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Age-specific mortality analysis of the dry forest kissing bug, *Rhodnius neglectus*

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Abstract

Age-specific mortality patterns can be very different across insects with different life histories. Some holometabolous insects (like mosquitoes, fruit flies) show a pattern where mortality rate decelerates at older ages, whereas other holometabolous insects (bruchid beetles) and hemimetabolous insects (cotton stainers, milkweed bugs, and kissing bugs) show an age-specific mortality pattern that increases through all ages. Kissing bugs are strictly hematophagous and are vectors of *Trypanosoma cruzi* Chagas, the etiologic agent of Chagas disease. Here, we tested whether cohort data from the dry forest kissing bug, *Rhodnius neglectus* Lent (Hemiptera: Reduviidae), supports an increase of mortality rate that decelerates with age. We analyzed the age-specific mortality pattern of a cohort of 250 individuals of *R. neglectus*. We used a suite of seven models with different degrees of complexity, to model age-dependent forms of change in mortality rate increase in *R. neglectus* in the laboratory. We used the Akaike model selection criterion to choose between models that consider absence or presence of mortality deceleration. Five of the seven models (logistic, Gavrilovs, Gompertz, DeMoivre, and exponential) showed a statistically significant fit to the mortality rate. Weak late-age mortality deceleration in *R. neglectus* was supported by the best fit (logistic model), and this result is consistent with predictions of the disposable soma theory of senescence.

Introduction

The understanding of senescence, that is, the increase in mortality rate with age because a decline in physiological functioning, is a major goal of the research agenda in evolutionary ecology (Williams, 1957; Abrams, 1993; Gavrilov & Gavrilova, 2001; Carey, 2003). The need to fully understand its underpinnings requires the use of the comparative approach (Carey, 2001) which builds upon observations of a wide variety of organisms. Most of the studies have been focused on a small subset of species, with the most comprehensive studies coming from insects, especially fruit flies (e.g., Carey, 2003). It has been observed that mortality patterns can be very different across insects with very different life histories. Holometabolous insects, such as mosquitoes (Styer et al., 2007), fruit flies (Curtsinger et al., 1992; Carey et al., 1992, 2005; Fukui et al., 1993), but not a bruchid beetle (Tatar et al., 1993), have shown a pattern where mortality decelerates at older ages, eventually reaching a plateau. Studies in hemimetabolous insects, including the cotton stainer (Dingle, 1966), the milkweed bug (Dingle, 1966), and two species of kissing bugs (Chaves et al., 2004a,b), have shown a pattern where mortality only increases with age.

Kissing bugs have strict hematophagy across all of their foraging ontogenetic stages, which is associated with a mechanism that avoids the accumulation of oxidative substances associated with aging (Graça-Souza et al., 2006). They are also vectors of *Trypanosoma cruzi* Chagas, the etiologic agent of Chagas disease (WHO, 1991). Thus, the study of senescence in this group may contribute to the study of senescence as an evolutionary process and prove useful for pest population management. Here, we present a cohort study of the dry forest kissing bug, *Rhodnius neglectus* Lent (Hemiptera: Reduviidae) focusing on the

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analysis of age-specific mortality and survivorship. As previous studies on kissing bugs (using *Rhodnius prolixus* Stål and *Rhodnius robustus* Larrousse) were focused on the Gompertz and the positive-shaped Weibull models (Chaves et al., 2004a,b) we compare these two models, which cannot account for mortality rate deceleration, to an additional suite of five models with various degrees of complexity, that can model mortality rate deceleration and different forms of mortality rate increase with age (Appendix A). Our aim is to test whether the instantaneous mortality rate in *R. neglectus* decelerates with age. We compare model fitting using Akaike Information Criterion model selection to test the absence or presence of instantaneous rate of mortality deceleration in this species.

Materials and methods

Background

Rhodnius neglectus is a species that has been observed in Northeast Brazil (Lent & Wygodzinsky, 1979; Carcavallo et al., 1999; Galvao et al., 2003) and in Venezuela in the State of Amazonas, until at least 1965 (Gamboa Cuadrado, 1973). This species is found in a region characterized by mean annual temperatures ranging between 19 and 30 °C (average 23.3 °C), and a mean (±SD) precipitation of 1 246 ± 429 mm year^{-1}. *Rhodnius neglectus* has been identified by Curto de Casas et al. (1999) to occupy the Holdridge Dry Forest and Very Dry Forest or Savannah Life Zones.

Laboratory conditions

The population of *R. neglectus* used was made available by Dr Rodolfo Carcavallo and came from the insectary of the Instituto Oswaldo Cruz (Rio de Janeiro, Brazil), but its exact geographical origin is not known. The study was carried out in Caracas, Venezuela, in a climate room with constant conditions of temperature (26 ± 1 °C) and humidity (60 ± 10% r.h.). Regulation of temperature is essential because of the strong effect it has on the developmental cycle and survival (Garcia da Silva & da Silva, 1988). The ambient photoperiod was used: this did not need to be controlled, because at the latitude of Caracas its seasonal variation is very small (day length in December is 11:29 hours, and in June 12:42 hours).

The design involved the follow up of five independent cohorts initiated simultaneously. Each cohort was started with 100 recently laid (0- to 48-h old) eggs, kept in 150-ml glass containers until all viable eggs hatched. The first instars were transferred to 3.8-l jars, covered with nylon mesh, and with vertically placed strips of paper inside, serving both as resting place and for climbing to the top at feeding time.

Each cohort was fed weekly using hens placed on a wooden box with holes at the bottom, through which the tops of the cohort jars could be tightly inserted. The insects climbed to the top and fed through the nylon mesh. Food was offered for 1 h, and during the following hour the jars were horizontally exposed to a fan, to avoid the accumulation of excessive moisture in the glass containers, which is common after the insects become engorged. The hens were sedated using an intramuscular dose of midazolam (0.25 mg kg^{-1}), a sedative commonly used to relax small animals before surgery. We allowed the hens to rest for 5–10 min after the sedative application, and then carefully plucked them on one side on a surface not larger than a diameter of 7 cm (the jars' mouth size). A towel was used to restrain the hens to minimize stress from being restrained. This procedure was carried out by two people who would gently wrap the towel around the hen, letting the head and legs stick out for better freedom of movements. This procedure was approved by the Animal Use Institutional Review Board of IVIC (Venezuelan Institute for Scientific Research) and is in conformity with Venezuelan law.

After being exposed to the fan, each jar was opened to check the number of dead individuals (identified by instar); identification by sex was performed only for adults, and only the female population was recorded. Thus, data on mortality during the nymphal stages reflect a mix of the two sexes, whereas the adult stage reflects only females. In two instances, the strict weekly feeding and counting schedule could not be maintained (although they were never out of phase for more than 3 days), and as the biodemographic methods require a constant time unit for analysis, the recorded information was subjected to linear interpolation to keep the week as the time unit for calculation of the age-specific mortality rates. Additionally, this procedure had to be applied at the beginning of the cohort study (when the number of individuals was still high). This interpolation is a sound procedure, because (1) when the cohorts are still in their first ages, all models predict an exponential mortality, which for small x values can be considered linear (due to the Taylor expansion; Demidovich, 1973), and (2) the various models do not discriminate mortality patterns at early ages of the cohort, that is, most of them coincide in their predictions for early ages, yet they can predict very different patterns for late ages.

Statistical analysis

The weekly recording of the numbers of bugs alive and dead was used to calculate, following Carey (2001), the basic parameters of a decrement life table: (1) survival as a function of age ($l_x = N_x/N_0$, or fraction alive at age x), (2) age-specific (period) survival ($p_x = l_{x+1}/l_x$, or fraction alive at age x surviving to x + 1), (3) age-specific (period) mortality ($q_x = 1 - p_x$, or fraction alive at x dying prior to x + 1).

The age-specific mortality, q_x, is a discrete quantity, representing mortality as the probability of dying over the 1-week interval used. The continuous analog of this measure is referred to as the force of mortality or instantaneous mortality rate and denoted as $\mu(x)$; it is defined as the mortality rate representing the limiting value of the age-specific mortality rate when the age interval to which the rate refers becomes infinitesimally short (Carey, 2001). The age-specific mortality rate is preferred over age-specific mortality, q_x, because it is not bounded by unity, it is independent of the size of the age intervals, and it is used in numerous mortality models (Carey, 2001). The analytical relationship between the discrete form of mortality, q_x, and its continuous expression is $\mu(x) = -\ln\ p_x$ and $p_x = e^{-\mu(x)}$.

We first analyzed the five cohorts independently, and carried out a Friedman ANOVA and a Kendall coefficient of concordance test to verify if the differences in mortality rates $\mu(x)$ among cohorts were statistically significant. Although mortality rates in older ages were not synchronized, results showed that $\mu(x)$ differences among the five cohorts were not statistically significant (ANOVA: $\chi^2 = 5.902$, $P = 0.21$; Coefficient of concordance = 0.0168, Average rank r = 0.00547). The further analysis of the age-specific mortality pattern was carried out after pooling the five cohorts, treating the data as a single cohort. The initial number of eggs in the pooled data was taken 250, because the average sex ratio was 0.44 [♀/(♀+♂)] (which is not significantly different from 0.5; two-sided t-test: P = 0.18; StatSoft, 2009). Similarly, unbiased 0.5 sex ratios have been found in other triatomine species (Ronderos, 1972; Paz Rodríguez, 1996). Despite the five cohorts being pooled for their fitting to mortality rate models, the results of the individual cohorts are also presented to provide visualization of the dispersion of the data of the cohorts around their pooled values.

In the analysis of the age-specific mortality pattern of the pool of the five cohorts, the following six mortality models were used: the exponential, deMoivre, Gompertz, Gompertz-Makeham, Weibull, and logistic models, using the algebraic presentation by Carey (2001), and a seventh three-parameter model based on reliability theory as proposed by Gavrilov & Gavrilova (2001); for simplicity the latter will be called hereafter 'Gavrilovs'. The reasons for using a mortality analysis and not a survival analysis, as well as a description of the models and parameters used, are given in detail in Appendix A.

Fitting models to data

Carey et al. (1992) claim that survival curves are poorly suited for summarizing age-specific mortality patterns as compared with mortality rate curves, so we fitted models to data using μ_x. For that purpose, the data used to fit the models were converted from their initially l_x format, into $p_x = l_{x+1}/l_x$, and then to mortality rates by $\mu_x = -\ln(p_x)$. All models were fitted using various procedures in the R language (R Development Core Team, 2007). Because of the small number of mortality rate observations between weeks 83 and 89, when the last bugs died, we only analyzed the first 83 weeks, because estimates of the mortality rate (μ_x) become unreliable given the small number of individuals that can face death (Carey et al., 1992). Despite the number of individuals decreasing with age, we did not weight the data because the models used are non-linear regression models that constitute an alternative to a weighing function in linear regression analyses. The resulting time series was analyzed both raw and smoothed; the reason for smoothing before model fitting is that the data become easier to handle by the models by removing irregularities and inconsistencies, thus capturing better the behavior of the mortality rate process being modeled (Carey, 2001). For smoothing we used the Kolmogorov–Zurbenko adaptive filter (KZA) (Chaves et al., 2008), with parameter q = 2 (half the size of the smoothing window). The exponential model was fitted using least squares (Faraway, 2005) and confidence intervals obtained from the standard errors. For this model no differences were observed when compared with a weighted regression, with weights proportional to the number of surviving individuals. The Gavrilovs model was fitted using the Nelder–Mead optimization algorithm (Nelder & Mead, 1965) implemented with the R command 'optim', and 95% confidence intervals were found using a non-parametric bootstrap (Faraway, 2005) on 1 000 replications. All other models were fitted by non-linear least squares (Bates & Watts, 1988), using the R command 'nls'. For these models, SEs were computed with a non-parametric bootstrap similar to the one described for the Gavrilovs model.

Goodness-of-fit model selection

To compare the performance of different models with various degrees of freedom (i.e., different number of fitted parameters), we used the Akaike Criterion (AIC) (Akaike, 1974), a method preferred over classical goodness-of-fit tests (such as the chi-squared or the G-tests) because it 'penalizes' models with a high number of parameters. The AIC evaluation of model performance (one of the most used goodness-of-fit indicators) represents the 'information content' of a given set of parameter estimates (Burnham & Anderson, 2002), and it is independent of the magnitude of the data points and of the number of observations. It is calculated as:

$$\mathrm{AIC} = n \ln \left(\sum_{i=1}^{n} (Y_{obsi} - Y_{calci})^2 \right) + 2p,$$

where Y_{obsi} and Y_{calci} are the observed and predicted mortality rates for the ith observation, 'p' is the number of parameters, and 'n' the number of observations. The smaller the value of the AIC, the more appropriate the model is.

Results

After converting the cohort age-specific survival (l_x) into q_x and $\mu(x)$, and despite the peaks of mortality in later ages not being synchronized, the differences between the age-specific mortality rate $\mu(x)$ among the five cohorts of *R. neglectus* are not statistically significant (Table 1), indicating that the five cohorts may be pooled.

The age-specific female survival (l_x) values for each cohort as well as the pool of the five cohorts are presented in Figure 1. There is a sharp decline in survival in the first developmental stages (egg, and instars 1 and 2), then the decline stabilizes until approximately the age of 45 weeks (about 30 weeks of adult female's age) when another sharp decline is observed; after age 45 there is a progressive decline in survival. Figure 2 shows the weekly evolution of the age-specific mortality rate $\mu(x)$ for the individual cohorts and for the pool among cohorts.

Some of the results of the fitting of the smoothed and non-smoothed data to the models are shown in Figure 3 and Table 2. The Weibull model is not shown graphically because it had a statistically non-significant fit to one of its parameters. Figure 3 shows the predictions of the remaining five models (Gompertz, exponential, DeMoivre, Gavrilovs, and logistic) with observed pooled laboratory data of the age-specific mortality rate $\mu(x)$ of *R. neglectus*. The parameter values of those five age-specific mortality rate models tested are shown in Table 2 for the raw data and the KZA smoothing. Figure 4 shows the maximum effort of egg laying. The quadratic fit for fecundity, given by the equation $y = -0.0085x^2 + 0.7386x - 5.4479$ ($r = 0.908$), is shown only to ease visualization of the peak of the reproductive effort of *R. neglectus* females. The method for calculating fecundity (eggs/female/week) is given in Rabinovich (1972). It is striking how the peak of reproductive effort of *R. neglectus* coincides with the second early peak in mortality (Figures 2–4).

Table 1 Friedman ANOVA and Kendall coefficient of concordance to test the differences in the weekly mortality rates [$\mu(x)$] sequence among the five cohorts of *Rhodnius neglectus*

	Mean ± SD
Cohort 1	0.0429 ± 0.1297
Cohort 2	0.0258 ± 0.0580
Cohort 3	0.0420 ± 0.1014
Cohort 4	0.0437 ± 0.1294
Cohort 5	0.0417 ± 0.1093

ANOVA $\chi^2 = 5.902$, d.f. = 4, P = 0.21; n = 88.
Coefficient of concordance = 0.0168, average rank r = 0.00547.

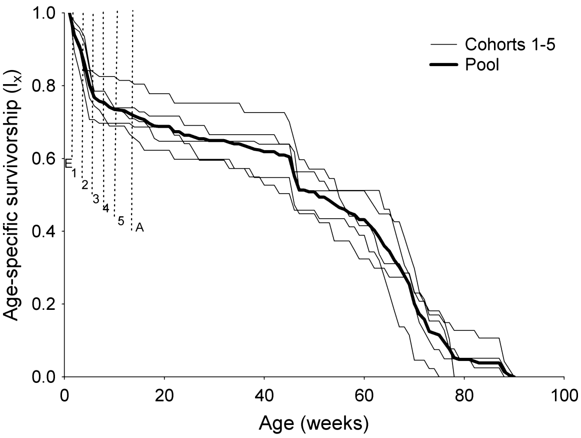

Figure 1 Age-specific survival (l_x) for each cohort of *Rhodnius neglectus* and the pool of the five cohorts. Vertical dashed lines indicate the average periods of immature development. E, eggs; 1–5, instars 1–5; A, female adults.

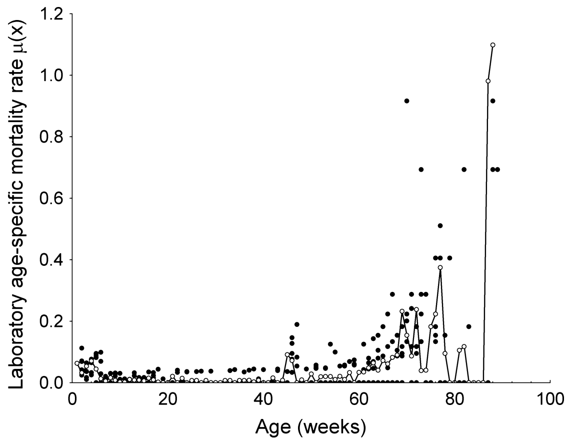

Figure 2 Age-specific mortality rate [$\mu(x)$] patterns of each cohort of *Rhodnius neglectus* (black dots) and the pool of the five cohorts (white dots and solid line). The individual cohorts were not identified separately to facilitate visibility.

To determine the model that represents best the age-specific mortality rate data of *R. neglectus*, the results of the application of the Akaike model selection criterion to the

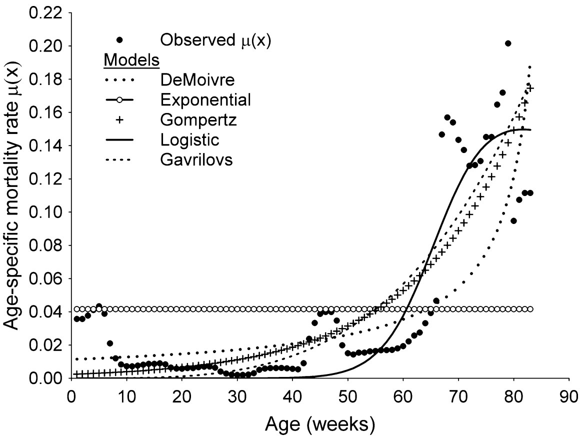

Figure 3 Age-specific mortality rate μ(x) of observed laboratory data (as pool of five cohorts) of *Rhodnius neglectus* and the predictions of the DeMoivre, exponential, Gompertz, logistic, and Gavrilovs models, for the smoothed data applied to μ(x).

seven mortality rate models are shown in Table 3 for the KZA smoothed μ(x) data. From the models that conformed significantly to the data (the Weibull and GM models did not have a significant fit), the logistic and the Gompertz conformed best, and DeMoivre and exponential the worst. Similar results (not shown) were obtained with the fit to the raw data.

Discussion

In recent years, some striking lack of fit to the Gompertz model has been found in large cohorts of experimental animals, primarily insects (Partridge & Mangel, 1999). The results of our study show weak evidence for late-age mortality deceleration in *R. neglectus*, in contrast to patterns reported for several Diptera, including the fruitfly *Drosophila melanogaster* Meigen (Curtsinger et al., 1992; Pletcher & Curtsinger, 1998), the medfly *Ceratitis capitata* (Wiedemann) (Carey et al., 1992, 1998), the mexfly *Anastrepha ludens* (Löw) (Carey et al., 2005), and the mosquito *Aedes aegypti* (L.) (Styer et al., 2007). *Rhodnius neglectus* age-specific mortality pattern seems to be more similar to that reported for other kissing bugs such as: *R. prolixus* (Chaves et al., 2004a), *R. robustus* (Chaves et al., 2004b), the milkweed bug, *Oncopeltus fasciatus* (Dallas) (Dingle, 1966), the cotton stainer, *Dysdercus fasciatus* Signoret (Dingle, 1966), and the bruchid beetle *Callosobruchus maculatus* (Fabricius) (Tatar et al., 1993). This result is consistent with the equilibrium predictions of the antagonistic pleiotropy and mutation accumulation models of senescence (decline in physiological functioning with age; Abrams & Ludwig, 1995). These models are known as the 'disposable soma' theory for the evolution of senescence (Kirkwood & Rose, 1991). This theory states that senescence arises from an optimal balancing of resources between reproduction and somatic repair. For instance, model simulation showed that only when reproductive output gradually approaches, but never reaches, a maximum value as the amount of diverted resources increases, senescence is consistent with the Gompertz model (Abrams & Ludwig, 1995).

Our results fit these predictions: we found a sharp decline in survival in the early developmental stages (egg and instars 1 and 2), then the decline stabilizes at the age of 45 weeks (about 30 weeks of adult female's age) when another sharp decline is observed, possibly related to the

Table 2 Parameter values of the five age-specific mortality rate [μ(x)] models (the Weibull and Gompertz-Makeham models were not included because their fit to the laboratory data was not statistically significant)

		No smoothing (raw data)	Smoothed data (KZA with q = 2)
Model	p	Parameter values	Parameter values
DeMoivre	1	ω = 89.06 [86.74–91.78]	ω = 88.25 [87.06–89.53]
Exponential	1	c = 0.0393 [0.02512–0.05346]	c = 0.0416 [0.0301–0.0531]
Gompertz	2	a = 0.00276 [0.0004–0.0093]	a = 0.00233 [0.0009–0.0047]
		b = 0.0485 [0.0317–0.0730]	b = 0.0520 [0.0425–0.0647]
Logistic	2	g = 67.359 [62.70–71.18]	g = 67.6185 [65.90–69.27]
		n = 12.275 [9.89–15.09]	n = 13.2362 [12.21–14.37]
Gavrilov	3	n = 4.3138 [3.044–4.841]	n = 4.5443 [3.845–4.816]
		k = 0.0154 [0.0154–0.0182]	k = 0.0154 [0.0153–0.0165]
		q = 1.0156 [1.014–1.016]	q = 1.0155 [1.014–1.016]

'p' is the number of parameters of each model. In brackets are the 95% confidence intervals; intervals that do not contain zero are statistically significant with P<0.05, and have been presented graphically in Figure 3. KZA refers to the Kolmogorov–Zurbenko adaptive filter smoothing method. The models are presented in increasing order of their number of parameters.

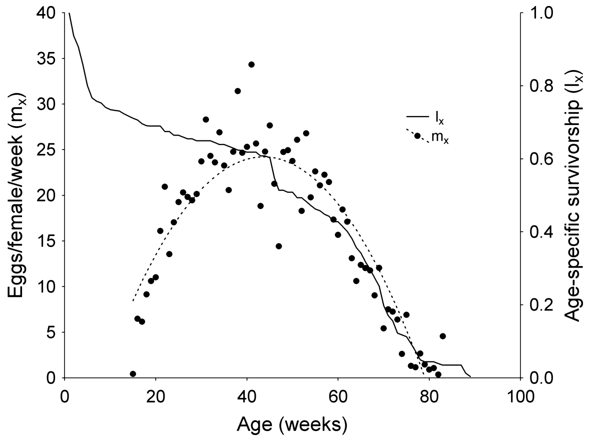

Figure 4 Age-specific survival of the pool of five cohorts of *Rhodnius neglectus* from the egg stage and the maternity function (eggs/female/week) as an indicator of egg laying effort. The maternity function was fitted to a quadratic equation ($y = -0.0085x^2 + 0.7386x - 5.4479, r = 0.908$) only to ease visualization of the peak of the reproductive effort of *R. neglectus* females.

maximum effort of egg laying; after 45 weeks there is a progressive decline in survival. These two declines in l_x correspond of course to the two strong earlier peaks of mortality rate $\mu(x)$ as shown in Figure 3. Figure 4 further supports the relationship between maximum effort of egg laying and the second early peak in the mortality rate. This apparent relationship between egg-laying effort and effects on the age-specific mortality rate is similar to the one found in the beetle *C. maculatus* (Tatar et al., 1993) and the kissing bug *R. prolixus* (Sulbaran & Chaves, 2006). Williams (1957) claimed that greater rates of 'extrinsic' (age- and condition-independent) mortality favored more rapid senescence. However, Abrams (1991) showed that the effects of the 'extrinsic' mortality differentially affect the rate of senescence according to density-dependence. Furthermore, Abrams (1991) showed that mortality patterns, contrary to Williams (1957) predictions, are possible when density-dependence is present, and acts primarily on the survival or fertility of later ages, or when most of the variation in mortality rates is due to variation in non-extrinsic mortality. No experimental evaluation exists on density-dependent processes in *R. neglectus*. However, Rodríguez & Rabinovich (1980) showed in *R. prolixus* that density had a significant effect on the developmental rate of second, third, and fourth instars, but not on first instars, adult survivorship, instantaneous population parameters, or age-specific parameters. If similar patterns were present in *R. neglectus*, then this would confirm that in this species Williams' (1957) rapid senescence may be a response to greater rates of 'extrinsic' mortality.

Table 3 Relative ranking (1 = best, 7 = worst) of the fit of seven mortality rate models to the KZA smoothed laboratory data of *Rhodnius neglectus* using Akaike model selection criterion (AIC)

$\mu(x)$ model rank	AIC
1. Logistic*	−23.013
2. Weibull	4.8909
3. Gompertz*	17.4839
4. Gavrilovs*	51.540
5. Gompertz-Makeham	53.374
6. DeMoivre*	71.714
7. Exponential*	79.795

Asterisks identify those models that had a statistically significant fit by the criterion of the 95% confidence intervals with the bootstrap method.

In *R. neglectus* there is no apparent reduction of the length of reproductive life as a consequence of high early fecundity (J.E. Rabinovich, unpubl.), unlike *R. prolixus* (Sulbaran & Chaves, 2006) and several strains of *D. melanogaster* (Giesel & Zettler, 1980; Partridge, 2001). On the contrary, *R. neglectus* shows a clear exponential increase in death rate in older ages, suggesting a Gompertz behavior, and possibly reflecting a minor effect of the trade-off between reproduction and somatic repair, confirming a possible larger effect of 'extrinsic' mortality.

In contrast with the Gompertz model, in the Weibull model the causes of death for young adults and old individuals are different, independent, and additive, and include death causes due to catastrophic intrinsic sources with a probability that increases with age, and not as a physiological function that declines with age as in the Gompertz model (Gavrilov & Gavrilova, 2001; Ricklefs & Scheuerlein, 2002). Partly for these reasons, the Gompertz model is considered by most demographers as an empirical model rather than a 'law' (Carey, 2003). Despite the Weibull function being usefully used in conjunction with failure-time models (in which failure depends on the occurrence of one or more rare events, such as genetic mutations or cell deaths; Ricklefs & Scheuerlein, 2002), the fit of this model to the *R. neglectus* mortality rate data was not statistically significant.

The only other instance of application of mortality models to kissing bug species are the studies of Chaves et al. (2004a,b). Only the Gompertz model was tested on the age-specific mortality rate of *R. robustus*, and the Gompertz and Weibull models were tested on the age-specific mortality rate of *R. prolixus*. The results from these two studies are different to those of *R. neglectus* in terms of the parameter values of both models. This may be partially due to demographic evolution of these taxa: *R. prolixus* has a population intrinsic rate of natural increase (r_m) of about 0.032 per week, almost double that of *R. neglectus* of 0.017 per week (J.E. Rabinovich, unpubl.) reflecting their adaptation as results of genetic selection to different environments. This result also confirms the soundness of the assumptions of Charlesworth (2000), supporting the use of r_m to explain the evolution of senescence and life-history patterns.

The weak late-age mortality deceleration in *R. neglectus* was observed only through the fit of the logistic model of mortality, the model selected as 'best' by the Akaike model selection criterion. However, this late-age mortality deceleration process is extremely sensitive to the sample size of the cohort study, and our initial cohort size of 250 individuals was probably too small to permit a strong assertion about late-age mortality deceleration in *R. neglectus*. As claimed by Gaines & Denny (1993), when mortality rate is close to zero it behaves as a threshold character; and to estimate the points 'below threshold mortality' becomes difficult and their measures are usually relatively inaccurate (Promislow et al., 1999). Carey (2003) considers that an estimate of the threshold mortality can be taken as 1/N, where N is the initial cohort size; in the case of *R. neglectus* the initial cohort size of 250 individuals would indicate a threshold mortality of 0.004, and mortality rate values below this threshold exist in our laboratory data, both raw and smoothed. An increase in the initial cohort size is difficult for kissing bugs, with a complex feeding behavior, the need of large containers to avoid problems of overcrowding, and requirements of important numbers of adequate and live avian or mammal hosts for feeding.

However, we strongly suggest future studies with kissing bugs to consider this threshold mortality factor, which will allow confirmation of whether hemimetabolous insects do not decelerate their mortality at old ages, as our results suggest. As stated by Chaves et al. (2004a) in relation to *R. prolixus*, the mortality rate of this triatomine vector has epidemiological importance through the demography of its populations. In the case of *R. neglectus*, despite being essentially sylvatic, it is considered to be in the process of becoming a truly domestic species (Lent & Wygodzinsky 1979). It has been shown in triatomines that important changes in life-history traits occur when reared under laboratory conditions (Perlowagora-Szumlewicz, 1976); for example, after only 4 years in the laboratory, the average reduction in the number of eggs within the first month of oviposition and the average female life span among *Triatoma infestans* Klug, *Triatoma brasiliensis* Neiva, and *Triatoma pseudomaculata* Correia & Espinola, and *Panstrongylus megistus* Burmeister was 37.5 and 45.9%, respectively. The adaptation to a domestic environment causes a similar effect on triatomines (Forattini, 1980) because conditions inside rural houses provide smaller predation risks, smaller fluctuations of temperature and relative humidity that exclude deleterious extreme values, and better chances of feeding. The values of the age-specific mortality rate parameters estimated under domiciliary conditions will prove of importance in the epidemiology of Chagas disease, and even decisive in the design of pest control measures.

In a recent review of mammal and bird comparative analyses of survival senescence by using life tables, Jones et al. (2008) have identified generalizations including the observation that mammals senesce faster than similar-sized birds. Furthermore, McCoy & Gillooly (2008) developed a model of natural mortality (relating body size and temperature to biological rate processes) and tested it with extensive field data from plants, invertebrates, fish, birds, and mammals; their results indicate that much of the heterogeneity in natural mortality rates can be predicted, explicitly and quantitatively, despite the many extrinsic sources of mortality in natural systems, suggesting that common rules govern mortality rates. We believe that comparative review is due for different orders of insects. Such a review not only has academic value, but it would also impinge on important pest population management and epidemiological areas. New age-dependent models of pathogen transmission show the importance of age and population age structure to transmission dynamics (Styer et al., 2007). These authors claim that, in the case of mosquitoes, a departure from the age-independent mortality paradigm is essential for an accurate understanding of mosquito biology and pathogen transmission.

Environmental heterogeneity, and particularly high environmental uncertainty (stochastic effects) affects environmental circumstances that are very important in insects, such as encounter rates with suitable oviposition sites, food availability, physiological state (e.g., reserves for producing oocytes, egg maturation rate, and somatic maintenance costs), and expected reproductive success that can lead to different patterns of behavior and rates of mortality and reproduction (Partridge & Mangel, 1999), and they should be incorporated in future studies for understanding senescence in insects.

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References

Abrams PA (1991) The fitness costs of senescence: the evolutionary importance of events in early adult life. Evolutionary Ecology 5: 343–360.

Abrams PA (1993) Does increased mortality favor the evolution of more rapid senescence? Evolution 47: 877–887.

Abrams PA & Ludwig D (1995) Optimality theory, Gompertz' law, and the disposable soma theory of senescence. Evolution 49: 1055–1066.

Akaike H (1974) A new look at statistical model identification. IEEE Transactions on Automatic Control 19: 716–723.

Bates DM & Watts DG (1988) Nonlinear Regression Analysis and its Applications. Wiley & Sons, New York, NY, USA.

Burnham KP & Anderson DR (2002) Model Selection and Multimodel Inference. A Practical Information - Theoretical Approach. Springer, New York, NY, USA.

Carcavallo RU, Curto de Casas SI, Sherlock IA, Galíndez Girón I, Jurberg J, et al. (1999) Geographical distribution and alti-latitudinal dispersion of Triatominae. Atlas of Chagas' Disease Vectors in the Americas, Vol. III (ed. by RU Carcavallo, I Galíndez Girón, J Jurberg & H Lent), pp. 747–792. Editorial Fiocruz, Rio de Janeiro, Brazil.

Carey JR (2001) Insect biodemography. Annual Review of Entomology 46: 79–110.

Carey JR (2003) Longevity. The Biology and Demography of Life Span. Princeton University Press, Princeton, NJ, USA.

Carey JR, Liedo P, Orozco D & Vaupel JW (1992) Slowing of mortality rates at older ages in large medfly cohorts. Science 258: 457–461.

Carey JR, Liedo P, Müller H-G, Wang J-L & Vaupel JW (1998) Dual modes of aging in mediterranean fruit fly females. Science 281: 996–998.

Carey JR, Liedo P, Müller H-G, Wang J-L, Senturke D & Harshmanf L (2005) Biodemography of a long-lived tephritid: Reproduction and longevity in a large cohort of female Mexican fruit flies, *Anastrepha ludens*. Experimental Gerontology 40: 793–800.

Charlesworth B (2000) Fisher, Medawar, Hamilton and the evolution of aging. Genetics 156: 927–931.

Chaves LF, Hernández MJ, Revilla TA, Rodríguez DJ & Rabinovich JE (2004a) Mortality profiles of *Rhodnius prolixus* (Heteroptera: Reduviidae), vector of Chagas disease. Acta Tropica 92: 119–125.

Chaves LF, Zamora E & Aldana E (2004b) Mortality profile of female *Rhodnius robustus* (Heteroptera: Reduviidae). Revista de Saúde Pública 38: 466–468.

Chaves LF, Kaneko A, Taleo G, Pascual M & Wilson ML (2008) Malaria transmission pattern resilience to climatic variability is mediated by insecticide-treated nets. Malaria Journal 7: 100–113.

Curto de Casas SI, Carcavallo RU, Galíndez Girón I & Burgos JJ (1999) Bioclimatic factors and zones of life. Atlas of Chagas' Disease Vectors in the Americas, Vol. III. (ed. by RU Carcavallo, I Galíndez Girón, J Jurberg & H Lent), pp. 793–838. Editorial Fiocruz, Rio de Janeiro, Brazil.

Curtsinger JW, Fukui HH, Townsend DR & Vaupel JW (1992) Demography of genotypes: failure of the limited life-span paradigm in *Drosophila melanogaster*. Science 258: 461–463.

Demidovich BP (1973) Problemas y Ejercicios de Análisis Matemático. Editorial Mir, Moscow, USSR.

Dingle H (1966) The effect of population density on mortality and sex ratio in the milkweed bug, *Oncopeltus*, and the cotton stainer, *Dysdercus* (Heteroptera). American Naturalist 100: 465–470.

Faraway JJ (2005) Linear Models with R. Chapman & Hall, Cambridge, UK.

Forattini OP (1980) Biogeography, origin, and distribution of triatominae domiciliarity in Brazil. Revista de Saude Pública 14: 265–299.

Fukui HH, Xiu L & Curtsinger JW (1993) Slowing of age-specific mortality rates in *Drosophila melanogaster*. Experimental Gerontology 28: 585–599.

Gaines SD & Denny MW (1993) The largest, smallest, highest, lowest, longest, and shortest: extremes in ecology. Ecology 74: 1677–1692.

Galvao C, Carcavallo RU, da Silva Rocha D & Jurberg J (2003) A checklist of the current valid species of the subfamily Triatominae Jeannel, 1919 (Hemiptera, Reduviidae) and their geographical distribution, with nomenclatural and taxonomic notes. Zootaxa 202: 1–36.

Gamboa Cuadrado J (1973) Distribución geográfica y prevalencia de la población de triatominos en Venezuela. World Health Organization Planning Meeting for the Unit for the Study of Vectors of Chagas Disease in Venezuela. Maracay 29–31 March, 1973. Dirección de Malariología y Saneamiento Ambiental, Maracay, Venezuela.

Garcia da Silva I & da Silva HH (1988) Influencia da temperatura na biologia de Triatomineos. II. *Rhodnius neglectus* Lent, 1954 (Hemiptera, Reduviidae). Revista Goiana de Medicina 34: 29–37.

Gavrilov LA & Gavrilova NS (2001) The reliability theory of aging and longevity. Journal of Theoretical Biology 213: 527–545.

Giesel JT & Zettler EE (1980) Genetic correlations of life historical parameters and certain fitness indices in *Drosophila melanogaster*: r_m, r_s, diet breadth. Oecologia 47: 299–302.

Gompertz B (1825) On the nature of the function expressive of the law of human mortality and on a new mode of determining life contingencies. Philosophical Transactions of the Royal Society, London 115: 513–585.

Graça-Souza AV, Maya-Monteiro C, Paiva-Silva GO, Braz GR, Paes MS, et al. (2006) Adaptations against heme toxicity in blood-feeding arthropods. Insect Biochemistry and Molecular Biology 36: 322–335.

Jones OR, Gaillard J-M, Tuljapurkar S, Alho JS, Armitage KB, et al. (2008) Senescence rates are determined by ranking on the fast-slow life-history continuum. Ecology Letters 11: 664–673.

Kirkwood TBL & Rose MR (1991) Evolution of senescence: late survival sacrificed for reproduction. Transactions of the Royal Society, London B 332: 15–24.

Kohler H-P & Kohler I (2000) Frailty modelling for adult and old age mortality: the application of a modified Demoivre hazard function to sex differentials in mortality. Demographic Research 3: 1–32.

Lent H & Wygodzinsky P (1979) Revision of the Triatominae (Hemiptera, Reduviidae), and their significance as vectors of Chagas' disease. Bulletin of the American Museum of Natural History 163: 123–520.

Makeham WM (1860) On the law of mortality and the construction of annuity tables. Journal of the Institute of Actuaries 6: 301–310.

Makeham WM (1867) On the law of mortality. Journal of the Institute of Actuaries 13: 325–358.

McCoy MW & Gillooly JF (2008) Predicting natural mortality rates of plants and animals. Ecology Letters 11: 710–716.

Nelder JA & Mead R (1965) A simplex algorithm for function minimization. The Computer Journal 7: 308–313.

Olshansky SJ & Carnes BA (1997) Ever since Gompertz. Demography 34: 1–15.

Partridge L (2001) Evolutionary theories of ageing applied to long-lived organisms. Experimental Gerontology 36: 641–650.

Partridge L & Mangel M (1999) Messages from mortality: the evolution of death rates in the old. Trends in Ecology & Evolution 14: 438–442.

Paz Rodríguez R (1996) Ciclo de Vida y Datos Biométricos de *Triatoma longipennis* (Usinger) (Hemiptera: Reduviidae, Triatominae). PhD Dissertation. Autonomous National University of Mexico, Mexico City, Mexico.

Perlowagora-Szumlewicz A (1976) Laboratory colonies of Triatominae, biology, and population dynamics. New Approaches in American Trypanosomiasis Research. Proceedings of an International Symposium, Belo Horizonte, Minas Gerais, Brazil, 18–21 March 1975, pp. 63–82. Pan American Health Organization, Scientific Publications No. 318.

Pletcher SD & Curtsinger JW (1998) Mortality plateaus and the evolution of senescence: why are old-age mortality rates so low? Evolution 52: 454–464.

Promislow DEL, Tatar M, Pletcher S & Carey JR (1999) Below-threshold mortality: implications for studies in evolution, ecology and demography. Journal of Evolutionary Biology 12: 314–328.

R Development Core Team (2007) R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria. http://www.R-project.org/

Rabinovich JE (1972) Vital statistics of Triatominae (Hemiptera: Reduviidae) under laboratory conditions. I. *Triatoma infestans* Klug. Journal of Medical Entomology 9: 351–370.

Ricklefs RE & Scheuerlein A (2002) Biological implications of the Weibull and Gompertz models of aging. Journal of Gerontology 57A: B69–B76.

Rodríguez D & Rabinovich JE (1980) The effect of density on some population parameters of *Rhodnius prolixus* (Hemiptera: Reduviidae) under laboratory conditions. Journal of Medical Entomology 17: 165–171.

Ronderos RA (1972) Biología de Triatominos. Simposio Internacional sobre Enfermedad de Chagas, 26 de Noviembre al 2 de Diciembre, pp. 321–326. Sociedad Argentina de Parasitología, Buenos Aires, Argentina.

StatSoft (2009) STATISTICA (Data Analysis Software System), version 9.0. Statsoft, Tulsa, OK, USA.

Styer LM, Carey JR, Wang J-L & Scott TW (2007) Mosquitoes do senesce: departure from the paradigm of constant mortality. American Journal of Tropical Medicine and Hygiene 76: 111–117.

Sulbaran JE & Chaves LF (2006) Spatial complexity and the fitness of the kissing bug, *Rhodnius prolixus*. Journal of Applied Entomology 130: 51–55.

Tatar M, Carey JR & Vaupel JW (1993) Long-term cost of reproduction with and without accelerated senescence in *Callosobruchus maculatus*: analysis of age-specific mortality. Evolution 47: 1302–1312.

Vaupel JW, Manton KG & Stallard E (1979) The impact of heterogeneity in individual frailty on the dynamics of mortality. Demography 16: 439–454.

WHO (1991) Control of Chagas disease. World Health Organization Technical Report Series 811. WHO, Geneva, Switzerland.

Williams GC (1957) Pleiotropy, natural selection and the evolution of senescence. Evolution 11: 398–411.

Appendix A

Survival vs. mortality analysis

As stated in section 'Statistical analysis' under 'Materials and methods', two basic parameters of a decrement life table are directly related: survival as a function of age ($l_x = N_x/N_0$), and age-specific mortality [$q_x = 1 - (l_{x+1}/l_x)$]. Thus we could have modeled survival (l_x) or mortality [q_x; actually the instantaneous mortality rate, denoted as $\mu(x)$; that is, the rate representing the limiting value of the age-specific mortality rate when the age interval becomes infinitesimally short]. There are several reasons why we preferred to use a mortality analysis and not the method of survival analysis: (1) survival analysis is based upon models like Kaplan–Meier or Cox, which estimate the death hazards but do not reflect any mechanism or process underlying the fitted curves as the models we use do (survival models can be viewed as ordinary regression models in which the response variable is time), (2) survival rates are mainly important for projections or prognosis, and to estimate the importance of cause-specific survival (e.g., to rank the effects of different causes of mortality), which is not the goal of our analysis, (3) when no truncation or censoring occurs, the Kaplan–Meier curve is equivalent to the empirical distribution, and (4) we were directly interested in the mortality and not the survival process.

In relation to the latter, despite the fact that mortality and survival are intimately related, there is an important difference: death is an event whereas survival is a 'non-event', that is, the absence of the mortality event (Carey, 2001). The advantage of the survival analysis method to deal with censored observations would not have improved our analysis, for in our study there were no censored observations (defining censored observations, as it is commonly used in survival analysis, as those individuals who are lost to follow up being still alive or that remained alive when the study ended). Although we deleted observations between weeks 83 and 89 because the mortality rate (μ_x) becomes unreliable given the small number of individuals that can face death at those ages (see section 'Fitting models to data' under 'Materials and methods'), those observations cannot be considered censored under our definition, but rather outliers.

Mortality models

Exponential mortality rate model

This is the simplest mortality model that assumes that the mortality rate grows linearly with age x. The formula for the mortality rate function is given by:

$$\mu_x = c.$$

DeMoivre model

The mortality rate in the DeMoivre model (1725, cited in Kohler & Kohler, 2000) attempts to describe mathematically the mortality patterns assuming that 'the number of lives existing at any age is proportional to the number of years intercepted between the age given and the extremity of old age', that is,

$$l_x = l_0\left(1 - \frac{x}{\omega}\right),$$

where ω is the maximum attainable age in the population. Then the hazard rate, or the age-specific mortality rate at age x, is defined as

$$\mu^D(x) = \frac{1}{\omega - x},$$

where $\mu^D(x)$ denotes the DeMoivre hazard function. The hazard rate increases towards infinity as x approaches the maximum attainable age ω. This maximum age implies that the age-specific mortality rate increases faster than in the Gompertz model (see below), and in particular as x approaches ω, when the mortality rate tends to approach unity.

Gompertz model

Gompertz (1825), cited in Olshansky & Carnes, 1997) set forth what is now recognized as the law of mortality. He noted that the differences between the common logarithm of the number of individuals living in successive equal age intervals were almost identical during a significant portion of their life span, in general at least after the age of sexual maturity or another predetermined age, such as the maximum reproductive effort (Carey, 2001, 2003). The mortality rate (μ_x) function in Gompertz's model has the following form:

$$\mu_x = a\,e^{bx},$$

where 'a' is a parameter representing the initial mortality rate, and 'b' is the Gompertz parameter that represents the rate of increase of mortality with age (also called the Gompertz aging parameter).

Gompertz-Makeham model

Makeham (1860) refined Gompertz's law of mortality, noting that the logarithms of the probabilities of living from Gompertz's formula, instead of proceeding in uniform geometrical progression, increased at a faster pace at higher ages than at younger ages. He solved this problem by adding a 'constant' term, and redefining the Gompertz law as 'the probabilities of living, increased or diminished in a certain constant ratio, from a series whose logarithms are in geometrical progression'. Later Makeham (1867) modified Gompertz's formula by developing 'a theory of partial forces of mortality'. The Gompertz-Makeham model is of the form:

$$\mu_x = ae^{bx} + c,$$

where 'c' is the additional parameter introduced by Makeham to represent the additional constant rate of mortality.

Weibull model

As with the Gompertz model, the Weibull model incorporates a minimum mortality rate suffered by young adults prior to the onset of their physiological decline. This is usually referred to as the initial mortality rate (μ_0). After this initial mortality the aging-related mortality rate increases exponentially as a multiple of the initial mortality μ_0. In the Weibull model, the aging-related component of mortality is a power function of age that is added to the initial mortality rate:

$$\mu_x = ax^n,$$

where ‘a’ and ‘n’ are the parameters of the Weibull model. The value of ‘a’ determines the scale, and ‘n’ determines the shape of the curve. The Weibull hazard function increases if $n>0$, decreases if $n<0$, and is constant if $n = 0$.

Logistic model

The logistic model has a Gompertz-type mortality rate as a baseline hazard, with hazard function (Carey, 2001):

$$\mu_x = \frac{nx^{n-1}}{g^n + x^n}$$

For this model mortality rates level off at older ages. This model is similar to a frailty model sensu Vaupel et al. (1979).

Gavrilov and Gavrilova model

Gavrilov & Gavrilova (2001) proposed a general model for aging based on reliability principles. In this model it is assumed that mortality rate $[\mu(x)]$ comes from the deterioration of redundant subunits (n) organized in blocks (m) that compose individuals, at a constant failure rate (k), where the probability of an element being initially functional can vary from fully functional ($q = 1$) to highly likely to be unfunctional ($q \sim 0$). In this model the initial number of functional elements has a binomial distribution:

$$\mu_x = mn(qk)^n\left(\frac{1-q}{qk} + x\right)^{n-1}.$$

Only the three-parameter version of this model was tested (where m remains constant, i.e., $m = 1$).