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Retarded senescence in an insular population of Virginia opossums (*Didelphis virginiana*)

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(With 4 figures in the text)

Evolutionary senescence theory predicts that genetically isolated populations historically subjected to low rates of environmentally-imposed mortality will ultimately evolve senescence that is retarded in relation to that of populations historically subjected to higher mortality rates. This prediction was evaluated in the field by comparing three general measures of senescence—age-specific mortality rate acceleration, age-related reproductive output and tail tendon collagen denaturation rate—in two radiocollared Virginia opossum (*Didelphis virginiana*) populations, one a mainland control and the other an insular population having a four- to five-thousand-year history of reduced exposure to predators. In comparison with those on the mainland, insular female opossums were found to exhibit greater survivorship and reduced litter sizes at all ages, as well as slower acceleration of age-specific mortality. Also, island females in their second reproductive year failed to show the senescent reduction in pouch young growth rate seen in mainland animals. Island opossums also manifested slower 'ageing' of tail tendon fibres, a generalized measure of physiological ageing. All these results are consistent with evolutionary senescence theory. Various environmental explanations (parasitism, disease, lessened food availability) for this populational difference are evaluated by physiological measures. No evidence for an environmental explanation is found.

Contents

Introduction

Senescence may be defined demographically as an age-related increase in adult mortality rate and/or decrease in reproductive output, or physiologically as the generalized organismic deterioration that leads to these demographic changes (Charlesworth, 1980; Finch, 1990; Rose,

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1991). More than three decades ago, evolutionary biologists (Medawar, 1952; Williams, 1957) developed a comprehensive theory of senescence, according to which organismic deterioration was due to the differential impact of natural selection on alleles with age-specific effects. According to this theory, which was later given a sound quantitative foundation (Hamilton, 1966; Charlesworth, 1980), natural selection will act strongly on traits manifested early in adult life because such traits will be expressed in virtually all reproductive individuals. On the other hand, natural selection will have less impact on traits expressed late in life, because environmentally-imposed mortality from such factors as predation, starvation, or infectious disease will ensure that fewer individuals have their reproduction curtailed by these late-acting traits. This age-related waning of the force of natural selection will lead to senescence as a consequence of: (1) the accumulation of late-acting deleterious alleles, and (2) positive selection for alleles with pleiotropically beneficial early-acting, and harmful late-acting, effects. This latter effect has been termed *antagonistic pleiotropy* (Williams, 1957).

Several authors (Edney & Gill, 1968; Charlesworth, 1980) have pointed out that a critical test of evolutionary senescence theory would be a comparison of genetically isolated populations evolving under conditions of relatively high versus relatively low environmentally-imposed mortality. In this case, the theory predicts retarded senescence in populations evolving in 'safer' environments, because natural selection wanes more slowly. And to the extent that antagonistic pleiotropy is present, retarded senescence will be accompanied by a reduction in early fitness components.

Ecologists have noted repeatedly that islands are commonly relatively benign environments (Carlquist, 1974; Williamson, 1981). They are frequently deficient in the number of predator species they support, and the surrounding water ameliorates seasonal temperature extremes. In addition, nonvolant mammals are poor dispersers across water barriers and insular populations are particularly prone to genetic isolation from their mainland ancestors once they are there (Brown, 1971; Lawlor, 1986). These factors in sum suggest that insular populations of mammals may often be expected to age more slowly than their ancestral mainland populations.

According to the above logic, I sought to evaluate critically the prediction that animals evolving in 'safer' environments will exhibit retarded senescence by comparing a mainland population of Virginia opossums (*Didelphis virginiana*) with an insular population which has lived in an environment historically lacking mammalian predators of opossums. A major source of mortality in wild opossums is predation by a variety of avian and mammalian predators (Fitch & Shirer, 1970; Hunsaker, 1977; Gardner, 1982). The importance of predation generally, and mammalian predation in particular, is shown by Ryser's (1990) data on radiocollared opossums, in which more than half (53·3%) of 30 naturally occurring deaths in the wild were due to predation and, in cases where the predator could be identified, two-thirds of the deaths were due to mammals.

Opossums are exceptionally suitable for demographic studies in the wild. They are sufficiently large to carry a long-lasting radiocollar, young can be aged precisely and marked during the more than two months they spend inside the maternal pouch, and overall lifespan is typically less than two years (Fitch & Shirer, 1970; Gardner, 1982; Seidensticker, O'Connell & Johnsingh, 1987; Sunquist & Eisenberg, In press). Thus individuals can easily be monitored throughout their lives.

Female opossums are also known to undergo manifest physical senescence both in captivity and in the wild during the second breeding year of life, as evidenced by atrophy of the reproductive organs and reduced fertility (Reynolds, 1952; Jurgelski & Porter, 1974; Harder & Fleming, 1982; Seidensticker *et al.*, 1987) in addition to senile weight loss and cataract formation (Austad, 1988; Henness, 1989).

Materials and methods

Study sites

The mainland study site was the Savannah River Site, an 81 000 ha U.S. Department of Energy National Environment Research Park near Aiken, South Carolina, USA. The island study site was Sapelo Island, Georgia, USA, a 4500 ha barrier island separated from the Georgia coast by approximately 5 miles of open water and *Spartina* salt marsh. Sapelo Island was chosen specifically because it met the 2 criteria for making a valid comparison with the mainland population with respect to the evolutionary hypothesis, namely that environmentally-imposed mortality was lower on the island (common mammalian opossum predators such as bobcats and feral canines were lacking: A. S. Johnson *et al.*, 1974) and that the island population was apparently genetically isolated from the mainland populations. Although no direct genetic evidence for isolation exists as yet, the indirect evidence is: (1) opossums are not known to be eager swimmers (although they will escape into the water when threatened and swim moderately well when pressed: Hunsaker & Shupe, 1977; Gardner, 1982), and (2) opossums have not recolonized other Georgia barrier islands a similar distance from the mainland since they were extirpated just after the U.S. Civil War. The only exceptions to this are islands that are connected to the mainland by bridges (A. S. Johnson *et al.*, 1974).

Demographic data collection

In January 1988, I began radiocollaring young opossums and individually marking and estimating the age of pouch young at both study sites. After 1988, all study animals had been marked as pouch young, and their birthdates were therefore known to within 5–10 days (see Petrides, 1949, for age estimation of pouch young from a suite of anatomical characters). When captured for radiocollaring all females were weighed and head, body and tail length were measured.

In 1988 itself, only virgin females were radiocollared, and ages of these females were estimated to the nearest month by using relative body size to determine in which of the 2 previous birth peaks (February and May) they had been born. Virgin females are distinguishable from non-virgins by the condition of their pouch (Reynolds, 1952). Radiocollars had mortality switches which altered pulse frequency when opossums had died. A total of 34 known-age adult females from Sapelo Island and 37 similar females from the mainland were radiocollared and monitored until their deaths. Females were monitored for survival at least every 6 weeks between May and September each year, and at least every 4 months during the remainder of the year. When a death was discovered, it was assumed to have occurred halfway between the time of its discovery and the last record that the animal was alive. Because the chief interest of this paper is senescence, only adult demography will be considered. All opossums are reproductive by the age of 1 year (Reynolds, 1952; Gardner, 1982), therefore demographic analyses begin at that age.

During the breeding season females were recaptured opportunistically, and the number and size of their pouch young were measured. Opossum pouch young are incapable of reattaching to the nipple between 3 and approximately 50 days of age (Reynolds, 1952). During this period, therefore, pouch young mass was estimated by measuring tail length. Previous studies have shown tail length to be linearly related at $r^2 > 0{\cdot}99$ to pouch young body mass (Sunquist & Eisenberg, In press). Ages of pouch young were also estimated from well-known developmental landmarks (Petrides, 1949).

'Ageing' of tail tendon fibres

Collagen, the most abundant protein in the vertebrate body, is metabolically inert once mature fibrils are laid down, yet it shows continuously advancing postranslational molecular changes throughout an animal's life. That is, it 'ages' (Verzàr, 1964). Collagen ageing is due to an increase throughout life in intra- and intermolecular crosslinks, which lead to progressive changes in the molecule's physical and chemical

properties (Hall, 1976). Several lines of evidence suggest that collagen ageing is a general measure of physiological ageing (Kohn, 1978). First, one measure of crosslinking—tendon fibre breaking time in buffered urea (Harrison & Archer, 1978)—increases consistently with increasing age in all species studied to date whether in the field or the laboratory (Verzàr, 1964; Hamlin & Kohn, 1972; Vancikovà & Deyl, 1973; Harrison & Archer, 1978; Harrison *et al.*, 1978; Bochantin & Mays, 1981; Sherman *et al.*, 1985). Secondly, *Peromyscus leucopus*, which lives more than twice as long as *Mus musculus* in the laboratory (Sacher & Hart, 1978), showed slower ageing of tendon fibre than *Mus* (Harrison *et al.*, 1978); and several strains of laboratory rats differing in maximum longevity showed parallel differences in tendon fibre ageing (Bochantin & Mays, 1981). Finally, food restriction, a demonstrated age-retarding treatment of rodents (Weindruch & Walford, 1988), slows tendon fibre ageing (Everitt, Giles & Gal, 1969; Harrison & Archer, 1978), as do hormonal manipulations thought to retard senescence (Everitt, Wyndham & Barnard, 1983; Everitt & Meites, 1989).

Beginning in 1989, 2–4 tendon fibres were surgically removed from each captured female and stored in physiological saline solution at −20 °C until analysed. Surgeries were performed using a 50/50 mixture of ketamine/valium anaesthesia; longitudinal incisions were made 60–80 mm from the base of the tail and closed with absorbable suture. Wounds heal in about a week with no apparent effect on tail function.

Collagen fibre diameter was measured in the laboratory and fibre denaturation rate was measured by recording the fibre breaking time in a standard temperature buffered urea bath when a standardized weight was suspended from the fibre. This method was modified from Harrison & Archer (1978), differing from their protocol in the mass of the weight used (14 g as contrasted with 2 g) and the temperature of the urea solution (58 °C instead of 45 °C). Values used in the analysis were means of all fibres measured from a given sample from an individual animal.

Blood chemistry parameters

In order to compare general health of individuals in the 2 populations, during the final 18 months of the study, blood samples were drawn from each opossum anaesthetized for removal of tendon fibres. The packed cell volume of each sample was determined by separation of cells from serum in a capillary haematocrit tube (Fischbach, 1988). Additionally, a subset of these opossums had 1 ml of blood drawn from the ventral tail vein. The samples were sent to Tufts Veterinary Diagnostic Laboratory (Grafton, Massachusetts, USA) for a complete clinical blood chemistry profile, which included concentrations of glucose, blood urea nitrogen (BUN), creatinine, albumin, globulin, alkaline phosphatase (APTase), serum glutamic oxalacetic transaminase (SGOT), cholesterol, total bilirubin, and serum glutamic pyruvic transaminase (SGPT). Blood chemistry values were compared with normal (i.e. non-diseased) opossum blood parameters taken from Henness (1989). Dr Henness is a veterinarian specializing in Virginia opossum medicine.

Results

Demographic comparison

Adult females from the island had a significantly greater mean and maximum longevity than mainland females (Table I). In addition, insular females exhibited greater survivorship at all ages (Fig. 1a). However, populations, or species, with substantially different mean and maximum longevities may still be undergoing senescence at similar rates if their incidence of environmentally-imposed mortality is sufficiently different (Finch, Pike & Witten, 1990).

One method for separating the effects of environmentally-imposed from those of senescent mortality is the use of a Gompertz (1825) mortality rate model:

$$q(t) = Ae^{at}$$

TABLE I

Opossum life-history differences between island and mainland (mean with S.D. in parentheses).

Trait	Mainland	Island	*P*-value
Mean longevity (months)	20·0 (5·04)	24·6 (7·09)	0·002 ($t_{69}=3·16$)
Maximum longevity (months)	31	45	—
Body mass index (kg/m^2)	8·30 (1·24)	8·22 (1·15)	0·767 ($t_{69}=0·30$)
Age at first reproduction (months)	10·55 (2·45)	11·56 (1·75)	0·297 ($t_{18}=1·07$)
Litter size (1st year)	7·63 (1·86)	5·86 (1·32)	0·002 ($t_{35}=3·39$)
Litter size (2nd year)	7·58 (1·31)	5·36 (1·01)	<0·001 ($t_{24}=5·00$)
Litter size (combined)	7·61 (1·62)	5·66 (1·22)	<0·001 ($t_{61}=5·47$)

All *t*-tests are one-tailed.

where $q(t)$ is mortality rate at adult age, t, a is the rate constant for age-related mortality change, and A describes the vulnerability of an organism to environmental hazards at the age of maturity. This model is an empirically derived function which provides a good fit to mortality rate data from a wide range of species in both the field and the laboratory (Finch, 1990; T. E. Johnson, 1990). Graphically, on a semilogarithmic plot of this model, the slope of the resulting line describes the rate of mortality rate increase, that is senescence, and the Y-intercept provides information on the relative degree of environmental 'riskiness'.

Adult mortality in both populations fits a Gompertz function well (Fig. 1b). These functions confirm that environmentally-imposed mortality is lower on the island (note the relative position of the intercepts), as was assumed because of the lack of predators, and also demonstrate that senescent mortality rate increases more slowly on the island (slope = 0·54 and 0·091 for island and mainland, respectively).

The two populations also differed in reproductive rates and reproductive ageing. Although age at first reproduction did not differ significantly between populations, and individuals from both populations had a modal production of two litters per year, litter size was significantly reduced at all ages on the island (Table I). Furthermore, although litter size did not decline between first and second reproductive years in either population (mainland: $t_{26}=0·07$, $P=0·47$; island: $t_{33}=1·21$, $P=0·119$), young grew significantly more slowly in the pouch during females' second reproductive year on the mainland—an effect that was lacking on the island (Fig. 2). Note also that, even with a smaller litter size, first-year island breeders' pouch young grew significantly more slowly than those of mainland breeders (general linear test, $P<0·01$), suggesting a lower overall reproductive investment. There was also a marginally significant tendency for old (i.e. second reproductive year) mainland females in a non-reproductive state (neither lactating nor with pouch young) to be captured more often during the breeding season (February–September) than young females, suggesting a higher rate of reproductive failure (Fisher's Exact Test, one tail, $P=0·054$). No similar difference is found in the insular population (Fisher's Exact Test, one tail, $P>0·9$).

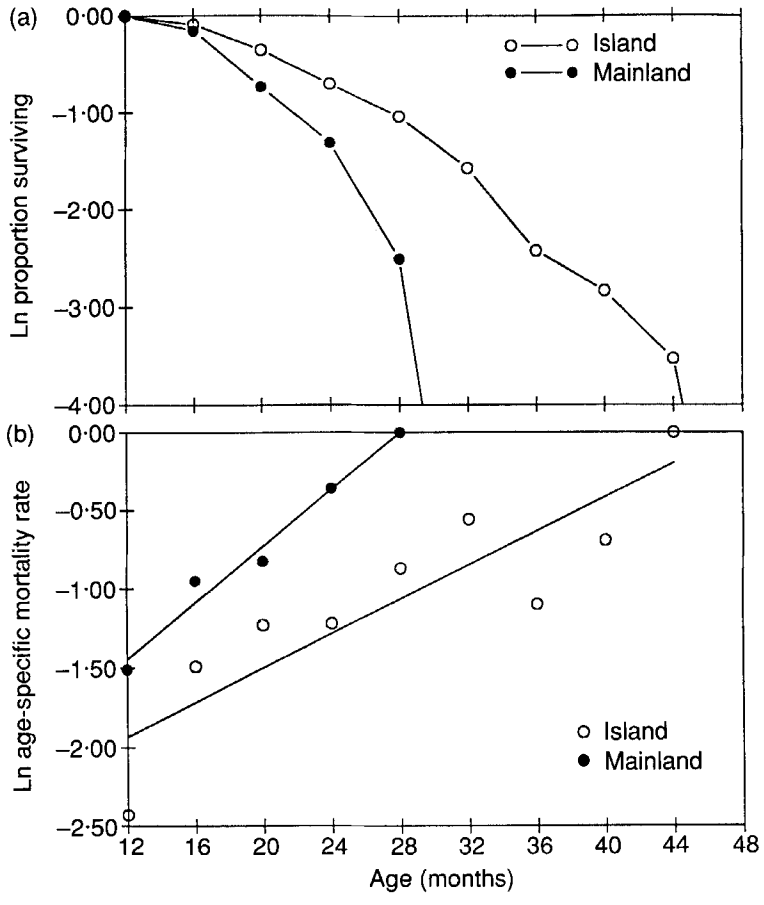

FIG. 1. (a) Survivorship compared in the two populations. The survivorship curve is begun at age 12 months, because all females are reproductively active by that age. (b) Age-specific mortality rates for the two populations. Regression lines reflect the fit of a Gompertz mortality function to the data. For the island and mainland populations, the lines account for 77% and 98% of the variation, respectively, and the slopes are both greater than zero ($P = 0{\cdot}002$ and $0{\cdot}001$). A general linear test concludes that the lines are significantly different ($F_{2,10} = 11{\cdot}505$, $P < 0{\cdot}005$), and the slope of the mainland population ($0{\cdot}091$) falls above the 95% confidence interval of the island population (mean $= 0{\cdot}054$, 95% confidence interval: $0{\cdot}080$–$0{\cdot}028$).

Collagen ageing

Ageing of tail tendon fibres, as measured by fibre breaking time, overlapped extensively in the two populations during the first 18 months of life; however, it was greatly different thereafter (Fig. 3). The overall rate of collagen ageing was more than double on the mainland what it was on the island. Fibre size did not correlate with adult age in either population (mainland: $r = 0{\cdot}009$, $P = 0{\cdot}969$; island: $r = -0{\cdot}249$, $P = 0{\cdot}229$) or differ between populations ($t_{45} = 0{\cdot}864$, $P = 0{\cdot}392$). Only its chemical properties were different.

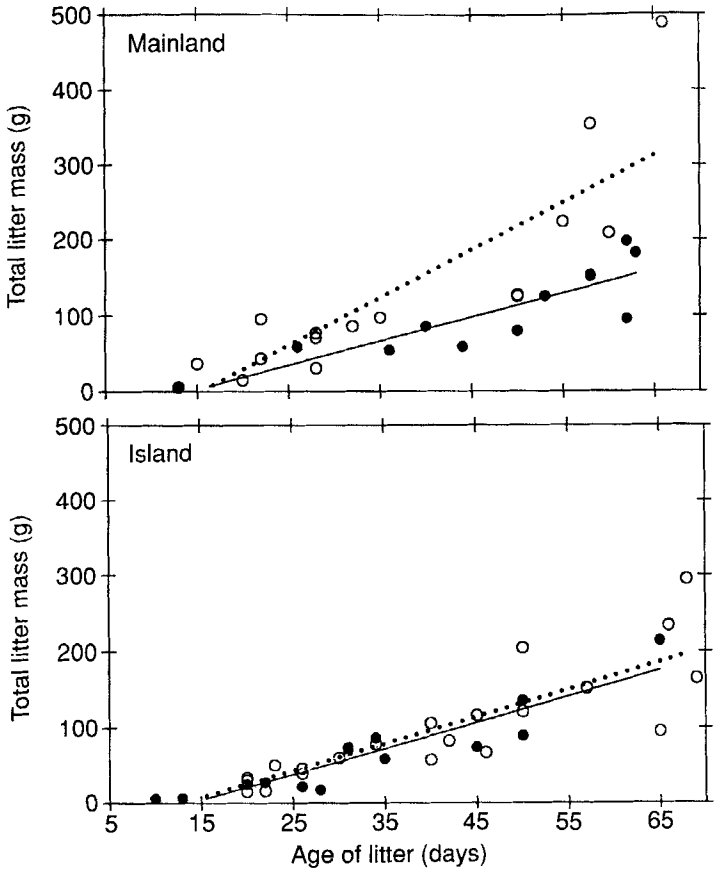

FIG. 2. Maternal age and pouch young growth rate in the two populations: ○ = first-year reproductive females, ● = second-year reproductives. Regression lines do not differ in the island population ($F_{2,31} = 0{\cdot}12$, $P > 0{\cdot}5$), but there is a significant age-related decline in litter growth rates on the mainland ($F_{2,24} = 14{\cdot}0$, $P < 0{\cdot}001$).

Discussion

Survival and reproductive parameters of mainland opossums in this study were roughly similar to those found in other demographic studies of Virginia opossums. For instance, in this study 27% of adult females survived into a second reproductive year and none survived into a third reproductive year, and signs of reproductive senescence were observed during the second reproductive year. In a radiotelemetric study in north-central Florida, USA, Sunquist & Eisenberg (In press) found that 26% of females survived to a second reproductive year, and second-year reproductives were more often infertile. Seidensticker *et al.* (1987) found that only 8% of females in Virginia lived into a second reproductive year, and these older females had smaller litter sizes than in their first reproductive year. Laboratory colonies of opossums also show manifold signs of senescence in their second reproductive year (Jurgelski & Porter, 1974; Harder & Fleming, 1982), especially reproductive senescence.

Litter sizes reported in Virginia opossums range from a mean of 6·3 to 10·0 and tend to increase

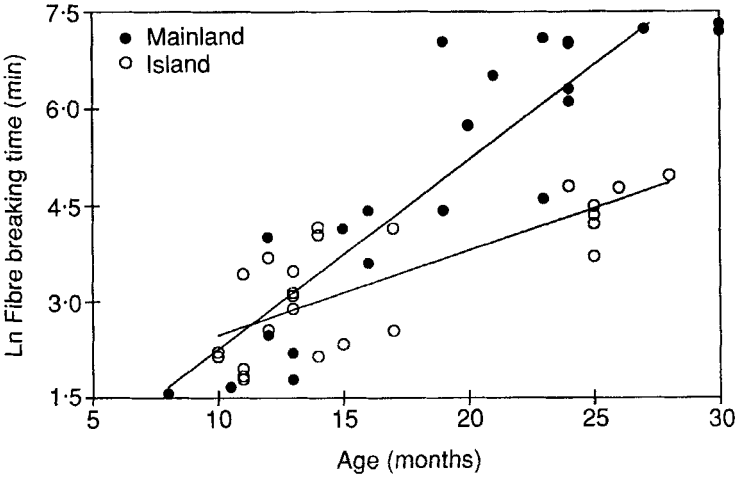

FIG. 3. Tail tendon fibre breaking time. Each point represents the mean breaking time of two to four fibres. The regression slopes are both significant at $P<0{\cdot}001$ and the lines are significantly different ($F_{2,43}=19{\cdot}312$, $P\ll0{\cdot}001$).

in size at more northerly latitudes (reviewed in Gardner, 1982). Therefore, the mainland litter sizes found in this study (mean = 7·6) are also comparable with other studies. Thus this opossum population does not seem in any way atypical of the species.

On the other hand, the insular population differed greatly from previously reported opossum populations. For instance, 50% of females survived into a second reproductive year, and 9% survived into a third reproductive year. Litter sizes were the smallest that have been reported in the species (mean = 5·7) and there was no sign of reproductive decline in the second reproductive year. None of the three females that lived into their third reproductive year was observed to have pouch young, however, raising the possibility that they were post-reproductive.

Life-history differences consisting of either longer life and/or smaller litter size in island populations than in mainland populations have been reported many times previously (e.g. Carlquist, 1974; Tamarin, 1978; Crowell & Rothstein, 1981). There is little evidence bearing upon whether such differences represent evolved genetic differences or simply phenotypic plasticity in the face of differing environments. However, it should be noted that genotypic differences in life-history traits have been discovered even without the physical separation that islands represent (Berven, 1982; Cameron & McClure, 1988), and that substantial life-history differences have been shown to evolve in the laboratory (Mueller, Pingzhong & Ayala, 1991) and field (Reznick, Bryga & Endler, 1990) in as few as 25–60 generations.

Insular opossums underwent senescence consistently more slowly than mainland opossums in terms of mortality rate acceleration, reproductive decline and changes in the chemical nature of tail tendon collagen, and also exhibited lower reproductive rates at all ages than mainland females. These differences are all consistent with the predictions of evolutionary senescence theory.

One of the mechanisms by which senescence is postulated to evolve is antagonistic pleiotropy between early beneficial and late deleterious fitness components (Williams, 1957). Such trade-offs are a central assumption of evolutionary life-history theory (Williams, 1966; Gadgil & Bossert, 1970; Schaffer, 1974; Charlesworth & Leon, 1976). Specifically, there is assumed to be a negative

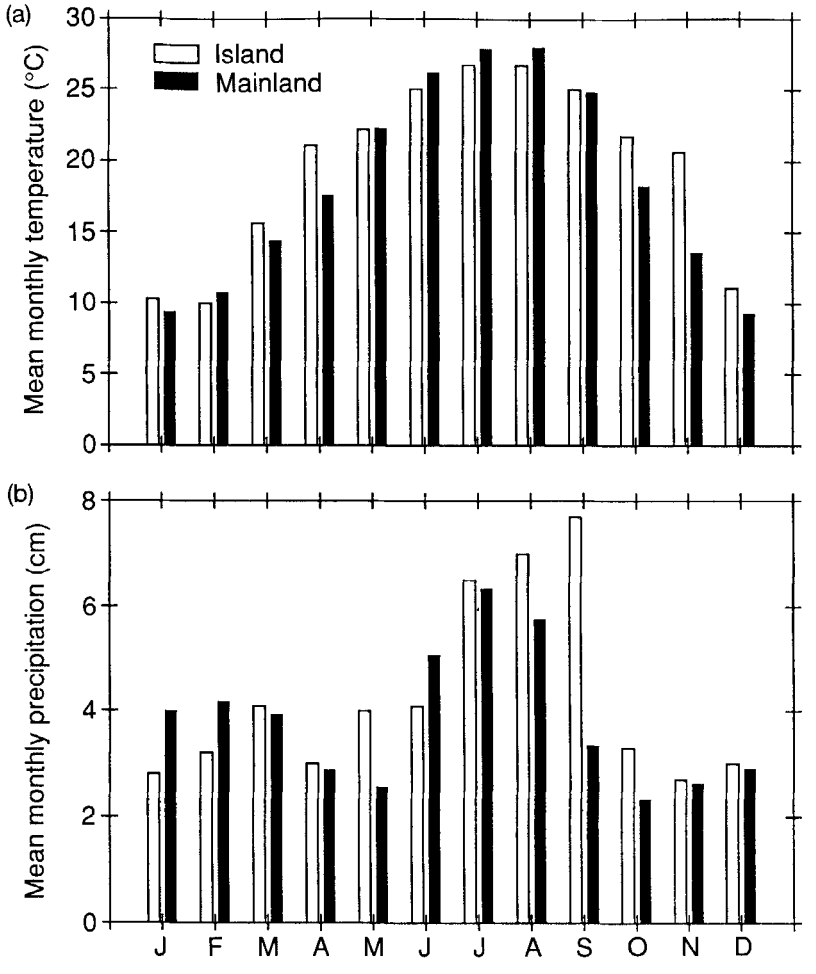

FIG. 4. Comparison of (a) monthly temperatures and (b) precipitation at the two study sites over a four-year period for the mainland and a 10-year period for the island. Paired t-tests reveal no statistical differences between these differences (precipitation: $t_{11} = 1{\cdot}045$, $P = 0{\cdot}312$; temperature: $t_{11} = 1{\cdot}526$, $P = 0{\cdot}155$).

genetic correlation between the speed and/or magnitude of reproduction and subsequent survival. Data from this study suggest that such trade-offs occur in nature. Opossums of the longer-lived island population had smaller litter sizes at all ages than the mainland opossums and they also had a suggestive, but not statistically significant, delay in the age of first reproduction (Table I).

However, because there is no direct evidence that these differences represent genetic differences, the hypothesis must be entertained that some environmental factor is responsible for them, in which case these results would not be relevant to evolutionary senescence theory. It is somewhat difficult to imagine exactly what sort of environmental factor would simultaneously increase longevity, a characteristic of a relatively benign or salubrious environment, and decrease reproductive rate, usually associated with a harsh or stressful environment. Three possible

TABLE II

Environmental differences between opossum (means with S.D. in parentheses)

Character	Island	Mainland	*P*-value
Temperature (°C)	19·7 (6·32)	18·5 (7·15)	—
Precipitation (cm/month)	10·9 (4·48)	9·7 (3·34)	—
Ear tick load	17·7 (12·50)	10·7 (7·85)	0·049 ($t_{34}=2·042$)
1st year breeders	14·2 (11·95)	10·8 (6·34)	0·454 ($t_{17}=0·766$)
2nd year breeders	22·7 (12·35)	10·6 (9·35)	0·036 ($t_{15}=2·308$)
Density (females/km^2)	3·85	0·90	
Range:	3·55–4·89	0·58–1·24	
Body mass index (kg/m^2)	8·22 (1·15)	8·30 (1·24)	0·767 ($t_{69}=0·297$)
Blood glucose (mg/dl)	102·3 (15·9)	91·0 (8·87)	0·126 ($t_{12}=1·644$)

TABLE III

Comparison of opossum blood profiles on mainland and island. Parameters stated as mean (S.D.)

Parameter	Island	Mainland
PCV	44·2 (5·09)	45·4 (7·61)
Glucose	97·3 (23·59)	91·13 (11·26)
BUN	39·83 (8·95)	41·75 (3·20)
Creatinine	0·60 (0·063)	0·70 (0·093)
Albumin	3·22 (0·217)	3·58 (1·02)
Globulin	3·94 (0·688)	4·30 (1·33)
APTase	284·8 (123·95)	271·43 (120·58)
Cholesterol	167·4 (32·07)	146·25 (18·65)
Total bilirubin	0·502 (0·160)	0·747 (0·425)
SGOT	351·2 (184·5)	322·14 (113·86)
SGPT	107·4 (68·79)	116·14 (91·51)

environmental sources of the observed demographic differences will be considered—climate, infection by pathogens or parasites, and resource availability.

The climate is moderately similar, although not identical, at the two sites (Fig. 4). The island has about a 1 °C higher annual mean temperature and 3 cm greater annual mean rainfall with roughly equal variances (Table II). However, the climate at both sites is mild in terms of temperature for opossums, which range as far north as southern Canada (Gardner, 1982) and both areas receive substantial rainfall throughout the year (Fig. 4). The forest at both sites is dominated by loblolly pine (*Pinus taeda*).

Differences as small as 3 °C in temperature have been demonstrated to affect tail collagen breaking time in mice (Harrison & Archer, 1978). However, in those experiments, the tail tendon

aged more slowly at cooler temperatures, whereas the results here show the opposite result if the slight ambient temperature difference is considered significant (Fig. 3). Thus climate seems unlikely by itself to have brought about these demographic and physiological changes.

Perhaps parasites or pathogens with different age-related effects were present on the island or mainland. Dog ticks were the most visible ectoparasite in both populations, although deer ticks and fleas were also abundant. Direct counts of ticks on opossums' ears (the preferred site of attachment) showed marginally higher tick loads in the island population and perhaps a marginally significant age-related difference in the island population (Table II). However, even the raw trends are in the opposite direction from that which might explain the observed differences in mortality rate. Furthermore, sanguinivorous ectoparasites (ticks, fleas) when serious can cause parasitic anaemia, depressing packed cell volume (PCV), a measure of erythrocyte density (Duncan & Prasse, 1986). Yet no differences in PCV were observed between populations ($t_{32} = -0{\cdot}569$, $P = 0{\cdot}573$). Both populations were comfortably within normal range PCV (Table III).

Opossums are subject to a host of diseases (Potkay, 1970), however, and it could be that some important diseases with age-related effects are absent from the island. General blood chemistry results again suggest that this is not so. General blood chemistry values, when compared with normal ranges for the species, are highly reliable indicators of parasitism or disease in a range of physiological systems, including liver, kidney, pancreas and bone, as well as endocrine and immune systems (Duncan & Prasse, 1986).

From the relatively few complete chemical profiles done (eight on the island, six on the mainland), there were no significant differences between populations in any of the measured blood chemistry parameters (Table III). Mean number of parameters outside normal ranges per sample was identical (4·5) on the mainland and island, and even the variance was similar (1·94 vs. 1·89). Thus limited evidence from the blood chemistry suggests no gross difference in disease between island and mainland.

Finally, it might be suggested that these results could potentially stem from food shortage on the island. Details of opossums' diet at the two sites are lacking, but opossums *do* live at much higher densities on the island (Table II), which implies that food may be in shorter supply there. Although food shortages are generally associated with increased mortality in nature (e.g. Balen, 1981; Sullivan & Sullivan, 1982), laboratory animals fed a calorically-restricted diet age more slowly (reviewed in Weindruch & Walford, 1988). Calorie restriction also reduces reproductive rate in both the laboratory and field (Holehan & Merry, 1985*a*, *b*; Bronson, 1989) and retards the ageing of tail tendon (Chvapil & Hruza, 1959; Giles & Everitt, 1967; Everitt *et al.*, 1969). This combination of traits is strikingly similar to those found in the island population.

Two lines of evidence suggest that naturally-occurring dietary restriction does not explain these results. First, a common measure of animal leanness, the body mass index (mass/length^2) (Burton *et al.*, 1985), has been found to differ substantially between rats fed *ad libitum* and calorically-restricted rats (McCay, Crowell & Maynard, 1935). However, this index does not approach a statistically significant difference in the two opossum populations (Table II). Secondly, restricted feeding regimes sufficient to cause retardation of senescence have been demonstrated to lower significantly the concentration of blood glucose (Masoro, Katz & Walton, 1989). Limited data on blood glucose in these two populations show no significant difference (Table II) and even the raw trend is in the wrong direction.

In sum, although it is impossible completely to discount the possibility that the retarded senescence found on this insular opossum population is due to some environmental factor, the

weight of evidence thus far gathered supports the notion that it has evolved a more *K*-selected, more slowly ageing, life history as evolutionary senescence theory suggests it should.

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