

LARVAL DISPERSAL AND CANNIBALISM IN A NATURAL POPULATION OF *ZOPHOBAS ATRATUS* (COLEOPTERA: TENEBRIONIDAE)

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Abstract. A natural population of the tenebrionid beetle *Zophobas atratus* was studied in an attic in El Zamorano, Honduras. Larvae were living in bat guano and were densest in the deepest part of the guano. No pupae or pharate pupae were found in the guano: they were always located a small distance away from the guano pile. Adults occurred on the guano surface and the beams and tiles of the roof. When larvae of three different ages and weights were marked and released in the guano, only the older, larger larvae were recaptured away from the guano, indicating a correlation between larval dispersal and increased weight and/or age. When 50 pharate pupae were exposed to a natural population of active larvae, only 4 (8%) survived after 5 days. This intense cannibalism of pharate pupae is consistent with the hypothesis that inhibition of pupation by crowding and the associated pre-pupal dispersal evolved, at least partly, in response to cannibalism.

Among insects, dispersal is commonly associated with high population density. For example, in aphids, crowding causes the production of alatae which fly to other sites and colonize them (Lees 1961). In the gypsy moth (*Porthetria dispar*) at high population density, first-instar larvae disperse aerially on silken threads (Leonard 1970) and a similar mechanism operates in the mite *Metatetranychus* sp. (Marle 1951). The phase change of migratory locusts at high densities is also essentially a dispersal response, for the gregarious-phase adults leave their home areas in large swarms (Uvarov 1977).

The dispersal of a pre-pupal stage from the area where it spent its larval life is another common phenomenon among insects (Price 1975). In many species, the apparent object of this dispersal is a sheltered spot for pupation, soil being the most common. The result of such dispersal is often a non-overlapping spatial distribution of life stages, with larvae and pupae occurring in different places.

The tenebrionid beetle *Zophobas rugipes* is unique, to date, in that if dispersal of the mature larva is prevented, metamorphosis is inhibited indefinitely, even until the larva dies of old age (Tschinkel & Willson 1971). The tendency to disperse increases as larvae grow, so that in a dispersal-measuring apparatus, young larvae disperse randomly, while old larvae overdisperse (Tschinkel & van Belle 1976). Overdispersal correlates with the ability to pupate upon isolation. At least in the laboratory, the observed overdispersion seems to be the result of fighting (Tschinkel 1978). Laboratory evidence suggests

that cannibalism of pharate pupae by active larvae may be the selective agent that has resulted in the evolution of the inhibition of pupation by crowding (Tschinkel & Willson 1971; Tschinkel 1978).

Among other tenebrionid beetles, the object of the dispersal is not always so clear. For example, *Tribolium* sp. larvae in complex habitats tend to disperse into warm regions of low larval density prior to pupation (King & Dawson 1973). Naylor (1965) found that larvae sporadically dispersed from high density vials, but the reason for the dispersal was obscure. Ghent (1966) found pupae to be more abundant near the tops of vials, evidence of possible site selection prior to pupation.

To date, all information on *Zophobas rugipes* has been the result of laboratory work. In order to show the importance of these findings to natural populations, it was necessary to establish at least two facts for wild populations: (1) that mature larvae actually disperse away from areas of high larval density, and (2) that cannibalism of pharate pupae actually occurs in wild populations when they are given the opportunity. The present study is the first that addresses both of these points in the field.

Materials and Methods

Locality of Study

The population was located in the attic of the Laboratory of Biology of the Pan American School of Agriculture in El Zamorano, Honduras (elevation 800 m). The attic was well ventilated by virtue of a tile roof, and this also provided

both access and roosts for several species of bats that produced the guano in which the beetle larvae live. The guano pile measured about 4×4 m and varied in depth from near 0 to about 8 cm, depending on the bats' favourite roosting sites (Fig. 1).

Population Sampling

The population was sampled by collecting all the guano within sample areas (414 cm^2) selected at random using a 0.5-m coordinate grid. The samples thus varied in volume depending upon the depth of the guano. All larvae in each sample were counted and weighed on a fibre balance calibrated with standard weights.

Species Identity

While my previous work has been carried out on *Z. rugipes*, it appears that this is probably not a biologically distinct species from *Z. atratus*. Cross-breeding experiments with both species have shown substantial to complete interfertility among the species from several localities throughout Central America (Tschinkel, unpublished). It is therefore likely that both names describe the same biological species.

Statistical Tests

In this report I utilize parametric statistical tests. However, I recalculated one set of tests (Fig. 2) using non-parametric statistics, and the conclusions were unchanged. I tested all other distributions for normality and found that they were all sufficiently close to justify using the *t*-test and its relatives.

Experiments and Results

The larval population was not evenly distributed, but was significantly concentrated ($P < 0.0001$) in the area of the deepest guano deposit (Fig. 1). In this area, the larval density varied between about 11 and 24 larvae/litre of guano. The state of guano pulverization was related to the population density. Areas with few larvae were generally characterized by intact faecal pellets, while those with many larvae showed partly to completely pulverized material. The attic received no rainwater, so all materials were dry.

The weights of the sampled larvae ranged from near zero to almost 900 mg, although few larvae weighed more than 650 mg (Fig. 2A). The shape of the weight-frequency curve (Fig. 2A) is

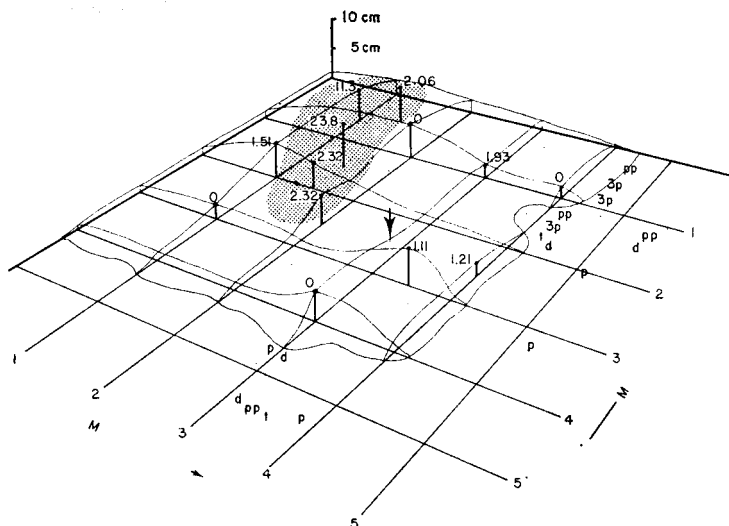


Fig. 1. Three-dimensional representation of the El Zamorano study site, showing the depth and distribution of guano, larval density (larvae/litre), and sites at which dispersing larvae (d), pharate pupae (pp), pupae (p), and teneral adults (t) were found. The horizontal grid is in metres with 0,0 at the southeastern corner of the attic. The vertical scale is exaggerated and in centimetres. The density of larvae per litre is noted next to the locations of the samples, and the area of highest density is shaded. The guano pile is approximately 4×4 m. The remaining attic floor is dusty, with much spider web and some leaf litter. The larvae were significantly clumped in the shaded area of the guano pile.

difficult to interpret, but suggests declining reproduction, since the smallest size classes are the least common. An alternative explanation might be cannibalism of smaller larvae and eggs by larger larvae. This occurs in laboratory populations (Tschinkel, unpublished).

Distribution of Life Stages

Pharate pupae, pupae, and teneral adults were not found in the guano deposit. On the other hand, 7 pharate pupae, 21 pupae, and 5 teneral adults were found within 1.5 m of the perimeter of the guano. Most of these metamorphosing individuals were found under loose boards or broken tiles, and within cracks in the attic floor (Fig. 1).

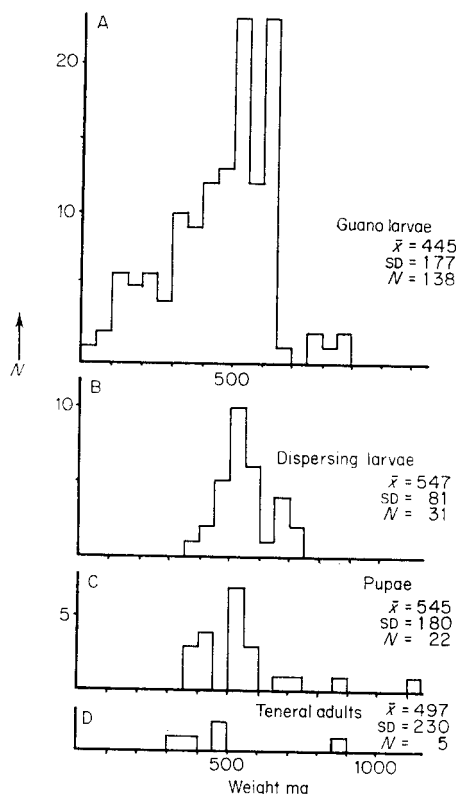


Fig. 2. The size-frequency distribution of larvae, dispersing larvae, pupae, and teneral adults from the natural population of *Zophobas atratus* in the attic of the Pan American School of Agriculture. The data for the dispersing larvae, pupae, and teneral adults are combined for January 1979 and August 1979. The guano larvae are from the random samples mapped in Fig. 1, while the remaining life stages are all those found. The weight of dispersing larvae is significantly less variable than that of guano larvae. The mean weights of dispersing larvae, pupae, and teneral adults are not significantly different.

In addition to metamorphic or post-metamorphic individuals, I captured 13 active larvae outside the guano pile, some of which showed signs of entering the pharate pupal stage (clearing a chamber, curling up, lying on one side). Figure 2B shows the weight-frequency distribution of all naturally dispersing larvae captured throughout this study. The population of dispersing larvae is significantly less variable (Ansari-Bradley Test, $P < 0.05$) than the general population. That these larvae are dispersing for pupation is supported not only by finding them at the same sites as pupae, but also by the similarity of the weight distributions of populations (NS, Ansari-Bradley Test) of dispersing larvae (Fig. 2B), pupae (Fig. 2C), and teneral adults (Fig. 2D). Some weight loss at each transformation has been observed in the laboratory (Tschinkel, unpublished), so that the mean weight declines from dispersing larvae to pupae to teneral adults.

Mature adults were found on the surface of the guano in the area of maximal larval density and hiding in the tiles and beams of the roof. Adults were never found burrowing in the guano.

The distribution of life stages suggests the following life cycle characteristics: the adults deposit eggs into the surface of the guano, the larvae feed on the guano and grow to some minimum size, whereafter they show an increasing probability of dispersing away from the crowded feeding population. Movement continues until the larvae come to rest in undisturbed spots, whereupon they metamorphose. The adults later return to the guano to reproduce, or possibly disperse to other localities. Tschinkel & Willson (1971) showed that crowding inhibits pupation in *Zophobas rugipes*, and hypothesized that larvae must therefore disperse in order to pupate. While the present evidence provides circumstantial support for this hypothesis, it would be desirable to show experimentally that large larvae disperse from natural populations, while small larvae do not. Such an experiment is described in the next section.

Dispersal of Marked Larvae in Relation to Weight and Age

Because the number of captured dispersing larvae in the attic population was never very large (possibly because of difficulty in searching many sites), I decided to use laboratory-reared larvae for this experiment. On my first visit to El Zamorano in January 1979, I returned several dozen adults to the laboratory at Tallahassee.

The adults were allowed to breed in boxes containing wheat bran, and were moved to a fresh box every month, thus creating a series of larval populations the ages of which differed by 1-month increments. Because dispersal from these culture boxes is impossible, and larvae continue to grow nevertheless, laboratory-reared larvae attain higher weights (up to 1300 mg) than larvae from natural populations.

Three laboratory groups were selected for the experiment carried out in August 1979: 200 small larvae approximately 2 to 3 months old, 120 larvae 4 to 5 months old, and 77 larvae 5 to 6 months old. The size distributions for these larvae are presented in Fig. 3. Each group was code-marked with different paint spots to allow recognition upon recapture. Marking was carried out in Tallahassee, 5 days before travel to and release in Zamorano. On the day of release in El Zamorano, the number of larvae in each group that had lost their paint spots (chiefly

through moulting) was counted in order to establish a rough estimate of the rate of mark loss (average about 2.5% per day). These larvae were then remarked.

All larvae of all three groups were released at a single spot near the northwest quarter of the guano pile (Fig. 1, arrow). The population of natural larvae in this area had increased noticeably since my January visit, so that a second, smaller population centre existed at this spot and the density was probably higher than Fig. 1 indicates.

In order to facilitate recapture of dispersing larvae, loose boards, broken tiles, and other litter were scattered about 0.5 m from the guano deposit margin. Larvae tend to pupate under such litter, and could thus be captured by daily inspections.

The results of 5 days of such recapture were unequivocal. Larvae from the youngest group were never recaptured away from the guano, while larvae from both older groups were regularly recaptured there. Furthermore, the highest rate of recapture was found for the oldest larvae (37%, corrected for mark-loss). Recapture for the middle group was still substantial (14%, corrected for mark-loss). These recapture patterns are significantly different from random ($P < 0.001$, χ^2 test).

The daily recapture rate is significantly higher for older than for middle larvae ($P < 0.05$, χ^2 test), but this difference declines over the 5-day experimental period (Fig. 4). The laboratory-reared dispersing larvae (Figs 3B and 3C) are much heavier than the naturally dispersing larvae (Fig. 2B) ($P < 0.001$, t -test). The fact that such a high proportion dispersed within 5 days indicates that the probability of dispersing increases with age and/or weight.

The population of 5- to 6-month-old larvae was significantly ($P < 0.05$, t -test) lighter upon recapture than it had been on release, but the variance remained the same (F -test). The weight loss probably resulted from dehydration. No significant weight loss was detectable for the 4- to 5-month-old larvae.

At the end of the 5-day recapture period, the entire guano pile was searched for marked larvae (Table I). Summing those recovered in the final search with those recaptured, and correcting for mark-loss, I was able to account for 59% of the oldest larvae, 26% of the middle larvae, and 31% of the youngest larvae. Of interest is the location of these larvae within the guano pile. Of the youngest larvae, 81% recovered were within 1.5

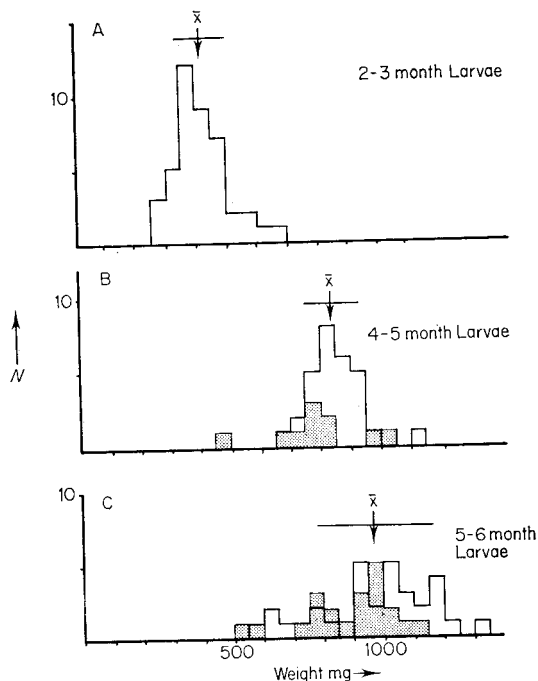


Fig. 3. Size-frequency distribution of the laboratory-reared larvae used in the dispersal experiment. Only a sample of larvae in the pre-release groups were weighed and histogrammed. The shaded histograms are the weights of all recaptured larvae of that group. The 5- to 6-month-old larvae were significantly lighter ($P < 0.05$, t -test) upon recapture than at release, probably as a result of dehydration during the intervening 5 to 10 days. Mean weights $\pm$ SD are indicated by arrows and bars.

m of the release point, while only 45% and 67% of the oldest and middle larvae, respectively, were within this area. However, a comparison (χ^2 test) of the final-search captures in the quarter of the guano in which they were released with the other three quarters showed that the age group composition just failed to attain significant difference ($P = 0.07$) between these areas.

Cannibalism of Pharate Pupae

Tschinkel & Willson (1971) noticed that, given the opportunity, cannibalism of pharate pupae

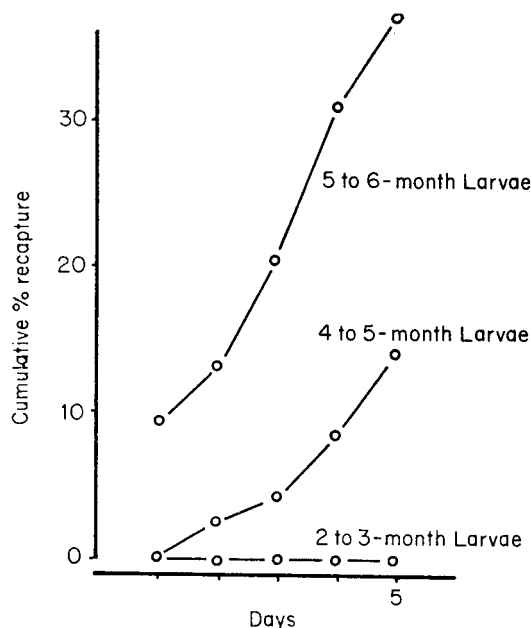


Fig. 4. Cumulative percentage of recaptured laboratory-reared marked larvae following release in a natural population. The percentage values are corrected for the estimated daily mark-loss. The oldest larvae disperse more rapidly and without a lag. No 2- to 3-month-old larvae were recaptured outside the guano. Daily recapture rates for 5- to 6-month-old larvae and 4- to 5-month-old larvae differ significantly ($P < 0.05$, χ^2 test).

by larvae was common in laboratory cultures and suggested that such cannibalism selected for inhibition of metamorphosis by crowding and the associated prepupal dispersal of larvae. Cannibalism is easy to demonstrate in the laboratory, but support of this hypothesis requires at least the demonstration of cannibalism in natural populations. The following experiment was therefore carried out.

Four- to five-month-old larval offspring of the adults collected in the Zamorano attic in January were isolated 5 days before travel to Zamorano in August. This is adequate time for them to have entered the pharate pupal stage irreversibly. About 70 such pharate pupae were taken into the attic: 20 were kept aside, while 50 were placed on the guano surface in the area of maximal larval density (Fig. 1). Both groups were checked daily, and any pupations or apparent disappearances were noted. The group of 20 kept aside allowed an estimate of the time of maximum pupal moulting (about 11 days after isolation). At the end of 5 days of exposure, the guano was searched carefully for any pupae that might have sunken below the surface.

There is a dramatic increase in unaccountable disappearances coincident with pupal moulting in the control group (Table II). By direct observation in the laboratory, cannibalism is maximal just around the time of the pupal ecdysis, the soft, young pupa or late pharate stage being most vulnerable. At the end of the field experiment, only 4 of 50 (8%) of the test animals could be accounted for. The rest had disappeared without trace.

It is possible that the pharate pupae were being taken by some mammalian predator, and small tracks were once seen on the guano. A sample of the presumptive cannibal larvae was therefore returned in guano to Tallahassee, where a similar experiment was repeated in plastic boxes (23×31 cm) about 10 days later. Twenty pharate pupae were placed on the guano surface of each of two boxes containing about 100 larvae each.

Table I. Recovery of Marked Larvae in the Final Search of the Guano Pile*

Larval group	No. released	Total recovered in search	Total recaptured away from guano	Total accounted for	% of released accounted for
2- to 3-month	202	59	0	59	31
4- to 5-month	119	27	16	43	26
5- to 6-month	77	11	26	37	59

*Percentages are corrected for mark-loss.

Table II. Cannibalism of Pharate Pupae by the Natural Population of Larvae at El Zamorano, Honduras*

Elapsed time (days)	Individuals found		Cumulative number of missing individuals (probably cannibalized)	% of control pupated
	Pharate pupae	Pupae		
0	50	0	0	0
1	48	0	2	0
2	48	0	2	0
3	43	3	4	25
4	14	6	30	50
5	1	3	46	85

*Fifty pharate pupae that had been isolated 7 days earlier were placed on the guano surface in the region of maximal larval density. The individuals remaining were counted daily. Proportion of control pupated was determined from a group of 20 pharate pupae not exposed to larvae.

Seven days later, only one pupa survived. A repetition of this experiment showed only 10 pupae surviving in each box after a day, and only 3 in each box after 4 days. Some of the pharate pupae were attacked almost immediately upon being placed on the guano, and five had disappeared in each box within 6 h. Most individuals moulted on the third day. All six survivors were pupae.

Thus, while it is difficult to give meaningful quantitative estimates of cannibalism by natural populations, it is obvious that larvae of *Zophobas atratus* are voracious cannibals of pharate pupae. It is consistent with the hypothesis that such cannibalism has selected against pupating under crowded conditions, and selected for movement to a secluded spot in order to pupate.

Discussion

The evidence is consistent with the hypothesis that prepupal larvae of *Zophobas atratus* disperse away from active larval populations because of the risk of cannibalism. The work of Tschinkel & van Belle (1976) and Tschinkel (1978) further suggests that larvae show an increasing tendency to overdisperse as their weight and/or age increase. Larval weight at dispersal determines adult weight, which in turn affects female fecundity (Tschinkel, unpublished). However, while an individual can increase fecundity by dispersing later at a larger body size, this simultaneously increases generation time (age at first reproduction). The two effects counteract one another and must be balanced by natural selection, but the balance point (i.e. dispersal age and weight) is probably highly dependent on the physical and biological environment, and varies from site to site. In this context, while most larvae seem to disperse at weights below 650 mg,

a number of much heavier individuals were found among larvae, pupae and teneral adults (Fig. 2). It is conceivable that in other contexts at other sites, natural selection would favour such larger individuals so that they would comprise a larger fraction of the population.

The importance of cannibalism in shaping the life history of an insect has been best characterized by the classic studies on the flour beetles, *Tribolium*, which are also tenebrionid beetles. Park (1934), Park et al. (1974a, b), Park et al. (1965), and Mertz & Cawthon (1973) have shown the overriding importance of egg and pupal cannibalism in the population dynamics of *Tribolium*. The intensity of cannibalism has been shown to vary with the age and sex of the victim as well as the age and sex of the cannibal. Ho & Dawson (1966) and Mertz & Robertson (1970) showed that cannibalism benefited larvae by resulting in higher fecundities as adults. In *Zophobas*, cannibalism of pharate pupae by larvae results in higher larval growth rates and higher fecundity (Tschinkel, unpublished).

Insofar as choice of pupation site affects chances of survival, *Zophobas* is similar to *Tribolium*. Park et al. (1968) have shown that location of pupae in the top or bottom half of the vial affected their chances of being cannibalized, and that *T. confusum* adults preyed more actively in the top while *T. castaneum* preyed more in the bottom half of the vial. While *Zophobas* larvae disperse to areas of low larval density for pupation, such behaviour has not been rigorously documented for *Tribolium*, though there are indications that dispersal may occur (King & Dawson 1973). For example, Ghent (1966) found pupae to be more abundant near the top of the vials and eggs near the bottom. Naylor (1965) found a sporadic, density-

dependent tendency for larvae to disperse from the densely populated flour. Likewise, King & Dawson (1973) reported that larvae in their complex experimental habitats leave regions of high larval density to pupate in warmer, less dense regions. Although the authors interpret this as movement into warmer regions to speed pupal development, the larvae may also be dispersing in direct response to density and in preparation for pupation. Ghent (1966) reports similar movement of what he refers to as 'prepupae' in *T. confusum*.

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REFERENCES

- Ghent, A. W. 1966. Studies of behavior of the *Tribolium* flour beetles. II. Distributions in depth of *T. castaneum* and *T. confusum* in fractionable shell vials. *Ecology*, **47**, 355-367.
- Ho, R. K. & Dawson, P. S. 1966. Egg cannibalism by *Tribolium* larvae. *Ecology*, **47**, 318-321.
- King, C. E. & Dawson, P. S. 1973. Habitat selection by flour beetles in complex environments. *Physiol. Zool.*, **46**, 297-309.
- Lees, A. D. 1961. Clonal polymorphism in aphids. *Insect Polymorphism*. London: Roy. Entom. Soc., Symposium, **1**, 68-79.
- Leonard, D. E. 1970. Intrinsic factors causing qualitative changes in populations of *Porthetria dispar* (Lepidoptera: Lymantriidae). *Can. Entomol.*, **102**, 239-249.
- Marle, G. 1951. Observations on the dispersal of the fruit tree red spider mite, *Metatetranychus ulmi* (Koch). *Ann. Rep. E. Malling Res. Sta.*, **150**, 155-159.
- Mertz, D. B. & Cawthon, D. A. 1973. Sex differences in the cannibalistic roles of adult flour beetles. *Ecology*, **54**, 1400-1402.
- Mertz, D. B. & Robertson, J. R. 1970. Some developmental consequences of handling, egg-eating and population density for flour beetle larvae. *Ecology*, **51**, 989-998.
- Naylor, A. F. 1965. Dispersal responses of female flour beetles, *Tribolium confusum*, to presence of larvae. *Ecology*, **46**, 341-343.
- Park, T. 1934. Observations on the general biology of the flour beetle, *Tribolium confusum*. *Q. Rev. Biol.*, **9**, 36-54.
- Park, T., Mertz, D. B., Grodzinski, W. & Prus, T. 1965. Cannibalistic predation in populations of flour beetles. *Physiol. Zool.*, **38**, 289-321.
- Park, T., Mertz, D. B. & Nathanson, M. 1968. The cannibalism of pupae by adult flour beetles. *Physiol. Zool.*, **41**, 228-253.
- Park, T., Mertz, D. B. & Holucek, H. 1974a. Factors affecting the susceptibility of flour beetle pupae to cannibalism by adults. *Physiol. Zool.*, **47**, 266-272.
- Park, T., Ziegler, J. R., Ziegler, D. L. & Mertz, D. B. 1974b. The cannibalism of eggs by *Tribolium* larvae. *Physiol. Zool.*, **47**, 37-58.
- Price, P. 1975. *Insect Ecology*. New York: Academic Press.
- Tschinkel, W. R. 1978. Dispersal behavior of the larval tenebrionid beetle, *Zophobas rugipes*. *Physiol. Zool.*, **51**, 300-313.
- Tschinkel, W. R. & van Belle, G. 1976. Dispersal of larvae of the tenebrionid beetle, *Zophobas rugipes*, in relation to weight and crowding. *Ecology*, **57**, 161-168.
- Tschinkel, W. R. & Willson, C. D. 1971. Inhibition of pupation due to crowding in some tenebrionid beetles. *J. exp. Zool.*, **176**, 137-146.
- Uvarov, B. 1977. *The Grasshoppers and Locusts*, Vol. 2. London: Centre for Overseas Pest Research.

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