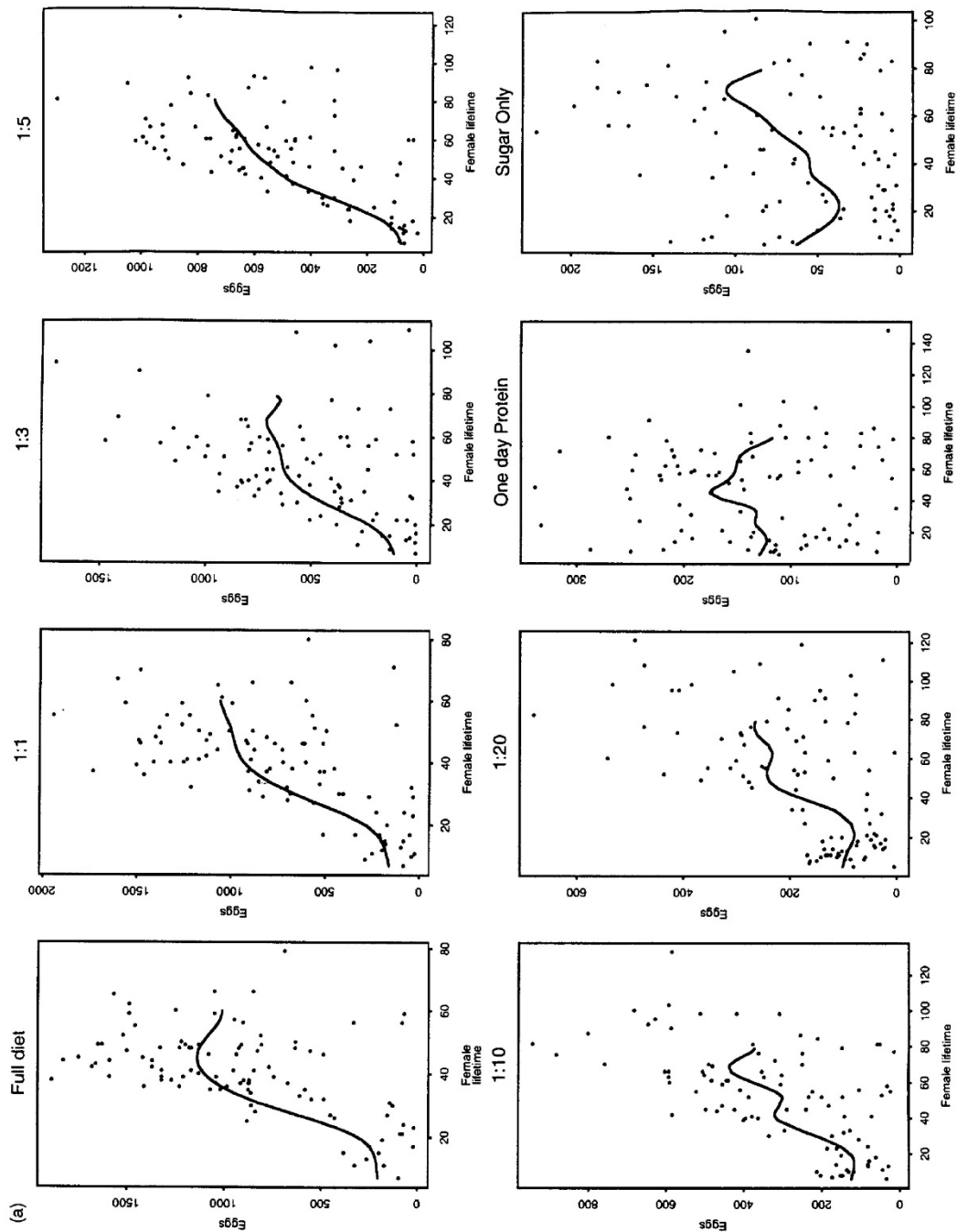


Figure 5. Relationship of lifetime egg production (response) and the predictors (a) female longevity, (b) age of initial peak of egg production, and (c) the height of the initial egg-laying peak. For each of these relations, a scatterplot is shown, indicating the observations made on each individual fly. The graph of a non-parametric regression fit is superimposed, indicating that these relationships are highly non-linear.

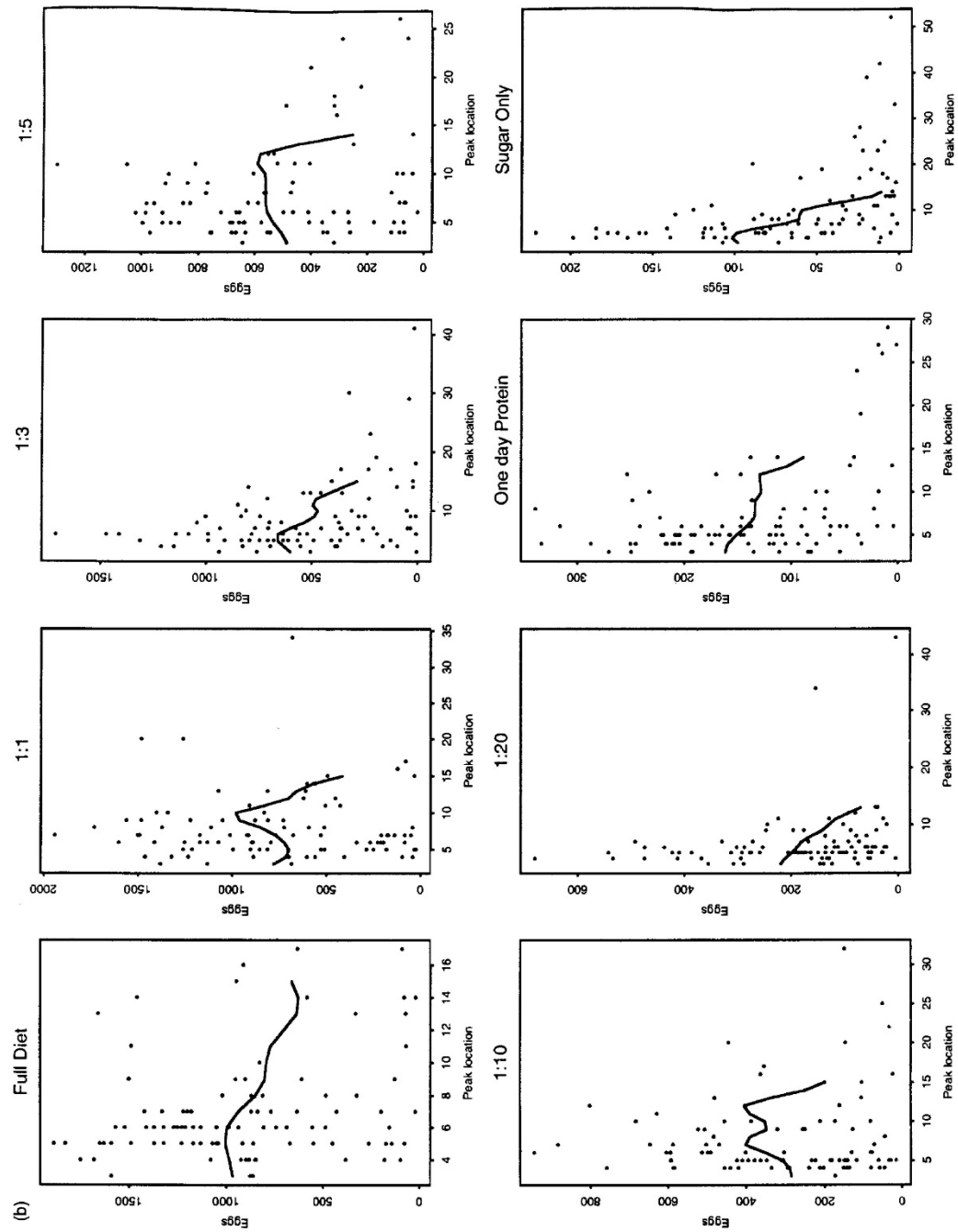


Figure 5. *Continued*

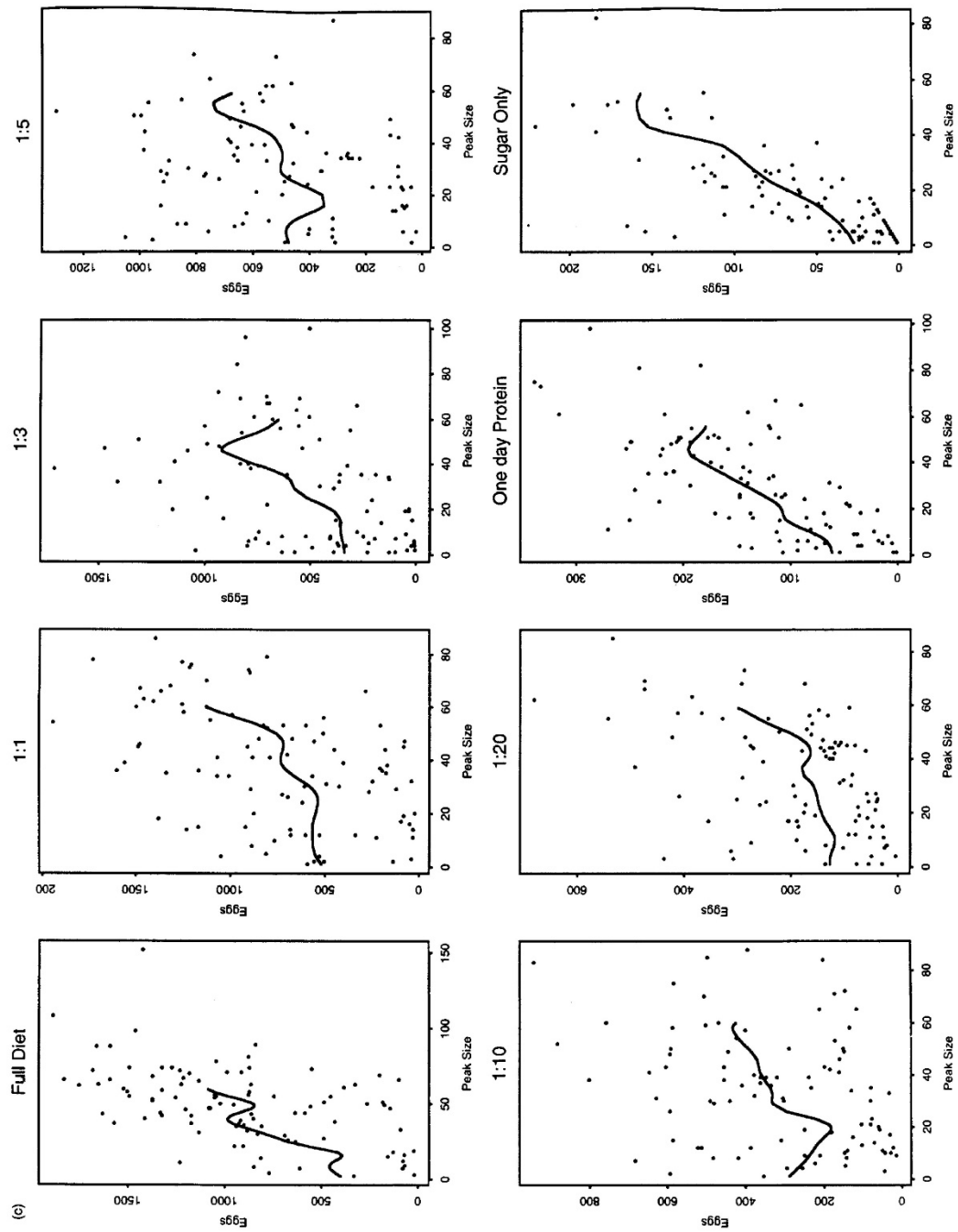


Figure 5. *Continued*

Second, our results demonstrate that the cost of reproduction has at least two different components, both of which compromise lifetime reproductive success: (1) risk of premature death which will prevent all reproduction at older ages, and (2) loss of potential eggs due to an apparent age-related degradation of oögonia (potential eggs). In other words, the level of reproduction at older ages in flies that laid few eggs at younger ages is higher than reproduction in flies that realized their full reproductive potential at younger ages. However, as a female ages, a fraction of her potential reproduction is completely lost due to aging alone, even if she manages to live to an extreme age. This same response to food manipulations was also observed when medfly females were deprived of ovipositional hosts (Carey et al. 1986).

Third, although the food restriction methods used in the current study with medflies (i.e., high-quality food either available or not available on a given day) differ from the food restriction methods used in mammalian caloric restriction studies (i.e., daily access to reduced amounts), the general response was similar to rodent studies (Masoro 1988; Masoro & Austad 1996)—extended life expectancies and reduced reproduction relative to *ad libitum* controls. Therefore we believe that the hypothesis introduced by Holliday (1989) regarding the evolutionary response of animals to reduced food availability is relevant to our medfly studies. Holliday noted that from all the experimental studies that have been carried out in which animals are maintained on a near-starvation diet, breeding is prevented during this period. However, breeding will be resumed when food supply becomes plentiful and females will retain the capacity to breed at significantly later ages than animals with a high-calorie diet throughout their life. These patterns led Holliday to postulate that Darwinian fitness will be increased in mammals and (by extension) other groups of organisms if animals cease breeding during periods of food deprivation and invest saved resources in maintenance. This would increase the probability of producing viable offspring during an extended life span.

Fourth, a great deal of interest in reproductive dynamics exists because egg load influences host foraging by parasitoids in natural populations (Casas et al. 2000), determines stability properties of host-parasitoid models (Rosenheim 1996; Rosenheim 1999), and affects the host finding behavior in a wide range of herbivorous insects (Prokopy et al. 1994). The results of the current studies suggest that egg load—the number of mature eggs present in a female at a given time—does not necessarily reflect the lifetime reproductive strategy. Rather egg load is likely determined by a combination of both feeding (i.e., access to dietary protein) and egg-laying history.

Conclusions

With a few notable exceptions (Boggs & Ross 1993; Chapman, Trevitt & Partridge 1994), surprisingly little research has been previously conducted on the reproductive response of insects to variable food environments. This study provides new insights into how insects respond to variable nutritional environment including their cyclical reproductive response to pulses of protein-rich food, their increased longevity in response to reduced reproductive effort, and their lifetime patterns of egg production relative to longevity and level and age of peak egg production.

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