Equal mating success among male reproductive strategies in a marine isopod

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THREE genetically discrete male morphs coexist in *Paracerceis sculpta*, a Gulf of California marine isopod[1–5]. The large α males defend harems within intertidal sponges, the smaller β males mimic female behaviour and morphology, and the tiny γ males invade and sequester themselves within large harems. If selection is responsible for maintaining this polymorphism, then the mean fitness of each male morph must be equal over time[6–9]. Here we report that average reproductive success is equivalent among the three male morphs in monthly population samples collected over two years. We have investigated the total opportunity for sexual selection within and among morphs, and find that <0.10% of the total opportunity for sexual selection occurs among morphs. Furthermore, alleles responsible for the expression of this polymorphism conform to the Hardy-Weinberg equilibrium, indicating the absence of differential natural selection among morphs. Conditions necessary for stable coexistence of three alternative male reproductive strategies seem to exist in nature.

When isolated with females, the three male morphs (Fig. 1) do not differ in their ability to sire young successfully[1,2]. Male fertilization success does depend, however, on the relative density of males and females within spongocoels. Experiments using genetic markers show that α males sire the majority of offspring when defending spongocoels containing a single female and one β, or one γ male[2] (S.M.S. and C. Sassaman, unpublished data). When more than one female is present, β males sire ~60% of all offspring within a spongocoel regardless of female density, although there is considerable variation about this average. The reproductive success of γ males increases linearly with harem size. Paternity rules derived from these data for other combinations of males and females within spongocoels are described in Table 1.

TABLE 1 Rules for assigning male mating success

Case	Rule
1	α male alone; sires all
2	β male alone; sires all
3	γ male alone; sires all
4	2 α males; each sires half
5	2 γ males; each sires half
6	3 α males; each sires one third
7	1 α +1β male; β sires 0.60, α sires 0.40*†
8	1α +1β +4γ males; β sires 0.60, α sires none, the remaining progeny are divided among γ males
9	1α +2β +3γ males; 0.60 of the progeny are divided between the β males and 0.40 among the γ males, the α male sires none
10	1α +1γ male; γ sires 0.08, α male sires 0.92*†
11	1α +2γ males; each γ follows rule 10‡
12	1α +3γ males; each γ sires 0.08, α male sires the rest
13	2α +1γ; γ follows rule 10‡, the remaining offspring are divided between the α males
14	2α +3γ males; offspring are evenly divided among the γ males

* Laboratory tests[2].
† Field data[3].
‡ Linear regression based on refs 2 and 3: $y = -0.047 + 0.113x$.

Using these rules, and the observed joint distribution of 825 females and 555 males collected in monthly samples from a natural population over two years (Table 2), we calculated the mean and variance of male mating success and the opportunity

FIG. 1 The three male morphs in *Paracerceis sculpta*. Left to right, γ male, β male, α male.

TABLE 2 Distribution of α, β and γ males and females observed in spongocoels

	Mating success rule													
	1	2	3	4	5	6	7	8	9	10	11	12	13	14
								$1\alpha+$			$1\alpha+$		$2\alpha+$	
Harem size	1α	1β	1γ	2α	2γ	3α	1β	$1\beta+4\gamma$	$2\beta+3\gamma$	1γ	2γ	3γ	1γ	3γ
0	121	4	5	1	1	0	2	0	0	5	1	0	0	0
1	101	0	5	2	1	0	4	0	0	8	1	1	0	0
2	55	0	2	2	0	1	4	0	0	7	1	0	0	0
3	33	0	1	1	1	0	2	0	0	3	1	0	1	0
4	22	0	0	2	0	0	0	0	0	6	1	0	1	0
5	17	0	0	0	0	0	1	0	0	2	0	0	0	0
6	6	0	0	1	0	0	0	0	0	2	0	0	0	0
7	0	0	0	2	0	0	0	0	0	0	0	0	0	0
8	2	0	0	0	0	0	0	0	0	0	0	0	0	0
9	1	0	0	1	0	0	0	0	0	0	0	0	0	0
10	1	0	0	0	0	0	0	0	0	0	0	0	0	0
11	1	0	0	0	0	0	0	0	0	1	0	0	0	0
12	0	0	0	0	0	0	0	0	0	0	0	0	0	1
13	0	0	0	0	0	0	0	0	0	1	0	0	0	0
14	0	0	0	0	0	0	0	0	0	0	0	0	0	0
15	0	0	0	0	0	0	0	1	0	0	0	0	0	0
$\sum$	360	4	13	12	3	1	13	1	1	35	5	1	2	1

Sponges containing isopod breeding aggregations were collected monthly between October 1983 and November 1985 from permanent tide pools in the midintertidal zone, 1.5 km SW of Puerto Peñasco, Sonora, Mexico. Within 15 randomly selected 0.25-m plots along a 100-m transect, all sponges were removed and spongocoels were examined for isopods which were then separated by sex and male type.

for sexual selection[10-12]. The opportunity for sexual selection (I) measures the sex difference in the variance of relative reproductive success[10-12]. In many species, the variance in male reproductive success is greater than that in females because of variations among males in the numbers of mates. This sex difference in the variance in fitness permits total selection on males to be stronger than on females, and allows the evolution of sexual dimorphism.

We partitioned the observed variation in relative male mating success into two components, that within and that among males. The within-male component measures the strength of sexual selection acting on males within the same strategy. The among-male component measures the strengh of sexual selection acting among the three different male reproductive strategies. When the different male reproductive strategies have equal average mating success, there is no opportunity for sexual selection among the male types.

We found that average mating success is statistically equivalent among the three male morphs, and that <0.10% of the total opportunity for sexual selection is acting among the male strategies (Table 3). Even this small fraction is probably an overestimate because the equal apportionment of paternity within spongocoels, for example by rules 4, 5 and 6 in Table 1, reduces the within-male variance in reproductive success.

Genetic experiments indicate that male morphology in this species is principally influenced by a single, autosomal locus (S.M.S. and C. Sassaman, unpublished data). The 'β allele' at this locus seems to be dominant to both the 'α allele' and the 'γ allele', and the 'γ allele' seems to be dominant to the 'α allele'. Morphological differences among males result from the influence these alleles exert on male growth and maturation rates. Gamma males mature most rapidly ($\bar{x} \pm$ s.e. $= 57.62 \pm 3.88$ days; $N = 21$); β males mature at an intermediate rate ($\bar{x} \pm$ s.e. $= 62.33 \pm 6.48$ days; $N = 9$); α males mature most slowly ($\bar{x} \pm$ s.e. $= 83.61 \pm 3.47$ days; $N = 18$). Differences in maturation rate (1/days to maturity) among the morphs could influence their relative contributions to the population. However, reproductive tenure (age at death minus age at maturity) varies inversely with maturation rate (Fig. 2). Life history differences that influence the relative contributions of α, β and γ males to the population seem, therefore, to cancel.

As a test of this hypothesis we calculated the expected population frequencies for α, β and γ males, assuming the three alleles at the male locus are in Hardy-Weinberg equilibrium. Morph

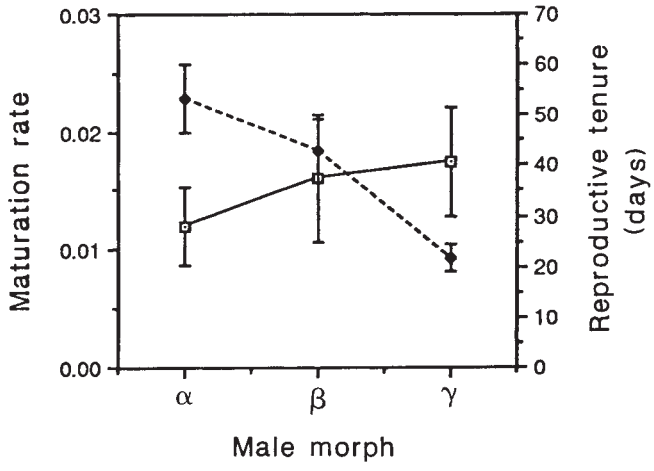

FIG. 2 The relationship between maturation rate (solid line; 1/(days from birth to maturity); $y = 1.87 - 0.19x$, $R^2 = 0.38$, $P < 0.001$) and reproductive tenure (dashed line; age at death minus age at maturity, in days; $y = 0.01 + 0.02x$, $R^2 = 0.36$, $P < 0.001$) for α ($N = 18$), β ($N = 9$) and γ males ($N = 21$). Bars represent one standard error.

TABLE 3 Mean and variance of male mating success and the opportunity for sexual selection

Male type	Mean (± s.e.) of number of mates	N
α males	1.51 ± 0.08	452
β males	1.35 ± 0.44	20
γ males	1.37 ± 0.23	83
Grand mean	1.49 mates per male	

The opportunity for sexual selection, I

Variance among-types	0.003
Variance within-types	3.075

I_{total} = (Total variance)/(Grand mean)^2 = 1.39

I_{among} = (Variance among-types)/(Grand mean)^2 = 0.001

frequencies (Table 1) are typical for this population[1,2,4,5] (α males, 0.814; β males, 0.036; γ males, 0.150; $N = 555$). As α males are recessive homozygotes, the frequency of the α allele is $(0.814)^{1/2} = 0.902$. The rarity of β males suggests that nearly all β males in the population are heterozygotes. Thus, to a first approximation, the frequency of the β allele is $1/2(0.036) = 0.018$. Because the sum of the three allele frequencies must equal unity, the frequency of the γ allele is 0.080. Therefore, the expected population frequencies for α males (α^2), β males ($2\alpha\beta + 2\beta\gamma$) and γ males ($\gamma^2 + 2\alpha\gamma$) are 0.814, 0.035 and 0.151. The observed frequency of the γ-male morph (0.150) conforms to the theoretical expectation ($\gamma^2 + 2\alpha\gamma = 0.151$).

Most polymorphisms in male mating behaviour seem to be 'condition dependent', in that males adjust their mating tactics to fit local environmental circumstances[6,7,13-27]. The presumed ancestral condition for male alternative strategies[9], however, and the model for the evolution of polymorphism in general, applies to male mating strategies that are genetically distinct[3,8,28-30]. Male polymorphism in *P. sculpta* provides such a model.

We have shown that average mating success is statistically equivalent among the three male morphs and that a tiny fraction of the total opportunity for sexual selection acts among the male strategies. Moreover, alleles responsible for the expression of male phenotype in this species conform to Hardy-Weinberg equilibrium. We conclude therefore that there is no significant selection favouring one male reproductive strategy, and that the necessary conditions for maintaining a polymorphism in male mating behaviour exist in this natural population. □

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Cloning of the gene for a human dopamine D_4 receptor with high affinity for the antipsychotic clozapine

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DOPAMINE receptors belong to the family of G protein-coupled receptors. On the basis of the homology between these receptors, three different dopamine receptors (D_1, D_2, D_3) have been cloned[1-7]. Dopamine receptors are primary targets for drugs used in the treatment of psychomotor disorders such as Parkinson's disease and schizophrenia[8,9]. In the management of socially withdrawn and treatment-resistant schizophrenics, clozapine[10] is one of the most favoured antipsychotics because it does not cause tardive dyskinesia[11]. Clozapine, however, has dissociation constants for binding to D_2 and D_3 that are 4 to 30 times the therapeutic free concentration of clozapine in plasma water[12,13]. This observation suggests the existence of other types of dopamine receptors which are more sensitive to clozapine. Here we report the cloning of a gene that encodes such a receptor (D_4). The D_4 receptor gene has high homology to the human dopamine D_2 and D_3 receptor genes. The pharmacological characteristics of this receptor resembles that of the D_2 and D_3 receptors, but its affinity for clozapine is one order of magnitude higher. Recognition and characterization of this clozapine neuroleptic site may prove useful in the design of new types of drugs.

Various tissues and cell lines were screened for the presence of dopamine-like receptors by northern blot analysis. RNA isolated from human neuroblastoma SK-N-MC cells (ATCC) was hybridized to a radiolabelled 300-base-pair (bp) *Bst*YI-*Bgl*II fragment of the rat D_2 receptor[1] which encodes the putative transmembrane domains VI and VII. Under low-stringency conditions, several bands were detected on the northern blots (data not shown), but under high-stringency conditions none of the bands hybridized with the probe. This suggested that the cell line contained other receptors homologous but not identical to the D_2 receptor.

A complementary DNA library of the neuroblastoma SK-N-MC was constructed in the vector λZAPII. About 500,000 independent clones were screened under low-stringency conditions, using the 300-bp *Bst*YI-*Bgl*II fragment and a 1.6-kilobase (kb) *Bam*HI-*Bgl*II fragment containing the complete coding sequence of the rat D_2 receptor[1]. Several different clones were identified by this procedure. One of the clones, called $D_2$10S, contained a 780-bp *Eco*RI-*Xho*I insert which has a high degree of homology to sequences encoding the putative transmembrane