

NOTES AND COMMENTS

SELECTION FOR CHANGES IN GENETIC FACILITATION: NEGATIVE RESULTS IN *TRIBOLIUM* AND *MUSCA*.¹

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1. INTRODUCTION

DIFFERENT species or strains competing for limited resources may diminish the utilisation of certain regions of their environmental hypervolume, resulting in increased fitness for one or more populations in the non-overlapping regions (*genetic facilitation*) as measured by survival, length of developmental period, body weight, and other characteristics. Lewontin (1955), Lewontin and Matsuo (1963), Sokal and Huber (1963), Sokal and Karten (1965), Sokal and Sullivan (1963), Sullivan and Sokal (1965), and Bhalla and Sokal (1964) provide evidence for *Drosophila*, *Tribolium* and *Musca*. Decreased overlap in environmental requirements may be achieved by preferential utilisation of fine differences in the environment by different genotypes, or indirectly by the life processes of one competitor beneficially affecting the other, *e.g.* through the enhancement of the substrate by desirable metabolites (Weisbrot, 1966; Bryant, 1969). The converse response to increased competition between species or strains may be reduced fitness of one or both competitors (*genetic disoperation*, Bhalla and Sokal, 1964).

Genetic facilitation or disoperation are probably controlled by natural selection and the reactions exhibited by two populations upon first encounter should change as a consequence. Such changes have been investigated in laboratory competition experiments between *Drosophila* species. Ayala (1966, 1969) was able to demonstrate changes in interactive capacities, but Futuyma (1970) did not find consistent trends.

Competition between strains of the same species is not of comparable evolutionary importance, since usually the adults at the end of the first generation would interbreed and distinct strains would disappear. But when the adults surviving competition are permitted to cross only with others of their own strain, such an experiment will serve as a model of interspecies competition. The initial overlap in environmental requirements between similar strains should be especially great and competition consequently intense. Thus natural selection acting on several generations of two competing strains should be more effective in producing increases in genetic facilitation or disoperation than corresponding interspecific models. Harper (1968) reported increases in genetic facilitation in seed mixtures and

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Seaton and Antonovics (1967) found marked increased genetic facilitation in a few generations of competition between a mutant and a wild type strain of *D. melanogaster*.

Their findings led directly to the present study intended to answer the following questions: (1) Can selection for changes in genetic facilitation between two strains of the same species be demonstrated for other organisms? (2) If such changes are found, can they be attributed to specific alterations in the ecology of one or both of the competing strains? This second question was of special interest, since recent studies in our laboratory have centred on the ecological components of natural selection and the mechanisms by which such interactions are brought about (Sokal and Sonleitner, 1968; Bryant, 1969).

2. DESIGN OF EXPERIMENTS

Parallel studies were carried out on *Tribolium castaneum* and *Musca domestica*, the design being kept as nearly identical as possible and similar to that of the Seaton and Antonovics experiment. Biological differences between the flour beetles and the houseflies and in laboratory handling procedures, led to differences in design shown in table 1. The general

TABLE 1
Design of experiments

	Tribolium	Houseflies
Strains	++, <i>bb</i>	OL, <i>bwb</i>
Quantity of medium	8 g.	18 g.
Input as number of eggs (replications in parentheses)		
low density	160 (4)	80 (10)
high density	800 (3)	320 (5)
Total number of generations of selection:		
low density	5	9
high density	6*	8
Assays parallel to generations	5	4, 8
Period of oviposition	3 days	6 hours
Survivors removed from culture	as pupae (daily)	as adults (every 8 hours)

* Only one replicate for each line in generation 6.

culture conditions of our previous work with these organisms prevailed and details of husbandry and origin of the strains can be consulted in Sokal and Karten (1965) for *Tribolium* and in Sullivan and Sokal (1963, 1965) and Bryant and Sokal (1967) for *Musca*.

Three kinds of cultures were set up, a wild-type strain, a mutant strain and 1 : 1 mixtures of the two. Eggs obtained from mass cultures were reared at a near optimal and a high density. "Pure" strains were handled identically to the mixed cultures, to allow for possible changes in fitness due to the handling procedure. Each type of culture was variously replicated depending on species and density (see table 1). Housefly results being inherently more variable than those from *Tribolium*, the former cultures had a greater number of replications. Survivors (pupae in *Tribolium*, virgin adults in *Musca*) were counted, separated by strain and sex, and kept in holding containers. Percent survival of egg input was computed. Egg hatchability tests were run for both species. After emergence was com-

pleted, adults from each type of culture were pooled over all replicates and permitted to mate *ad libitum* (in mixed cultures, of course, only with members of their own strain). Eggs from these adults were used to set up the next generation, following the same design. Adult dry weight (in *Tribolium* only) and duration of developmental period were also recorded.

Special assay tests were run during some generations (see table 1) to examine changes in the strains in mixed culture by comparison with the same strain in the parallel pure cultures. Thus in addition to the mixed cultures of that generation, there were three other mixtures at the two densities with the same number of replicates. These were 1 : 1 mixtures of wild type (pure) \times mutant (pure), wild type (mixed) \times mutant (pure), and wild type (pure) \times mutant (mixed). The adjective in parentheses refers to the experience of the strain in earlier generations. The same measures of fitness were recorded in the assay generations.

3. RESULTS

Tribolium

No genetic facilitation for survival was evident in the first-generation mixed cultures. Positive linear regression of percent survival (angular transformation) on generations was significant only at the low density for *bb* ($P < 0.05$) from both pure and mixed cultures. At the high density, the slopes of the regression lines of survival (in degrees) on time for ++ and *bb* in the mixed cultures are not significant, nor are their differences. However, the differences between the slopes for pure and mixed cultures within each strain are significant (++ : $P < 0.01$, *bb*: $P < 0.05$). In ++, this is caused by a significant decrease in survival in the pure cultures with time, rather than by improvement in the mixed cultures. In *bb*, the mixed lines show nonsignificant reduction of survival with time, while survival in pure lines increased slightly with time (fig. 1). There is therefore no indication of improvement among the mixed cultures.

In the assay test at generation 5 at high density, mean percentage of survival to pupation of ++ from mixed cultures (77.7 per cent.) was moderately, but significantly, higher than that of ++ from pure cultures (64.0 per cent.; $P = 0.05$ by the Mann-Whitney *U*-test). The pure culture *bb* used for testing the two types of ++, also survived significantly better ($P = 0.05$) when combined with ++ from mixed (66.1 per cent.) rather than pure cultures (53.0 per cent.). This resulted in higher total survival in vials containing ++ eggs from mixed cultures. Trends of high-density *bb* and at low density were not significant.

No important or significant changes were detected in mean body weight in the strains for the generations tested.

There was a slight trend towards faster development from generation 1 to 5 in the low-density cultures and in *bb* in the high-density pure cultures. At the high density, mixed cultures showed a similar though more erratic trend. These results are probably due to a bias in the handling procedure. In an egg farm the early enclosed adults contributed proportionately more of the individuals of the next generation. However, the high-density pure ++ were considerably delayed in their pupation (median pupation times: generation 1 = 26.8 days, generation 5 = 36.2 days).

The cumulative distributions of pupation times of the two ++ lines for

generation 5 plotted together with that of pure *bb* (fig. 2) simulate the assay combinations. While ++ (mixed) has an emergence curve similar to *bb* (pure), the ++ (pure) develops much slower and most individuals pupate after their competitors. Since the quality of the food is diminishing with time and waste materials accumulate, these slow-developing individuals

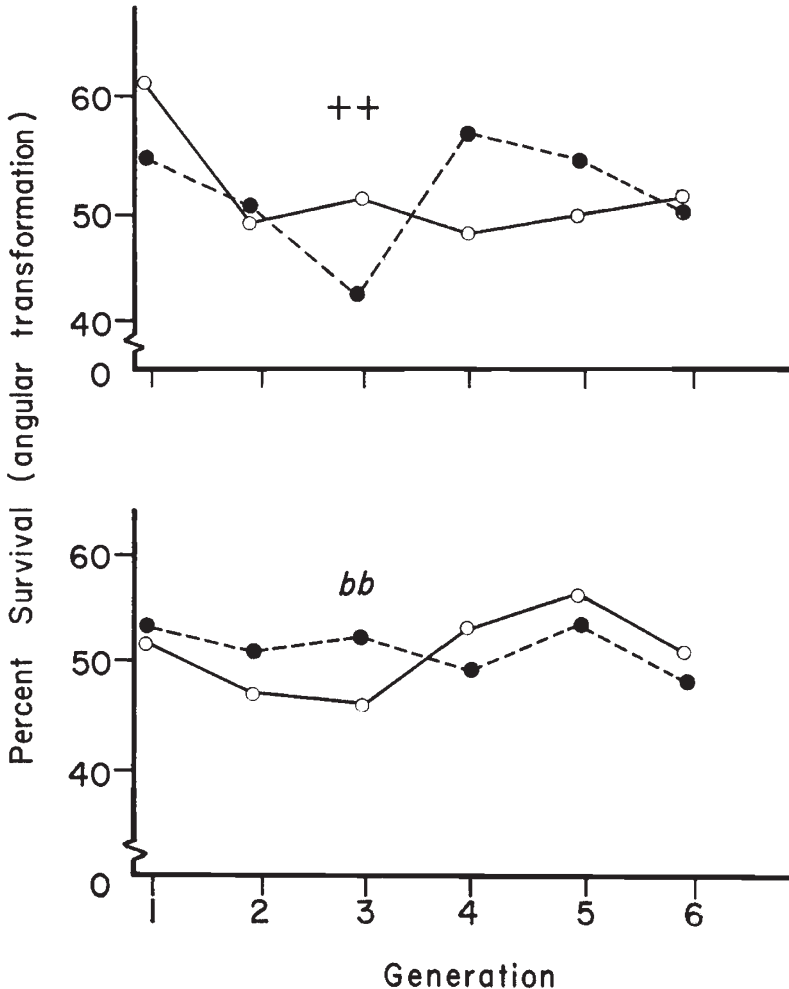


FIG. 1.—Percent survival in degrees ($\arcsin \sqrt{\%}$) for *Tribolium* in dense cultures for the 6 generations of the experiment. Survival in pure (hollow circles) and mixed cultures (solid circles) is contrasted for ++ in the upper graph and for *bb* in the lower graph.

have a much reduced chance of survival by generation 5 as shown above. Thus the apparent improvement due to selection in mixtures—the better survival of mixed-culture beetles—is an artificial result of the changes in developmental period in the ++ pure cultures.

Musca

Survival in the first generation of mixed culture showed no genetic facilitation. We could not detect any systematic trends in percentage

survival to adulthood during the experiment. Illustrative of the absence of trend is fig. 3 showing the survival of the strains in pure and mixed cultures at the low density. Neither strain showed any significant differences in survival between pure and mixed lines (all regressions and differences nonsignificant with $P > 0.05$). A slight hint of decreasing survival of OL was impossible to establish statistically due to erratic fluctuations in percentage survival in a few generations. The fluctuations did not result from coincident environmental changes as the covariance for survival between pure lines was not significant ($P > 0.05$). Furthermore these fluctuations

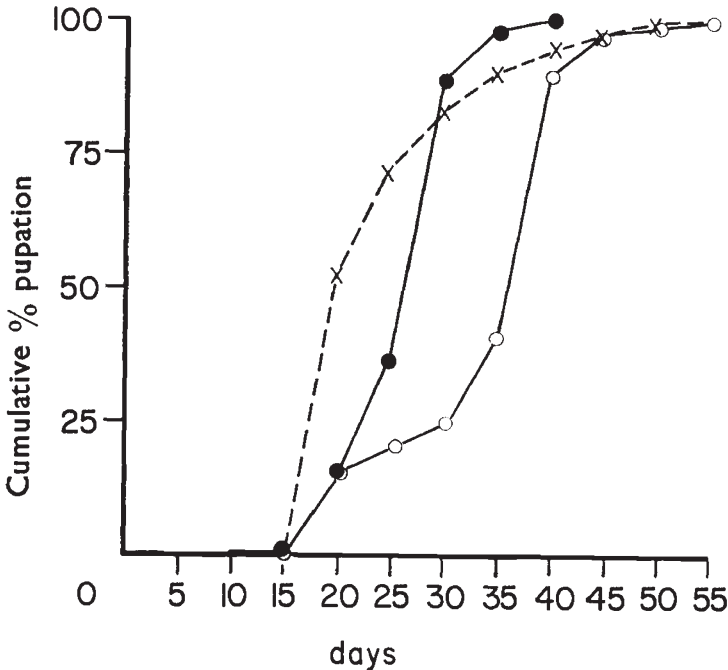


FIG. 2.—Cumulative distribution of pupation times at generation 5 of ++ *Tribolium* from the pure cultures (hollow circles) contrasted with ++ from mixed cultures (solid circles) and with *bb* from pure cultures (crosses) which was used as a reference strain against both ++ strains in the assay generation. For easier inspection, 5-day totals have been plotted. Ordinate: Cumulative percent pupation; Abscissa: days since beginning of culture.

were from two sources. In generations 6 and 7 of the pure OL cultures they were due to low egg hatching, while in generation 8 of the mixed cultures of OL, low larval survival was responsible.

The other measure of fitness, developmental period, also lacked consistent trends and differences in the study (body weight was not measured in *Musca*).

4. DISCUSSION

In the two species studied here no significant improvement in fitness arose during competition between pairs of strains. The housefly results were unequivocal. Changes in *Tribolium* can be explained by a deterioration of fitness in the pure ++ cultures, rather than by an improvement in the

mixed cultures. Why this deterioration should have occurred is of some interest in terms of the population biology of *Tribolium*, but is irrelevant to the questions raised in the present paper.

Tribolium and housefly strains were chosen which from previous experiments were known to lack initial genetic facilitation, to permit an increase in facilitation to develop. Thus competition in *Musca* was between OL and *bwb* rather than OL and *ge* to avoid the known genetic facilitation of the latter (Bhalla and Sokal, 1964; Bryant, 1969). *Tribolium* mixtures in which *bb* was rare were similarly eschewed (Sokal and Karten, 1965).

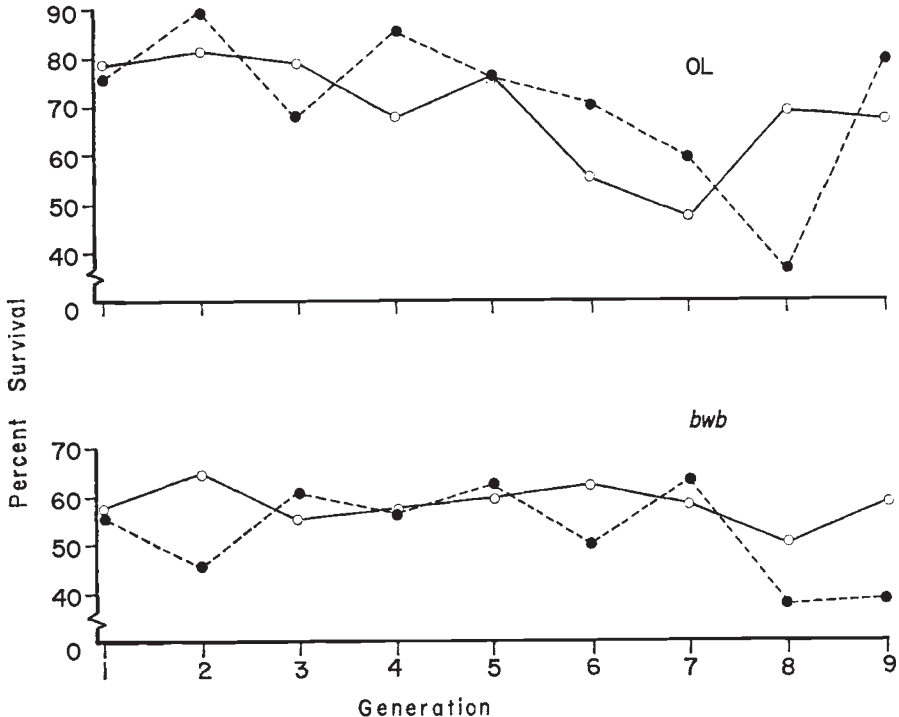


FIG. 3.—Percent survival for *Musca* in low density cultures for the 9 generations of the experiment. Survival in pure (hollow circles) and mixed cultures (solid circles) is contrasted for OL in the upper graph and for *bwb* in the lower graph.

These results differ from those of Seaton and Antonovics (1967), who demonstrated development of significant genetic facilitation between two strains of *D. melanogaster* in three generations. No obvious reasons for this discrepancy come to mind. The populations in the Seaton and Antonovics experiment derived from fewer parent pairs, hence selection should have been more successful in our studies but it was not. Our strains were not inbred, having repeatedly given rise to changes in fitness, especially duration of developmental period. In *Tribolium*, selection clearly took place even in this experiment, but unexpectedly in the pure cultures. It is somewhat difficult to compare the intensity of selection between our experiments and those of Seaton and Antonovics since the latter do not give numbers of emerged flies. However, assuming average fecundities in *Drosophila*, and given the volume of medium specified by these authors, it would appear

that selection was more intense in our experiments, certainly at the high densities. Although, as indicated earlier, some authors have found genetic facilitation between competing species, others have been unable to observe important and directed changes. Thus Futuyma (1970) was unable to find such changes between two species of *Drosophila* after 10 generations.

We hesitate to claim any generality for our findings as contrasted with those by Seaton and Antonovics, before further studies at the interspecies and interstrain levels have been made. Until general statements can be made about the outcome of intense competition between two genetically variable strains of the same species, caution should be exercised about evolutionary models involving rapid changes in interaction between two strains. If it is in fact true that competing strains show more overlap in their resource requirements than different species, even more caution should reign at the interspecies level.

5. SUMMARY

1. Two separate series of experiments, one with *Tribolium castaneum* and one with *Musca domestica* followed the general design of the Seaton and Antonovics experiments with two strains of *Drosophila melanogaster*.

2. Two strains of each species were competed at low and high densities, control cultures being maintained in parallel under similar handling procedures. Adult survivors of strains in mixed culture were permitted to mate with their own kind only.

3. No clear evidence of increase in genetic facilitation was observed for survival, weight (studied in *Tribolium* only) and duration of developmental period.

4. The implications of these findings lead us to caution in the development of models relying on selection for increased genetic facilitation in competing strains or species.

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