13-*cis*-retinoic acid had such lesions with accompanying epithelial atypia. Moreover, the total number of proliferative lesions, including cancers, was lower by one-half in the animals fed 13-*cis*-retinoic acid. Finally, feeding of the retinoid also diminished the severity of atypical cellular changes throughout the epithelium. Although some epithelial atypia was found in all animals treated with OH-BBN, there were significantly fewer animals with severe atypia in the group fed 13-*cis*-retinoic acid after the treatment with OH-BBN.

This study confirms our earlier findings that 13-*cis*-retinoic acid can inhibit both the incidence and the severity of neoplastic alterations induced in the bladders of rats with carcinogens. The animals were not started on the diet containing the 13-*cis*-retinoic acid until after the carcinogen exposures, which precludes the possibility that the inhibition was exerted during the initiation phase of carcinogenesis. As in our previous study, it is also unlikely that the inhibition was due to a generalized toxic effect, as no signs of retinoid toxicity (*6*) were observed.

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7. We thank Hoffmann-La Roche Inc., Nutley, N.J., for 13-*cis*-retinoic acid, L. Keefer and M. Litwack for assistance in obtaining OH-BBN, and M. O'Boyle and D. Little for help with the manuscript.

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Sex-Ratio Adjustment in the Common Grackle

Abstract. *From the nestling period through maturity, female grackles are distinctly smaller than males and presumably cost less to rear. Individual birds nesting early in the season lay more female eggs than those nesting later, and in large broods, mortality after hatching consistently favors female fledglings. The first result suggests an adaptive nonrandom meiosis that anticipates seasonal conditions of food availability; the second implies a brood reduction strategy consistent with Fisher's prediction that differential mortality in sexually dimorphic species should favor the less expensive sex.*

Fisher (*1*) hypothesized that natural selection will favor equal parental expenditure on each sex until the end of parental care. If individual males cost as much to rear as individual females, selection should result in population sex ratios of unity at the end of parental care. If one sex costs less to raise than the other, a numerical excess of that sex would be expected at independence. Unless extreme inbreeding occurs (*2*), the hypothesis should apply to all diploid organisms in which reproductive investment in one offspring diminishes parental ability to invest in another.

Despite its generality, Fisher's hypothesis has not been adequately tested with birds. This is surprising because females contribute the sex-determining chromosome in this group (*3*). Nonrandom deviations in primary sex ratio must result from maternal control and cannot reflect conflicting maternal and paternal influences. In principle it is possible to compare initial with subsequent sex ratios produced by heterogametic females, thereby separating the mechanisms of segregation distortion from differential mortality. Dual maternal and paternal influences obscure such comparisons in mammals and other organisms in which males contribute the sex-determining chromosome (*4*). I now report that adjustment of both primary and subsequent sex ratios occurs in a species of bird and interpret these results in an evolutionary context.

Sex-ratio control consistent with Fisher's hypothesis may come about through parental manipulation of either primary or later sex ratios. The least interesting case concerns organisms in which it costs as much to raise one sex as the other. In such species, the primary population sex ratio should be unity and should be maintained until the end of parental care. More interesting are organisms among which one sex costs more for the parents to raise than the other, as might occur in species with strong sexual size dimorphism. The simplest situation is a hypothetical population for which future conditions of parental care are the same for all adults about to breed: the population sex ratio should favor the less expensive sex from conception through independence. A more likely circumstance is that different breeding adults face different conditions of parental care, so that some are more capable of rearing the "expensive" sex than others. Proximate factors might be differences in physical condition, weather, or assistance from mates. If individuals can predict the conditions under which they will later care for young, primary sex ratios should anticipate their future ability to invest. Such anticipation may occur when females know their physical condition relative to others in the population (*5*) or, more likely, when individuals breeding early in a highly seasonal environment consistently face circumstances different from those breeding later. If parents cannot predict future conditions when they commence breeding, the sex ratio after conception may be adjusted by selectively resorbing embryos or by killing the young in a manner adaptive for the parents (*1*, *5*). Such a method might be expected in a bird in which an adaptive pattern of brood reduction permits parents to cope with seasonal conditions that frequently deteriorate during breeding attempts (*6*, *7*).

Table 1. Embryonic sex ratios among common grackles from two seasons at Dexter, Michigan.

Clutch size	Nests (No.)	Males (No.)	Females (No.)
3	4	4	8
4	25	51	43
5	63	151	143
6	4	5	5
Total	96	211	199

Common grackles (*Quiscalus quiscula*, Icteridae) are likely candidates for studies of sex-ratio adjustment. Female fledglings weigh 82 percent as much as males of a similar age, a dimorphism maintained through maturity (*8*). Thus, one expects a sex ratio at the end of parental care that favors female young requiring less food than males. Preliminary evidence indicates that fledgling males of female size suffer disproportionate mortality after leaving the nest (*7*). Parents should be selected to produce large healthy males under particularly favorable food conditions but to allocate resources to smaller females when food is scarce. Finally, the species breeds early in the spring in a highly seasonal environment. Individuals nesting earlier than others consistently face severe conditions of freezing temperatures, snow, and cold rains when nestlings require invertebrate food collected by parents. Under such circumstances of predictable food shortage, early breeders should produce female young less costly to rear than males. Birds nesting later in the season sometimes face food shortage due to climatic changes, but at other times encounter early flushes of insects and other invertebrates used as food for young. A late female probably cannot predict which extreme her nestlings are most likely to encounter at the time she lays eggs (2 weeks before hatching); under such conditions, a brood reduction strategy is likely to be adaptive (*6, 7*).

Sex ratios were determined by inspecting the gonads of embryos, newly hatched young, and nestlings about to fledge (12 to 13 days after hatching) at two sites in southern Michigan. At Dexter, 50 clutches (220 birds) in 1976 and 46 clutches (200 birds) in 1977 were collected for dissection 12 days after the onset of incubation (*9*). In addition, sex ratios at fledging were determined by surgery and gonadal inspection [23 nests (81 birds) in 1975 at Dexter and 22 nests (55 birds) in 1976 at Ann Arbor] (*10*).

The proportion of females declines with laying date for clutches of five eggs for which the laying date of the first egg of the clutch and the sex of each egg are known (Fig. 1) (1976: 24 clutches, $r = -.45$, $P < .03$; 1977: 18 clutches, $r = -.43$, $P = .07$); pooled data: $r = -.44$, $P < .005$) (*11*). Trends are similar in samples including nests in which some eggs failed to develop (*12*). No trends are evident in smaller clutches. In both years, these seasonal trends balanced to produce overall sex ratios slightly in favor of males, but not significantly so. Embryo sex-ratio data are summarized in Table 1.

Sex ratios near the end of parental care (at fledging) are shown in Table 2 (*13*). Differential mortality consistently favors females in the most common clutches of five and in the overall sex ratio. Pooled data show that sex ratios deviate from unity unequivocally in the direction predicted by Fisher's hypothesis. As fledgling ratios also differ from embryo ratios ($\chi^2 = 6.81$, d.f. = 1, $P < .01$), this deviation may be attributed to differential mortality rather than to unbalanced primary sex ratios.

Table 2. Fledgling sex ratios among common grackles.

Clutch size	Nests (No.)	Males (No.)	Females (No.)	P*
		Dexter, 1975		
3	3	2	7	
4	2	1	3	
5†	16	24	37	.062
6	2	3	4	
Total†	23	30	51	.013
		Ann Arbor, 1976		
3	2	2	4	
4	9	11	12	
5	10	8	17	.055
?	1	0	1	
Total	22	21	34	.053
		Pooled data		
3	5	4	11	
4	11	12	15	
5	26	32	54	.012
6	3	3	4	
?	1	0	1	
Total	45	51	85	.005

*One-tailed binomial test. †Corrected from Howe (*7*).

The results of this study lead to several intriguing evolutionary interpretations.

1) A seasonal pattern of primary sex ratio change suggests adaptive control. The mechanism is probably nonrandom segregation of the sex chromosomes, a cytological phenomenon well documented in some insects (*2, 14*) but hitherto undetected in vertebrates. With regard to common grackles, the most plausible evolutionary explanation for the seasonal pattern is that females nesting at different times during the spring face different conditions of food abundance. Early breeders favor smaller females when the chances of raising larger males are remote.

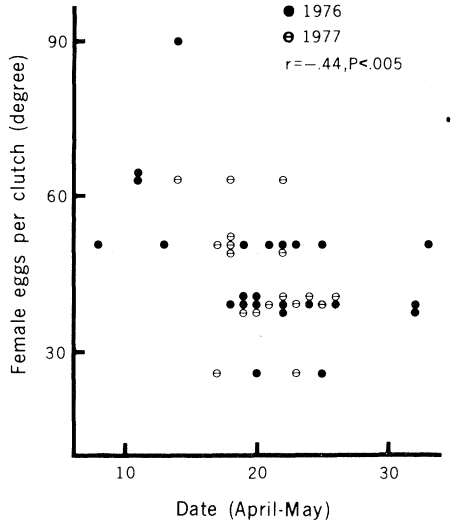

Fig. 1. Proportion of females per complete clutch of five plotted against the date the first egg was laid. The proportion of females per clutch significantly decreases as the season progresses from early April into May. Proportions are expressed in degree units of the angular transformation $\Theta = \arcsin \sqrt{p}$, where p is a proportion from 0 to 1. $\Theta_{1.0} = 90.00$, $\Theta_{.8} = 63.43$, $\Theta_{.6} = 50.77$, $\Theta_{.4} = 39.23$, $\Theta_{.2} = 26.57$ (*11*).

2) The overall primary sex ratio of unity suggests a statistical mean produced by conflicting selective pressures on individuals rather than a population "optimum." Where such variation exists, the mean must be considered an effect of selection and not an evolved adaptation itself (*15*).

3) Differential mortality of male young occurs as part of a conditional brood reduction strategy (*7*). The proximate reason for male-versus-female starvation is that males cannot sustain high growth rates under the stress of food shortage. From an evolutionary perspective, the key point is that this shortage is largely under parental control. A considerable body of theory suggests that parents should select which of their offspring receive resources (*16*); evidence from the common grackle indicates that parents do allocate investment differentially to their young (*17*). The result of differential mortality is a population sex ratio favoring the less expensive sex, as predicted from Fisher's hypothesis (*18*).

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8. At fledging, females average 60.0 ± 1.0 g and males 73.5 ± 1.7 g ($N = 51$ and 30, respectively). The same relative dimorphism occurs in adults. Of the first 20 of each sex netted in Ann Arbor in 1976, females weighed 102 ± 10.5 g and males 124.7 ± 6.2 g.
9. The 1976 set includes two nests of five and one of four for which clutch size, but not laying date, are known. The 1977 set includes one nest of five and two of four for which laying dates are unknown. Nestlings from these nests were dissected. No nests of unknown initial clutch sizes are included.
10. Laparotomies were performed on etherized nestlings by means of an 8-mm incision anterior to the left femur. Gonadal inspection is the essential method of sexing nestlings where malnutrition renders size comparisons unreliable [R. K. Selander, *Condor* **62**, 34 (1960)]. Small popu-

lation sizes precluded sampling both embryo and fledgling sex ratios at the same place during a season.

11. The angular transformation is required for parametric analysis of proportional data [R. R. Sokal and F. J. Rohlf, *Biometry* (Freeman, San Francisco, 1969), p. 386; F. J. Rohlf and R. R. Sokal, *Statistical Tables* (Freeman, San Francisco, 1969), p. 129]. Correlations are also significant for 1976 and for combined analyses without the extreme clutch of five females in 1976 ($r = -.42$, $P < .05$ and $r = -.40$, $P < .01$, respectively).
12. For samples including clutches in which some eggs failed to develop, negative correlations are significant for 1976 ($r = -.34$, $P = .05$, $N = 33$), for 1977 ($r = -.41$, $P < .03$, $N = 29$), and for pooled data ($r = -.33$, $P < .01$). It is not known whether embryonic mortality favors either sex, as data from domestic fowl are inconsistent [W. Landauer, *The Hatchability of Chicken Eggs as Influenced by Environment and Heredity* (Univ. of Connecticut Agricultural Experimental Station, Storrs, 1961), p. 170].
13. Parental care decreases after the young fledge, but it is not known how long or variable this period is in this or comparable species. Trends set during maximal parental effort may be accentuated after fledging but are unlikely to be reversed.
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18. Trivers and Willard (*5*) hypothesized correlations between female condition and investment in offspring and between this investment and the future reproductive success of potentially polygynous male offspring. This grackle is sometimes polygynous, but early breeders (likely to be the healthiest in a bird population) are most likely to produce female young. In this species, a correlation between differential investment in the sexes and future reproductive success of offspring is likely to be an artifact of parental need to ensure that males attain a fast enough rate of growth and a high enough fledging weight to make survival to reproductive age possible. This is a separate hypothesis. It does not require that female condition and parental investment covary with the reproductive success of that small subset of males that does survive to reproductive age. Nor does it confuse the origin of sexual dimorphism—which may well have evolved in response to sexual selection (*1*)—with selection on sex ratio. Reproductive consequences of parental condition have been discussed by D. Lack [*Population Studies of Birds* (Oxford Univ. Press, London, 1966)], C. M. Perrins [*Ibis* **112**, 242 (1970)], and R. E. Ricklefs [in *Avian Energetics*, R. Paynter, Jr., Ed. (Publ. No. 15, Nuttall Ornithological Club, Cambridge, Mass., 1974), pp. 152-297]. Polygyny in this species has been reported by R. H. Wiley [*Z. Tierpsychol.* **40**, 59 (1976)], and Howe (*7, 17*).
19. I thank Dr. and Mrs. W. Burns of Dexter, Michigan, for the use of their property and the staff of the University of Michigan Botanical Gardens for their cooperation. This research was accomplished in partial completion of the Ph.D. requirements at the University of Michigan. I am grateful for useful criticisms of the manuscript from R. D. Alexander, D. De Steven, K. Fiala, D. H. Janzen, L. Masters, J. Maynard Smith, R. B. Payne, and D. W. Tinkle. This work was supported by a Walker Scholarship of the Museum of Zoology and the Graduate School, University of Michigan.

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A Mutant of *Paramecium* Defective in Chemotaxis

Abstract. *In an effort to study the sensory-motor pathway of chemotaxis in* Paramecium tetraurelia, *I have generated mutants defective in their responses to chemicals. One mutant in particular, d4-530, is repelled by sodium acetate, which attracts normal paramecia by klinokinesis. The mutant is repelled by the mechanism of orthokinesis. To my knowledge, this is the first report of orthokinesis in chemotaxis of paramecia.*

Swimming and avoiding reaction are the two main components of behavior in *Paramecium*. Jennings (*1*) first described the avoiding reaction as a transient backing away from a stimulus, turning, and renewed forward swimming in a randomly chosen direction. This corresponds to normal ciliary beating, transient reversal of the ciliary beat, and a return to normal beating, respectively. We now understand a great deal about the membrane potential control of this behavior in the wild type (*2*) and in behavioral mutants defective in membrane electrogenesis (*3*).

Paramecia combine these two behaviors, swimming and avoiding reaction, in the more complex behavior of chemotaxis, or more accurately, chemokinesis (*4*). In chemokinesis, paramecia accumulate near or escape from the vicinity of certain chemicals. They accomplish this by modulating either the frequency of avoiding reaction (klinokinesis) or the velocity of forward swimming (orthokinesis) (*4*): an increase in frequency of avoiding reaction or increased velocity in a solution will cause repulsion from that solution and a decreased frequency of avoiding reaction or decreased velocity will cause attraction.

A genetic approach was used to dissect the chemosensory pathway. I describe here d4-530, a mutant of *Paramecium tetraurelia* that is repelled by sodium acetate, which attracts normal paramecia (*5, 6*). Previously, the mechanism of chemokinesis in *Paramecium* was believed to be solely klinokinesis (*1, 7*). However, d4-530 is repelled from sodium acetate by orthokinesis, and, to my knowledge, this is the first report of orthokinesis in *Paramecium* chemokinesis.

The response of paramecia to chemicals was measured by a T-maze assay, designed to present a test and control solution to a population of animals (*5*). The number of animals swimming into the arm with the test solution divided by the number of animals swimming into both test and control solution arms of the T gives an index of chemokinesis (I_{che}). An $I_{\text{che}} > 0.5$ indicates attraction to the test solution relative to the control; $I_{\text{che}} < 0.5$ indicates repulsion from the test solution into the control solution. Mutant d4-530 is defective in chemokinesis, and by this assay, it was similar to the wild type in response to all chem-

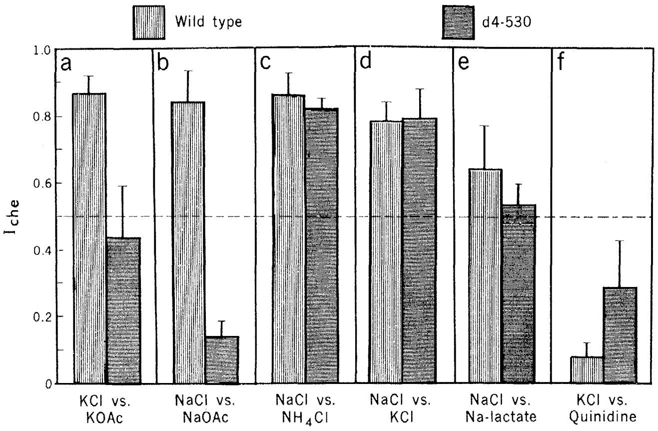

Fig. 1. Chemokinesis assayed by means of a T-maze (*5*). The index of chemokinesis (I_{che}) is defined as the number of animals in the test arm divided by the number of animals in both the test and control arms of the T. An $I_{\text{che}} > 0.5$ denotes attraction; $I_{\text{che}} < 0.5$ denotes repulsion from the test solution. All solutions included salts indicated in the chemokinesis buffer described in Table 1. The first solution under the histogram fills the control arm and all of the maze except the test arm; the second solution fills the test arm. (a) to (e) Concentrations are 5 mM of salt indicated; (f) concentrations are 0.1 mM quinidine hydrochloride or KCl. *KOAc*, potassium acetate; *NaOAc*, sodium acetate.