

PROCEEDINGS OF THE SECOND
INTERNATIONAL CONFERENCE
ON QUANTITATIVE GENETICS

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1988



Sinauer Associates, Inc. • Publishers
Sunderland, Massachusetts

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Quantitative Genetics and Selection in
Natural Populations: Microevolution of
Vertebral Numbers in the Garter Snake
Thamnophis elegans

The microevolution of vertebral numbers in garter snakes is modeled using quantitative genetic theory and data on inheritance and selection from natural populations in California. Coastal and inland populations of the garter snake *Thamnophis elegans* have diverged more than four phenotypic standard deviations in number of body vertebrae and two standard deviations in number of tail vertebrae. The two traits are highly heritable but show only a weak, positive genetic correlation. Coastal and inland populations had similar genetic parameters. Local demes within each of the regions also yielded similar parameter estimates. The net forces of directional selection required to account for the observed divergence in means were reconstructed on the assumption that the genetic system remained relatively constant on a microevolutionary time scale. That computation suggests that selection on the body was three times more forceful than selection on the tail. Despite the weak genetic correlation, correlated responses to selection on the body apparently played a major role in the microevolution of tail vertebral number.

INTRODUCTION

Ecologists and evolutionary biologists are showing a resurgence of interest in quantitative genetics since it provides the necessary tools to study the inheritance of those polygenic traits (e.g., body size, lifespan, behavioral responses to prey) that typically concern them. In company with empirical developments, a series of theoretical models by Lande and colleagues has invoked genetic covariance in a variety of evolutionary processes (Lande, 1976a, 1979, 1980a, 1981, 1982a,b; Via and Lande, 1985). By developing quantitative genetic

models for the evolution of allometry, sexual dimorphism, mating preference, reproductive isolation, and niche polymorphism, Lande has made quantitative inheritance relevant to a wider audience of biologists. These models have also made biologists more aware of the importance of quantitative genetic effects in long-term evolutionary dynamics and kindled new interest in estimating genetic parameters in natural populations.

The aim of this chapter is to present a case study of inheritance, selection and microevolution in garter snakes. Two traits are considered: the number of vertebrae in the body and in the tail. Results of inheritance studies conducted in two differentiated geographic races of the garter snake *Thamnophis elegans* are given. Within each of these races, differences between local populations in heritability and genetic correlation were tested and the two races compared. Because homogeneous estimates of genetic parameters were found, pooled results were used to estimate the pattern of directional selection that might have been responsible for the observed geographic differentiation in vertebral numbers, and the reconstructed pattern of selection was compared with natural selection measured in one of the populations. Finally, because vertebral numbers may be affected by temperature during embryonic development the possibility of maternal effects was explored. Preliminary results from ongoing studies of maternal thermoregulation are given and a graphical model that integrates various themes from the empirical work is presented.

A CASE STUDY

Background

About 20 species of garter snakes (*Thamnophis*) are distributed in the New World from northern Canada to Central America (Ruthven, 1908). Most garter snakes are riparian animals that forage in the vegetation that lines streams and ponds. Some species forage under water. Annelids, frogs and fish are the most common prey, but there is much differentiation among species and populations (Arnold, 1981b). All species in the genus are viviparous. Typically, young are born in late summer after an 80–100 day gestation. Litter sizes range from a few to a few dozen, depending on species and body size (Fitch, 1985). Litter mates are almost certainly full sibs (Ayres and Arnold, 1983).

The number of vertebrae in the body and tail of a snake can be assayed by counting external scales (Figure 1). Ventral scales correspond to body vertebrae and subcaudal scales correspond to tail vertebrae (Alexander and Gans, 1966; Voris, 1975). Vertebral numbers do not change during postnatal life, except for tail loss. When the tip of the tail is lost by accident or predatory encounter, it is not regenerated.

There is a wide range of variation in average vertebral numbers among species ranging from about 100 to 300 in colubrid snakes (Marx and Rabb, 1972). Because differences between species often vastly exceed the variation within species, vertebral numbers have been used by snake systematists since the time of Linnaeus to diagnose species, genera, and higher taxa. Coefficients of variation within populations are usually only a few percent (Kerfoot, 1969).

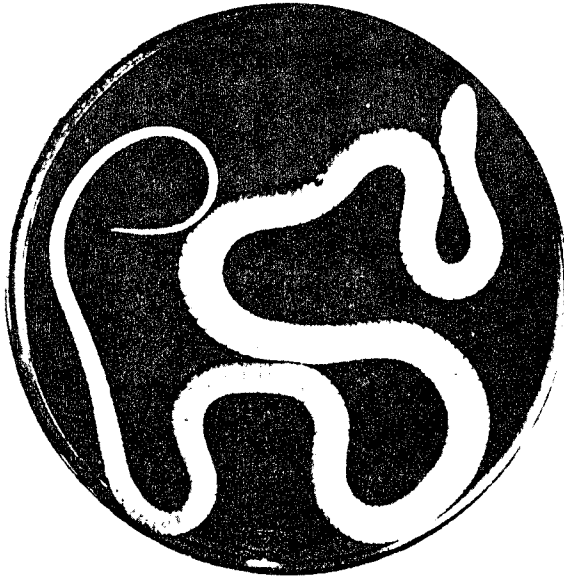


Figure 1. Ventral surface of *Thamnophis elegans* from the inland study site in northern California. The solid arrowhead indicates the anal scale that marks the transition from the body to the tail. The ventral scales, anterior to the anal scale, are usually undivided and correspond to individual body vertebrae. The subcaudal scales, posterior to the anal scale, are usually divided and correspond to individual tail vertebrae. The open arrowhead indicates an abnormal vertebral scale that corresponds to a vertebra with one rib on one side and two ribs on the other. Scales are clipped in the right anterior region of the snake to provide a code for individual recognition in the mark and recapture study of selection.

Often vertebral counts within a population show a normal or log-normal distribution (Kerfoot and Kluge, 1971).

Vertebral numbers often show dramatic microevolution within snake species. For example, Klauber (1941) found that 12 out of 13 broadly distributed species in southern California showed parallel clines in numbers of body vertebrae in one or both sexes. Populations along the coast had fewer vertebrae than populations found in the inland deserts. Other aspects of scalation did not show such clines, and even the number of caudal vertebrae did not show clear coastal-to-inland clines. Only four species showed such clines, and in one of these, the gradient was opposite in direction to that of body vertebrae. This suggests that the two vertebral counts may evolve relatively independently.

Samples and Geographic Variation

Pregnant females of the main study species (*Thamnophis elegans*) were collected from a coastal and an inland site located 282 km apart in northern

California (Arnold, 1981a,b,c). After transport to the laboratory, females were maintained for about the second half of pregnancy. During this interval females had access to a thermal gradient (about 20 to 40°C, produced by substrate heating). The gradients were turned on in phase with daytime of a natural (Chicago) photoperiod. At night cages were kept at a spatially uniform 20 to 22°C. Females were collected over a 10-year period in an effort to obtain approximately 100 dams and 1000 offspring from each of the main geographic sites in Humboldt and Lassen Counties, California.

Within each of the main sites, pregnant females were collected at two local populations. In inland California, the two populations were located along the southern shore of Eagle Lake, about 3 km apart (Pikes Point and Wildcat Point). Some gene flow probably occurs between these populations, even though the intervening shoreline is largely uninhabited by *T. elegans*. Of several hundred snakes marked and recaptured, only a single male moved between the two sites sometime during a 3-year interval. The variance effective population number at one of these localities (Pikes Point) is a few hundred (Kephart, 1981). The two populations in coastal California are situated about 4 km apart, north and south of the mouth of the Mad River in coastal sand dunes. No mark-recapture work has been conducted there.

Numbers of body vertebrae vary slightly between local populations but differ greatly between the two regions (Table 1). Divergence among local populations averages less than 1% of the mean or about 0.4 phenotypic standard deviation. Divergence between the coastal and inland regions confirms Klauber's (1941) observation that inland snakes showed more body vertebrae but the divergence is slightly larger than he noticed, amounting to a 10% difference in the mean or more than four phenotypic standard deviations.

Numbers of tail vertebrae generally show less differentiation than numbers of body vertebrae (Table 1). Differentiation between the coastal populations is comparable to differentiation in body vertebrae, but the inland divergence is only 0.02–0.07 phenotypic standard deviation. Between the coastal and inland regions the tail vertebrae show about half as much differentiation as body vertebrae, 1.9–2.4 phenotypic standard deviations.

Sexual dimorphism in vertebral numbers is conspicuous in many snakes, including *Thamnophis*. *T. elegans* males have about five more vertebrae in the body and about 10 more vertebrae in the tail than do females. Because the degree of sexual dimorphism is relatively invariant among populations and between the two regions, we concentrate on the analysis of divergence in the sexual average number of vertebrae. Accordingly, sex-corrected data are employed in the analyses that follow.

Temperature during pregnancy is known to affect vertebral numbers in snakes (Fox, 1948; Fox et al., 1961; Osgood, 1978), so the coastal-inland difference might reflect the warmer climate experienced by inland snakes and their embryos. Three considerations, however, argue against this possibility. First, the regional divergence is four to eight times larger in magnitude than the largest effects of temperature effects produced in the laboratory. In the best study, Osgood (1978) was able to shift the mean number of body vertebrae by less than one standard deviation. Second, crosses between the coastal and

Table 1. Phenotypic means (\pm phenotypic standard deviation) for the two traits in newborn *Thamnophis elegans* from each of two local populations within each of the two regions in northern California. Sample sizes are shown in parentheses. Tests for differences in means between local populations were conducted by analysis of variance using litter means. Within each region, local populations differ in number of body vertebrae in both sexes ($P < 0.05$ or < 0.01). Males from the two coastal populations differ in number of tail vertebrae ($P < 0.01$) but other local differences in tail vertebrae are not significant ($P > 0.05$).^a

		Coastal California			Inland California			Divergence between coastal and inland California in units of average phenotypic standard deviation
		Local population ^b		Population average ^d	Local population ^c		Population average ^d	
		A	B		C	D		
Number of body vertebrae	Males	156.0 \pm 2.45 (100)	157.0 \pm 3.28 (293)	156.5 \pm 2.86	172.2 \pm 3.62 (422)	173.6 \pm 5.21 (336)	172.9 \pm 4.42	4.5
	Females	151.2 \pm 3.63 (124)	152.8 \pm 3.39 (357)	152.0 \pm 3.51	167.1 \pm 3.24 (388)	168.8 \pm 4.78 (340)	167.9 \pm 4.01	4.2
Number of tail vertebrae	Males	79.4 \pm 6.57 (100)	81.4 \pm 3.67 (291)	80.4 \pm 5.12	90.2 \pm 4.83 (423)	90.6 \pm 6.48 (332)	90.4 \pm 5.61	1.9
	Females	70.2 \pm 4.21 (129)	71.6 \pm 3.48 (366)	71.2 \pm 3.73	80.6 \pm 3.84 (390)	80.7 \pm 4.41 (334)	80.7 \pm 4.11	2.4
Phenotypic correlation	Males	0.08 ^{ns} (96)	0.20 ^{**} (286)	0.14	0.20 ^{**} (362)	0.06 ^{ns} (325)	0.13	
	Females	0.34 ^{**} (121)	0.11 [*] (353)	0.22	0.21 ^{**} (417)	0.19 ^{**} (325)	0.20	

^a ns, $P > 0.05$; *, $P < 0.05$; **, $P < 0.01$.

^b A, population 4 km north of the mouth of the Mad River; B, population just south of the river mouth, Humboldt Co., CA.

^c C, Wildcat Point; D, Pikes Point, Eagle Lake, Lassen Co., CA.

^d Computed as the mean of the two population averages when the populations showed statistically significant differences. Computed from pooled data when local populations did not show significant differences.

inland sites yield F_1 with intermediate vertebral numbers and no sign of a maternal effect. Third, females thermoregulate during pregnancy by selecting microhabitats and so may buffer their embryos against differences in climate, and in the present study they were allowed access to a thermal gradient. Thus, environmental temperature can be excluded as the proximate cause of the divergence.

Vegetation is more dense on the coast in both northern and southern California and may be a major selective factor in promoting coastal-inland divergence. Larger vertebral numbers may promote greater flexibility of the vertebral column (Jayne, 1986) but the functional connection to vegetation density is unknown.

The analyses that follow leave the identity of selective agents unresolved and concentrate instead on the magnitude of the putative selective forces that might have produced the observed divergences. In order to make those computations we first need to consider the systems of inheritance that prevail within populations.

Inheritance

The main issues are (1) the magnitudes of genetic variances and covariances, (2) whether genetic parameters differ among local demes, and (3) whether genetic parameters differ among geographic races.

Genetic parameters were estimated from regressions of offspring on dam's value, weighted by litter size (equal weighting of litters gave similar estimates). All computations were based on sex-corrected data. The genetic correlations in Table 2 were computed from the average of the two possible mother-offspring cross covariances (Bulmer, 1980; Becker, 1984). Tests for heterogeneity of estimates between local populations and between the two geographic regions were conducted by analysis of covariance. Pooled estimates for each region were made after first adjusting local populations to a common mean.

Numbers of body and tail vertebrae showed moderate to high heritabilities within local populations (Table 2). Genetic correlations were positive, moderate in magnitude, and somewhat larger than corresponding phenotypic correlations. Within both geographic regions, local populations yielded statistically indistinguishable genetic parameter estimates.

Pooled data for both of the geographic regions gave moderate to high heritabilities for the two traits and moderate positive genetic correlations between them (Table 2). Thus, despite considerable divergence between the two regions in the phenotypic means of both traits, genetic parameter estimates were statistically indistinguishable.

Selection and Performance

Growth rate is likely to be a major fitness component in both sexes because faster growth shortens the interval to sexual maturity and, in females, because clutch size is strongly size-dependent. If so, part of the adaptive significance

Table 2. Heritability and genetic correlation estimates for each of two local populations within each of the two regions in California. Standard errors are shown after each estimate. These were computed using approximations given by Falconer (1981). Sample sizes are indicated as (number of litters, number of offspring).

	Coastal California			Inland California		
	Local population		Pooled data	Local population		Pooled data
	A	B		C	D	
Heritability of number of body vertebrae	0.52 ± 0.29 (22,153)	0.70 ± 0.16 (72,627)	0.65 ± 0.14 (94,780)	0.61 ± 0.13 (72,795)	0.96 ± 0.18 (87,664)	0.79 ± 0.11 (159,1459)
Heritability of number of tail vertebrae	0.50 ± 0.20 (25,202)	0.44 ± 0.14 (59,509)	0.46 ± 0.11 (84,711)	0.64 ± 0.15 (58,625)	0.64 ± 0.20 (67,502)	0.64 ± 0.13 (125,1127)
Genetic correlation between number of body vertebrae and number of tail vertebrae	0.48 ± 0.25 (25,198)	0.30 ± 0.05 (72,632)	0.36 ± 0.14 (97,830)	0.37 ± 0.03 (72,798)	0.22 ± 0.04 (87,653)	0.28 ± 0.02 (159,1451)

of minor differences in vertebral numbers can be discerned by their effects on growth. A longitudinal study of growth in the field was therefore conducted at the inland California study site. In addition, the relationship between crawling performance and vertebral numbers was evaluated in a second species, *T. radix*.

In the growth study, nearly 1000 *T. elegans* were captured along a 600-meter section of shoreline at the inland site (Eagle Lake) over a 6-year period. This study site lies 1 km east of the easternmost population sampled in the inheritance study and is separated from that site by uninhabited sandy beach. Each snake was individually marked, measured, and weighed. Vertebral numbers were counted, a variety of scalation and coloration traits were scored, and then the snake was released. Seventy-one males and 69 females were recaptured. Due to sexual dimorphism, the sexes were analyzed separately.

A multivariate, curvilinear regression was computed to see if relative growth rate could be predicted by numbers of body and tail vertebrae (as well as the numbers of abnormal body and tail vertebrae), the squared number of body vertebrae, the squared number of tail vertebrae, and the product of the numbers of body and tail vertebrae using procedures outlined by Lande and Arnold (1983). The coefficients of these multiple regressions are the so-called directional and stabilizing selection gradients (Lande and Arnold, 1983). These gradients reflect the direct forces of directional and stabilizing selection acting through growth rate provided that some correlated, unmeasured trait is not the actual target of selection.

In males, only the number of abnormal body vertebrae was a statistically significant and negative predictor of growth. Surprisingly, this relationship did not hold in females. Instead, females with intermediate numbers of body and tail vertebrae grew at the fastest rates (directional selection gradients for both traits were zero; stabilizing selection gradients were negative and statistically significant). The correlational selection gradient, corresponding to the coefficient for the product term, was positive and significant. Figure 2 portrays a two-dimensional projection of the complete multiple regression equation. The set of gradients corresponds to a growth surface consisting of a level ridge oriented as shown in the figure. The population mean is situated on the crest of this ridge. The ridge implies that there is no unique combination of body and tail vertebrae that confers optimal growth. Rather, any of a large number of vertebral combinations is associated with optimal growth. In other words, the optimal number of tail vertebrae depends on the number of body vertebrae.

The conclusion that selection might favor an intermediate number of body vertebrae is supported by previous work on related natricine snakes showing that samples of juveniles typically have greater variance in number of body vertebrae than do adult samples, but the same mean (Inger, 1942, 1943; Beatson, 1976).

We can gain some insight into the patterns of stabilizing and correlational selection revealed in the field study by considering the crawling performance of snakes in the laboratory. Each of 100 newborn *T. radix* was tested on two consecutive days for ability to crawl over a 1-meter stretch of racetrack while being chased by an investigator (Arnold and Bennett, 1984). The test is eco-

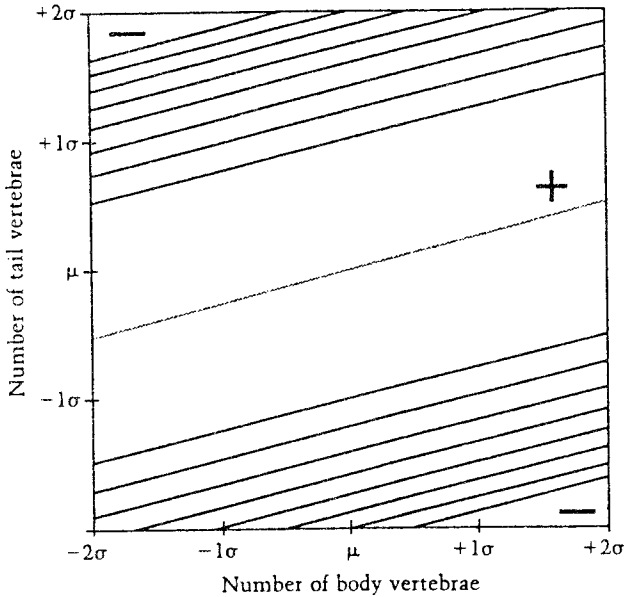


Figure 2. Growth surface fitted to longitudinal data from the inland site. The surface portrays the fitted surface for female *T. elegans* ($n = 69$): residual growth = $\alpha + \gamma_{11} = (z_1^2/2) + \gamma_{22}(z_2^2/2) + \gamma_{12}z_1z_2$, where α is a constant, z_1 is the number of vertebrae in the body, z_2 is the number of vertebrae in the tail, $\gamma_{11} = -0.706$, $\gamma_{22} = -11.938$, and $\gamma_{12} = 5.891$. The coefficients are for independent variables standardized to unit standard deviations (see Lande and Arnold, 1983).

logically meaningful because garter snakes often bask within 1 m of a retreat and speed over that distance can determine the outcome of attacks by raptors and other predators. Speed in the 1-m dash was moderately repeatable ($r = 0.55$) and the average of two trials was used as each snake's performance score. Vertebral counts were also made on each snake and body size was measured in order to test for effects on performance. The stabilizing performance gradient for body vertebral number was negative and the corresponding gradient for tail vertebral number was positive but both were not significantly different from zero. The correlational performance gradient, however, was positive and statistically significant. This result suggests that there is a premium in having properly proportioned numbers of body and tail vertebrae. The two-dimensional (body and tail vertebrae) projection of the complete multiple regression is portrayed in Figure 3.

Reconstructing the Net Selection Gradient

Using the observed genetic variances and covariance within populations, we can compute the cumulative force of directional selection needed to account for the observed divergence in population means (Lande, 1979), assuming that

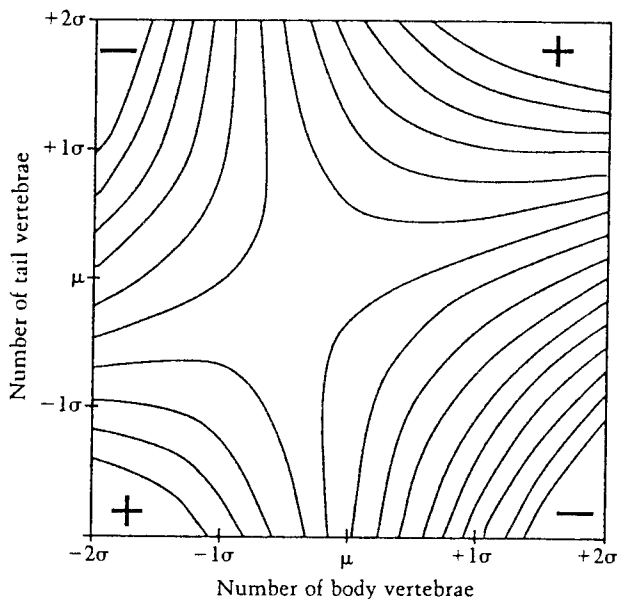


Figure 3. Crawling performance surface for newborn *T. radix* ($n = 100$). The surface portrays the fitted equation: relative burst speed = $\alpha + \beta_1 z_1 + \beta_2 z_2 + \gamma_{11}(z_1^2/2) + \gamma_{22}(z_2^2/2) + \gamma_{12}z_1z_2$, where α is a constant, z_1 is the number of body vertebrae, z_2 is the number of tail vertebrae, $\beta_1 = -0.013$, $\beta_2 = 0.029$, $\gamma_{11} = -0.040$, $\gamma_{22} = 0.016$, and $\gamma_{12} = 0.072$. Variables are standardized as in Figure 2. (Data from Arnold and Bennett, 1984.)

the genetic parameters have remained relatively constant during the process of differentiation. In the present case this assumption of constancy is buttressed by the result that genetic parameters are not detectably different among local populations or between the two regions in northern California. The method yields only the net force of selection needed to account for phenotypic divergence. The sister populations may actually have traversed a complicated trajectory that included evolutionary reversals. The method yields a minimal estimate of directional selection, not necessarily the actual sum of the forces that acted each generation.

The two vertebral numbers have been treated in isolation from other traits, because preliminary calculations showed either no genetic correlation with other meristic traits (head scalation and scales around the body) or extremely weak coupling.

Sexes have been treated separately in these computations and genetic correlations between the sexes were ignored. Since there is relatively little divergence in sex dimorphism among the populations, suggesting that the populations have completed the slow evolutionary phase of elaborating sexual differences (Fisher, 1958; Lande, 1980a), the estimates of net selection on the sexual averages should be taken as averaging the selection acting on both sexes.

The differentiation between local populations within each region in vertebral numbers amounts to less than half a phenotypic standard deviation and might have been produced by selection and/or drift. Nevertheless, we can inquire about the pattern of selection that would be required to account for the local divergence, under the hypothesis that a deterministic process of selection was the main force.

Divergence between the two inland populations can be accounted for largely by directional selection acting only on the number of body vertebrae (Appendix 1). Differentiation between the two coastal populations could be accounted for by comparable forces of selection acting on both the body and tail. The small magnitude of all these gradients can be appreciated by contrasting them with the net selection gradient for the two regions.

To account for the divergence between inland and coastal California, we must invoke cumulative forces of selection that are 7 to 11 times stronger than the forces needed to account for differentiation of local populations. The net selection gradient for regional divergence was computed using the average of the coastal and inland genetic variance-covariance matrices (Appendix 2). The net gradient for body vertebrae is more than three times stronger than the net gradient for tail vertebrae (Figure 4). We can use these results to estimate how much of the divergence in number of body vertebrae was due to selection acting directly on body vertebrae and how much arose as a correlated response to selection acting on tail vertebrae. Ninety-one percent of the total response can be attributed to direct selection. In the case of numbers of tail vertebrae, however, only 52% of total divergence can be attributed to selection acting directly on tail vertebrae. Thus correlated responses to selection apparently played a major role in the observed evolution

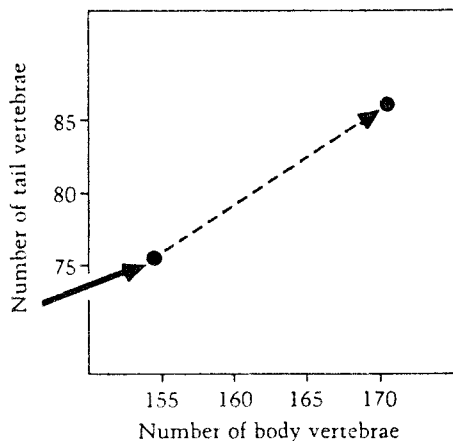


Figure 4. Net selection gradient (solid arrow) that accounts for coastal to inland divergence in the vertebral numbers of *T. elegans* in light of the observed genetic variances and covariance for the traits. Means of the coastal and inland populations are shown with dots. The length of the net selection gradient is arbitrary.

of tail vertebral number. The result is striking because the genetic correlation between the two traits is only 0.32.

The Possibility of Maternal Effects

Maternal effects could complicate our interpretation of maternal-offspring covariance (interpreted simply as a genetic variance in Table 2) and our reconstructions of selection (Appendices 1 and 2), which are based on that interpretation. A genetic element to maternal effects on vertebral numbers would mean that maternal-offspring covariance is composed of contributions from additive genetic variance for both direct and maternal effects, additive genetic covariance between direct and maternal effects, dominance covariance between direct and maternal effects (Willham, 1963), as well as environmental covariance between direct and maternal effects. Thus the estimates of selection response might be under- or overestimated, depending on the signs of the three contributing covariances.

Work on vertebral inheritance in garter snakes has not proceeded far enough to yield estimates of the zygotic and maternal genetic parameters. The main goals in current work with C.R. Peterson on maternal thermoregulation in garter snakes are (1) to determine the body temperatures of free-ranging pregnant mothers using radiotelemetry in the field, (2) to determine whether mothers differ in their thermoregulatory performance and whether that variation is heritable, and (3) to determine the impact of that maternal variation in thermoregulation on the vertebral numbers of progeny. So far, we know most about the impact of temperature on embryos.

Temperature during development affects vertebral number and other meristic traits in all classes of vertebrates, but the phenomenon has been studied most in fish (Hubbs, 1922; Fox, 1948; Tåning, 1952; Yntema, 1960; Barlow, 1961; Lecyk, 1965; Lindsey and Moodie, 1967; Fowler, 1970; Lindsey and Harrington, 1972; Peabody and Brodie, 1975). The population norm of reaction to temperature is commonly U-shaped in fish, with the lowest number of vertebrae at intermediate temperatures and with higher numbers of vertebrae at both cold and warm temperatures (Fowler, 1970). Linear norms of reaction also are common, with vertebral numbers decreasing with increasing temperature (Lindsey and Harrington, 1972). Osgood (1978) held sets of pregnant water snakes (*Nerodia fasciata*) at three different constant temperatures and obtained a V-shaped response in the vertebral numbers of progeny. The impact of temperature on vertebral numbers was modest (the average count of 130 vertebrae was changed by 1–3 vertebrae, or 35–92% of a standard deviation by the extreme temperatures) but statistically significant. In preliminary experiments with *T. elegans*, we also found curvilinear norms of reaction to temperature by meristic traits.

The temperature at the inflection point for the norms of reaction in *T. elegans* might correspond to the average temperature experienced by embryos during pregnancy. Part of that average temperature is actively created by female thermoregulation (Peterson, 1987). It is intriguing to speculate that the inflection point of the population reaction norm might correspond to the

average embryonic temperature. A major goal of our ongoing studies is to test this possibility by holding pregnant females at a series of temperatures.

A Graphical Model for Selection on Reaction Norms in Equilibrium Populations

A graphical model helps illustrate the relationships between selection on vertebral numbers, temperature effects, and thermoregulation. A simplified model, shown in Figures 5 to 7, supposes that the population is in evolutionary equilibrium. A hypothetical population average reaction norm is shown in Figure 5. Individual variation about that average, the frequency distribution of vertebral numbers in a population experiencing an average environment \hat{E} , and stabilizing selection on vertebral numbers are illustrated in Figure 5.

We can use the model to explore how the population will evolve in response to a small deviation from the optimum or to a gradual change in conditions. In particular, consider the buffering effect of the U-shaped reaction norm. If the environment varies in the neighborhood of the inflection point, there is virtually no change in progeny phenotype, z . Variation about \hat{E} seems inevitable, even when the embryo's environment is buffered by active thermoregulation. Consequently, the reaction norm whose inflection point corresponds to the intersection of Θ and \hat{E} will be the most buffered against environmental variation (Figure 7). If the population reaction norm evolves simply by changing position in the phenotype–environment space without changing shape, selection acting on the trait (e.g., vertebral number) will move

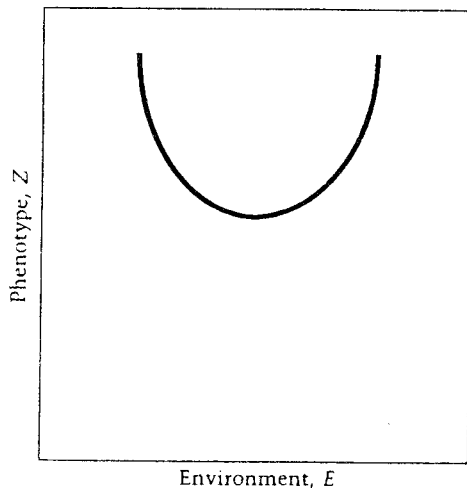


Figure 5. Hypothetical population norm of reaction of a type commonly found in experimental studies of meristic traits in fish and snakes. Each point on the curve represents the mean of many embryos developing in a particular environment (e.g., at a particular temperature).

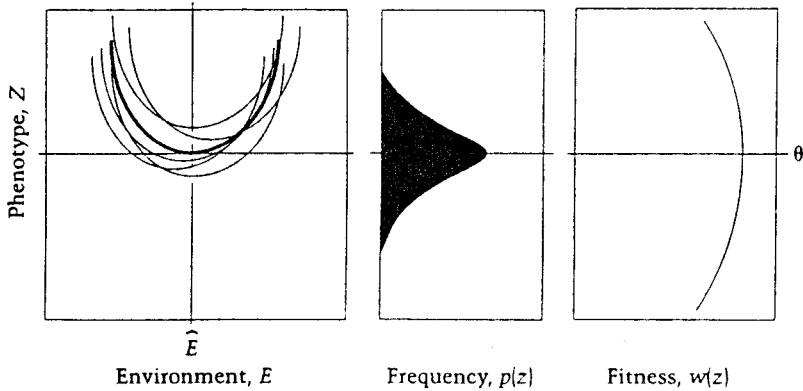


Figure 6. Model for reaction norms in an equilibrium population under stabilizing selection for phenotypic trait, z . The prevailing average environment is denoted \bar{E} . (a) The average norm of reaction is shown as a heavy curve; a representative sample of individual reaction norms are shown as light curves. (b) The frequency distribution of the quantitative trait, z . (c) Stabilizing selection toward an intermediate optimum, Θ .

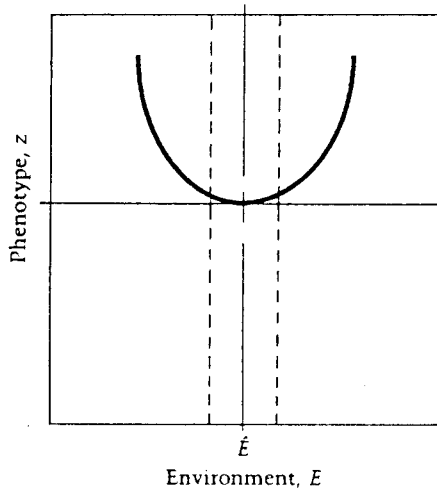


Figure 7. Hypothetical population norm of reaction showing the relative invariance of phenotype, z , within the range of environmental variation indicated by the dashed lines.

the distribution of reaction norm toward the optimal reaction norm shown as a heavy curve in Figure 6.

In the case of temperature-dependent vertebral numbers in garter snakes, the temperature environment is not fixed but is instead created in part by maternal thermoregulation, and so is presumably capable of evolving. Consequently, the equilibrium value for average thermoregulatory performance might be a function of selection acting directly on thermoregulation as well

as selection on vertebral numbers. For example, there might be a premium on relatively high body temperatures that shorten the length of the gestation period. Thus, the equilibrium thermoregulatory performance might be a compromise between selection acting directly on maternal thermoregulation and opposing selection acting on vertebral numbers.

The joint evolution of maternal thermoregulation and temperature-sensitive phenotypes will be affected by genetic parameters as well as by selection. In particular, negative (positive) covariance between direct genetic effects and maternal genetic effects on vertebral numbers can retard (accelerate) the short-term response to selection (Dickerson, 1947; Cheverud, 1984). A negative genetic covariance of sufficient magnitude could cause the population to evolve toward lower values of vertebral number and maternal thermoregulation, even though selection favors higher values for both variables. Whether the genetic covariance is negative or positive, the long-term evolutionary effect is likely to be curvature of the trajectory of evolving trait means and temporary maladaptation of one of the traits, as Lande (1980a) found in the case of evolving sex dimorphism. Thus a crucial goal for our empirical work is to measure the correlation between direct genetic and maternal genetic effects.

DISCUSSION

The most general conclusion we can reach is that it is feasible to study quantitative inheritance in natural populations. Even in organisms in which it is difficult or impossible to implement standard breeding designs (e.g., birds, reptiles), it may be feasible to assemble large series of parent-offspring sets and achieve good parameter estimates. Using garter snakes, this was accomplished by repeatedly sampling a set of populations over a series of years. Investigators working with hole-nesting passerine birds (e.g., great tits, flycatchers) have saturated the environment with nest boxes and so gained access to numerous families (van Noordwijk et al., 1981; Alatalo and Lundberg, 1986; Gustafson, 1986). Alternatively, researchers have worked on small islands where passerines were either tame or nests were easily accessible (Grant, 1986; Smith and Zach, 1979; Smith and Dhondt, 1980). Quantitative genetic studies in nature will undoubtedly expand as workers find new techniques that enable them to master the key step of getting the large sample sizes needed for parameter estimates.

At least two conceptual models of selection are consistent with the *T. elegans* results. The reconstruction of net selection gradients suggests that selection acted more strongly on the body than on the tail in producing coastal-inland differentiation in vertebral numbers, but different pictures of selection within populations are consistent with that result. On the one hand, the coastal and inland populations might both be exposed to bivariate stabilizing selection but for different, intermediate numbers of body and tail vertebrae. Thus contemporary divergence might have evolved because daughter populations tracked hill-shaped adaptive surfaces whose hilltops moved farther and farther apart. The longitudinal study of inland female *T. elegans* revealed downward curvature in the growth component of fitness for both the body

and tail vertebral counts. On the other hand, the coastal and inland means might reside at different points on the same adaptive surface and might rest at two far-flung points along the crest of an adaptive ridge. This possibility is suggested both by the longitudinal study of female growth in *T. elegans* and by the study of crawling performance in *T. radix*, which revealed positive correlational selection on numbers of body and tail vertebrae. Under this view, populations might differentiate along a line of equilibria under the combined action of drift and selection. Such lines of equilibria and differentiation have been found in evolutionary models in which selection acts on only one of two traits (Lande, 1981, Kirkpatrick, 1982). In the present formulation, *both* traits might experience stabilizing selection. A level ridge could prevail if selection also favored a particular ratio in numbers of body and tail vertebrae.

Two improvements in our understanding of selection would be needed to resolve these two alternatives. First, it would be highly desirable to obtain estimates of lifetime fitness so that total selection on the two traits could be measured (such estimates may be impractical in *Thamnophis* because of their long lifespans). Is the topography of total selection similar to the selection topographies for growth and locomotory performance, or is it completely different? Second, it would be useful to compare the selection surfaces estimated for differentiated populations to see if they do indeed reside at different local maxima.

The rate of evolution of genetic variances and covariances is an outstanding empirical issue. These genetic parameters might evolve so slowly that they can be considered as constants in relation to the more rapid evolution of phenotypic means (Lande, 1976b, 1980b, 1984). Turelli (1984, 1985), however, has argued that the input of genetic variation from mutation may not be enough to yield appreciable genetic variance and covariance. Equilibrium variances and covariances under a mutation-selection balance critically depend on unknown distributions of allelic effects. Nevertheless, evolutionary constancy of genetic parameters can be viewed as an empirical issue separate from the controversy over the magnitude of mutational input (Turelli, Chapter 54). Constancy can be tested with comparative studies (Arnold, 1981a; Atchley et al., 1981; Ayres and Arnold, 1983; Lofsvold, 1986). Thus, one tentative conclusion emerging from the few studies done so far, including the present one, is that genetic parameters seem to be relatively constant among geographic races of the same species. Furthermore, the *Thamnophis* results are strikingly similar to the heritability estimate of 0.75 for body vertebral number that Beatson (1976) obtained for a species in the closely related genus *Nerodia*.

APPENDIX 1

Reconstructing the Net Selection Gradients for Divergence between Local Populations

Denote the change in phenotypic means (evaluated before selection) from one generation to the next due to selection as

$$\Delta \bar{z} = G\beta$$

where $\Delta\bar{z}$ is a column vector, \mathbf{G} is the additive genetic variance-covariance matrix, and β is a column vector of selection gradients representing the direct forces of directional selection on the traits (Lande, 1979). If \mathbf{G} remains constant over time, then the net change in phenotypic means over a period of generations is

$$\Sigma \Delta\bar{z} = \mathbf{G}\Sigma\beta$$

with summation over generations. The net selection gradient is denoted $\Sigma\beta$. The net divergence in the means of two sister populations, $\Sigma\Delta\bar{z}$, is a summation over the same period as the summation in selection. Consequently, we can take the inverse of \mathbf{G} and solve for the net selection gradient

$$\Sigma\beta = \mathbf{G}^{-1}\Sigma \Delta\bar{z}$$

(Lande, 1979).

For example, consider the divergence between the two coastal populations. Arbitrarily, let the divergence be from smaller to large values, the number of body vertebrae be the first trait in each matrix, and the number of tail vertebrae be the second. Then the average divergence for the two sexes is

$$\Sigma\Delta\bar{z} = \begin{pmatrix} 1.30 \\ 1.70 \end{pmatrix}$$

and the pooled estimate of genetic parameters is

$$\mathbf{G} = \begin{pmatrix} 25.7549 & 10.7756 \\ 10.7756 & 35.0388 \end{pmatrix}$$

with inverse

$$\mathbf{G}^{-1} = \begin{pmatrix} 0.0446 & -0.0137 \\ -0.0137 & 0.0328 \end{pmatrix}$$

Solving for the net selection gradient, we obtain

$$\Sigma\beta = \mathbf{G}^{-1}\Sigma\Delta\bar{z} = \begin{pmatrix} 0.035 \\ 0.038 \end{pmatrix}$$

The average divergence for the two sexes between the two inland populations is

$$\Sigma\Delta\bar{z} = \begin{pmatrix} 1.54 \\ 0.25 \end{pmatrix}$$

Using the pooled sex-corrected data to estimate the genetic variance-covariance matrix as

$$\mathbf{G} = \begin{pmatrix} 45.1663 & 11.9303 \\ 11.9303 & 39.5558 \end{pmatrix}$$

we obtain

$$\Sigma\beta = \begin{pmatrix} 0.035 \\ -0.004 \end{pmatrix}$$

as the net selection gradient.

If we had measured divergence from larger to smaller trait values, we would obtain the same net selection gradients, but with reversed signs.

APPENDIX 2

Reconstructing the Net Selection Gradient for Regional Divergence

Employing the same conventions as in Appendix 1, the average divergence for the two sexes is

$$\Sigma\Delta\bar{z} = \begin{pmatrix} 16.21 \\ 9.69 \end{pmatrix}$$

Taking the average of coastal and inland variance-covariance matrices reported in Appendix 1, we obtain

$$\bar{\mathbf{G}} = \begin{pmatrix} 35.4606 & 11.3530 \\ 11.3530 & 37.2973 \end{pmatrix}$$

which has the inverse

$$\bar{\mathbf{G}}^{-1} = \begin{pmatrix} 0.0312 & -0.0095 \\ -0.0095 & 0.0297 \end{pmatrix}$$

Solving for the net selection gradients as before, we obtain

$$\Sigma\beta = \begin{pmatrix} 0.414 \\ 0.134 \end{pmatrix}$$

Acknowledgments

The work reported here could not have been done without the untiring devotion of Jean Gladstone to her laboratory work. Collaborations with A. Bennett, R. Lande, C.R. Peterson and M. Wade profoundly affected the direction of the work. L. Houck, C.R. Peterson, D. Kephart and many snake campers helped with the field work. N. Lopes made many of the scale counts, P. Phillips drew the performance/selection surfaces and J. Steadman curated the preserved animal collection. I am grateful to J.S.F. Barker, G. Namkoong, D.S. Falconer, T. Garland, Jr., B. Jayne, R. Lande, C.R. Peterson and P. Phillips for helpful discussions and comments on the manuscript. This work was supported by grants from the National Science Foundation and the Public Health Service.

Literature Cited

- Alatalo, R.V. and A. Lundberg. 1986. Heritability and selection on tarsus length in the pied flycatcher. *Evolution* 40: 574-583. [7, 55]
- Alexander, A.A. and C. Gans. 1966. The pattern of dermal-vertebral correlation in snakes and amphisbaenians. *Zool. Mededelingen. Leiden* 41: 171-190. [55]
- Arnold, S.J. 1981a. Behavioral variation in natural populations. I. Phenotypic, genetic and environmental correlations between chemoreceptive responses to prey in the garter snake, *Thamnophis elegans*. *Evolution* 35: 489-509. [7, 53, 55]
- Arnold, S.J. 1981b. The microevolution of feeding behavior. In *Foraging Behavior: Ecological, Ethological and Psychological Approaches*, A. Kamil and T. Sargent (eds.), Garland STPM Press, New York. pp 409-453. [55]
- Arnold, S.J. 1981c. Behavioral variation in natural populations. II. The inheritance of feeding response in crosses between geographic races of the garter snake, *Thamnophis elegans*. *Evolution* 35: 510-515. [55]
- Arnold, S.J. and A.F. Bennett. 1984. Behavioural variation in natural populations. III. Antipredator displays in the garter snake, *Thamnophis radix*. *Anim. Behav.* 32: 1108-1118. [55]
- Atchley, W.R., J.J. Rutledge and D.E. Cowley. 1981. Genetic components of size and shape. II. Multivariate covariance patterns in the rat and mouse skull. *Evolution* 35: 1037-1055. [7, 55]
- Ayres, F.A. and S.J. Arnold. 1983. Behavioural variation in natural populations. IV. Mendelian models and heritability of a feeding response in the garter snake, *Thamnophis elegans*. *Heredity* 51: 405-413. [55]
- Barlow, G.W. 1961. Causes and significance of morphological variation in fishes. *Syst. Zool.* 10: 105-117. [55]
- Beatson, R.R. 1976. Environmental and genetic correlates of disruptive coloration in the water snake, *Natrix s. sipedon*. *Evolution* 30: 241-252. [7, 55]
- Becker, W.A. 1984. *Manual of Quantitative Genetics*. 4th Ed., Academic Enterprises, Pullman. [55]
- Bulmer, M.G. 1980. *The Mathematical Theory of Quantitative Genetics*, Clarendon Press, Oxford. [6, 7, 8, 9, 11, 37, 54, 55]
- Cheverud, J.M. 1984. Evolution by kin selection: A quantitative genetic model illustrated by maternal performance in mice. *Evolution* 38: 766-777. [55]
- Dhondt, A.A. 1982. Heritability of blue tit tarsus length from normal and cross-fostered broods. *Evolution* 36: 418-419. [7]
- Dickerson, G.E. 1947. Composition of hog carcasses as influenced by heritable differences in rate and economy of gain. *Iowa Ag. Exp. Sta. Res. Bull.* 354: 489-524. [55]
- Falconer, D.S. 1981. *Introduction to Quantitative Genetics*, 2nd ed. Longman, London. [6, 7, 9, 19, 20, 29, 50, 54, 55]
- Fisher, R.A. 1958. *The Genetical Theory of Natural Selection*, 2nd Ed., Dover, New York. [7, 55]
- Fitch, H.S. 1985. *Variation in Clutch and Litter Size in New World Reptiles*. Univ. Kansas Mus. Nat. Hist. Misc. Pub. 76, Lawrence. [55]
- Fowler, J.A. 1970. Control of vertebral number in teleosts—an embryological problem. *Quart. Rev. Biol.* 45: 148-167. [55]
- Fox, W. 1948. Effect of temperature on development of scutellation in the garter snake, *Thamnophis elegans atratus*. *Copeia* 1948: 252-262. [55]
- Fox, W., C. Gordon and M.H. Fox. 1961. Morphological effects of low temperatures during the embryonic development of the garter snake, *Thamnophis elegans*. *Zoologica* 46: 57-71. [55]
- Grant, P.R. 1986. *Ecology and Evolution of Darwin's Finches*. Princeton Univ. Press. Princeton. [7, 55]
- Gustafson, L. 1986. Lifetime reproductive success and heritability: Empirical support for Fisher's fundamental theorem. *Am. Nat.* 128: 761-764. [55]
- Hubbs, C.L. 1922. Variations in the number of vertebrae and other meristic characters of fishes correlated with temperature of water during development. *Am. Nat.* 56: 360-373. [55]
- Inger, R.F. 1942. Differential selection on variant juvenile snakes. *Am. Nat.* 76: 527-528. [55]
- Inger, R.F. 1943. Further notes on differential selection of variant juvenile snakes. *Am. Nat.* 77: 87-90. [55]
- Jayne, B.C. 1986. Kinematics of terrestrial snake locomotion. *Copeia* 1986: 915-927. [55]
- Kephart, D.G. 1981. Population ecology and population structure of *Thamnophis elegans* and *Thamnophis sirtalis*. Ph.D. dissertation. University of Chicago, Chicago. [55]
- Kerfoot, W.C. 1969. Selection of an appropriate index for the study of the variability of lizard and snake body scale counts. *Syst. Zool.* 18: 53-62. [55]
- Kerfoot, W.C. and A.G. Kluge. 1971. Impact of the lognormal distribution on studies of phenotypic variation and evolutionary rates. *Syst. Zool.* 20: 459-464. [55]
- Kirkpatrick, M. 1982b. Sexual selection and the evolution of female choice. *Evolution* 36: 1-12. [55]
- Klauber, L.M. 1941. The correlation between scalation and life zones in San Diego County snakes. *Bull. Zool. Soc. San Diego* 17: 73-79. [55]

- Lande, R. 1975. The maintenance of genetic variation by mutation in a polygenic character with linked loci. *Genet. Res., Camb.* 26: 221-235. [6, 7, 11, 33, 37, 39, 53, 54, 55]
- Lande, R. 1976. Natural selection and random genetic drift in phenotypic evolution. *Evolution* 30: 314-334. [7, 54, 55]
- Lande, R. 1979. Quantitative genetic analysis of multivariate evolution applied to brain: Body allometry. *Evolution* 33: 402-416. [7, 29, 54, 55]
- Lande, R. 1980a. Genetic variation and phenotypic evolution during allopatric speciation. *Am. Nat.* 116: 463-479. [7]
- Lande, R. 1980b. Sexual dimorphism, sexual selection, and adaptation in polygenic characters. *Evolution* 34: 292-305. [7, 55]
- Lande, R. 1980c. The genetic covariance between characters maintained by pleiotropic mutation. *Genetics* 94: 203-215. [6, 53, 54, 55]
- Lande, R. 1981a. Models of speciation by sexual selection on polygenic traits. *Proc. Natl. Acad. Sci. USA* 78: 3721-3725. [7, 55]
- Lande, R. 1982b. A quantitative genetic theory of life history evolution. *Ecology* 63: 607-615. [7, 55]
- Lande, R. 1982c. Rapid origin of sexual isolation and character divergence in a cline. *Evolution* 36: 213-233. [7, 55]
- Lande, R. 1984. The genetic correlation between characters maintained by selection, linkage and inbreeding. *Genet. Res.* 44: 309-320. [7, 55]
- Lande, R. and S.J. Arnold. 1983. The measurement of selection on correlated characters. *Evolution* 37: 1210-1226. [7, 37, 53, 55]
- Lecyk, M. 1965. The effect of hypothermia applied in the given stages of pregnancy on the number and form of vertebrae in the offspring of white mice. *Experientia* 21: 452-453. [55]
- Lindsey, C.C. and G.E.E. Moodie. 1967. The effect of incubation temperature on vertebral count in the chicken. *Canad. J. Zool.* 43: 99-104. [55]
- Lindsey, C.C. and R.W. Harrington, Jr. 1972. Extreme vertebral variation induced by temperature in a homozygous clone of self-fertilizing cyprinodontid fish *Rivulus marmoratus*. *Canad. J. Zool.* 50: 733-744. [55]
- Lofsvold, D. 1986. Quantitative genetics of morphological differentiation in *Peromyscus*. I. Tests of the homogeneity of genetic covariance structure among species and subspecies. *Evolution* 40: 559-573. [7, 53, 55]
- Marx, H. and G.R. Rabb. 1972. Phyletic analysis of fifty characters of advanced snakes. *Fieldiana* 63: 1-321. [55]
- Osgood, D.W. 1978. Effect of temperature on the development of meristic characters in *Natrix fasciata*. *Copeia* 1978: 33-37. [55]
- Peabody, R.B. and E.D. Brodie, Jr. 1975. Effect of temperature, salinity and photoperiod on the number of trunk vertebrae in *Ambystoma maculatum*. *Copeia* 1975: 741-746.
- Ruthven, A.G. 1908. Variations and genetic relationships of the garter snakes. *Bull. U.S. Nat. Mus.* No. 61. [55]
- Smith, J.N.M. and A.A. Dhondt. 1980. Experimental confirmation of heritable morphological variation in a natural population of song sparrows. *Evolution* 34: 1155-1158. [7, 55]
- Smith, J.N.M. and R. Zach. 1979. Heritability of some morphological characters in a song sparrow population. *Evolution* 33: 460-467. [55]
- Taning, A.V. 1952. Experimental study of meristic characters in fish. *Biol. Rev.* 27: 169-193. [55]
- Turelli, M. 1984. Heritable genetic variation via mutation-selection balance: Lerch's zeta meets the abdominal bristle. *Theor. Pop. Biol.* 25: 138-193. [6, 7, 11, 33, 54, 55]
- Turelli, M. 1985. Effects of pleiotropy on predictions concerning mutation-selection balance for polygenic traits. *Genetics* 111: 165-195. [6, 7, 54, 55]
- van Noordwijk, A.J., J.H. van Balen and W. Scharloo. 1981. Genetic and environmental variation in the clutch size of the Great Tit (*Parus major*). *Neth. J. Zool.* 31: 342-373. [7, 55]
- Via, S. and R. Lande. 1985. Genotype-environment interaction and the evolution of phenotypic plasticity. *Evolution* 39: 505-522. [7, 53, 55]
- Willham, R.L. 1963. The covariance between relatives for characters composed of components contributed by related individuals. *Biometrics* 19: 18-27. [55]
- Yntema, C.L. 1960. Effects of various temperatures on the embryonic development of *Chelydra serpentina*. *Anat. Rec.* 136: 305-306. [55]