

Heritability and physiological correlates of migratory tendency in the grasshopper *Melanoplus sanguinipes*

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Abstract. Durations of tethered flights by the North American migratory grasshopper *Melanoplus sanguinipes* Fabricius are bimodally distributed: most individuals either will not fly, or else will fly for many hours. This observation suggests a simple measure (the ‘one-hour rule’) for distinguishing migrants from non-migrants. This measure is repeatable (repeatability = 0.6–0.7). Using laboratory-reared offspring of grasshoppers from an Arizona population of mixed migratory tendency, a breeding experiment was conducted to determine the heritability of migratory tendency and possible correlated responses to selection on migratory behaviour. When migratory tendency is considered as a threshold trait, the heritability of liability is in the range 0.5–0.6. Most families in the breeding experiment had at least some migrants among their offspring; selection on migratory incidence had a correlated effect on the durations of flights by these individuals. The magnitude of thoracic lipid reserves showed a modest correlated response to selection on migratory behaviour. Thoracic and abdominal lipid reserves in identified migrants are reduced by flight, indicating that lipid is mobilized and consumed during flight in this species.

Key words. Genetic correlates, heritability, lipid mobilization, *Melanoplus sanguinipes*, migration, threshold trait.

Introduction

The North American migratory grasshopper *Melanoplus sanguinipes* Fabricius (Orthoptera: Acrididae) exhibits within- and between-population variation in migratory tendency, as determined both from field observations and from tethered-flight assays (McAnelly, 1985). This variation evidently has a substantial genetic component. McAnelly (McAnelly, 1985; McAnelly & Rankin, 1986) compared grasshoppers derived from three natural populations that differed in migratory incidence. Laboratory colonies maintained the migratory incidences of the source populations over several generations of culture. Group crosses between a sedentary population and a highly migratory population yielded offspring with migratory incidence similar to the more sedentary population. Results from reciprocal between-population crosses provided no evidence of maternal effects. Furthermore, colonies of grasshoppers identified as either migrants or non-migrants by tethered-flight assays produced offspring with migratory incidence similar to their parents.

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In the locusts *Locusta migratoria* and *Schistocerca gregaria*, migratory behaviour, like much of locust behaviour and physiology associated with phase polymorphism, is strongly influenced by environmental factors. This is not the case in *M. sanguinipes*. McAnelly (1985) examined the effect on flight behaviour of varying environmental factors, including rearing temperature, food moisture, photoperiod and larval density. None of these manipulations had a significant effect on migratory incidence.

The migratory physiology of *M. sanguinipes* is similar in key respects to that of *Locusta* and *Schistocerca*, the well-established models for migratory energetics (Kent *et al.*, 1997). McAnelly’s results suggest that *M. sanguinipes* may serve, more so than the locusts, as a model system for relating migratory physiology to genetic factors.

In some insect species, migration is closely tied to specific stages in the life cycle, and in these cases a natural measure of the behaviour suggests itself. For example, the armyworm moth *Spodoptera exempta* makes a long dispersing flight on a night soon after emergence. Consequently, in this species, duration of the (tethered) emergent flight has been used as a measure for artificial selection (Parker & Gatehouse, 1985). The flight behaviour of *M. sanguinipes*, however, is not closely tied to a particular stage of the life cycle. Although oviposition does tend to reduce the frequency of flights in females

(McAnelly & Rankin, 1986), flights can occur at any time during adulthood. This species can be thought of as comprising 'opportunistic migrants' that can be stimulated to fly by some (not yet well-understood) interaction of external and internal cues.

Individual *M. sanguinipes* in tethered-flight experiments may not fly on every trial, yet some individuals are clearly more likely to fly than others when tested repeatedly. Furthermore, individuals tend either to fly < 30 min (or not at all) or to make uninterrupted flights of several hours (Fig. 1). This bimodal distribution of flight behaviour led McAnelly (1985) to propose a one-hour rule: an individual that makes at least one 60-min flight in several trials (in her study, 3–5) can be classified as a migrant. Parker *et al.* (1955) estimated the ground speed of migratory swarms in the field, with a light tailwind, to be 16–19 km/h; at this rate, even one hour of flight would move a grasshopper far from its original habitat.

McAnelly's one-hour rule has been used to identify presumed migrants and non-migrants in subsequent studies (Rankin & Burchsted, 1992; Kent *et al.*, 1997). The one-hour rule is convenient for many sorts of experimental work, because it allows determination of migratory tendency without lengthy flights to voluntary cessation. (When flights to cessation are permitted, some last longer than 10 h, and the median duration in one set of experiments was 232 min; Kent *et al.*, 1997). The variance of durations of flights to cessation, both between and within individuals, is very large.

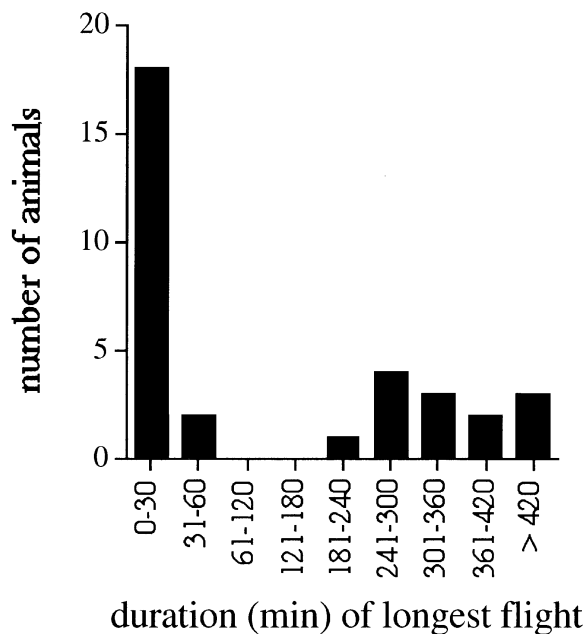


Fig. 1. Distribution of durations of tethered flights by sons of field animals collected in 1992. Grasshoppers were given three opportunities to fly to voluntary cessation. Each duration counted represents the longest flight by a different individual.

The utility of the one-hour rule as a basis for genetic analysis depends in part on the repeatability of the measure. The repeatability,

$$rep = 1 - \frac{\text{var}_{\text{within individuals}}}{\text{var}_{\text{total}}}$$

is the proportion of phenotypic variance exclusive of each individual's variation in performance in repeated trials. Because within-individual variation is a component of environmental variance that is least susceptible to experimental control, the repeatability defines a theoretical upper limit to the heritability ($= \text{var}_{\text{additive genetic}} / \text{var}_{\text{total phenotypic}}$) (Falconer, 1989; p. 140).

The present study used *M. sanguinipes* derived from an Arizona U.S.A. population that exhibits mixed migratory tendency. Tethered-flight data from McAnelly (1985) were used to estimate the repeatability of the one-hour rule. Two breeding experiments, beginning with the offspring of two different cohorts of field collected adults, were performed using the one-hour rule as the measure of selection. In each experiment, pairs of grasshoppers of known migratory tendency were mated, and the migratory tendency of their offspring was used to estimate the realized heritability of the trait in the Arizona population. In addition, in the first breeding experiment, a number of physiological characters were compared in the offspring to identify any correlated responses to selection on migratory tendency.

Among the characters examined for evidence of correlated response were lipid reserves in the thorax and abdomen. Lipid is likely to be the primary fuel for long-duration flight in *M. sanguinipes*, as in locusts; however, experimental evidence in this species of lipid consumption during flight has been ambiguous, perhaps because of the large variance of whole-body lipid reserves (Kent *et al.*, 1997). In the present study, the mobilization of lipid reserves in different compartments of the grasshopper body during flight is examined, and the magnitude of these reserves is compared in resting migrants and non-migrants.

Materials and Methods

Collection, rearing, and tethered-flight assay

In late June of most years of this study (July in 1994), approximately 900 adult *M. sanguinipes* were collected on the San Carlos Apache Reservation, Arizona. These insects were maintained in caged colonies of 100–150 grasshoppers per cage in an environmental chamber at the University of Texas at Austin, as described (Kent *et al.*, 1997). Eggs were collected every 4 days, held at 31 °C for 10 days, then transferred to storage in moist vermiculite at 8–12 °C. A small percentage of these eggs are capable of hatching without diapause, but most require at least 6 weeks of cold exposure to break diapause. Eggs remain viable in cold storage for at least 12 months. The median time to non-diapause hatch in laboratory conditions is

21 days; because eggs were transferred to cold within 10–14 days after oviposition, any eggs predisposed to hatch without diapause should have had their development arrested by the cold exposure. All experiments in this study began with the laboratory-reared offspring of field-collected animals.

Breeding experiments: mated pairs

During the period November 1992 to January 1993, a breeding experiment was begun using the offspring of field animals collected in June 1992. Twenty-four pairs of grasshoppers previously identified as migrants or non-migrants, based on their performance in 80-min flight tests on the 4th, 6th and 8th day after eclosion, were maintained in 46 cm³ cages as described previously (Kent *et al.*, 1997). Each female was allowed to oviposit in a small plastic cup filled with moist sand. Egg cups were replaced every 10 days; egg pods (encapsulated clutches) were transferred to moist vermiculite, held in the rearing chamber (31 °C) for 4 days, then stored at 8–12 °C for 9–12 months. At the end of the chilling period, the egg pods in vermiculite were allowed to hatch in an environmental chamber at 31 °C and a LD 12 : 12 h photocycle. Adult survivors were obtained from 10 of the 24 crosses. One of these families had only one surviving offspring, which was not used. Some difficulty was experienced in breaking the egg diapause in laboratory conditions, even with cold treatment, which reduced egg hatch. Hatching success was not related to migratory tendency of the parents (see Results). At eclosion, the adult offspring were removed to individual cages maintained in a LD 16 : 8 h photocycle. These grasshoppers were flight-tested on the 4th, 6th and 8th days after eclosion, and were allowed to fly until voluntary cessation on each trial. Although total flight duration was recorded for each trial, the migratory tendency of each offspring was determined using the one-hour rule (a score of 1 for any flight > 60 min in at least one trial, or 0 otherwise).

A second breeding experiment was begun in December 1994, using the progeny of animals collected in July of that year. The late collection in this year may have had a negative effect on the laboratory colony: migratory tendency was unusually low in the offspring of these animals, both in this breeding experiment and in unrelated experiments using the same colony. In retrospect, this lower performance may have been due to departure of migrants from the field population associated with weather systems in late June. Twenty-seven crosses were made, including migrant × migrant, non-migrant × non-migrant, and reciprocal migrant × non-migrant crosses.

Maintenance and egg collection were as described above. Eggs were transferred to hatching conditions in March 1995 (after only 3 months in cold storage). Adult offspring were obtained from only six of the 27 crosses, because many nymphs died in the first instar. The reason for this mortality is unclear: there were no external signs of *Malameba* infection or other parasitism. As in the first breeding experiment, however, hatching success and survival did not appear to be correlated with migratory tendency. Both parents and offspring in this

experiment were flight-tested for 80 min on the 4th, 6th and 8th days after eclosion.

Anatomical characters

At the conclusion of the 1992–93 breeding experiment, grasshoppers were maintained in pairs in 46 cm³ cages until natural death. At death, most of these animals were frozen in labelled tubes at –80 °C (some individuals were lost haphazardly). In 1998, these specimens were measured for six characters potentially correlated with migratory tendency: body size, wing length, thoracic circumference, thoracic muscle mass, thoracic lipid mass and abdominal lipid mass.

Processing and measurement of grasshoppers were carried out over several days as follows: each grasshopper was thawed and weighed. The lengths of a hind femur and the folded wings were measured with a micrometer (Manostat, New York). Thoracic circumference was measured with a piece of waxed ribbon dental floss (Johnson & Johnson Dentotape) wrapped snugly across the fold in the pronotum and immediately behind the prothoracic legs. Lipid was extracted from the abdomen and the eviscerated thorax by incubation of each overnight in 3 mL of 2 : 1 v/v chloroform : methanol. Lipid extracts were dried on tarred aluminium foil pans and weighed on a Mettler microbalance (Mettler, Hightstown, NJ).

The defatted 'flight apparatus' (eviscerated thorax with wings) was dried in an oven at 60–70 °C, then weighed; the residue was incubated for 48 h in two 4 mL portions of 2% sodium hydroxide, then washed, dried and re-weighed. The mass of gross thoracic muscle and connective tissue (primarily flight muscle) was measured as the difference in weight between the dried, defatted tissue and its cleared exoskeleton.

Analysis of lipid reserves by body compartment

This comparison used the laboratory-reared offspring of grasshoppers collected in various years. Males were flight-tested for 90 min at 4–5 days after eclosion; those that did not fly for at least 60 min on the first trial were re-tested 2 days later. Individuals that flew for at least 60 min in either trial were classified as migrants, and those that flew < 60 min in either trial were classified as non-migrants. To maximize observable differences between migrants and non-migrants, those few males that flew longer than 10 but less than 60 min were removed from the experiment.

Results

Repeatability of the one-hour rule

McAnelly (1985) reported the flight behaviour of 17 female and 19 male grasshoppers flight-tested for 60 min every other day from the 4th to the 28th day after eclosion (Fig. 2). In these data, the repeatability of any single tethered-flight test is

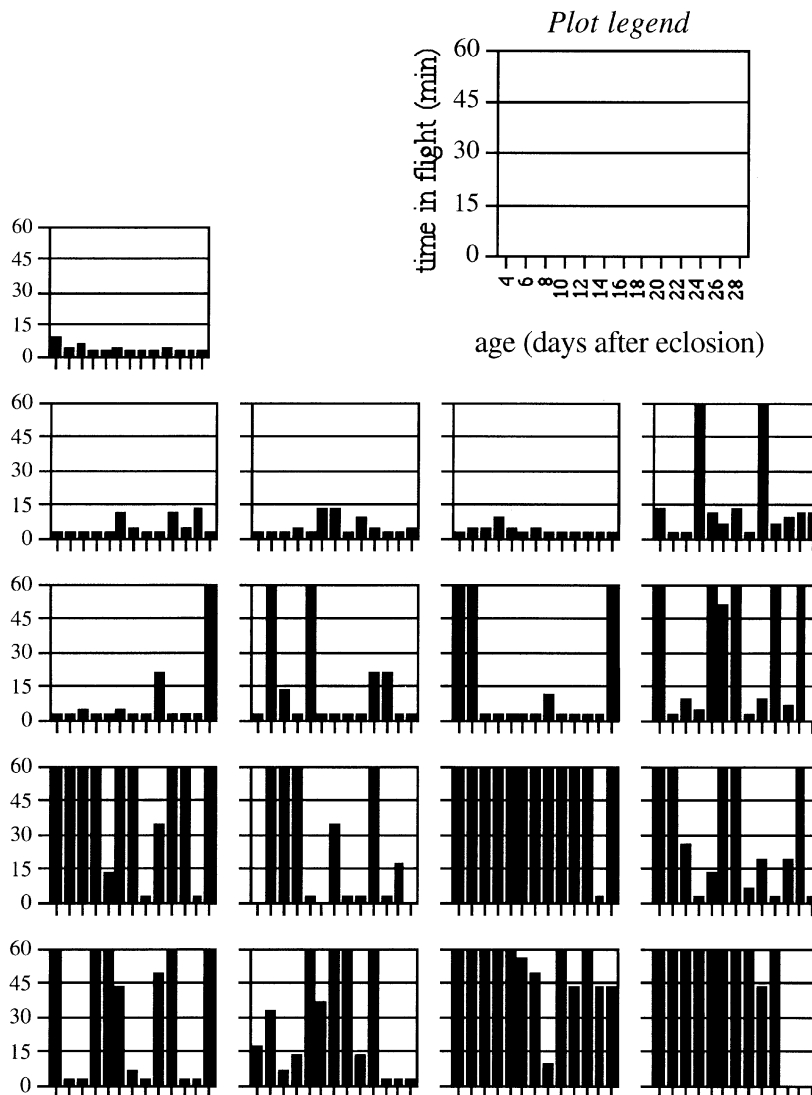


Fig. 2. McAnelly's flight records for 17 females assayed every other day from 4 to 28 days after eclosion. Flight performance by each individual varied from trial to trial, but an individual that made one 60 min flight was likely to make another (see text for discussion). Each shaded bar represents the total minutes of flight during a 60-min assay (re-drawn from McAnelly, 1985).

Table 1. Estimate of repeatability (mean \pm SE) of 'one-hour rule' using repeated flight measures reported in McAnelly (1985). Repeatability is estimated for individual tests (1 = flight \geq 60 min, 0 otherwise); and pairs or triplets of successive tests (1 = at least one flight \geq 60 min, 0 otherwise). Repeatability is estimated as the intraclass correlation of repeated measures (class = individual) (Falconer, 1989). Standard errors are calculated as in Becker (1984).

	<i>n</i>	Single tests	Pairs of tests	Triplets of tests
Females	17	0.300 \pm 0.091	0.471 \pm 0.115	0.618 \pm 0.111
Males	19	0.524 \pm 0.093	0.653 \pm 0.090	0.661 \pm 0.097

modest; however, the repeatability of at least one 60 min performance in three successive tests is about 0.62 in females and 0.66 in males (Table 1).

Pooling repeated measures in this way creates a new measure with a higher repeatability. This would not be the case if 60-min flights were performed at random with respect to individuals. To demonstrate this, 'pseudodata' were prepared, consisting of flight records for 17 hypothetical grasshoppers. The value of each hypothetical flight test was assigned using a random number table, with a score of 1 assigned if the random number was 0, 1 or 2; or a score of 0 assigned otherwise. The result was a randomized set of simulated flight records with

Table 2. Estimate of repeatability (mean \pm SE) of randomly assigned test scores (1 or 0) with the same overall frequency of scores (approx. 0.3) as the female flight data in Table 1. For single scores and pairs of scores, the estimated repeatabilities are not significantly different from zero. The repeatability of successive triplets of scores is actually less than zero (i.e. the sample estimate of within-individual variance is greater than the sample estimate of total variance).

	<i>n</i>	Single tests	Pairs of tests	Triplets of tests
Pseudodata	17	0.019 \pm 0.034	-0.012 \pm 0.051	-0.163 \pm 0.061

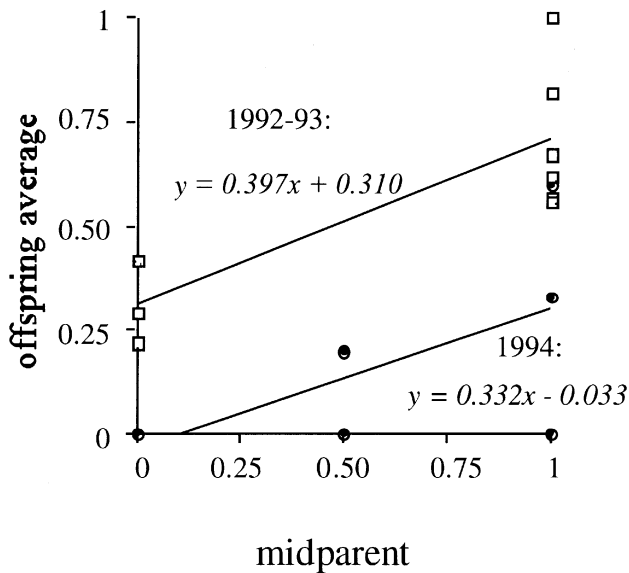


Fig. 3. Offspring mean on midparent regression of migratory incidence for breeding experiments begun in 1992-93 (open symbols) and 1994 (closed symbols).

approximately the same overall frequency of 60-min flights (c. 30%) as in McAnelly's data for females. The repeatability of single flight tests is not significantly different from zero in these pseudodata, and is not improved when successive flights are pooled (Table 2).

Breeding experiments: hatching success

As noted in Materials and Methods above, a substantial proportion of the mated pairs in the two breeding experiments had no surviving adult offspring. This was due less to infertility than to low hatching success (perhaps because our conditions did not break the egg diapause consistently). In the first breeding experiment, for example, 24 pairs were mated, 20 produced eggs and 10 had offspring that survived to adulthood. Reproductive failure was not associated with migratory tendency. Even though no migrant \times non-migrant pairs ($n = 5$) in the first experiment had surviving offspring, all

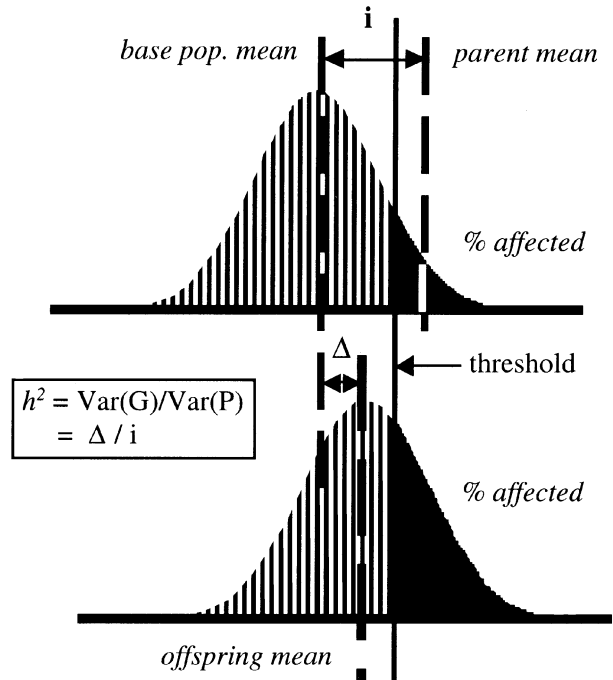


Fig. 4. Model of threshold selection: affected individuals (dark area) have more than a threshold value of an inferred, normally distributed trait, the liability. If the incidence of affected individuals in the parent population is $x\%$, then the threshold is the cut-off point, in standard deviation units, for the portion of the normal distribution holding $x\%$ of the probability mass. If the variances of the parent and offspring populations are equal (so that both distributions are on the same scale), and if the threshold is the same for both populations, then the intensity of selection, i , and the response to selection, Δ , can be calculated from the parent and offspring incidences. The heritability (h^2) is estimated as Δ/i (i = mean of affected parents - parental population mean; Δ = offspring population mean - parental population mean.) (After Falconer, 1965).

of these crosses produced eggs. Infertility was not significantly associated with the type of cross (contingency table: d.f. = 2, $\chi^2 = 2.667$, $P = 0.264$), and the various types of crosses did not differ significantly in number of egg pods produced (ANOVA: $F_{2,21} = 0.276$, $P = 0.761$) or number of surviving offspring (ANOVA: $F_{2,21} = 1.971$, $P = 0.164$).

Heritability of migratory incidence

Heritability (h^2) of migratory incidence was estimated by regression of average offspring values on midparent values, using data from the breeding experiments begun in 1992 and 1994 (Fig. 3). For the 1992 experiment, h^2 (= slope of the regression line) is 0.397 (95% CI: $0.139 \leq h^2 \leq 0.656$). Flight performance was lower in all families in the 1994 experiment. However, the estimate of h^2 is similar to that obtained in the earlier experiment (0.332; this estimate is not significantly

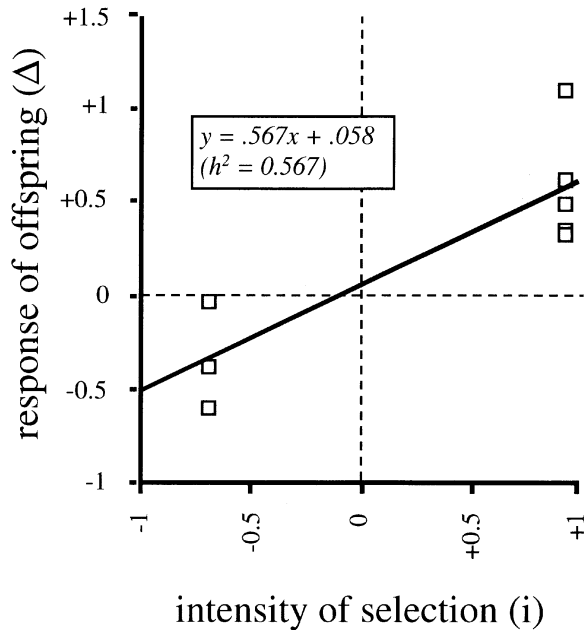


Fig. 5. Offspring mean on midparent regression of migratory liability for 1992–93 breeding experiment. Family Q (number of offspring = 4) had incidence = 1, or infinite liability when transformed to standard deviation units; it has been excluded from this analysis.

different from zero (95% CI: $-0.355 \leq h^2 \leq 1.019$), perhaps because of the small sample size).

Heritability of migratory liability

Whereas the heritability of migratory behaviour can be estimated from the incidences as above, dichotomously defined polygenic traits can be analysed as threshold traits (Falconer, 1965, 1989). The underlying (unknown) physiological basis of the trait, termed the liability, is assumed to be continuously distributed; individuals possessing more than some threshold value of the liability express one of the two possible phenotypes (Fig. 4). Threshold analysis requires that incidence be transformed to liability (mean standard deviation of subpopulations from a population mean), which may afford a better fit to the linear assumptions of the heritability model.

The liability transformation calculated by Falconer's method requires an independent estimation of the source population mean. In addition, the transformation gives infinite values when the incidence is either zero or unity, as is likely to occur when family sizes are small. For both of these reasons, h^2 of liability was estimated only for the breeding experiment that began in 1992–93. Some animals from the offspring of the 1992 field collection were flight-tested in an unrelated experiment (B.V. Sorenson, J.C.A. Burchsted & M.A. Rankin, unpublished observations), providing an independent estimate of the incidence of migratory tendency in this population. In this experiment ($n = 37$ males + 41 females),

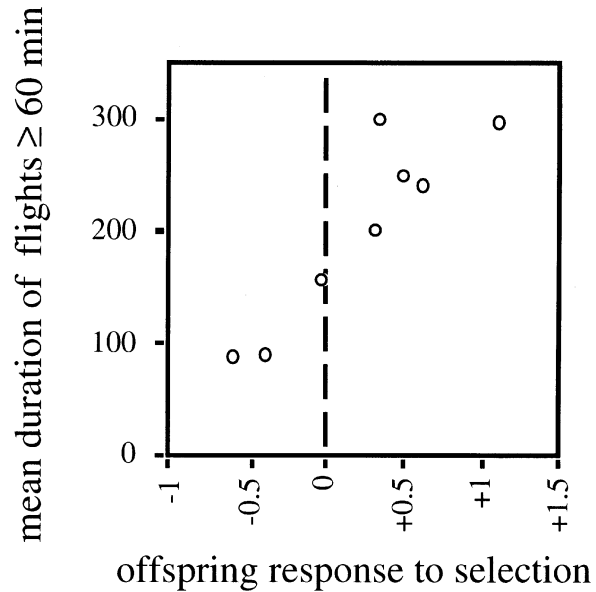


Fig. 6. Correlation between each family's response to selection on migratory liability and the mean duration of longest flights = 60 min by offspring in each family. Correlation coefficient $r = 0.936$; $P = 0.0006$. Family Q excluded from analysis (see legend to Fig. 4).

43% of grasshoppers performed at least one flight = 60 min in three trials.

Based on deviations from this independent estimate of migratory incidence in the parental laboratory population, h^2 can be estimated as the slope of the regression of offspring mean liability on midparent liability for each family (as suggested by Fig. 3 of Falconer, 1965). This regression (Fig. 5) estimates h^2 of migratory liability to be 0.567 (95% CI of slope: $0.229 \leq h^2 \leq 0.906$).

Correlated effect on flight duration

Offspring in the 1992–93 experiment were allowed to fly to voluntary cessation. Among those grasshoppers in each family that flew ≥ 60 min, the mean duration of longest flights by individuals was significantly correlated with the mean liability of their respective families (Fig. 6).

Correlated response of anatomical characters

Female *M. sanguinipes* are significantly larger than males, but there was no significant within-sex difference in body size among offspring of migratory vs. non-migratory parents. Other anatomical characters were analysed on a size-corrected basis. The sexes were not significantly different for these characters, and consequently data for both sexes were pooled (Table 3). Four of these characters did not differ significantly with respect to selection regime; however, size-corrected residual thorax

Table 3. Correlated responses of anatomical characters. Comparison of offspring of migrants (six families) and non-migrants (three families) for body size (as hind femur length) and residuals of linear regression of five other anatomical characters on body size. Units of measurement: mm (body size, wing length, thorax circumference) and mg (thoracic lipid and muscle, abdominal lipid). Comparisons by ANOVA.

Character	Migratory families Means \pm SE (<i>n</i>)	Non-migratory families Means \pm SE (<i>n</i>)	<i>F</i>	<i>P</i>
Body size	13.3 \pm 0.2 (42)	13.4 \pm 0.2 (17)	0.122	0.728
Size-corrected residuals:				
Wing length	0.1 \pm 0.2 (41)	- 0.3 \pm 0.3 (17)	1.935	0.170
Thoracic circumference	- 0.0 \pm 0.2 (41)	0.0 \pm 0.3 (17)	0.012	0.913
Thoracic lipid	1.26 \pm 1.11 (41)	- 3.44 \pm 1.35 (15)	5.429	0.024*
Thoracic muscle	- 0.22 \pm 0.80 (41)	0.54 \pm 2.31 (17)	0.159	0.692
Abdominal lipid	0.23 \pm 0.79 (40)	- 0.55 \pm 1.12 (17)	0.303	0.584

Table 4. Comparison of resting lipid reserves in thorax and abdomen of identified migrants and nonmigrants. ANOVA: dependent variable: size-corrected lipid content (mg) (corrected by regression on hind femur length).

	Migrants	Non-migrants	<i>F</i>	<i>P</i>
Thoracic lipid: mean \pm SE (<i>n</i>)				
Cohort I	14.43 \pm 0.55 (6)	15.53 \pm 0.94 (11)	0.640	0.436
Cohort II	17.46 \pm 1.60 (3)	15.06 \pm 1.06 (17)	0.833	0.373
Abdominal lipid: mean \pm SE (<i>n</i>)				
Cohort I	8.74 \pm 0.96 (6)	10.19 \pm 0.70 (11)	1.514	0.237
Cohort II	10.37 \pm 2.12 (3)	8.19 \pm 0.64 (17)	1.578	0.225

Table 5. Change of thoracic reserves during flight. Males from three separate eclosion cohorts, previously identified as migrants by the one-hour rule, were either killed at rest (time = 0, *n* = 15) or allowed to fly to voluntary cessation (three individuals were arbitrarily stopped after 90 min of flight) and then killed within 15 min (*n* = 14). Data are analysed by multiple linear regression to account for effects of body size (measured as length of a hind femur) and differences in lipid reserves between cohorts (represented by indicator variables).

Variable	Partial regression coefficient	Partial slope	SE of slope	<i>t</i>	<i>P</i>
Thoracic lipid: R^2 (adjusted for sample size) = 0.839					
Time in flight	1.711	- 0.017	2.064	2.921	0.0075
Femur length	0.060	0.592	0.833	0.711	0.4839
Cohort A = 1	- 1.005	- 12.316	1.199	10.275	0.0001
Cohort B = 1	- 0.165	- 2.193	1.293	1.696	0.1028
Abdominal lipid: R^2 (adjusted for sample size) = 0.765					
Time in flight	1.711	- 0.014	2.064	2.906	0.0078
Femur length	0	0.001	0.698	0.002	0.9986
Cohort A = 1	- 0.945	- 8.032	1.005	7.992	0.0001
Cohort B = 1	- 0.188	- 1.737	1.084	1.602	0.1222

lipid mass was greater in offspring of migrants than in offspring of non-migrants.

Comparison of resting lipid reserves in identified migrants and non-migrants

Identified migrants and non-migrants were allowed to feed at will from flight-testing until 14 days after eclosion. They

were then killed (by freezing), dissected and assayed for lipid contents of eviscerated thorax and eviscerated abdomen. Lipid extraction and gravimetric analyses were performed as described in Materials and Methods.

Although offspring of migrants and non-migrants differed in magnitude of thorax lipid reserves, suggesting a correlated response to artificial selection on migratory tendency, resting thoracic and abdominal lipid reserves did not differ significantly in identified migrant and non-migrant males drawn from

the general laboratory population (Table 4). Presumably the magnitude of thoracic lipid reserves in these animals is influenced by non-genetic as well as genetic factors. (Laboratory-reared grasshoppers have less opportunity to be active than their conspecifics in the field, and also have access *ad libitum* to a more constant food source in the semiarid San Carlos habitat.) Lipid reserves were highly variable in these animals, but the variances did not differ significantly with respect to migratory tendency (*F*-test: thoracic lipid, $F_{[27,8]} = 2.774$, $< F_{0.05 [27,8]} = 3.10$; abdominal lipid, $F_{[8,27]} = 1.055$, $< F_{0.05 [8,27]} = 2.31$).

Change of lipid reserves during flight

Identified migrant males were assigned randomly to one of two treatments: to be flight-tested to voluntary cessation at 14 days after eclosion, and killed within 15 min of ceasing to fly; or to be killed at the same age without flight-testing. Each grasshopper was subsequently dissected and assayed for lipid contents of eviscerated thorax and eviscerated abdomen.

This analysis was carried out using three groups of grasshoppers from three different cohorts reared at different times; in each cohort, nymphs were reared together and eclosed within a week of one another. Each group was small, and the mean resting lipid reserves in each compartment differed in the three groups. Consequently, combined results from the three groups were analysed by multiple linear regression with the following independent variables: time in flight, length of a hind femur (a reliable measure of body size), and two indicator variables (Snedecor & Cochran, 1980) to distinguish the groups.

There was a significant decline of both thoracic and abdominal lipid reserves during extended flight (Table 5). Interestingly, in the identified migrants held as controls, there was no significant correlation between body size (measured as hind femur length) and the magnitude of lipid

reserves ($n = 15$; thoracic lipid, $r = 0.009$; abdominal lipid, $r = -0.103$).

Discussion

Migratory behaviour is limited to a particular time during the adult stage in many insect species; it may, for example, be closely tied to the reproductive cycle. *Melanoplus sanguinipes* appears to be more of an 'opportunistic migrant' in that migratory flights can occur at any time during at least the first month of adulthood if environmental conditions are favourable for flight initiation.

Melanoplus sanguinipes populations exhibit a bimodal distribution of migratory behaviour: individuals tend either to not fly (or make very short flights) or to fly > 1 h. Individuals that fly > 1 h are likely to do so again if the appropriate cues are present, although they may not fly at every opportunity. As noted earlier, this is the basis for McAnelly's (1985) one-hour rule for identifying presumed migrants and non-migrants. Single flight tests are modestly repeatable; the repeatability is increased to 0.6 or more in both sexes when the results of two or three successive tests are pooled. For single flight tests or pairs of tests, repeatability is somewhat greater in males than in females, perhaps because migrant females tend to show an interruption in flight behaviour around the time of first oviposition (15–23 days after eclosion in McAnelly's experiment) (see also McAnelly & Rankin, 1986). Application of the one-hour rule provides a convenient way of identifying presumed migrants and non-migrants for physiological and genetic experiments.

McAnelly's crosses of groups of grasshoppers within and between populations of different migratory incidence suggested that the behaviour has a substantial genetic component in this species. In our study, artificial selection on flight behaviour, measured by the one-hour rule, produced substantial change in migratory incidence within a single generation.

Table 6. Heritability estimates for migratory traits in several arthropod species.

Species	Behaviour or trait	h^2	Reference
<i>Melanoplus sanguinipes</i> (migratory grasshopper)	Migratory incidence	0.33–0.40	This study
	Migratory liability	0.53–0.77	
<i>Lygaeus kalmii</i> (milkweed bug)	Flight duration	0.20–0.41	Caldwell & Hegmann (1969)*
<i>Spodoptera exempta</i> (African armyworm)	Flight duration	0.50–0.88	Gatehouse (1986)*
<i>Epiphyas postvittana</i> (light brown apple moth)	Flight duration	0.43–0.57	Gu & Danthanarayana (1992)*
<i>Heliothis armigera</i> (cotton bollworm moth)	Total flight duration	0.39	Colvin & Gatehouse (1993)
	Longest single flight	0.15	
<i>Mythimna separata</i> (Oriental armyworm)	Flight duration	0.27	Han & Gatehouse (1993)
<i>Cydia pomonella</i> (codling moth)	Flight distance	0.37–0.57	Schumacher <i>et al.</i> (1997)
<i>Oligonychus pratensis</i> (Banks grass mite)	Initiation of wind dispersal	0.09	Margolies (1993)
<i>Tetranychus urticae</i> (two spotted spider mite)	Initiation of wind dispersal	0.28	Li & Margolies (1994)
<i>Laodelphax triatellus</i> (small brown planthopper)	Wing shape	0.27–0.36	Mori & Nakasuji (1990)*
<i>Dysdercus bimaculatus</i> (cotton stainer bug)	Wing length	0.51	Derr (1980)*
<i>Oncopeltus fasciatus</i> (milkweed bug)	Wing length	0.49–0.87	Dingle <i>et al.</i> (1988)*

*Reviewed in Dingle (1996).

Regression of offspring incidence on parental incidence yielded an estimate of realized heritability in the range 0.3–0.4.

The heritability estimates obtained in the present study suggest that there is substantial additive genetic variance for migratory tendency within a population (San Carlos) of mixed tendency. Heritability estimates obtained here are comparable to those found for other measures of migratory capacity in several species (Table 6). McAnelly (1985) had previously found evidence of a substantial genetic basis for migratory tendency in *M. sanguinipes* from reciprocal grouped crosses made between a highly migratory population (New Mexico) and a sedentary population (Colorado). The offspring in her cross-population study had a migratory tendency closer to the sedentary population, suggesting between-population dominance effects (Dingle, 1996; p. 356). Estimates of additive genetic variance within the New Mexico and Colorado populations were not made in her study.

Because our one-hour measure is dichotomous, migratory behaviour can be modelled as a threshold trait, and heritability can be estimated for another measure, the liability. This estimate is higher, in the range 0.5–0.8. The liability measure, being normally distributed, offers a better fit to the linear assumptions of the heritability model, reducing a component of phenotypic variance (in other words, liability is a transformation of incidence).

Interestingly, selection on liability produced a correlated change in the duration of flight by those individuals in any family that flew > 60 min. Similarly, in the blackcap warbler *Sylvia atricapilla*, migratory incidence is genetically correlated with the duration of a pre-migratory behaviour, *Zugunruhe* or nocturnal restlessness (Pulido *et al.*, 1996). Such correlation could not be assumed a priori. In the case of *M. sanguinipes*, it is possible to imagine a physiological mechanism underlying the behaviour (say, for example, some variation in the sensitivity of the insects' sensory system to the environmental cues that stimulate flight) that would have allowed a change in the incidence of migrants and non-migrants without a correlated change in flight capacity of migratory individuals. The very close correlation between migratory liability and flight duration in *M. sanguinipes* suggests that variation in migratory tendency is physiologically linked to variation in migratory capacity.

It would be interesting to undertake field studies to determine if migratory tendency in this species is a threshold trait with respect to natural selection (and not just for laboratory convenience), or if selection on flight duration predominates. The effect on the evolution of grasshopper populations should differ substantially, because threshold selection, unlike selection on duration, would be frequency dependent. Because threshold selection does not distinguish degrees of liability above the threshold limit, the intensity of selection is greatest when few individuals exceed the threshold; as more members of the population exceed threshold liability, their mean liability approaches that of the population as a whole, and intensity of selection approaches zero (see also Becker, 1984; Roff, 1996).

A number of studies in other species have examined anatomical and physiological characters correlated with

migratory tendency or flight capacity. In the crickets *Gryllus firmus* and *G. rubens*, the brachypterous morph exhibits a lower respiratory rate, lower triglyceride content and lower flight muscle mass than macropters (Zera & Mole, 1994). In four genera of planthoppers (*Javesella*, *Nilaparvata*, *Prokelisia* and *Sogatella*), body size is positively correlated with migratory incidence (Denno, 1994). Flight muscle degeneration occurs upon reproductive onset in both sexes of many species (e.g. the pine engraver beetle *Ips pini*; Robertson, 1998), resulting in a life-stage-specific correlation between muscle mass and flight behaviour. Palatable butterflies, which have more need for evasive manoeuvres than do aposematic unpalatable ones, exhibit morphological adaptations related to flight performance, including greater thoracic diameter (Chai, 1996) and location of the centres of body mass to optimize wing efficiency (Srygley & Dudley, 1993).

In our study, offspring of the 1992–93 breeding experiment were allowed to live out their natural lives. Subsequently, six characters were measured on these individuals: body size (represented by hind femur length; Burchsted, 1990), size-corrected wing length, thoracic circumference, thoracic lipid, thoracic muscle mass and abdominal lipid. One of these (thoracic lipid) was somewhat greater in offspring of migrants than in offspring of non-migrants. This correlated response makes sense if, as expected from physiological studies with locusts, lipid is the principal flight fuel in *M. sanguinipes*. In the present study, lipid content of both thorax and abdomen declined during flights to voluntary cessation, proportional to the duration of flight. This result does indeed suggest that lipid is a flight fuel in *M. sanguinipes*. (This evidence had eluded us in an earlier study (Kent *et al.*, 1997), perhaps because of the large variance of the whole-body lipid reserves analysed there.)

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