Evolutionary Divergence of the Genetic Architecture Underlying Photoperiodism in the Pitcher-Plant Mosquito, *Wyeomyia smithii*

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ABSTRACT

We determine the contribution of composite additive, dominance, and epistatic effects to the genetic divergence of photoperiodic response along latitudinal, altitudinal, and longitudinal gradients in the pitcher-plant mosquito, *Wyeomyia smithii*. Joint scaling tests of crosses between populations showed widespread epistasis as well as additive and dominance differences among populations. There were differences due to epistasis between an alpine population in North Carolina and populations in Florida, lowland North Carolina, and Maine. Longitudinal displacement resulted in differences due to epistasis between Florida and Alabama populations separated by 300 km but not between Maine and Wisconsin populations separated by 2000 km. Genetic differences between New Jersey and Ontario did not involve either dominance or epistasis and we estimated the minimum number of effective factors contributing to a difference in mean critical photoperiod of 5 SD between them as $n_E = 5$. We propose that the genetic similarity of populations within a broad northern region is due to their more recent origin since recession of the Laurentide Ice Sheet and that the unique genetic architecture of each population is the result of both mutation and repeated migration-founder-flush episodes during the dispersal of *W. smithii* in North America. Our results suggest that differences in composite additive and dominance effects arise early in the genetic divergence of populations while differences due to epistasis accumulate after more prolonged isolation.

UNDERSTANDING the genetic basis of evolutionary divergence between populations is a fundamental question in the study of evolution. Many models of adaptive change within populations consider only additive effects of genes (*e.g.*, LANDE 1980a,b; CHEVERUD 1984; ROSE 1985; VIA and LANDE 1985; GILLESPIE and TURRELLI 1989; CHARLESWORTH 1990) although nonadditive effects can make an important contribution to fitness (ROBERTSON 1957; FISHER 1958; WRIGHT 1968, 1977; BARTON and TURELLI 1989; WHITLOCK *et al.* 1995) and genic (epistatic) interaction can contribute to the additive genetic variance and, consequently, the heritability of traits (CROW and KIMURA 1970; FALCONER 1981). During the founding of populations, the joint effects of isolation and drift may disrupt genic interactions; mass selection during subsequent population growth may then result in novel genetic architectures in the descendant population, even if the mean phenotype remains the same (MAYR 1954; CARSON 1968; WRIGHT 1977; TEMPLETON 1980; GOODNIGHT 1987; HARD *et al.* 1993b; SLATKIN 1996). Hence, the genetic differentiation of populations through their evolutionary history can result in reorganized genetic architectures that differ in net or composite pleiotropic, dominance, or epistatic, as well as additive effects.

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Herein, we examine the extent to which dominance and epistasis have contributed to the genetic divergence of photoperiodic response among populations of the pitcher-plant mosquito, *Wyeomyia smithii* (Coq.). Photoperiodic response is a crucial trait for the establishment and diversification of insect populations at temperate latitudes (ANDREWARTHA 1952; DANILEVSKII 1965; MASAKI 1983; TAUBER *et al.* 1986; DANKS 1987). As the favorable season declines with increasing latitude or altitude, the median day length (critical photoperiod) that triggers hibernal dormancy (diapause) also increases. Populations of insects living at higher latitudes or altitudes where winter arrives earlier, respond to longer days and enter diapause earlier than populations at lower latitudes or altitudes (DANILEVSKII 1965; TAYLOR and SPALDING 1986; DANKS 1987, Table 25), including populations of *W. smithii* (BRADSHAW 1976).

The immature stages of *W. smithii* develop only within the water-filled leaves of the purple pitcher plant, *Sarracenia purpurea* L. The range of the mosquito in North America follows that of its host from the Gulf of Mexico to north-central Canada (30–54°N Lat.). Present-day distribution, physiology, morphology, and behavior all indicate that *W. smithii* has invaded North America from South America and that the evolution of this species in North America has proceeded from south to north (ROSS 1964; BRADSHAW and LOUNIBOS 1977; ISTOCK and WEISBURG 1987; BRADSHAW and HOLZAPFEL 1990). Throughout its range, *W. smithii* overwinters in the

leaves in a state of larval diapause initiated, maintained, and terminated by photoperiod (BRADSHAW and LOUNIBOS 1977). The critical photoperiod for the induction and maintenance of diapause in *W. smithii* increases linearly with latitude and altitude but not longitude of origin (BRADSHAW 1976: $R^2 = 0.96$). Hence, photoperiodic response in *W. smithii* is closely correlated with the climatic gradient that parallels their historical biogeography in North America.

Wyeomyia smithii is the sole temperate species of a large neotropical genus (LANE 1953; STONE *et al.* 1959), and only two species from this genus, *Wyeomyia mitchelli* and *W. vanduzeei,* are found in subtropical Florida (DARSIE and WARD 1981). Phenotypically, populations of *W. smithii* group into four geographic regions (Figure 1). Like most mosquitoes, larval *W. smithii* from the Gulf Coast possess four long anal papillae; coastal and Piedmont North Carolina populations possess two long ventral and two short dorsal papillae; all more northern and alpine populations possess but two moderately lengthened ventral papillae (BRADSHAW and LOUNIBOS 1977). Gulf Coast and lowland North Carolina females are obligately nonbiting for the first ovarian cycle but require a blood meal for the second and subsequent ovarian cycles. Northern and alpine females produce repeated egg batches without blood feeding, a highly unusual trait in mosquitoes without predatory larvae (O'MEARA *et al.* 1981; O'MEARA and LOUNIBOS 1981; BRADSHAW 1980, 1986). Gulf Coast populations diapause in the fourth instar; lowland North Carolina populations are polymorphic for third or fourth instar diapause; northern and alpine populations diapause in the third instar but may enter a second, fourth instar diapause (LOUNIBOS and BRADSHAW 1975; BRADSHAW and LOUNIBOS 1977). Hence, anal papillae morphology, diapause characteristics, and reproductive physiology all distinguish populations of *W. smithii* from the Gulf Coast, from lowland North Carolina, and from alpine North Carolina and the northern region (New Jersey and northwards) as distinct and separate groups. Although the alpine North Carolina populations are phenotypically identical to northern populations, their relatedness to the northern populations is uncertain because the distribution of *S. purpurea,* and therefore *W. smithii,* is disjunct in the Appalachian Mountains between North Carolina and Pennsylvania (MCDANIEL 1966). These alpine populations may then constitute a geographic region that is genetically distinct from populations in the northern region.

HARD *et al.* (1992, 1993a) examined the contribution of nonadditive genetic effects to the latitudinal divergence of critical photoperiod in *W. smithii* along their evolutionary trajectory in North America. They compared populations from the southern region with populations from the low elevation North Carolina and the northern regions. They did not examine genetic differences between the southern alpine and the other regions or differences due to longitudinal or latitudinal displacement within a region. Furthermore, since the presence of nonadditive genetic effects biases estimates of the number of genetic factors contributing to genetic differences between populations (LANDE 1981; MATHER and JINKS 1982; ZENG *et al.* 1990), HARD *et al.* (1992, 1993a) were not able to estimate the number of loci potentially contributing to the evolved differences in critical photoperiod. In this article, we first examine whether the phenotypic similarity of the southern alpine to the northern populations is the product of a similar underlying genetic architecture. Second, we examine whether longitudinal isolation-by-distance independently of broad-scale climatic variation can result in nonadditive genetic differences between populations. Finally, we estimate the number of loci (minimum number of effective factors) that contribute to genetic divergence of critical photoperiod between populations that differ in critical photoperiod only in additive effects.

MATERIALS AND METHODS

Collection and maintenance of populations: Approximately 2000 *W. smithii* larvae were collected from each of eight localities in eastern North America (Figure 1): two southern populations in Alabama (AL: 30°N, 87°W, 15 m elev) and Florida (FL: 30°N, 85°W, 10 m elev); four northern populations in Ontario (ON: 50°N, 94°W, 400 m elev), Wisconsin (WI: 46°N, 90°W, 500 m elev), Maine (ME: 46°N, 68°W, 250 m elev), and New Jersey (NJ: 40°N, 75°W, 10 m elev); and two populations in North Carolina at alpine (Alp: 35°N, 83°W, 900 m elev) and lowland (Low: 35°N, 80°W, 90 m elev) localities. These populations correspond to localities LI, WI, DL, ML, KC, PB, DB, and SH, respectively, in previous articles from this laboratory. Collections were made in the spring of 1993 before adult emergence so that 100% of the genotypes were still available for sampling as larvae in the pitcher-plant leaves. Samples were taken from 50 (lowland AL-NJ) to 200 (northern and alpine) plants over 100–500 m distance within each population. Samples were maintained on ice, transported to the laboratory, and reared through four to six generations before the start of experiments as described in HARD *et al.* (1992).

Line crosses: The contribution of nonadditive genetic effects to the genetic differentiation of populations can be evaluated from parent and hybrid phenotype means and variances (CAVALLI 1952; HAYMAN 1958, 1960a,b; LANDE 1981; COCKERHAM 1986; HARD *et al.* 1992). We set up our crosses to reflect differences or similarities in latitude, longitude, and altitude (Figure 1; Table 1). First, to evaluate the genetic divergence of the alpine from the other regions, we crossed the alpine North Carolina population (Alp) and the Florida population (FL) from the southern region, the alpine population (Alp) and a lowland population (Low) at the

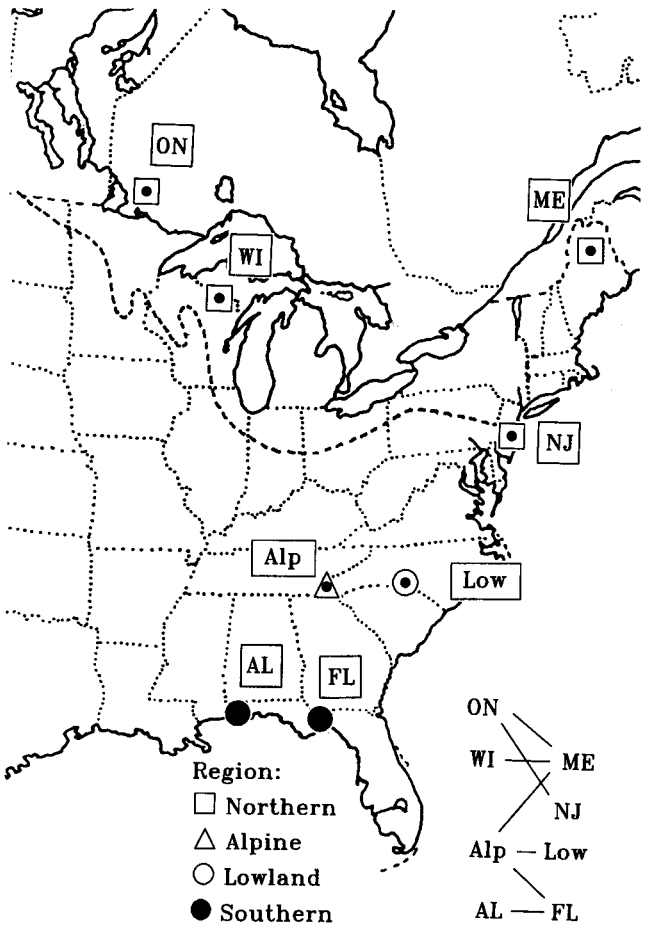

FIGURE 1.—Origin of populations and crosses between them. Different symbols show the different geographic regions identified by differences and similarities in morphology, diapause, and reproductive physiology. Lines connecting populations in the inset show the crosses performed. The thick dashed line shows the southern maximum of the Laurentide Ice Sheet.

same latitude in North Carolina, and the alpine population (Alp) and a Maine population (ME) from the northern region. Second, to evaluate genetic divergence due to isolation-by-distance within a region, we crossed latitudinally similar but longitudinally displaced populations within the southern region (FL·AL) and within the northern region (ME·WI). Third, to evaluate extreme latitudinal and longitudinal displacement within a region, we crossed the Ontario population (ON) and the Maine (ME) or New Jersey (NJ) population, all within the northern region.

Crosses included the F_1 and F_2 hybrids and first backcrosses (B_1 and B_2), with all of their reciprocals nested within each reciprocal F_1, which yielded a total of 14 lines per cross (92 total lines). For each cross, the sexes were separated as pupae. Only male pupae from one population or generation were placed in adult cages with only female pupae from the other population or generation. The resulting adults were allowed to mate *en masse* and females to oviposit within this single adult cage. Each reciprocal cross was set up in a separate adult cage. Because each cohort of parents had been maintained on unambiguous long-day photoperiod (L:D = 18:6) since they had been larvae, we assumed that mating between adults was random with respect to larval critical photoperiod. In the first generation of the experiment, parent populations were split into two halves, one half being used to generate new parent populations and the other half to create the F_1 and reciprocal F_1 lines. The F_1s and reciprocal F_1s were each split into thirds: one third to generate the F_2, one third to generate the B_1 and its reciprocal, and one third to generate the B_2 and its reciprocal. At the same time, the parental lines were split into quarters: one quarter to generate new parent lines, one quarter to generate new F_1s and their reciprocals, and one half to generate the backcrosses and their reciprocals. The progeny of this second generation were reared on short-day photoperiod (L:D = 8:16) to induce diapause and, for each of the 92 lines, critical photoperiod and its variance were determined concurrently from 125 individuals in each line exposed to linearly increasing (3 min/day) photoperiod as in HARD *et al.* (1992). For each of the 92 lines, the grandparental generation of the experi-

TABLE 1

Geographic separation of parent populations and joint scaling tests for the adequacy of the five-parameter ADM model and the eight-parameter ADME model

Cross	Difference in geography			ADM[a]	ADME[a]	Figure 3
	°N Lat	°W Lon	m Alt	χ^2 (0 d.f.)	χ^2 (1 d.f.)	
Alp·FL	5.0	1.8	890	59.19***	3.32°	B
Alp·Low	0.1	3.7	810	51.06***	1.35°	C
Alp·ME	11.1	14.9	530	54.57***	15.42***	D
FL·AL	0.4	2.4	5	25.96***	1.19°	E
ME·WI	0.2	21.2	130	10.74°	—	F
ME·ON	3.4	25.8	40	48.93***	0.24°	G
NJ·ON	9.7	19.8	400	5.77°	—	H

[a] After column-wide sequential Bonferoni: ° $P > 0.05$, *** $P < 0.001$. —, the ADME model was not tested since the ADM model was not rejected. ADM, additive-dominance-maternal; ADME, additive-dominance-maternal-epistasis.

mental larvae consisted of 95–284 males and 78–298 females; the parent generation of the experimental larvae consisted of 72–153 males and 54–120 females. Within each generation, all of the progeny from each line were pooled in a large pan and then the parents for the next generation or the experimental larvae were withdrawn haphazardly from the pooled larvae.

Operationally, diapausing larvae are exposed to short days that are incremented 3 min/day by Chrontrol CD-4 electronic timers. At some point, the increasing photoperiod triggers resumed development and each individual larva eventually pupates. We then define the critical photoperiod for an individual as the daylength of the day on which it pupates and for the population as the mean daylength of pupation in a cohort of individuals.

Under the experimental conditions, development is both positively skewed and slightly protandrous. To reduce skewness, stabilize coefficients of variation, and eliminate any significant sexual dimorphism, we $\log_{10}$ transformed individual critical photoperiods and pooled the data from both sexes (HARD *et al.* 1992) without scoring sex. Raw data were $\log_{10}$ transformed and used to calculate means and variances on an Excel spreadsheet. The means and variances of critical photoperiods of the fourteen lines resulting from crosses between any two parental populations were used to derive estimates of the composite additive, dominance, additive maternal, dominance maternal, and digenic epistatic effects for this trait. By eliminating the scoring of sex, we were able to determine critical photoperiod in all 92 lines (12,000 larvae) concurrently. We were therefore not able to test for sex linkage, but HARD *et al.* (1993a) found no evidence for sex linkage in crosses involving even greater geographic distances and phenotypic divergence of critical photoperiod than in the present study. Initial tests for maternal effects revealed these effects scattered throughout the crosses. Rather than report and discuss each of these several tests, we combined estimates of maternal effects into a least-squares model containing the information from all of the generations within each cross. Since we were interested primarily in the contribution of epistasis to genetic differences between populations, we first tested for the adequacy of a five-parameter model including additive, dominance, and maternal effects (ADM) and then an eight-parameter model including additive, dominance, maternal, and digenic epistatic effects (ADME). All matrix manipulations and tests for goodness of fit were programmed and performed using Mathcad 4.0.

The weighted least-squares model that incorporates these composite effects is (HAYMAN 1958; MATHER and JINKS 1982) as follows:

$$\hat{\mathbf{x}} = (\mathbf{C}^T\mathbf{E}^{-1}\mathbf{C})^{-1}\mathbf{C}^T\mathbf{E}^{-1}\mathbf{y} \quad (1)$$

$$\mathrm{Var}(\hat{\mathbf{x}}) = (\mathbf{C}^T\mathbf{E}^{-1}\mathbf{C})^{-1} \quad (2)$$

$$\hat{\mathbf{y}} = \mathbf{C}\hat{\mathbf{x}} = \text{the fitted mean values} \quad (3)$$

where $\hat{\mathbf{x}}$ is the vector of mean ($\hat{m}$), additive ($\hat{d}$), dominance ($\hat{h}$), additive-maternal ($\hat{d}_m$), dominance-maternal ($\hat{h}_m$), additive × additive epistasis ($\hat{i}$), additive × dominance epistasis ($\hat{j}$), and dominance × dominance epistasis ($\hat{l}$) parameters. $\mathbf{C}$ is the matrix of coefficients for these parameters from the equations for predicted line means (Table 2), $\mathbf{C}^T$ its transpose, $\mathbf{E}$ is a diagonal matrix of error variances (squared phenotypic standard errors) of the line means, and $\mathbf{y}$ is the vector of observed line means. $\mathrm{Var}(\hat{\mathbf{x}})$ is a diagonal matrix of the sampling errors of the respective elements in $\hat{\mathbf{x}}$. $\hat{\mathbf{y}}$ is the vector of predicted line means representing, respectively, the parents (P_1 and P_2), the F_1 and its reciprocal (F_1 and rF_1), the combined F_2 and reciprocal F_2, the first backcross and its reciprocal ($\sigma \times \varphi$:$B_1 = P_1 \times F_1$ combined with $P_1 \times rF_1$ and $rB_1 = F_1 \times P_1$ combined with $rF_1 \times P_1$), and the second backcross and its reciprocal ($\sigma \times$

TABLE 2

Expected generation means of an additive-dominance-digenic epistasis model with maternal effects

Generation	Expected contribution to generation mean	Coefficients of $\mathbf{C}^a$							
		m	d	h	d_m	h_m	i	j	l
P_1	$m + d + d_m + i$	1	1	0	1	0	1	0	0
P_2	$m - d - d_m + i$	1	−1	0	−1	0	1	0	0
F_1	$m + h + d_m + l$	1	0	1	1	0	0	0	1
rF_1	$m + h - d_m + l$	1	0	1	−1	0	0	0	1
F_2	$m + \frac{1}{2}h + h_m + \frac{1}{4}l$	1	0	$\frac{1}{2}$	0	1	0	0	$\frac{1}{4}$
B_1	$m + \frac{1}{2}d + \frac{1}{2}h + d_m + \frac{1}{4}i + \frac{1}{4}j + \frac{1}{4}l$	1	$\frac{1}{2}$	$\frac{1}{2}$	1	0	$\frac{1}{4}$	$\frac{1}{4}$	$\frac{1}{4}$
rB_1	$m + \frac{1}{2}d + \frac{1}{2}h + h_m + \frac{1}{4}i + \frac{1}{4}j + \frac{1}{4}l$	1	$\frac{1}{2}$	$\frac{1}{2}$	0	1	$\frac{1}{4}$	$\frac{1}{4}$	$\frac{1}{4}$
B_2	$m - \frac{1}{2}d + \frac{1}{2}h + -d_m + \frac{1}{4}i - \frac{1}{4}j + \frac{1}{4}l$	1	$-\frac{1}{2}$	$\frac{1}{2}$	−1	0	$\frac{1}{4}$	$-\frac{1}{4}$	$\frac{1}{4}$
rB_2	$m - \frac{1}{2}d + \frac{1}{2}h + h_m + \frac{1}{4}i - \frac{1}{4}j + \frac{1}{4}l$	1	$-\frac{1}{2}$	$\frac{1}{2}$	0	1	$\frac{1}{4}$	$-\frac{1}{4}$	$\frac{1}{4}$

These data were compiled from MATHER and JINKS (1982), Tables 31 and 117. m, mean; d, additive; h, dominance; d_m, additive maternal; h_m, dominance maternal; i, additive by additive epistasis; j, additive by dominance epistasis; l, dominance by dominance epistasis.

[a] Equations 1–3.

♀:B_2 = $P_2 \times F_1$ combined with $P_2 \times rF_1$ and $rB_2 = F_1 \times P_2$ combined with $rF_1 \times P_2$). Hence, the analyses consist of eight parameters to be estimated ($\hat{m}$ to $\hat{l}$) from nine generation means and variances (P_1, P_2, F_1, rF_1, F_2, B_1, rB_1, B_2, and rB_2), allowing tests for the goodness-of-fit of the line means to sequential models incorporating additive-dominance-maternal effects (five-parameter ADM model) and additive-dominance-maternal-epistatic effects (eight-parameter ADME model). Under the assumption of normality, goodness-of-fit of the line means to the ADM model, incorporating *m*, *d*, *h*, d_m, and h_m effects, was tested with the chi-square statistic with 4 d.f. (9 generation means − 5 parameters) derived from HAYMAN (1958) and MATHER and JINKS (1982). If the five-parameter model was rejected, we then tested for goodness-of-fit to the full eight-parameter model, incorporating *m*, *d*, *h*, d_m, h_m, *i*, *j*, and *l* effects, with 1 d.f. (9 generation means − 8 parameters). The chi-squared statistic was calculated as

$$\chi^2 = \mathbf{y}^T\mathbf{E}^{-1}\mathbf{y} - \mathbf{y}^T\mathbf{E}^{-1}\hat{\mathbf{y}} \tag{4}$$

with 4 or 1 d.f., as appropriate.

Since we performed these tests with seven different geographical crosses, we applied the sequential Bonferoni (RICE 1989) to assure a model-wide protection level of $\alpha = 0.05$ for testing of the five-parameter model across all seven crosses and for testing the eight-parameter model across all crosses for which the five-parameter model had been rejected. Acceptance of the five-parameter model would indicate that composite additive, dominance, and maternal effects alone were adequate to account for genetic divergence in critical photoperiod between the parental populations. Rejection of the five-parameter model would indicate that at least some form of epistasis was required to account for this difference. Acceptance of the eight-parameter model would indicate that additive, dominance, maternal, and digenic epistatic effects alone were adequate to account for genetic divergence in critical photoperiod between the parental populations. Rejection of the eight-parameter model would indicate that genetic divergence between the two parental populations also involved linkage, trigenic or higher order epistasis, or maternal by offspring genotype interaction.

Estimates of the eight composite effects $\hat{m}$ to $\hat{l}$ were obtained from the vector $\hat{\mathbf{x}}$ (Equation 1) after the appropriate five- or eight-parameter genetic model was fitted by the method of least squares. The composite effects are sums of weighted line means; therefore, their sampling errors (Var $\hat{\mathbf{x}}$, Equation 2) were calculated as the square root of the sum of the error variances (**E**) of the contributing line means, weighted by the corresponding coefficients (**C** in Table 2). The significance of each of the parameters $\hat{m}$ to $\hat{l}$ was assessed as

$$t_i = \frac{\hat{x}_i}{\sqrt{\mathrm{Var}(\hat{x}_i)}} \tag{5}$$

for the *i*th parameter with d.f. >200 in each case.

Number of loci: We estimated the minimum number of effective factors (n_E) contributing to the divergence between populations and its standard error from the estimated line means and segregational variances by the formulas derived in LANDE (1981) and COCKERHAM (1986):

$$n_E = \frac{(\bar{y}_1 - \bar{y}_2)^2 - E_1 - E_2}{8 \cdot \mathrm{Var}S} \tag{6}$$

where $\bar{y}_1$ and $\bar{y}_2$ are the phenotypic means of the two populations, E_1 and E_2 are their respective error variances, and Var*S* is the segregational variance calculated using the method of least squares. Var*S* is obtained from the vector of variance components, **a**, whose elements represent the least-squares estimates of the phenotypic variances of the two parents and the segregational variance, respectively, under an additive model:

$$\mathbf{a} = (\mathbf{X}^T\mathbf{V}^{-1}\mathbf{X})^{-1}\mathbf{M}^T\mathbf{V}^{-1}\mathbf{v} \tag{7}$$

where **v** is the vector of phenotypic variances for the P_1, P_2, F_1, F_2, B_1, and B_2 generations, respectively, **V** is a vector of the sampling variances of the phenotypic variances based on n_i observations each:

$$V_i = \frac{2\nu_i^2}{n_i} \tag{8}$$

and **X** is the matrix of coefficients for the predicted line variances under an additive model (COCKERHAM 1986, p. 660). We used COCKERHAM's (1986) maximum likelihood procedure for the estimation of Var*S* but we found that **a** was consistently stable to three decimal places at the first iteration. Finally, we calculated the standard error of n_E as (COCKERHAM 1986):

$$s_{n_E} = n_E\left[\frac{4(E_1 + E_2)}{\bar{y}_1 + \bar{y}_2} + \frac{\mathrm{Var}S}{(\mathrm{Var}S)^2}\right]^{1/2} \tag{9}$$

where Var*S* is the variance of the segregational variance, the third element in the variance vector $\mathbf{Va} = (\mathbf{X}^T\mathbf{V}^{-1}\mathbf{X})^{-1}$. Because this estimation of n_E assumes unlinked loci and equal allelic effects, it also provides an estimate of the minimum number of genetic loci contributing to population divergence (ZENG *et al.* 1990).

RESULTS

There was a close association between the latitude and altitude of a population and its critical photoperiod (Figure 2). The critical photoperiod of the low-elevation populations increased consistently with latitude and the critical photoperiod of the alpine (high-elevation) population in North Carolina (35°N) was substantially higher than the critical photoperiod of the lowland population, indicating an equivalent latitude just north of the New Jersey population. We used the average mean and standard deviation of the FL and AL populations as an estimate of the mean and standard

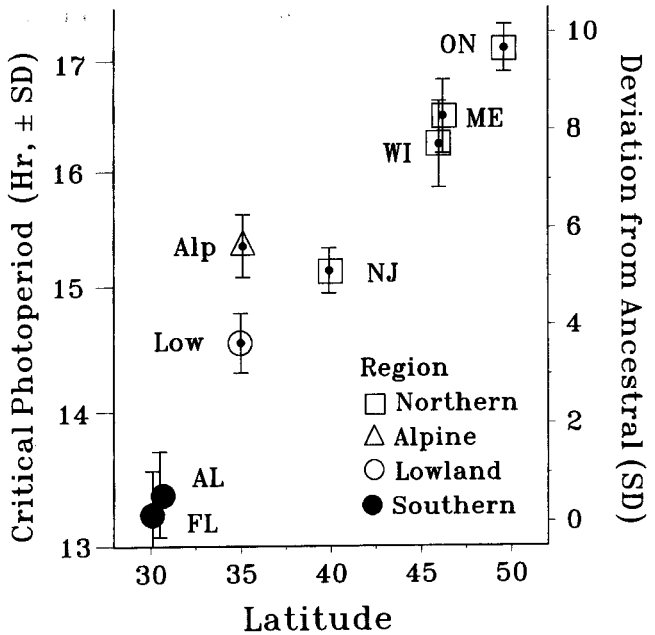

FIGURE 2.—Geographic variation in the mean (±1 SD) critical photoperiod on a scale of hours (left axis) or on a scale of deviation from the ancestral (Al and FL) mean in units of the average ancestral standard deviation. Regional symbols as in Figure 1.

deviation in critical photoperiod of the ancestral population and then plotted divergence of mean critical photoperiod from the "ancestor" in units of standard deviation of the ancestral population. On this scale, critical photoperiod has diverged from the ancestral mean by about 4 SD in the lowland population, 6 SD in the alpine population, and 5–10 SD in the northern populations (Figure 2), confirming the relationship between geography and critical photoperiod found in previous studies (BRADSHAW 1976; BRADSHAW and LOUNIBOS 1977; HARD *et al.* 1992, 1993a).

Figure 3A shows the additive and additive-dominance expectations from the line crosses. All crosses showed a clear departure from the additive expectation (Figure 3B–H) except the ON · NJ cross (Figure 3H) for which the ADM model was not rejected (Table 1) and for which there was also no significant difference due to dominance (Table 3). The northern longitudinal cross (ME · WI, Figure 3F) conformed to the general pattern expected for the additive-dominance model and the ADM model was not rejected in this case (Table 1), but the hybrid generations still showed the presence of significant differences in dominance (Table 3). For all the other crosses, the presence of epistasis was suggested (1) because the F_2 fell on the opposite side of the F_1 from the mid-parent expectation (Figure 3B, E, and G), (2) because the F_2 mean fell on the opposite side of the additive expectation from the F_1 (Figure 3D), or (3) because one (Figure 3C) or both (Figure 3E) of the backcross means fell on opposite sides of the additive expectation than did the F_2 mean. Indeed, the ADM model was rejected for all these five crosses (Table 1), and there was a significant contribution of at least one component of digenic epistasis to genetic differences between the parents (Table 3: Alp · Fl, Alp · Low, Alp · ME, ME · ON, FL · AL).

In the five crosses for which the ADM model was rejected, the eight-parameter additive-dominance-maternal-epistasis (ADME) model was rejected only for the alpine-northern cross (Alp · ME); composite additive, dominance, maternal, and digenic epistatic effects were adequate to account for genetic differences in critical photoperiod in the other four crosses. The FL · AL cross showed evidence for additive by additive (A × A) and dominance by dominance (D × D) but not additive by dominance (A × D) epistasis. The Alp · FL cross showed evidence for A × D and D × D but not A × A epistasis. The Alp · Low cross showed evidence of D × D but not A × D or A × A epistasis. The ME · ON cross showed evidence of D × D but not A × A or A × D epistasis. In the Alp · ME cross, there was evidence of A × A but not A × D or D × D epistasis; in this case, the ADME model was also rejected (Table 1), indicating a contribution of linkage, higher order epistasis, or maternal-offspring genotype interaction to genetic divergence between the Alp and ME populations. These results show that, in the five crosses where epistasis contributed to genetic differences in critical photoperiod between populations, the pattern of epistatic effects was unique in three of them (Alp · Fl, Alp · ME, and FL · AL) and identical in the other two (Alp · Low and ME · ON).

Estimates of the minimum number of effective factors involved in differences in critical photoperiod between populations ranged from zero to six and were positively correlated with the difference in mean critical photoperiod between populations (Figure 4). Of the seven estimates, only the highest four differed significantly from zero after application of the sequential Bonferoni: Alp · Low, Alp · ME, NJ · ON, and Alp · FL. One estimate (ME · WI) was less than, but not significantly different from, zero because the phenotypic variance was actually lower in the F_2 than in the F_1 or in either parent, leading to a negative segregational variance (Var$S$) and, consequently, a negative estimate of n_E (Equation 6).

DISCUSSION

First of all, we want to reiterate that the line-cross approach only identifies *differences* in composite additive, dominance, or epistatic effects *between* the parent populations. Elements of the genetic architecture within populations or that are common to both populations will not be detected by this methodology. Furthermore, multiple differences in additive, dominance, or epistatic effects that differ in sign will tend to cancel each other out. We therefore view our results as a conservative estimate of the changes in genetic architecture

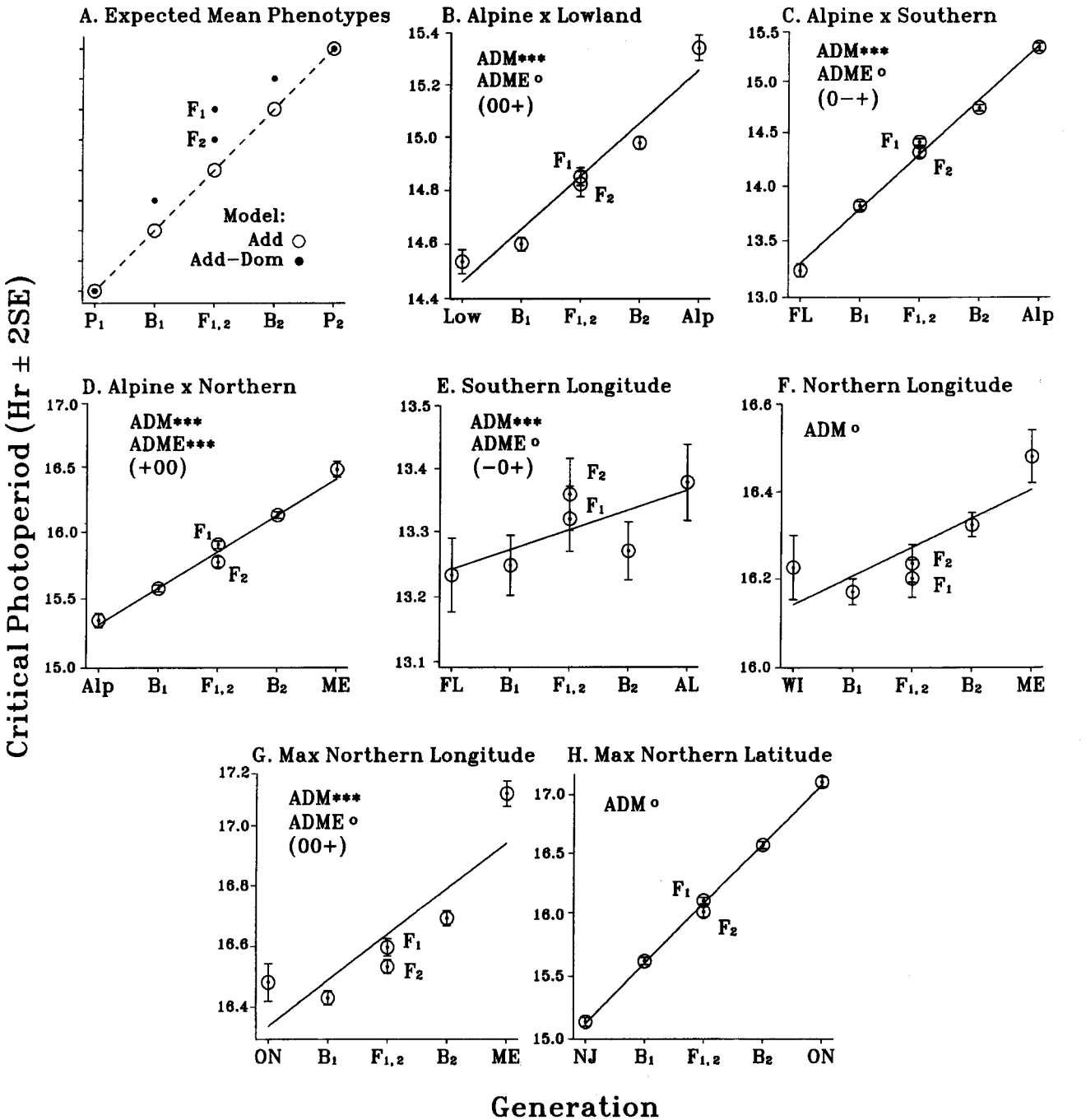

FIGURE 3.—Parental and hybrid critical photoperiods (mean hours ± 2 SE). (A) Additive (○) and additive-dominance (●) expectations. If populations differ only in additive effects, then all generation means should fall on a straight line connecting the parents. Differences due to dominance produce a deviation of the hybrid generations above or below this line. Because of independent assortment in the F_1, the F_2 and backcross generations lose half of their heterozygosity. Therefore, the F_2 should fall half the distance from the F_1 and the additive expectation, and the backcross means should deviate from the additive expectation by the same amount as the F_2. Deviations from the additive-dominance expectation indicate differences due to epistasis. (B–H) Experimental results with the diagonal line showing the additive expectation. This expectation is determined by the method of least squares, incorporating all of the generation means; consequently, the line need not pass through the parental means. The inset shows the goodness-of-fit to the ADM and ADME models from Table 1; the symbols in parentheses indicate the significance and sign of A × A, A × D, and D × D epistasis, respectively: 0, not significant; +, significant and positive; −, significant and negative from Table 3.

following adaptive divergence of photoperiodic response. Our results do, however, indicate that evolution of critical photoperiod over a climatic gradient sufficient to select for a difference in mean phenotype of 4–6 SD probably involves a minimum of four to six loci. At least one component of photoperiodic time measurement in *W. smithii* includes an underlying circadian pacemaker and the contribution of this pacemaker

TABLE 3

Composite effects (parameter estimates, $\hat{X}$) contributing to differences in critical photoperiod between populations

Cross	$\hat{m}$	$\hat{d}$	$\hat{h}$	$\hat{d}_m$	$\hat{h}_m$	$\hat{i}$	$\hat{j}$	$\hat{l}$
Alp · FL	1.154	0.032	0.009	-3.12×10^{-4}	0.003	-6.41×10^{-4}	−0.004	0.014
	315.54	46.19	1.18	−0.81	3.60	−0.18	−4.03	3.02
	***	***	°	°	**	°	***	*
Alp · Low	1.177	0.011	−0.021	9.93×10^{-5}	5.92×10^{-4}	−0.003	-7.55×10^{-4}	0.016
	399.53	20.03	−3.47	−0.29	1.25	−0.96	−1.12	4.66
	***	***	***	°	°	°	°	***
Alp · ME	1.191	−0.014	0.013	−0.002	0.001	0.011	4.61×10^{-4}	−0.002
	460.78	−22.05	2.36	−6.53	2.48	4.2	0.64	−0.68
	***	***	°	***	*	***	°	°
FL · AL	1.147	-5.68×10^{-4}	−0.022	−0.002	−0.005	−0.022	0.002	0.021
	243.49	0.61	−4.22	−2.77	−4.34	−4.81	1.27	3.46
	***	°	***	*	***	***	°	**
ME · WI	1.213	0.004	−0.004	-2.88×10^{-4}	4.62×10^{-4}			
	2.36×10^{3}	7.54	−4.42	−0.75	−1.13	—	—	—
	***	***	***	°	°			
ME · ON	1.225	−0.007	−0.016	−0.001	−0.001	8.91×10^{-5}	0.001	0.011
	540.11	−12.62	−3.52	−3.80	−3.41	0.04	2.00	3.90
	***	***	**	**	**	°	°	***
ON · NJ	1.207	0.026	7.03×10^{-5}	6.67×10^{-5}	−0.001			
	3.58×10^{-3}	61.99	0.14	0.22	−3.54	—	—	—
	***	***	°	°	**			

Entries for each cross show the parameter estimate (Equation 1), the associated value of t (Equation 5) with d.f. > 200, and the associated probability after sequential Bonferoni was applied row-wide within each cross for the top, middle, and bottom row, respectively. ° $P > 0.05$, * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$. —, No estimates were made of composite epistatic effects since the five-parameter ADM model was not rejected. $\hat{m}$ mean; $\hat{d}$, additive; $\hat{h}$, dominance; $\hat{d}_m$, maternal additive; $\hat{h}_m$, maternal dominance; $\hat{i}$, additive by additive epistasis; $\hat{j}$, additive by dominance epistasis; $\hat{l}$, dominance by dominance epistasis.

to the evolution of critical photoperiod involves its amplitude but not its period (WEGIS *et al.* 1997). These observations suggest that the photoperiodic adaptation of *W. smithii* to the ecoclimatic gradient of North America has involved multiple, interacting loci that affect primarily the amplitude of the circadian component of photoperiodic time measurement.

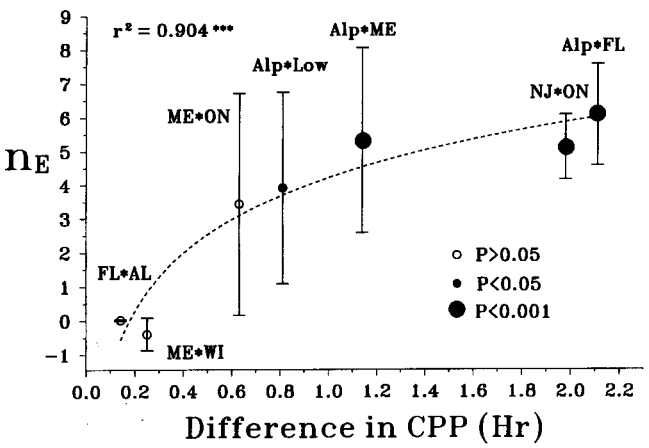

FIGURE 4.—Estimated mean number of effective factors (n_E ± 2 SE) by which two populations differ, fitted to a logarithmic curve. Note that some of these estimates do not differ from zero after a figure-wide sequential Bonferoni and that all but the NJ · ON estimate are potentially biased by nonadditive effects.

Epistasis makes a significant contribution to genetic divergence between some, but not all populations (Table 1). In the presence of substantive epistatic effects, the contribution of additive and dominance effects cannot be established reliably (HAYMAN 1960b). Consequently, we discuss differences in additive and dominance effects only in the absence of significant differences due to epistasis. The pattern of epistasis, or lack thereof, enabled us to answer our initial questions about the evolutionary divergence of critical photoperiod in *W. smithii*. First, epistasis was involved in the genetic divergence of the alpine from southern, lowland, and northern populations and the Alp · ME was the only cross involving higher order epistasis, linkage, or maternal-offspring genotype interaction (Table 1). Hence, although alpine populations are indistinguishable from northern populations in terms of morphology, diapause, and reproductive physiology (BRADSHAW and LOUNIBOS 1977; BRADSHAW and HOLZAPFEL 1990), they clearly differ markedly in the genetic architecture underlying critical photoperiod.

Second, longitudinal displacement of populations at similar latitudes resulted in differences in epistasis in

the southern (AL · FL) but not in the northern (ME · WI) cross, even though the southern populations were geographically closer to each other (Table 1). Similarly, ARMBRUSTER *et al.* (1997) found greater genetic distance between two populations in north Florida than between the same ME and ON populations as in this study. The results of both studies indicate that isolation-by-distance is not adequate to explain the involvement of epistasis in the genetic divergence of populations of *W. smithii.*

Third, two of the three crosses involving exclusively populations from within the northern region (Figure 1) did not differ in epistatic effects (Table 1), and one of them did not differ in epistatic or dominance effects despite the fact that the parent populations (NJ, ON) differed dramatically in latitude, longitude, and altitude (Table 1) and by 5 SD in mean critical photoperiod (Figure 2). These results, in combination with those immediately above, lead us to conclude that populations within the broader northern region share more similar genetic architectures than do populations between any two regions, even though the latter may be geographically closer.

We propose that the genetic similarity of populations within the northern region is a consequence of their more recent origin. Populations near to and north of the maximum southern extent of the Laurentide Ice Sheet (Figure 1) must have been established after the recession of the ice 21–7 K years ago. At the same time, these more recently founded populations complete only one to three generations per year while more southern, older populations of *W. smithii* complete five to seven generations per year (BRADSHAW 1983; BRADSHAW and HOLZAPFEL 1986). The lack of differences in epistasis between distant northern but not relatively nearby southern populations may then be explained by the total number of generations elapsed since divergence of the parent populations. Our results suggest that differences in composite additive and dominance effects arise early in the genetic divergence of populations (as within the northern region) while differences due to epistasis accumulate after more prolonged isolation (as within the Gulf Coast region or between regions).

The specific components of digenic epistasis by which populations differ in critical photoperiod are virtually unique to each cross and unrelated to the historical range expansion of *W. smithii* in North America (Table 3; HARD *et al.* 1993a). The two exceptions (Table 3: Alp · Low, ME · ON) are in different geographic regions and are therefore likely convergent, rather than simultaneously or sequentially evolved genetic differences. Similarly, ARMBRUSTER *et al.* (1997) found that the genetic architecture underlying fitness (r_c) and its components varied in a pattern that was unique to each cross within or between the northern (ME, ON) or southern (western and eastern FL) regions. Genetic differences between populations of *W. smithii* have accumulated in a stochastic, rather than deterministic manner.

Isolation and drift have probably contributed to this uneven adaptive landscape. First, pitcher plants are often locally abundant but regionally scarce. *Wyeomyia smithii* is a small, weak-flying mosquito, "extremely prone to death by desiccation" (ISTOCK and WEISBURG 1987). Allozyme variation indicates pervasive gene flow within local habitats but limited gene flow between even nearby bogs (ISTOCK and WEISBURG 1987). The opportunity for isolation and drift therefore clearly exists. Second, average gene diversity at 10 allozyme loci declines with increasing latitude within the northern region (≥40°N) (P. A. ARMBRUSTER, W. E. BRADSHAW and C. M. HOLZAPFEL, unpublished results), suggesting that drift after sequential colonizations of progressively more northern bogs has depleted genetic variation at structural gene loci. Because of their strict host specificity, *W. smithii* cannot invade a new locality until *Sarracenia purpurea* has become established. We therefore view the northward adaptive radiation of *W. smithii* as the successive colonization by a few individuals into a wide-open habitat, followed by drift, rapid population growth, and mass selection.

This process of migration, drift, and selection can promote the rapid adaptation of populations under favorable environmental conditions by (1) increasing the efficacy of natural selection in expanding populations (SLATKIN 1996), (2) reordering the average effects of alleles (LANDE 1980a; GOODNIGHT 1995), (3) releasing additive from dominance and epistatic variances (MAYR 1954; CARSON 1968; WRIGHT 1977; TEMPLETON 1980; GOODNIGHT 1987; WILLIS and ORR 1993; BRYANT and MEFFERT 1995), (4) facilitating major changes in some traits while alleviating possible negative pleiotropic effects through the action of modifier loci (WEBER 1996), or (5) facilitating the independent evolution of formerly genetically correlated traits (HARD *et al.* 1993b).

Critical photoperiod of *W. smithii* has undergone rapid adaptive evolution in North America. Within the northern region previously occupied by the Laurentide Ice Sheet (>40°N), pitcher plants have become established, *W. smithii* has invaded the new habitats, and critical photoperiod has evolved about 5 SD (Figure 2) during the last 21 K years. The high heritability of critical photoperiod (HARD 1993a: $h^2 = 0.15–0.80$) and the lack of differences due to dominance or epistasis in the cross between NJ and ON (Tables 1 and 3) provide evidence that pre-existing additive genetic variance, mutation, and selection alone may have been sufficient to generate this degree of difference in mean phenotype. At the same time, the additive genetic variance for critical photoperiod increases with increasing latitude, especially within this same northern region, despite a history of directional selection on a latitudinal scale and similar stabilizing selection at the local scale (HARD *et al.* 1993a). This increase in variance cannot be ex-

plained by mutation-selection balance (HARD *et al.* 1993a) and therefore provides evidence for the release of additive from previously covert dominance or epistatic variance. We therefore conclude that the independent genetic architectures among populations of *W. smithii* have probably arisen both through mutation and through the genetic reorganization of pre-existing allelic and genic interactions following founder events. Regardless of the relative contribution of these two processes, our results underscore the fundamental role of nonadditive genetic effects to the adaptive diversification of populations.

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LITERATURE CITED

ANDREWARTHA, H. G., 1952 Diapause in relation to the ecology of insects. Biol. Rev. **27:** 50–107.

ARMBRUSTER, P., W. E. BRADSHAW and C. M. HOLZAPFEL, 1997 Evolution of the genetic architecture underlying fitness in the pitcher-plant mosquito, *Wyeomyia smithii.* Evolution **51:** 451–458.

BARTON, N. H., and M. TURELLI, 1989 Evolutionary quantitative genetics: how little do we know? Annu. Rev. Genet. **23:** 337–370.

BRADSHAW, W. E., 1976 Geography of photoperiodic response in a diapausing mosquito. Nature **262:** 384–386.

BRADSHAW, W. E., 1980 Blood-feeding and capacity for increase in the pitcher-plant mosquito, *Wyeomyia smithii.* Environ. Entomol. **9:** 86–89.

BRADSHAW, W. E., 1983 Interaction between the mosquito *Wyeomyia smithii,* the midge, *Metriocnemus knabi,* and their carnivorous host *Sarracenia purpurea,* pp. 161–189 in *Phytotelmata: Terrestrial Plants as Hosts for Aquatic Insect Communities,* edited by J. H. FRANK and L. P. LOUNIBOS. Plexus Publishing, Medford, NJ.

BRADSHAW, W. E., 1986 Variable iteroparity as a life-history tactic in the pitcher-plant mosquito, *Wyeomyia smithii.* Evolution **40:** 471–478.

BRADSHAW, W. E., and C. M. HOLZAPFEL, 1986 Geography of density-dependent selection in pitcher-plant mosquitoes, pp. 48–65 in *The Evolution of Insect Life Cycles,* edited by F. TAYLOR and R. KARBAN. Springer-Verlag, New York.

BRADSHAW, W. E., and C. M. HOLZAPFEL, 1990 Evolution of phenology and demography in the pitcher-plant mosquito, *Wyeomyia smithii,* pp. 47–67 in *Insect Life Cycles: Genetics, Evolution, and Coordination,* edited by F. GILBERT. Springer-Verlag, London, UK.

BRADSHAW, W. E., and L. P. LOUNIBOS, 1977 Evolution of dormancy and its photoperiodic control in pitcher-plant mosquitoes. Evolution **31:** 546–567.

BRYANT, E. H., and L. M. MEFFERT, 1995 An analysis of selectional response in relation to a population bottleneck. Evolution **49:** 626–634.

CARSON, H. L., 1968 The population flush and its genetic consequences, pp. 123–138 in *Population Biology and Evolution,* edited by R. C. LEWONTIN. Syracuse University Press, Syracuse, NY.

CAVALLI, L. L., 1952 An analysis of linkage in quantitative inheritance, pp. 135–144 in *Quantitative Inheritance,* edited by E. C. R. REEVE and C. H. WADDINGTON. Her Majesty's Stationery Office, London, UK.

CHARLESWORTH, B., 1990 Optimization models, quantitative genetics, and mutation. Evolution **44:** 520–538.

CHEVERUD, J. M., 1984 Quantitative genetics and developmental constraints on evolution by selection. J. Theor. Biol. **110:** 155–171.

COCKERHAM, C. C., 1986 Modifications in estimating the number of genes for a quantitative character. Genetics **114:** 659–664.

CROW, J. F., and M. KIMURA, 1970 *An Introduction to Population Genetics Theory.* Harper & Row, New York.

DANILEVSKII, A. S., 1965 *Photoperiodism and Seasonal Development in Insects.* Oliver and Boyd, Edinburgh, UK.

DANKS, H. V., 1987 *Insect Dormancy: An Ecological Perspective.* Biological Survey of Canada (Terrestrial Arthropods), Ottawa, Canada.

DARSIE, R. F., JR., and R. A. WARD, 1981 *Identification and Geographical Distribution of the Mosquitoes of North America, North of Mexico.* Mosq. Syst. Suppl. **1:** 1–313.

FALCONER, D. S., 1981 *Introduction to Quantitative Genetics.* Longman, New York.

FISHER, R. A., 1958 *The Genetical Theory of Natural Selection.* Dover Publications, New York.

GILLESPIE, J. H., and M. TURELLI, 1989 Genotype-environment interactions and the maintenance of polygenic variation. Genetics **121:** 124–138.

GOODNIGHT, C. J., 1987 On the effect of founder events on epistatic genetic variance. Evolution **41:** 80–91.

GOODNIGHT, C. J., 1995 Epistasis and the increase in additive genetic variance: implications for phase 1 of Wright's shifting-balance process. Evolution **49:** 502–511.

HARD, J. J., W. E. BRADSHAW and C. M. HOLZAPFEL, 1992 Epistasis and the genetic divergence of photoperiodism between populations of the pitcher-plant mosquito, *Wyeomyia smithii.* Genetics **131:** 389–396.

HARD, J. J., W. E. BRADSHAW and C. M. HOLZAPFEL, 1993a The genetic basis of photoperiodism and evolutionary divergence among populations of the pitcher-plant mosquito, *Wyeomyia smithii.* Am. Nat. **142:** 457–473.

HARD, J. J., W. E. BRADSHAW and C. M. HOLZAPFEL, 1993b Genetic coordination of demography and phenology in the pitcher-plant mosquito, *Wyeomyia smithii.* J. Evol. Biol. **6:** 707–723.

HAYMAN, B. I., 1958 The separation of epistatic from additive and dominance variation in generation means. Heredity **12:** 371–390.

HAYMAN, B. I., 1960a The separation of epistatic from additive and dominance in generation means II. Genetica **31:** 133–146.

HAYMAN, B. I., 1960b Maximum likelihood estimation of genetic components of variation. Biometrics **16:** 369–381.

ISTOCK, C. A., and W. G. WEISBURG, 1987 Strong habitat selection and the development of population structure in a mosquito. Evol. Ecol. **1:** 348–362.

LANDE, R., 1980a Genetic variation and phenotypic evolution during allopatric speciation. Am. Nat. **116:** 463–479.

LANDE, R., 1980b The genetic covariance between characters maintained by pleiotropic mutations. Genetics **94:** 203–215.

LANDE, R., 1981 The minimum number of genes contributing to quantitative variation between and within populations. Genetics **99:** 541–553.

LANE, J., 1953 *Neotropical Culicidae.* University of Sao Paulo, Sao Paulo, Brazil.

LYNCH, M., and B. WALSH, 1997 *Genetics and Analysis of Quantitative Traits.* Sinauer Associates, Sunderland, MA (in press).

LOUNIBOS, L. P., and W. E. BRADSHAW, 1975 A second diapause in *Wyeomyia smithii:* seasonal incidence and maintenance by photoperiod. Can. J. Zool. **53:** 215–221.

MASAKI, S., 1983 Introduction, pp. 1–5 in *Diapause and Life Cycle Strategies in Insects,* edited by V. K. BROWN and I. HODEK. Dr. W. Junk Publishers, The Hague, Netherlands.

MATHER, K., and J. L. JINKS, 1982 *Biometrical Genetics: The Study of Continuous Variation,* Ed. 3. Chapman & Hall, New York.

MAYR, E., 1954 Change of genetic environment and evolution, pp. 157–180 in *Evolution as a Process,* edited by J. S. HUXLEY, A. C. HARDY and E. B. FORD. Allen & Unwin, London, UK.

MCDANIEL, S. T., 1966 A taxonomic revision of *Sarracenia* (Sarraceniaceae). Doctoral Dissertation, Florida State University, Tallahassee, FL.

O'MEARA, G. F., and L. P. LOUNIBOS, 1981 Reproductive maturation in the pitcher-plant mosquito, *Wyeomyia smithii.* Physiol. Entomol. **6:** 437–443.

O'MEARA, G. F., L. P. LOUNIBOS and R. A. BRUST, 1981 Repeated

egg clutches without blood in the pitcher-plant mosquito. Ann. Entomol. Soc. Am. **74:** 68–72.

RICE, W. R., 1989 Analyzing tables of statistical tests. Evolution **43:** 223–225.

ROBERTSON, F. W., 1957 Studies in quantitative inheritance. XI. Genetic and environmental correlation between body size and egg production in *Drosophila melanogaster.* J. Genet. **55:** 428–443.

ROSE, M. R., 1985 Life history evolution with antagonistic pleiotropy and overlapping generations. Theor. Popul. Biol. **28:** 342–358.

ROSS, H. H., 1964 The colonization of temperate North America by mosquitoes and man. Mosq. News **24:** 103–118.

SLATKIN, M., 1996 In defense of founder-flush theories of speciation. Am. Nat. **147:** 493–505.

STONE, A., K. L. KNIGHT and H. STARKE, 1959 *A Synoptic Catalog of the Mosquitoes of the World (Diptera, Culicidae).* Entomological Society of America, Washington, DC.

TAUBER, M. J., C. A. TAUBER and S. MASAKI, 1986 *Seasonal Adaptations of Insects.* Oxford University Press, New York.

TAYLOR, F., and J. B. SPALDING, 1986 Geographical patterns in the photoperiodic induction of hibernal diapause, pp. 66–85 in *The Evolution of Insect Life Cycles,* edited by F. TAYLOR and R. KARBAN. Springer-Verlag, New York.

TEMPLETON, A. R., 1980 The theory of speciation *via* the founder principle. Genetics **94:** 1011–1038.

VIA, S., and R. LANDE, 1985 Genotype-environment interaction and the evolution of phenotypic plasticity. Evolution **39:** 505–523.

WEBER, K. E., 1996 Large genetic change at small fitness cost in large populations of *Drosophila melanogaster* selected for wind tunnel flight: rethinking fitness surfaces. Genetics **144:** 205–213.

WEGIS, M. C., W. E. BRADSHAW, T. E. DAVISON, and C. M. HOLZAPFEL, 1997 Rhythmic components of photoperiodic time measurement in the pitcher-plant mosquito, *Wyeomyia smithii.* Oecologia **110:** 32–39.

WHITLOCK, M. C., P. C. PHILLIPS, F. B.-G. MOORE and S. J. TONSOR, 1995 Multiple fitness peaks and epistasis. Annu. Rev. Ecol. Syst. **26:** 601–629.

WILLIS, J. H., and A. H. ORR, 1993 Increased heritable variation following population bottlenecks: the role of dominance. Evolution **47:** 949–956.

WRIGHT, S., 1968 *Evolution and the Genetics of Populations,* Vol. 1. *Genetic and Biometric Foundations.* University of Chicago Press, Chicago, IL.

WRIGHT, S., 1977 *Evolution and the Genetics of Populations,* Vol. 3. *Experimental Results and Evolutionary Deductions.* University of Chicago Press, Chicago, IL.

ZENG, Z.-B., D. HOULE and C. C. COCKERHAM, 1990 How informative is Wright's estimator of the number of genes affecting a quantitative character? Genetics **126:** 235–247.

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