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Aging and Extended Longevity in Wild Medfly Populations

by

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ABSTRACT

Age structure and longevity in populations of wild medflies in Greece were estimated using a new methodology that we refer to as the ‘captive cohort model’. The underlying concept is that the age structure of the wild population from which flies are captured and subsequently monitored in the laboratory and, by extension, the ages of the longest-lived individuals at the time of their capture, can be deduced from their distribution of deaths in the laboratory. Biodemographic data (birth and death rates) were gathered in Chios, Greece on approximately 1,000 wild flies (captive cohorts) captured on 1-of-16 sampling dates during the 2003-04 field seasons and from a (reference) cohort of 134 medfly females reared from infested hosts. Analysis and modeling of these data combined with results of previous studies on the field ecology of the medfly contradicts the conventional wisdom that the survival of most insects is low and supports the hypothesis that medflies are long-lived in nature. Implications for demographic and gerontological research on aging in the wild regarding both the methodology and the empirical results are discussed.

INTRODUCTION

Understanding aging in the wild is one of the most important problems in gerontology because empirical data derived from field studies can be used to frame and test theories of aging, inform research concerned with aging mechanisms, and establish baselines for the natural history of aging. Using a simple model that incorporates laboratory survival rates from both *wild-caught* individuals of *unknown* age and *wild-type* individuals of *known* age, we show that wild populations of the Mediterranean fruit fly (*Ceratitis capitata*), commonly known as the medfly, experience major shifts in age structure of both sexes and contain a substantial number of individuals that have survived to relatively advanced ages. The underlying concept is that the age structure of the wild population from which flies were captured and subsequently monitored in the laboratory and, by extension, the ages of the longest-lived individuals at the time of their capture, can be deduced from their distribution of deaths in the laboratory.

The study was motivated by questions regarding the relative abundance of older individuals in field populations of medflies and, in turn, the need to develop a method to estimate age structure in field populations of insects. Referred to as the *captive cohort method*, this technique eliminates the need to mark, release, and recapture individuals in the wild as do other ecological methods (Caughley, 1977; Krebs, 1999) or to estimate the age of the individual. The approach is based on the assumption that survival differences in captive cohorts are due to different age distributions of captured flies with early deaths being attributable mostly to flies that are older and later deaths attributable mostly to flies that were young when captured. Estimates are independent of the relationship between captive and wild environments but rather depend on the assumption that death rate of

captured flies will be the same as wild flies of known age maintained under same laboratory conditions.

The investigation consisted of three components including: (1) the capture and transfer of wild flies from the field to individual cages in the laboratory where the date of death was recorded for both sexes and daily reproduction was recorded for females. The data gathered from these *captive cohorts* was used to construct *captive life tables* indexed according to the *captive age* (number of days from date of capture); (2) creation of wild-type *reference cohorts* in which wild adult flies were reared from infested hosts collected in the field and their age-specific reproduction and life spans recorded in the laboratory; and (3) data analysis, modeling and age structure estimation using the distributions of births and deaths in both the captive and reference cohorts to answer the question: What is the age distribution of the wild population that will yield the distribution of deaths observed in the captive cohort?

METHODS

Empirical

Live adult medflies of both sexes were collected on the Greek island of Chios starting in July in 2003 and in June in 2004 and continuing through November both years. The collections were made using a total of 30 McPhail traps (McPhail, 1939) baited with food attractants (ammonium acetate; trimethylamine) installed in 10 trapping locations (3 traps/location) in two different citrus orchards 2 km apart. The flies captured within one day were placed alive in separated cages for each trapping station and air-shipped for same-day arrival to the University of Thessaloniki where they were placed individually in 3x6x6 cm cages with water, adult food, and oviposition devices (perforated hollow domes

in Petri dish lid with water and fresh orange juice). Survival of both sexes was monitored daily and the number of eggs laid by individual females was recorded each day from time of capture through their death. A total of 6 collections were made in 2003 (6/8; 8/28; 9/18; 10/9; 11/3 and 11/27) with a total capture of 578 individuals (358 females and 220 males) and a total of 11 collections were made in 2004 (15-day intervals from June through November) with total capture of around 700 individuals. From mid September through mid November wild alive flies were captured in a daily basis from an apple orchard in Thessaloniki, using the same technique as in Chios, and sent to the University of Volos for life span determination. Individual flies were placed in small transparent cages consisting of two plastic containers (9.5cm high, 7.0 cm upper diameter and 5.0 cm base diameter) that fitted into one another. The upper container housed the flies, food and a single dome fitted in a hole at the bottom. The space between the two bottoms of the containers contained water that was made available to the flies by a cotton wick.

To construct reference life tables, medfly pupae were obtained from infested figs (*Ficus carica*) and mandarin oranges (*Citrus aurantium*) in Chios on 10 different sampling dates during the 2004 field season. The newly-emerged flies were maintained in individual cages (identical to the field-caught adults for the captive cohorts) and their reproduction and survival recorded daily until death. These data were used to construct the cohort life table as well as to compute summary statistics for maturation and reproduction.

Analytical Framework

Life Tables

The data gathered from the reference cohorts were used to construct a complete cohort life table. The notation for this life table follows demographic convention in which x denotes age, l_x is the probability of a newborn surviving to age x, p_x is the probability of surviving in the interval x to x+1, q_x is the probability of dying in the interval x to x+1, d_x is the fraction of the cohort that die in the interval x to x+1 and e_x is remaining life expectancy at age x (Carey, 2003). Assuming that the population is stable, stationary and closed, the number of subjects of age x is $c_x = l_x / \sum l_y = c_0 l_x$.

The data derived from the captive cohort (field-caught individuals) were used to construct the *captive life table*—a complete cohort life table in which the cohort is defined by all individuals captured on the same day that we refer to as 'marked age' denoted x'. Thus $l^*_{x'}$ denotes the fraction of wild individuals that survive from the day of capture to marked age x'. The death rates in the marked sample life table at age x' are by definition $d^*_{x'} = l^*_{x'} - l^*_{x'+1}$. These death rates are generated by subjects that enter the marked sample life table at various (unknown) ages, survive to 'marked age' x' but die prior to marked age x'+1. For all subjects that enter the marked sample cohort at age z, the contribution to $d^*_{x'}$ is therefore $c_0 = l_{z+x'} - l_{z+x'+1}$ where l_z refers to the survival function or survival schedule of the wild population at age z (Müller et al., 2004).

The relationship between the survivorship in a captive cohort derived from a hypothetical replacement-level population subject to the same survival rates as the reference cohort is $l^*_{x'+1} = l^*_{x'} - c_x$ where $l^*_{x'+1}$ and $l^*_{x'}$ denote the fraction of the captive cohort that survives to captive ages x'+1 and x', respectively. The radix of the captive life

table consists of individuals from all (chronological) age classes that die off are the age-specific rates observed in reference cohort but which are all lumped into a single captive age-specific rate as described by Müller and co-workers (Müller et al., 2004).

Age Structure Estimation

We define the following functions: f_A is the probability density function for the unknown age distribution in the wild, and is the target. A basic assumption is that flies are sampled from the wild proportionally to their age density f_A. Each fly that is captured is reared out in captivity and the time from captivity to death is recorded and denoted as X. The life table of remaining lifetimes X and the corresponding cumulative distribution function $\overline{F}_X$ and survival function $\overline{F}_X = 1 - F_X$ can be estimated from these data. In addition, a separate sample of flies is reared from eclosion in captivity, giving rise to a captive life table and cumulative distribution function F_C, as well as survival function $\overline{F}_C = 1 - F_C$, which can be estimated from the data of the captive cohort. One can then show that the following relationship holds:

$$\overline{F}_X(x) = \int_0^\infty P(X > x | A = a) f_A(a) da \tag{3a}$$

$$= \int_0^\infty \overline{F}_C(x + a) \frac{f_{A(a)}}{\overline{F}_C(a)} da \tag{3b}$$

All quantities appearing Eqn(3b) are empirically represented by histograms with bin width one day. This leads to a discrete linear equation where the unknown density $f - A$ is represented as

$$f_A(x) = \sum_{j=1}^{M-1} c_{jA} I_{(x_j, x_{j+1})}(x), \tag{4}$$

where x_j are the endpoints of the histogram bins and the density estimate is determined by the unknown constants $c_{jA.}$ These are obtained by solving the discrete convolution equation by penalized least squares, where the penalties are defined in terms of integrated second derivative squared, deviation from the integral over the estimated density from 1, and negative estimates.

If we integrate both sides of Eqn(4) over x, we obtain

$$E(X) = \int \overline{F}_X(x)dx \tag{5a}$$

$$= \iint \frac{\overline{F}_C(x+a)}{\overline{F}_C(a)} dx f_A(a)da \tag{5b}$$

$$= \int E(X-a \mid X > a) f_A(a)da \tag{5c}$$

$$= \int r(a) f_A(a)da, \tag{5d}$$

Where E(X) is the remaining life expectancy of the captured flies and r(a) is the remaining life expectancy at age a in the reference cohort; these quantities can be estimated from the available data. This provides some information for , $f_A(a)$, but not enough to related man age E(A) in the wild to mean remaining lifetime E(X) of the captured cohort. It holds that

$$E(X) - E(A) = \int f_A(a)(r(a) - a)da, \tag{6}$$

And the difference depends intrinsically on the shape of f_A.

Assumptions and Sources of Bias

Estimations of the age structure of wild populations using data from both the reference and captive cohorts are based on two assumptions. *Assumption #1: Age-independent probability of medfly capture*. This concept means that adult flies of each

age class are captured in proportion to their abundance in the wild. Although we could not find any literature on age-patterns of capture and/or attraction to protein bait on the medfly in particular or tephritids in general, a study on food intake in a tephritid fruit fly related to the medfly, the apple maggot fly, *Rhagoletis pomonella*, revealed that consumption in a wild strain decreased by 30% from the initial level during the first 2-3 weeks after eclosion and then remained constant whereas in a laboratory strain there was no change in consumption with age (Webster et al., 1979). These results suggest that if food consumption is related to food-seeking tendencies in tephritids, young (though not necessarily newly-emerged) flies might be captured in slightly greater frequency than older flies. Inasmuch as adult tephritids of all ages and both sexes require both carbohydrates for energy and protein sources (Prokopy and Roitberg, 1984), we believe that attraction to traps will be less affected by age than by the constant need to find high-quality sources of food every day.

Assumption #2: Identical age-specific birth and death rates of reference and captive cohorts. The underlying concept here is that no deleterious effects (either acute or chronic) are experienced by captured wild flies while they are in the trap or in transit, that there are no differences in the birth and death rates of female medflies that emerged from larval hosts relative to those flies reared from the hosts (figs; citrus) used in the reference cohort, and adjustment periods of captured wild flies to the cage environment (e.g. egg laying) are similar to that of the reference cohort. The results of a study designed to assess fertility in wild-caught flies in Chios (Katsoyannos et al., 1999) revealed that the cumulative percentage of wild medflies that oviposited after 15 days was nearly 100% when they had access to a natural host within a cage (i.e. pears), only 35% of wild flies

oviposited by this age when they had access to the artificial domes as used in the current study. It is clear that females need a period of adaptation to start oviposition into artificial substrates. However, there is no evidence that the death rates of wild flies that are captured in the wild and maintained in the laboratory are different from wild-type flies that are maintained in the laboratory from eclosion.

RESULTS

Reference Cohort

Survival and age-patterns of reproduction in the reference cohort of wild medflies that reared from field-infested hosts are presented in the event history chart given in Fig. 1. Several aspects of these patterns merit comment. *First*, survival of wild flies at young ages was extraordinarily high with no mortality for 28 days and only 10% mortality at 40 days. Survival to older ages was also quite high with 10% of the original cohort still alive at 3 months and the last fly dying at 140 days. Expectation of life of the wild flies at eclosion was 66 days. For perspective, an earlier study laboratory medflies maintained under similar conditions (individually) reported that 10% of the medflies were dead after only 14 days, 90% were dead after around 55 days and the oldest fly died at about 90 days (Carey et al., 1998a). Expectation of life at eclosion for this laboratory strain of medflies was 37 days. *Second*, the vast majority of wild medfly females maintained in the laboratory on a full diet did not begin laying eggs for nearly 3 weeks as reflected in the pre-reproductive period shown in the left-most region (green-coded) of Fig. 1. This time to maturity is in sharp contrast to the laboratory strains of medflies which begin laying eggs in 5-7 days (Carey et al., 1998a). The length of the maturation period in wild flies suggests that in the field conditions maturation may be even longer since it is virtually

certain that both the quality and availability of food in the field are much reduced relative to the food in the laboratory. *Fourth*, net reproduction (average number of eggs laid by females in their lifetimes) was 368.1 eggs/female. The overall levels of reproduction in wild medflies ranged from low to moderate with less than half of all females flies laying over 500 eggs. Only 2-of-134 flies laid no eggs in their lifetimes. In contrast, the average level of reproduction in laboratory strains of medflies ranges from 600 to 1,200 eggs/female with lifetime reproduction in some flies exceeding 2,000 eggs (Carey et al., 1998a).

{Fig. 1 around here}

Captive Cohorts

Captive Life Expectancies and Survival

Life expectancy of captive medfly females was conditional on sampling date (Fig. 2) with the captive life expectancies of only 23.6 and 16. 2 days in the flies captured on October 17 and 31, 2004, respectively, and with captive life expectancies exceeding 55 days on three sampling dates including one in 2003 (October 8) and two sampling dates in 2004 (June 29 and October 2). The life expectancies for all medfly females captured for each of the two field seasons differed by over one week (47.5 days for 2003 versus 40.0 days for 2004) although medflies were sampled on only 5 dates 2003 but were sampled on 11 dates in 2004.

Age-specific survival rates (Fig. 2) and captive expectation of life (Fig. 3) for each of the 16 sampling date during both the 2003 and 2004 field seasons reveal the variation in both within- and between-season survival of captured flies as well the relationship of survival and life expectancy relative to the reference cohort. Several

aspects of these graphs merit comment. *First*, survival patterns of captive cohorts ranged from sigmoidal (high early survival followed by gradual decrease) to convex (rapid drop at young ages and leveling off at middle and older ages) as shown in Fig. 2 (top panel). These differences are presumably due to differences in the ages of capture with the convex captive survival patterns due to a larger fraction of older flies and the sigmoidal patterns the outcome of a skew towards younger flies in the captive cohort. *Second*, the pattern of the composite survival curve of all captive flies for both seasons is similar to the captive survival curve predicted from the life table population of the reference cohort (Fig. 2 bottom panel). Although the similarity in pattern does not provide definitive proof that field populations are experiencing the mortality rates observed in the laboratory reference populations, the close affinity strengthens the conceptual and empirical linkages between reference and captive cohorts. *Third*, the range of captive life expectancies shown in Fig. 3 reflect the variation in captive survival rates with average longevity in captivity ranging from 35 to 58 days in 2003 (i.e. 1.7-fold difference) and from 16 to 57 days in 2004 (i.e. 3.6-fold difference). Inasmuch as rearing conditions in the laboratory were similar both within and between seasons, the differences in life expectancies must be attributable to differences in frailty of the cohorts due to age differences.

{Figs. 2 & 3 around here}

Frequency Distributions of Death

Three age distributions of cohorts deaths are presented in Fig. 4. The first is the age distribution of deaths in the reference cohort showing i) the absence of deaths to age 24 days, the more-or-less flat distribution from around 40 to 80 days, and finally the

residuals deaths at around 3 months and beyond. This is the distribution of deaths in the reference cohort of known age. The second age distribution of deaths shown in Fig. 4 is one predicted from the reference cohort if this cohort constituted a stable, stationary, closed population in which individuals were captured in proportion to their abundance to live out their lives in this constituted captive cohort. This distribution is constant through 24 days, begins to taper downward slightly through about 40 days and then decreases more-or-less linearly through 100 days and beyond. This distribution is plotted relative to captive age. The third age distribution of deaths shown in Fig. 4 is the observed captive age distribution of deaths in the field-captive, composite cohort of 1,033 medfly females. A comparison of this observed distribution to the other two distributions shows both similarities and differences. For example, there are fewer deaths observed deaths at younger captive ages than predicted from the reference cohort and more deaths at ages 25 to 40 days. However, the distribution of deaths at older captive ages between the predicted and the observed are fairly similar.

{Fig. 4 around here}

Reproduction in Captive Cohorts

Summary statistics for reproduction in the captive cohorts of medfly females are given in Table 1. Net reproduction for the captive flies ranged from less than 20 eggs/female for the 90 females captured on October 17, 2004 to nearly 400 eggs/female for the 4 flies captured on June 29, 2004. Average captive lifetime reproduction for both field seasons was similar with the average captive female producing 146.7 and 140.5 eggs for 2003 and 2004, respectively. Thus the net reproduction of captive cohorts of 140-148 eggs was less than half of the net reproduction in the reference cohort where the

average female laid 368.1 eggs in her life time. Whereas the percentage of days in which +females laid no eggs was nearly identical for the reference and captive cohorts (62.9 versus 63.1%), the percentage of days in which females laid only one egg was strikingly different with reference cohort females producing one egg on only 0.3% of the fly-days but captive cohort females producing one egg nearly 30% of the time—a 100-fold difference. This tendency to lay one of a few eggs each day accounts for the large disparity in net reproduction between reference and captive cohorts. The earliest egg laying occurred between 5 and 6 days post-capture both seasons with a range of 3 to 11 days.

{Table 1 around here}

Event history charts for medfly female survival and reproduction observed for 11 different sampling dates (both seasons) as well for all flies combined are shown in Fig. 2. The charts reveal two important patterns of reproduction in captive flies. *First*, egg laying is delayed virtually all flies as they require a period of acclimation post-capture. *Second*, unlike fertility patterns observed in earlier studies of laboratory flies (Carey et al., 1998a) in which females at older ages exhibited distinct periods of post-reproduction, most wild flies continued to lay at least a few eggs right up to their age of death.

The fertility schedule relative to their captive age for the composite cohort is given in Fig. 6 relative to both the captive cohort fertility schedule predicted from the reference cohort (relative to captive age) and the original fertility schedule observed by known age in the reference cohort. Differences between the observed and predicted captive cohort fertility schedules at young ages reflect the adjustment period needed by wild flies that are captured in the wild and maintained under laboratory conditions

relative to wild flies that eclosed in the laboratory. However, the similarity between the predicted and observed captive cohort fertility schedules at captive age 24 days and beyond is striking.

{Figs. 5 & 6 around here}

Characteristics of Wild Populations

Interpretation of Empirical Patterns

Support for the argument that captive cohort reflects wild age structure. *First*, the most compelling argument that older medflies are present in the wild is the similarity between the predicted and composite observed captive cohort survival schedule, not merely for one or two of the species for which field data was collected but for all of the species. *Second*, the 3-week maturation period for the wild flies that was observed in the reference cohort provides a longevity requirement for population renewal. Given that food in the wild is usually often scarce (Drew et al., 1983; White, 1978), wild flies might need to live a month or more just to mature and acquire adequate protein to produce eggs. *Third*, large differences were observed in the survival of captive cohorts suggesting that they differed widely in the age composition. *Fourth*, the extreme ages of the captive and the reference cohorts were similar at around 150 days. This is predicted from the captive cohort since some of the wild flies were young when captured and some would be expected to attain these extreme ages. *Fifth*, small percentage of flies laying in the first 10 days in the reference cohort relative to the captive cohorts. Indicates that older, more mature flies captured in wild.

Estimates of Age Structure

Estimates of the age distributions of wild flies on 11 of the individual sampling dates plus for the entire group of medflies for both seasons are presented in Fig. 7. Several aspects of this composite figure merit comment. *First*, the changes in the estimated age distributions of the populations on the different sampling dates lack clear within-season trends. This absence of trends are probably due to a variety of reasons including medfly generation overlap, small sample size (<100 individuals), local capture bias (e.g. many flies captured in small number of traps), and length of sampling period (bi-weekly) masking age structure shifts of short periodicity. Similar biases were outlined in an earlier paper on Drosophila (Begon, 1976). Therefore comparisons of nuanced changes in age structure over different are probably not valid. *Second*, the majority of populations appeared to consist of flies ranging in age from newly-emerged to 20 days. However, large variation with life expectancy of medflies captured on September 14 and October 2, 2004 of less that 20 days to medflies captured on October 31st estimated at 110 days. Many of the distributions are bimodal and/or discontinuous. Third, the composite captive cohorts of over 1000 individuals flies probably is the best indicator of the age structure of the average population over the two field seasons—the average females in the field population was approximately one month old. Thus if requires she requires an average of 10 to 20 days to mature, she is fertile for an average of 10 to 20 days in the field.

Cumulative age distributions (predicted) for both the reference cohort and the composite captive cohorts (Fig. 8) show the large relative difference between the age structure of the hypothetical reference population and the wild population based on the

composite cohort estimates. Whereas over 90% of the population in the wild is estimated to be aged 40 days or less, this equivalent percentage in the hypothetical reference population would be aged around 70 days or less. In other words, estimates of the age structure of the medfly population in Chios, Greece during 2003-04 based on the captive cohort methods indicates that it was much younger than would be predicted from the life table reference cohort.

{Figs. 7 & 8 around here}

DISCUSSION

{Rough drafts by sub- sections; still needs development and integration}

Studying aging in the wild using conventional capture-recapture methods poses some of the greatest technical challenges in ecology and gerontology because of the extraordinary difficulty of recapturing or tracking large numbers of marked (or radio-collared) individuals. Methods of capture-recapture introduce two types of bias (Service, 1993; Slade, 1995; Smith, 1995). First, the methods underestimate longevity because persistence is less than longevity—the first and last captures occur at some time between birth (eclosion) and death. Thus the extent of this bias is conditional on the probability of capture. For example, many single captures can be attributed to either low survival or low probability of re-capture. Second, heterogeneity in the probability of capture of certain individuals can introduce major errors in age-related estimates of survival probabilities. For example, individuals with a propensity for re-capture will skew survival estimates towards higher values and propensities for capture avoidance will skew them towards lower values.

The technologies developed to age insects are also fraught with problems and thus are of little use for estimating either age structure or longevity in insect populations. For

example, all three of the functional categories of age-grading (e.g. changes in the reproductive system such as follicular relics, the soma such as cuticular hydrocarbons, and in structural integrity such as wing fray) (Tyndale-Biscoe, 1984) are relatively accurate at young ages, imprecise at middle ages, and nearly useless at older ages.

Captive Cohort Method in Context

Although there have been a small number of previous studies where the researchers gathered demographic information from wild-caught Drosophila maintained in the laboratory (Begon, 1976; Boesiger, 1968; Bouletreau, 1978), the current study appears to be the first attempt to use the data derived from captive cohorts to estimate the age structure of the field populations from which they were captured. The results of these field studies differ from other field studies of aging in insects in two respects. The first is methodological in that we use the age pattern of deaths observed in the laboratory relative to a reference cohort to estimate the age structure of the field population. The second way in which the current study differs from other investigations is demographic—estimates are of chronological age rather than physiological stage.

Studies concerned with aging in the wild have traditionally focused on either survival estimates using mark-recapture techniques (Austad, 1993) or life table differences between of cohorts that were subjected to different selection pressures in the field (Reznick et al., 2004; Tatar et al., 1997). The current research appears to be the first to use estimates of age structure to gain insights into aging and longevity in the wild. No method is without constraints and the captive cohort approach is no exception. The approach assumes that age-bias of captures in the wild is minimal and that the actuarial response of captured flies is identical to same-aged flies of the same wild strain that are

reared throughout their lives in the laboratory. Data interpretation requires the availability of life table data on reference cohorts. However, there are several advantages of this method compared to other approaches. *First*, unlike mark-recapture methods which require the capture, marking, and re-capture of large numbers of individuals of known age (Buckland, 1982), the captive cohort method requires that individuals be captured only once. Therefore the assumptions in mark-recapture studies that the exact age of marked individuals is known (Pradel, 1996) and that the behavior of tagged or radio-collared animals is representative of those in the population at large (Moorhouse and MacDonald, 2005) are rendered moot. *Second*, the pool of source material for gathering demographic information using the captive cohort approach is potentially large and, during some times of the season limited less by the number of individuals that can be captured but by the number that can be monitored in the laboratory. For example, in the current study demographic information was gathered on over 1,000 individual wild females. *Third*, whereas capture-mark-recapture methods can be used for large insects such as butterflies (Boggs et al., 2004), these methods have limited usefulness for monitoring survival and longevity in populations of very invertebrates because of both low recapture rates and the likelihood of injury. Thus the captive cohort method provides a method for studying the age structure and longevity in wild populations of organisms for which no methods were previously available.

Alternative Explanations

We considered two alternative explanations for the earlier mortality that was observed in the wild flies maintained in the laboratory. The first possibility was that wild flies placed in a captive environment experience greater stress and thus die younger. This

explanation is unlikely, not only because we did not observe any overt differences in the behavior of the captured flies relative to the wild flies reared from infested hosts, but because survival rates several weeks post-capture in several of the captive cohorts were nearly as high as was observed at younger ages in the reference cohort. This high survival rate in many of the captive cohorts eliminates greater stress as an explanation for higher mortality at young ages in captive cohorts. A second hypothesis for why captive flies experienced higher mortality during the first 3-4 weeks post capture is that their reduced life spans were an outcome of having developed on larval hosts of lower nutritional quality than the flies collected from infested figs for the reference cohort. Although there is evidence that larval host does affect adult survival (Krainacker et al., 1987), there is no evidence that adult life span can be reduced to the extent necessary to explain the higher mortality rates observed in the captive cohort. Moreover, high-quality larval hosts are extremely abundant throughout the season in Chios, all of which almost certainly served as source hosts for the captured medflies. Although the scarcity of field research on aging stems partly from gerontologist's lack of interest and proficiency in ecology, the main reason for this empirical dearth involves constraints on the gathering of field data. For example, survival data derived from radio-collar technologies developed to track medium-sized and large vertebrates are costly and usually based on small numbers of individuals, and the mark-recapture methods designed to estimate survival of small invertebrates are inefficient and imprecise.

Implications and Importance

Results of several important implications. First, force population biologists to revisit the question of whether short life span of all individuals follows from

extraordinarily high mortality in nature. Second, small number of long-lived individuals may account for majority of recruitment. Third, evolutionary factors may favor selection for long life. Highlights the incongruity between the extraordinarily long life spans observed in captive environment and the putative short life span in nature. Three possible explanations including overengineering, seasonal or periodic bottlenecks which select for long-lived individuals and thus maintain long life, or that is a trait that is required in day-to-day living for species. Evolutionary ecology implications. Implies that longevity shaped by selection for feast and famine conditions in aboriginal home; maintains through seasonal bottlenecks. Extraordinarily long life span indicates selection for extended longevity in wild. This is particularly evident in the long reproductive life, not post-reproductive.

Biodemography of the medfly in the wild

The results of the current study as well as of earlier studies on the medfly in both the field and the laboratory provide three sources of both direct and indirect evidence of extended longevity in natural populations of the medfly including: (1) *Greater longevity of wild strains of medflies*. The longevity of a laboratory strain of the medfly has been extensively documented by Carey and co-workers (Carey, 2003) with life expectancies in females ranging from 30 to 45 days and record life spans typically in the range of 100 days. In contrast, the longevity of wild strains of medflies in Greece consistently show life expectancies of at over 2 months as in the current results and 3 months in previous studies of wild medflies that emerged in early spring (Papadopoulos et al., 1996; Papadopoulos et al., 2002). (2) *Host ecology and phenology*. Ecological studies of the medfly overwintering in both northern (Chios) and southern (Crete) regions of Greece

indicate that the medfly must be relatively long-lived during certain times in the season for population maintenance and replacement. For example, since no larval hosts are available until late May or early June for medflies that emerge from in March or April from hosts that had been infested the previous fall, adult females must survive for at least 2 months before they can lay eggs (Papadopoulos et al., 1996). This host-free period in early and late spring probably selects for long-lived adults in this part of the Mediterranean. Medfly populations in Crete experience slightly different environmental conditions that favor the maintenance and evolution of extended longevity in this species (Mavrikakis et al., 2000). Because winter conditions in this more southern region of Greece are much more mild, evidence suggests that low-level populations of adults survive throughout the winter. Although some host material is available throughout this period, it is sparse and it is likely that the adults that enter the late fall conditions are the individuals that replenish the population the following spring when conditions improve. (3) *Estimates of age structure*. The age estimates for the medfly populations in the wild indicate that 30-day old medflies are relative common in nature during different times in the season and that a small fraction of individuals probably survive for several months.

These results establish a demographic foundation for considering questions regarding the evolution and maintenance of extended longevity in natural populations of animals. What environmental factors favor the evolution of extended longevity in the medfly in particular and fruit flies more generally? This question must be considered in the context of the ecology of the fruit hosts on which medfly larvae feed as well as of the adult medfly. Medfly larvae must exploit mature fruit (Bateman, 1972; Christenson and Foote, 1960) which are often extraordinarily abundant in contemporary agricultural and

urban environments but which were scattered and highly seasonal in the primitive (pre-domesticated) environment in which medflies evolved. These host characteristics required the rapid exploitation of the larval food substrate and reduced the period of contact of the medfly with its host. These conditions favored the evolution of long-lived adults capable of surviving unfavorable conditions (Zwolfer, 1983). It is likely that the medfly and other tephritids as well as different Drosophila species adapted to feast-and-famine host conditions in which adults were selected for that possessed two main life history traits: i) the ability to survive for extended periods of time when conditions were unfavorable for reproduction (i.e. absence of hosts); and ii) the ability to lay large numbers of eggs over a short period when larval hosts became available.

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Table 1. Summary of reproductive traits in captive cohorts[a] (females only) by sampling date in Chios, Greece. n denotes number of captive individuals, and R_0=captive net reproductive rate (average captive lifetime number of eggs per female).

Sample		Percent Days Laying		First Egg (days)		
Date	**n**	**Zero Eggs**	**Single Egg**	**Min**	**Average**	**R_0**
2003						
5-Aug	25	70.4	18.4	4	11.9	221.7
26-Aug	84	78.3	14.3	3	12.1	198.3
17-Sep	42	67.2	25.9	9	26.1	160.1
8-Oct	99	61.1	32.2	6	28.7	117.4
4-Nov	100	72.5	21.9	6	23.6	108.0
Subtotal	**350**	**69.9**	**23.2**	**5.6**	**21.8**	**146.7**
2004						
16-Jun	15	60.9	30.5	4	17.7	186.9
29-Jun	4	47.4	29.8	11	14.8	378.3
14-Jul	87	56.8	31.6	3	18.8	196.0
30-Jul	26	58.9	28.7	8	16.3	182.1
14-Aug	11	58.6	27.1	3	11.6	257.4
31-Aug	52	48.3	43.5	3	18.7	97.5
14-Sep	100	61.3	29.4	3	18.5	217.7
2-Oct	100	43.9	46.6	5	21.7	187.5
17-Oct	90	72.6	25.6	5	23.4	19.2
31-Oct	98	81.4	16.2	5	18.0	30.0
11-Nov	100	48.0	40.9	6	20.1	167.4
Subtotal	**683**	**59.4**	**32.5**	**5.1**	**19.6**	**140.5**
TOTAL	**1033**	**62.9**	**29.4**	**5.3**	**20.3**	**142.6**

[a] Reference cohort: n= 134; percent of days females laid zero and one egg was 63.1 and 0.3%, respectively, minimum and average age of first reproduction was 8 and 19.4 days, respectively, and the net reproduction rate, R_0=368.1

FIGURE LEGENDS

Fig. 1. Event history chart (Carey et al., 1998b) of the 134-medfly reference cohort reared from infested hosts in 2004. The chart shows the inter-individual variation in age of onset of reproduction and both the inter- and intra-individual variation in timing and intensity of reproduction relative to age of death.

Fig. 2. Captive survival in medflies for each of the 16 sampling dates in the 2003-04 field seasons in Chios, Greece (top panel). Bottom panel shows composite captive survival (survival in all flies from both seasons) and predicted from the reference life table.

Fig. 3. Average longevity of medfly females captured on each 16 sampling dates in 2003-04 in Chios, Greece. Reference lines A, B, and C indicate life expectancy in the reference cohort, captive life expectancy in the composite cohort of all captured flies, and the predicted captive life expectancy from the reference life table, respectively.

Fig. 4. Frequency distributions of deaths for reference medfly life table, the captive cohort predicted from the reference life table, and the observed captive cohorts from field captures (composite of captures in both 2003 and 2004 in Chios, Greece).

Fig. 5. Event history charts showing captive survival and captive reproduction in 8-of-16 captive cohorts (plus composite chart in lower right) derived from medfly females trapped in the 2003-04 field seasons in Chios, Greece. Only cohorts with 25 or more individuals were plotted.

Fig. 6. Daily reproduction in the reference medfly cohort for females of known age, in the captive cohort predicted from the reference life table populations, and actual

field-derived captive cohort of wild flies (composite of all sampling dates from both 2003 and 2004). The X-axis refers to either chronological (actual) age of reference cohort flies or time from capture (captive age) for wild-caught flies.

Fig. 7. Estimates of the age distributions of medfly females in the wild derived from the captive cohort model (see text) incorporating knowledge of age-specific mortality of wild-strain medflies and the distribution of deaths in captive flies. Estimates of the average age of each population (x*) are indicated below date.

Fig. 8. Estimated cumulative age structure in medfly populations based on the observed reference cohort, the reference cohort in which mortality rate is increased at all ages by 5-fold, and the composite captive cohort (all field captures). Dashed lines show variation between estimates for ages containing cumulative totals of 50 and 90% of all age classes.

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