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THE ECOLOGY AND GENETICS OF FITNESS IN *CHLAMYDOMONAS*. IV. THE PROPERTIES OF MIXTURES OF GENOTYPES OF THE SAME SPECIES

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Abstract. — The properties of mixtures of genotypes of *Chlamydomonas reinhardtii* were investigated by growing them in monoculture and in all possible pairwise combinations in chemically defined axenic medium. Two sets of genotypes produced by crossing wild-type isolates were cultured in each of two physical environments. Mixtures were consistently more productive and less variable over environments than were their constituent monocultures. The average performance of a genotype in mixture was tightly correlated with its performance in monoculture. Reisolation of spores from mixtures at the end of growth showed that the mixtures became dominated by the component with the greater performance in monoculture, so that the properties of mixtures were attributable to replacement rather than to complementation. These results differ from those of similar trials using a range of different species of *Chlamydomonas*, where genetic interactions were found to be important. They are discussed in relation to theories of diversity and diversification, and related to the agronomic use of crop mixtures.

Key words.—Competition, complementation, evolution of sex, genetic interaction, selection, sib-competition.

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When genotypes of *Chlamydomonas* are grown as pure cultures in a range of nutrient media their relative fitness varies widely among environments (Bell, 1990a, 1990c). This observation suggests that mixed cultures may be more productive than pure cultures of their components, since the growth of each component may be limited by different environmental factors or combinations of factors. This paper describes an experiment in which pairwise mixtures of sibs are compared with the corresponding pure cultures, using crosses among unrelated wild-type isolates of Chlamydomonas reinhardtii. The results are relevant to several propositions in evolutionary ecology.

The first is that the production of arbitrary mixtures may consistently exceed the average production of their components in monoculture, as the consequence of complementary resource utilization. Such an effect would be important in applied fields such as agronomy, since it offers a low-input method of improving crop yields. The properties of mixtures of crop plants have often been studied (see reviews by Trenbath, 1974; G. Bell, unpubl.), and the work with *Chlamydomonas* can be viewed as a cheap and convenient model system for investigating the feasibility of such agronomic practices. It is possible that any general superiority of cultivar mixtures can be attributed largely to lower levels of disease (see reviews by Browning and Frey, 1969; Wolfe, 1985); any such effect would not occur in the experiment described here, where cultures are maintained axenically and no disease has been observed.

Second, mixtures may be more stable than monocultures, even if the mean performance of mixtures and monocultures does not differ. Greater stability implies a lower variance among environments, which might be the consequence of the substantial genotype-by-environment interaction known to occur among pure cultures of *Chlamydomonas* genotypes. This possibility is again interesting from an economic point of view, since it implies that the use of cultivar mixtures would reduce the frequency of disastrously low harvests (see Allard, 1961; Browning and Frey, 1969; G. Bell, unpubl.).

Third, the effect of mixtures on either the mean or the variance of production may be relevant to the evolution of mechanisms that regulate the quantity of variation, such as sexuality. In a spatially heterogeneous environment, it may be advantageous to produce genetically diverse offspring for two reasons. The first is that resource competition will be reduced by phenotypic differences, so that the overall production of a diverse brood may exceed that of a phenotypically uniform brood (the Tangled Bank theory of Bell, 1982). The second is that a diverse brood is more likely to include a genotype that performs well in a random environment (the Lottery models of Williams, 1975). In a temporally varying environment, a lower variance over environments implies greater geometric mean fitness and thus the long-term success of genetic mixtures (e.g., Treisman, 1976).

Fourth, genotypes may vary in the nature and magnitude of their interactions with other genotypes in mixture. This would imply that competing ability can be selected, and therefore that evolved mixtures may come to exceed monocultures, either in the mean or in the stability of production, even if arbitrary mixtures do not (Seaton and Antonovics, 1967; Donald and Hamblin, 1978). Artificial selection for performance in monoculture would then be an inefficient method of developing lines for use as mixture components, unless monoculture performance and competing ability are highly correlated. Furthermore, the genetic variance of fitness in natural populations could not be reliably estimated from measurements of pure cultures.

Finally, the conditions that production be maximized by resource complementation in mixtures and that mixtures be stable in composition over time are related, since both require positive interactions among genotypes. Mixtures will be generally more productive than monocultures if a genotype generally produces more when competing with alien genotypes than when competing with itself; mixtures rather than monocultures are likely to be evolutionarily stable if a genotype generally produces more when competing with an alien genotype than that alien genotype does when competing against itself. The properties of mixtures are therefore relevant to the wider question of the maintenance of genetic variance of fitness in self-sown natural populations.

EXPERIMENTAL DESIGN

Two trials were run. The first was a mixture diallel designed to test the null hypothesis that pairwise mixtures of sibs express the average performance of their components in monoculture, and is referred to as the Random Spore Diallel Mixture Experiment, or RSDME. The second was a spore monoculture trial designed to test the null hypothesis that there is no genetic variance in mixtures at the end of the period of culture, and is referred to as the Diallel Mixture Check Trial, or DMCT.

The material used to set up RSDME was a selection from that used in a Random Spore Pure Culture Experiment (RSPCE) designed to investigate the magnitude of genotype-by-environment interaction in pure cultures, reported in a previous paper in this series (Bell, 1990a). Briefly, four unrelated wild-type isolates of Chlamydomonas reinhardtii with minus mating type (mt⁻) were each crossed with a fifth plus mating-type (mt⁺) isolate. The recurrent mt⁺ parent and three of the mt⁻ parents were obtained from the Chlamydomonas Genetics Center at Duke University (strain designations CC-1010 mt⁺, CC-410 mt⁻, CC-1009 mt⁻, and CC-1418 mt⁻) and the remaining mt⁻ parent (strain designation S1D2) from Dr. P. Lefebvre of the University of Minnesota. CC-1009 and CC-1010 were originally isolated from the same zygote; all other combinations of isolates are unrelated. Few other wild-type isolates are known. Mating and single-spore isolation followed standard procedures (see Harris, 1989). From each cross, two mt⁻ and two mt⁺ spores were chosen at random; these spores are among those investigated in RSPCE. The eight spores of each mating type were used to set up two series of trials, the "minus" set and the "plus" set, in order to exclude any possibility of mating taking place within the mixtures. Within each set, all 28 pairwise combinations and all 8 monocultures were established; the mixture components are thus either full sibs (4 cases) or half-sibs (24 cases). Each set of 28 mixtures and 8 monocultures was grown in each of two culture environments, constructed from the basic recipe for Bold's medium (Nichols, 1973) by reducing the concentrations of either nitrogen (N) or phosphorus (P) to 25% of their standard values, forming the low-N and low-P environments, respectively. These culture environments represent chemically defined mixtures of simple inorganic salts, the only carbon source being supplied by the diffusion of atmospheric carbon diox-

FIG. 1. Overall behavior of pairwise mixtures and their components in monoculture in a diallel experiment. Plotted points are means over all combinations within a given category, each mean being itself the average of six replicate cultures. A "combination" is a given genotype or pair of genotypes, from either the plus or the minus mating-type set, in either the low-N or the low-P environment. The categories are monocultures (32 cases), full-sib mixtures (16 cases), or halfsib mixtures (96 cases). Bars extending up from the half-sib means and down from the monoculture means are one standard error.

ide. The experiment as a whole therefore consists of four separate trials that can be analyzed separately-minus/low-N, minus/ low-P, plus/low-N, and plus/low-P. Cultures were initiated in the following manner. Each genotype was grown up in standard Bold's medium for 10 days. The resulting log-phase cultures were diluted to the same optical density, and one loop then transferred to 40 ml of sterile culture medium in a culture tube of 1 cm internal diameter. Culture tubes representing mixtures each received one loop of each of the two components; monocultures were set up with two loops of their single component. The cultures remained axenic through the course of the experiment. Tubes were placed in racks and put on two shelves of a growth cabinet beneath two fluorescent tubes, which were rotated weekly. All the tubes representing the same culture environment were placed

on the same shelf, plus and minus sets being mingled together. There were 6 replicates of each combination of mixture and culture environment, so that each shelf contained 2 sets \times 36 combinations of strains \times 6 replicates = 432 cultures, laid out as a single randomized block, and the experiment as a whole comprised 864 cultures. The cultures were allowed to grow for 35 days, being vortexed twice a week to prevent the cells adhering to the glass at the top or bottom of the tube. They were then scored on each of six successive weeks by recording optical transmittance T at 665 nm and using 1,000-10T as a measure of production (for justification see Bell, 1990c). These 6×864 = 5,184 readings constitute the data analyzed below.

The final composition of the mixed cultures in RSDME is unknown because the two components cannot be distinguished. It is possible that each mixture develops into a monoculture as the result of selection during the course of the experiment. To investigate this possibility, two combinations (a mixture and its two component monocultures) were chosen from each of the four combinations of set and environment after the conclusion of RSDME. From each combination, a single replicate tube of the mixture and each monoculture was used to spread agar plates. From these plates, 10 colonies for each monoculture and 20 colonies from the mixture were used to inoculate culture tubes containing the appropriate culture environment. Each tube therefore contained a single genotype, and the variance among the spores isolated from the mixture replicate can be compared with that among the spores isolated from the monocultures. The 40 cultures from each combination were laid out as a single randomized block in a 40-tube rack, and the 8 racks comprising DMCT maintained under the same conditions and scored on the same schedule as RSDME.

RANDOM SPORE DIALLEL MIXTURE EXPERIMENT

Overall Mixture Performance

The overall growth of monocultures and mixtures is shown in Figure 1. There is a clear and highly significant tendency for





FIG. 2. The regression of pairwise mixtures on the mean performance of their components in monoculture. The four graphs refer to (a) minus mating-type set, low-N environment; (b) minus mating-type set, low-P environment; (c) plus mating-type set, low-N environment; (d) plus mating-type set, low-P environment. The 28 points on each graph are the 28 possible pairwise combinations of the 8 genotypes in each set; solid circles are the 4 full-sib mixtures and open circles are the 24 half-sib mixtures. Each point represents the mean over six replicates of the mixture and the difference between the means over six replicates each of the two components in monoculture. The solid line is the least-squares regression, the cross within a circle marking the bivariate mean; the broken line is the line of equality.

mixtures to be more productive than monocultures. The between-treatment effect has F(1,858) = 11.4, P = 0.0008, by repeatedmeasures analysis of variance. The excess of mixtures over monocultures remains the same through time, and amounts, very roughly, to about 10% of monoculture production. Half-sib mixtures were more productive than full-sib mixtures, as anticipated by theories of resource complementation, but this effect was initially small and decreased through time.

If mixtures are stable in composition and if there is no interaction between genotypes then the expected production of a mixture is the mean of the two component genotypes



FIG. 3. Variance of monocultures and mixtures. The height of bars on the histogram indicates the magnitude of variance components estimated from two-way analyses of variance. See text for discussion of variance attributable to combination (C), environment (E), their interaction (CE), and error (e). Histograms to the left of the scale bar refer to the minus mating-type set of genotypes, those to the right of the scale the plus mating-type set, with monocultures shown to the left and mixtures to the right in both cases. The six values refer to the six consecutive readings of optical density.

in monoculture. The excess production of mixtures can thus be evaluated by regressing mixtures on component means. Figure 2 shows these regressions for the four setenvironment combinations. In all four cases the mean production of all mixtures exceeds the mean of component means. In three of four cases, the regression lies above the line of equality over the range of the data. Altogether, 25/112 mixtures produced less than the mean of their components, while 87/112 produced more. There is therefore little doubt that the sib mixtures used in this experiment are more productive than expected. It follows that either the mixtures are not stable in composition, tending to become dominated by the more productive component, or that there are generally complementary interactions between genotypes.

Variance of Mixtures and Monocultures

Within either set, the variance of performance can be analyzed in terms of "combination" (the genotype of monocultures, or the pair of genotypes constituting a mixture) and, environment. The four variances estimable from a two-way analysis—the two main effects, their interaction, and the residual variance among replicates—can then be used to describe the effect of mixture on variation; different variances and ratios of variances represent different aspects of "stability." They are shown in Figure 3.

The variance attributable to combination falls through time, both in monocultures and in mixtures, as cultures approach their carrying capacities. It is always much less for mixtures than for monocultures, showing that pairwise mixtures vary less among themselves than do their constituent monocultures. If mixtures remained equal mechanical mixtures of monocultures, their variance should be one-half as great as that of monocultures. In fact, it is somewhat less, being on average about one-third that of monocultures, so that mixtures are more similar to one another than would be expected. This could arise from genetic interactions, since a negative correlation between monoculture performance and competing ability would reduce the variance of a set of equal mixtures. However, it is also consistent with a biased shift in mixture composition, since if mixtures were generally to come to be dominated either by the less or by the more productive genotype, they would express only a part of the variation seen among monocultures.

The macroenvironmental variance is large throughout the experiment in both mixtures and monocultures, although it may tend to fall somewhat through time. It is generally, but not always, slightly greater for monocultures than for mixtures. It is possible,

TABLE 1. Analysis of variance for mixture effects in RSDME. The design follows Federer et al. (1982); cf. Bell (1990b) for a similar analysis of interspecific mixtures. Convention for indicating probability that an effect differs from zero by chance alone is no asterisk, 1 > P > 0.1, * 0.1 > P > 0.01, ** 0.01 > P > 0.001, *** 0.001 > P > 0.01.

Source	Mean squares					
	df	Minus/low-N	Minus/low-P	Plus/low-N	Plus/low-P	
Genotype of monoculture	7	293,144***	197,030***	193,334***	90,439***	
Monoculture vs. mixture	1	20,264**	135,308***	182,436***	168,185***	
Genetic effects in mixtures	27	78,192***	90,322***	99,692***	45.589***	
General competing	7	220,964***	278,979***	344,025***	121,413***	
Specific competing	20	28,221***	24,292*	14,175	19.051*	
Error (replication)	180	12,305	14,777	11,364	12,223	
Total	215			,	,	

therefore, that the average of a set of mixtures varies less over environments than does the average of their constituent monocultures.

Perhaps the most important parameter of stability is the combination-by-environment interaction, since this represents the heritable part of environmental variance. The interaction variance rises through the experiment, roughly balancing the loss of variance among combinations. It is always less for mixtures than for monocultures, showing that individual mixtures vary less relative to one another than do monocultures when the environment changes. In this sense, mixtures are more stable than are monocultures. On the other hand, consistency is often expressed as the intraclass correlation coefficient $t = \sigma^2_{\rm comb}/(\sigma^2_{\rm comb} +$ $\sigma^2_{\text{comb}\times\text{env}}$), with values closer to +1 indicating consistency of performance over environments. This is considerably greater for monocultures than for mixtures, because the main effect of combination is decreasing faster through time among monocultures than among mixtures.

The variance among replicates expresses average sensitivity to microenvironmental differences. It declines slowly through time but does not differ consistently between mixtures and monocultures, being slightly greater among monocultures in one set but slightly greater among mixtures in the other.

Competing Ability

The performance of a genotype in mixture can be defined from the average performance of mixtures in which it is a component, and can be called general competing ability (GCA), by analogy with the concept of general combining ability in a diallel set of crosses. The specific competing ability (SCA) of a genotype in a particular mixture is then the difference between the actual performance of a mixture and the performance that would be predicted from the sum of the mean over all mixtures and the GCAs of its components. An analysis that partitions the variance among mixtures into components attributable to GCA and SCA is given in Table 1. This shows that by far the greater part of the variance among mixtures is attributable to GCA. In this material, therefore, specific genotype-by-genotype interactions are weak or absent.

Performance in Monoculture and in Mixture

The relationship between the performance of a genotype when grown alone and its performance when mixed with other genotypes depends on the magnitude of interactions among genotypes. If interactions among genotypes in mixture were sufficiently extensive, then the performance of a mixture cannot be predicted by summing the monoculture performances of its components, and GCA will be uncorrelated with monoculture performance. If interactions are weak, then GCA and monoculture performance will be highly correlated. Figure 4 shows that the regression of GCA on monoculture performance in RSDME was positive and highly significant, with a slope of 0.41 and $r^2 = 0.8$. There is little indication of genetic interaction, and the most reasonable interpretation of the result is that the



FIG. 4. The relationship between monoculture performance and competing ability. The general competing ability (GCA) of a genotype is calculated from the average performance of mixtures in which it is a component, as described by Falconer (1981 pp. 250-253). Monoculture performance is expressed as the deviation from the mean of the eight monocultures in each of the four combinations of sets and environments. The symbols represent the combinations of sets and environments: open symbols represent the minus matingtype set and solid symbols the plus mating-type set; circles represent the low-N environment and triangles the low-P environment. Slopes do not differ significantly (P > 0.05) among these four combinations. The regression equation relating competing ability GCA to monoculture performance MP is GCA = 0.41 MP, r^2 = 0.80, P < 0.0001.

genotype with superior performance in pure culture comes to predominate in mixture.

THE DIALLEL MIXTURE CHECK TRIAL

If the superior performance of mixtures arises from complementary interactions among genotypes, then it will be lost when genotypes are reisolated from the mixture and cultured separately. Figure 5 shows that the average of isolated genotypes exceeds the average of the two components in monoculture; the average excess of the mixtures was about the same whether the components were cultured together or separately. Therefore the superior performance of mixtures must be attributed to the increase in frequency of the component with



FIG. 5. Results of the Diallel Mixture Check Trial (DMCT). Open circles represent the performance of pairwise mixtures of genotypes grown in RSDME; closed circles represent average performance of the spores isolated from these mixtures at the end of growth and cultured separately in DMCT. These performances are regressed on the mean performance of the constituent monocultures. The analysis of covariance is

Source	df	MS	F	Р
Mixture vs. separated spores	ĺ	16.4	0.0	0.96
Mean of constituent monocultures	1	226,314.7	33.5	< 0.0001
Interaction Error	1 12	907.8 6,750.3	0.1	0.72

The interaction term refers to the way in which the performances of combinations of genotypes depend on whether these combinations are physically separated or physically mixed. The lack of interaction shows that mixture superiority does not depend on whether the constituents are grown together or apart. The overall regression yields the following estimates: intercept +93.2 (SE 71.6), slope +1.02 (SE 0.16), $r^2 = 0.75$, P < 0.0001.

superior monoculture performance during the growth of the mixture, and not to complementary interactions among genotypes.

The frequency of either component in the mixture can be estimated from the data. Let the mean performance of the N spores isolated from the mixture be m and their variance be σ^2 . The less productive monoculture has mean m_1 and variance σ_1^2 , while the more productive has mean m_2 and variance σ_2^2 . If the N spores isolated from the mixture comprise N_1 spores from the less productive and N_2 spores from the more productive monoculture, then $(N_1 + N_2)m = N_1m_1 + N_2m_2$ and $(N_1 + N_2)\sigma^2 = N_1\sigma_1^2 + N_1(m_1 - m)^2 + N_2\sigma_2^2 + N_2(m_2 - m)^2$, so that an estimate of N_1 is

$$N_1 = N[(\sigma^2 - \sigma_2^2) - (m_2 - m)^2] \\
\div [(\sigma_1^2 - \sigma_2^2) - (m_2 - m)^2 \\
+ (m_1 - m)^2]$$

From the eight mixtures tested, the mean of estimates of N_1 was 1.05 from the N = 20 spores tested in each case, with standard error 1.22. This is consistent with the conclusion that the component that is less productive in monoculture tends to be eliminated from mixtures.

DISCUSSION

Similar trials using mixtures of different species of Chlamydomonas were described in a previous paper in this series (Bell, 1990b). They showed that there was an average mixture excess in some environments but not in others, and that when a mixture excess was observed it could be magnified by increasing the number of components in the mixture. I concluded that there were strong interactions among genotypes, and that these could be either complementary or antagonistic. No reisolation of spores from mixtures was attempted; the experimental evidence for the existence of interactions was that the growth of a genotype was promoted or inhibited by filtrates from cultures of a second genotype in a manner consistent with their behavior when grown together as a mixture. Moreover, in contrast to the result obtained in RSDME, GCA was uncorrelated with monoculture performance.

The overall properties of mixtures thus seem to depend on the similarity of their components. When the components are dissimilar (different species) there can be strong positive or negative interactions between them, and the performance of the mixture cannot reliably be predicted from their separate performances in monoculture. On the other hand, when the components are similar (spores from crosses within the same species) there is little interaction, the more productive component in monoculture replaces the less productive, and monoculture performances are thus an accurate predictor of mixture yield. Complementarity (or antagonism) is manifested only among genotypes that are sufficiently dissimilar.

It is true that the behavior of very simple mixtures under highly controlled, axenic conditions of growth may differ from the behavior of much more complex populations growing in natural environments; model systems such as Chlamydomonas batch culture are useful only in detecting very general effects, but they are correspondingly powerful in negating the proposed general operation of an effect. It may also be argued that an arbitrary collection of genotypes developed from strains that have not evolved together may be expected to show replacement rather than complementarity, primarily because many inferior recombinants may be produced by crossing. However, mixtures of sibs from the CC- $1010 \times \text{CC-1009 cross}$, in which both parents descend from the same zygote, did not differ from mixtures of sibs whose parents were unrelated.

The source of mixture excess may be of little immediate interest in agronomic practice, where the crucial issue is whether mixtures have greater yield, rather than how this superiority arises. The components of a mixture of crop plants may be complementary, in the sense that one or both grow larger than they would in a monoculture; or the larger and more vigorous type may suppress the growth of the smaller and less vigorous type. It is, however, the overall yield of the mixture that is of primary economic interest. It may be argued that if replacement rather than complementarity is the source of mixture excess then a still greater yield could be obtained by sowing a monoculture of the more productive type. However, this is practicable only if the more productive type can be identified in advance. It will not be possible to do this with confidence if there is substantial genotypeby-environment interaction, such that the relative merit of a cultivar varies widely among localities or years. It may therefore be advantageous to sow mixed crops, even if average mixture excess arises solely from replacement rather than from complementarity. The Chlamydomonas results suggest that complementarity is more likely to be displayed by mixtures of crop species, and replacement by mixtures of cultivars or lines of the same species. However, these results were obtained in strictly axenic conditions, and new advantages for mixtures may arise in agronomic conditions where pathogens. herbivores, and competitors are always present. Furthermore, the source of mixture excess, while it may not be immediately important, is crucial in developing schemes to improve the yield of mixtures; it would be pointless to select for greater GCA or SCA in mixture components if mixture performance is determined by replacement. The properties of agronomic mixtures and the nature of interactions among crop species and cultivars have been reviewed by Trenbath (1974) and Bell (1985, 1990b).

By contrast, the source of mixture excess is of great interest in theoretical population biology. To explain the maintenance of diversity through environmental heterogeneity, we suppose either that selection proceeds in different directions in different places, or that there are complementary interactions among genotypes. The former view requires that the physical environment itself varies from place to place, and refers to stationary or sedentary organisms that inhabit more or less fixed locations (e.g., Levene, 1953); the latter view requires only that resource requirements vary among organisms, and therefore applies even to physically mixed environments and highly motile organisms (e.g., Clarke, 1972). The results presented here are compatible with the former view but not with the latter. Previous results from similar trials using a range of different species (Bell, 1990b) are compatible with both interpretations. In this simple axenic system, therefore, it seems that species diversity could be maintained even in physically uniform environments through interactions among genotypes, whereas genetic diversity within a species can be maintained only by disruptive selection in heterogeneous environments. Experiments designed to test these predictions are in progress.

There is a parallel between theories of diversity and theories of sexuality in spatially heterogeneous environments. Williams (1975) suggested that sexual diversification of progeny may be favored when

only a single genotype survives at a given site (see also Williams and Mitton, 1973; Taylor, 1979; Bulmer, 1980; Young, 1981; Barton and Post, 1986). Bell (1982) argued that complementary interactions among genotypes will favor sexual diversification even in uniform environments (see also Young, 1981; Price and Waser, 1982). The outcome of DMCT shows that the superiority of mixtures arises through replacement, falsifying Bell's hypothesis but not Williams'. If diversity is advantageous because selection favors a different genotype at each site, then two conditions must be satisfied: genotypeby-environment interaction must be very great, so that the relative fitness of genotypes is inconsistent over sites; and the interaction with environment should be less for mixtures than for monocultures, as the consequence of mixtures becoming dominated by a superior component. Both conditions are now known to hold in cultures of Chlamvdomonas. Further, if the diversity under selection is created sexually, then the peculiar characteristic of sexual broods is that they will include many different combinations of genes, and a third condition is therefore that the relative fitness of different combinations of genes should vary among environments. This can be expressed as the complex interaction of additive genotype with additive genotype with environment, which despite its complexity is readily detected and is known to contribute to the variance of fitness in Chlamydomonas (Bell, 1990c). Lottery models are therefore consistent with what is known about genotypeby-environment interaction and the properties of mixtures in *Chlamvdomonas*. They should be taken seriously as theories of diversification in organisms that are immobile, or that inhabit discrete habitats such as ponds or the insides of other organisms.

Several recent studies have investigated sib competition in flowering plants. Antonovics and Ellstrand (1984) compared the performance of majority and minority genotypes in manipulated swards of *Anthoxanthum* grown under quasinatural conditions. There was a tendency for minority types to have the better performance, but for statistical reasons the results were somewhat ambiguous. Schmitt and Antonovics (1986) found little difference between *An*-

thoxanthum seedlings surrounded by sibs and those surrounded by unrelated plants, except that those surrounded by sibs seemed to suffer more severely from a fortuitous aphid infestation. Kelley (1989b) found no evidence for sib competition in greenhouse experiments with Anthoxanthum; in field experiments, sexual plants were more successful than clonal plants, but the difference was nearly as great for plants growing alone as for groups of plants (Kellev, 1989a). It should be noted that in Anthoxanthum sexual propagules are seeds and the asexual propagules are tillers, so that in many cases the comparison of uniform and diverse progenv is confounded with a comparison between seed reproduction and vegetative reproduction. Schmitt and Ehrhardt (1987) and McCall et al. (1989) compared the performance of self-fertilized with that of crossfertilized seeds in Impatiens. Neither report describes consistent evidence for sib competition. Willson et al. (1987) compared groups of sibs with groups of unrelated individuals in four genera of herbs, and found that growth and survival were largely independent of group composition. Tonsor (1989) showed that groups of full sibs, half sibs, and nonrelatives grow to about the same size in Plantago lanceolata. In short, attempts to demonstrate complementary interactions by comparing groups of close relatives with groups of unrelated individuals have generally failed, and the Chlamvdomonas results are therefore consistent with those obtained from terrestrial plants. Resource complementation within species therefore seems unlikely to be a mechanism of great general interest. It remains possible that mixtures may suffer less damage from pathogens than do monocultures, but the only direct evidence on this point comes from agronomic trials. The only experiments designed to test Lottery models were performed using Anthoxanthum by Kelley (1989a, 1989b), and did not support the hypothesis that severe truncation selection commonly eliminates all genotypes except one. In summary, it seems reasonable to conclude that experimental attempts to show that the crucial advantage of sexuality is gained by diversifying progeny, as distinct from making progeny different from parents, have not yet been successful.

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