U, uracil; abbreviations of the amino acid residues are: Ala, alanine; Asn, asparagine; Asp, aspartic acid; Arg, arginine; Cys, cysteine; Glu, glutamic acid; Gln, glutamine; Gly, glycine; His, histidine; Ile, isoleucine; Leu, leucine; Lys, lysine; Met, methionine; Phe, phenylalanine; Pro, proline; Ser, serine; Thr, threonine; Trp, tryptophan; Tyr, tyrosine; Val, valine.
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34. We thank Berge Hampar and Martin Zweig of the National Cancer Institute, Frederick, Md., for providing HSV-1 monoclonal antibodies used in this study, Tom Silhavy for the *E. coli* strain NF1829, and Bob Stallard for assistance with DNA sequencing. The collaboration of Anamaris Colberg-Poley, Carol Marcus-Sekura and Barrie Carter of the National Institutes of Health, Bethesda, Md., in the oocyte injection experiments is gratefully acknowledged.

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Heritable True Fitness and Bright Birds: A Role for Parasites?

Abstract. *Combination of seven surveys of blood parasites in North American passerines reveals weak, highly significant association over species between incidence of chronic blood infections (five genera of protozoa and one nematode) and striking display (three characters: male "brightness," female "brightness," and male song). This result conforms to a model of sexual selection in which (i) coadaptational cycles of host and parasites generate consistently positive offspring-on-parent regression of fitness, and (ii) animals choose mates for genetic disease resistance by scrutiny of characters whose full expression is dependent on health and vigor.*

Whether mate choice could be based mainly on genetic quality of the potential mate has been a puzzle to evolutionary biologists (*1, 2*). Population genetic theory predicts that any balanced polymorphism for a selected trait ends with zero heritability of fitness, so that no one mate is better for "good genes" than any other. However, females of many species act as if they are choosing males for their genes; thus "good genes" versions of sexual selection have been frequently, albeit tentatively, suggested (*2–4*). Here we propose a way out of the difficulty via a previously unconsidered mechanism of sexual selection, and give preliminary evidence for the operation of the principle in North American birds.

There may exist a large class of genes with effects on fitness that always remain heritable. The genes are those for resistance to various pathogens and parasites. The interaction between host and parasite [parasite here being interpreted in a broad evolutionary sense (*5*)] is unusual because it so very readily produces cycles of coadaptation. These cycles can ensure a continual source of fitness variation in genotypes.

To illustrate, imagine a host and parasite population, each with the two alternative genotypes *H*, *h* and *P*, *p*, respectively. For simplicity, assume the organisms are haploid, although diploid models can easily and realistically be made to work in a similar fashion. An *H* individual is resistant to pathogen type *p*, but susceptible to *P*, and vice versa for *h* individuals. The parasite, of course, flourishes in an individual host that is susceptible and dies (or is less productive) in a host that is resistant.

If a female chooses an *H* male when *p* will be the more common parasite genotype in the next generation, she is obviously getting a selective advantage, since her offspring will be more likely to be resistant to disease. As selection proceeds, both by the basic advantage of *H* when *p* is common and by the enhancement through any preference for *H*, the usual problem of variation damping out as all individuals become resistant might be envisioned; but meanwhile selection has been operating within the parasite population and has been favoring *P*. As the proportion of *P* individuals increases, the advantage of *H* falls; *h* then begins to increase in frequency and becomes the better genotype for females to choose.

Such a system usually has an equilibrium point where all four genotypes could occur together. But theory predicts that, given the pattern of host-parasite genotype interactions outlined above, this equilibrium point is unlikely to be stable (*6*). If it is unstable, then a limit cycle, or at least a permanently dynamical behavior of some kind, is instead the probable outcome. Cyclical selection affecting one locus in the host implies that there must be two generations per cycle where heritability is negative—those where advantage switches from *H* to *h* and vice versa. But it also implies that as cycles lengthen the *mean* parent-offspring correlation in fitness must become positive, with .5 as asymptotic upper limit. If cycles are very short, then trying to choose mates for the "right" genes for resistance is a perverse task; an animal might even do best to seek a "worst-looking" mate. Despite theoretical possibility (*7*), it is not clear yet if extremely short cycles (for example, period 2) are likely to occur in nature. Nevertheless, cycling could be relatively rapid under, in general, conditions involving intense selection pressure and pathogens that are short-lived and highly mobile and infectious (*7*). On the other hand, weak selection, approximate equality of generation time of host and parasite, and also any lag in the feedback (such as long-dormant infective eggs or a long-lived vector) tend to create long cycles. Up to a point (*8*), these should favor sexual selection.

Broadening an a priori case for cycles generally it may be noted (i) that epidemic rather than steady occurrence of disease can be involved without losing the tendency to cycle, and (ii) that existence of two or more species of parasite differently virulent to host genotypes hardly differs conceptually from the case of two or more genotypes in an asexual parasite species. In any case when several cycles

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of differing periods are in progress at once, care in choice of a mate should be eugenically rewarding. Then a male (for example) who is unmistakably outstanding in health and vigor offers females that mate with him an inherited healthiness in their offspring that is reliably well above average.

No direct evidence exists for coadaptational cycles of the kind suggested, but they have never really been looked for. Since the search requires study over several generations of a host, cycles may be hard to show even if common. The possibility of cycles is suggested by differing varietal susceptibilities to particular diseases within host species (*9, 10*), by differing virulences of parasite strains (*9, 11*), and by both kinds of variation within single species-pair systems (*12*). It has been claimed that any domestic species can have its resistance to almost any disease rapidly improved by selection (*13*). This again suggests that disease resistance may be permanently heritable in nature and speaks against common occurrence of static equilibria due to fitness overdominance.

Suitable parasites for tests of the theory are those that debilitate their host rather than either kill it or allow total recovery after brief sickness. Death of potential mates preempts the need for choice, while, if recovery is so complete that affected animals are indistinguishable from those which were never susceptible, a selecting mate has no usable basis for discrimination. Admittedly some kill-or-recover diseases may leave records of their severity in the form of stunted growth or shabby plumage, and thus may be able to provide cues of the required kind, but again such diseases are likely to be too rapidly evolving, as mentioned above, to give the longish cycles favorable to sexual selection. The ideal disease is one that can be acute and cause heavy juvenile mortality, but persists in chronic form in survivors either as an infection actually latent or as prolonged aftereffects (such as autoimmune disease) throughout later life.

How could animals choose resistant mates? The methods used should have much in common with those of a physician checking eligibility for life insurance. Following this metaphor, the choosing animal should unclothe the subject, weigh, listen, observe vital capacity, and take blood, urine, and fecal samples. General good health and freedom from parasites are often strikingly indicated in plumage and fur, particularly when these are bright rather than dull or cryptic (*14*). The incidence of bare patches of skin, which may expose the color of the blood in otherwise furry or feathered animals, and the number of courtship displays involving examination of male urine (*15*), are of interest in this regard. Vigor is also conveyed by success in fights and by the frequently exhausting athletic performances of many displaying animals (*16*). Display characters in polygynous species often seem to go far beyond the obvious ways of advertising health—huge tail feathers, for example, or wattles pigmented almost so as to conceal blood color rather than reveal it. Already proposed processes of exaggeration through sexual preference may account for this (*17–20*).

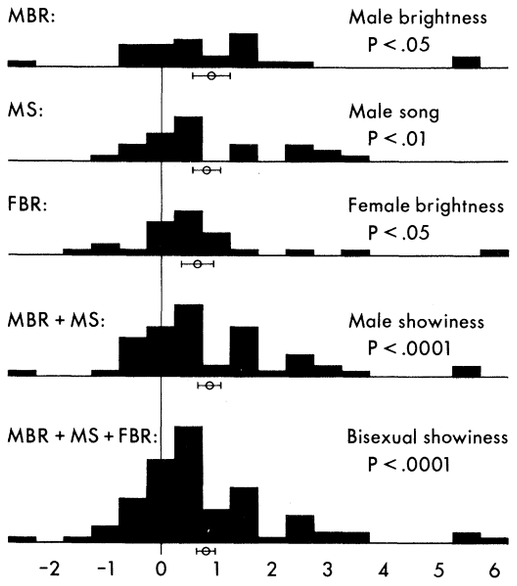

Fig. 1. Distributions of measures of association of display characters with levels of incidence of six blood diseases in North American passerine bird species. Each of first three histograms distributes the measures from 26 tables obtained from the following schema: 1 character × [5 diseases × (1 place × 2 ages + 2 places × 1 (all) ages) + 6 diseases × 1 place × 1 (all) ages]. The two lower histograms are accumulations of those above. Measure distributed is Goodman-Kruskal G divided by standard error. It is grouped for intervals −.25 to .25, .25 to .75, with 0, 1, and so on marked on base line scale. On null hypothesis of no association a standard normal distribution on this scale is expected. Bars show observed means ± standard errors.

If susceptibility to parasites is as important in sexual selection as this idea suggests, animals that show more strongly developed epigamic characters should be subject to a wider variety of parasites (except for purely acute pathogens). In species where disease is relatively unimportant, or where only acute diseases occur, sexual selection should be less apparent and the animal less showy. Whichever sex does the choosing picks individuals with the fewest parasites and the highest resistance; the point is that such choice by one sex and advertisement of good health by the other is needed most in species where chronic parasites are common to begin with. Our hypothesis is contradicted if *within* a species preferred mates have most parasites (*21*); it is supported if *among* species those with most evident sexual selection are most subject to attack by debilitating parasites.

A test we have conducted supports the last point. We used data already in the literature on comparative parasitemias of North American birds. In fairly comparable studies in different places, blood smears were taken from mist-netted and trapped birds. Fairly standard numbers of microscopic fields were searched for the presence of various hematozoa and usually also for microfilarial worms (*22*). Data from two locations were substantial and particularly well suited to our analysis: Algonquin Park in Canada (Alg) (*23*), and Cape Cod (CC) (*24*). Four small surveys of birds in South Carolina and Georgia were included as one set (SCG) (*25*); these did not completely record microfilariae. Finally, a small set from the District of Columbia (DC) was included (*26*). The DC set alone recorded *Toxoplasma*. Each of the six generic "diseases"—*Leucocytozoon, Haemoproteus, Plasmodium, Trypanosoma, Toxoplasma*, and *Microfilaria*—was recorded when discovered as having P_{ij} cases of disease among n_j birds, where i is disease and j is bird species. Multiple infections were counted as one case for each disease involved. The diseases all seemed such as could contribute to the driving of our model. Most tend to be severe and fatal in young birds and then chronic or with long latent periods in the survivors; for some, hints of both variation in resistance (*27*) and epidemic cycles (*28*) have been reported.

From the species lists, each sex of each passerine bird species was ranked by one of us who had not yet seen the disease surveys on a showiness scale of 1 to 6, with 1 being very dull and 6 very striking. Male scarlet tanagers were thus assigned a 6, while most male warblers rated a 3 or 4. No passerines were dull enough to rate 1 (chimney swifts, however, would fall here). The songs of male passerines were ranked similarly, on the basis of variety and complexity, by an expert in bird songs who was supplied

with only the list of species and not their disease frequencies. Adult body weights for each species were also added to the data file. Since nonpasserines were in very varying proportions both among themselves and with respect to the passerines, and were always a rather small minority, they were discarded from the analysis (*29*). The passerines surveyed included 109 species and 7649 individual birds.

For each location (and for Algonquin juveniles and adults separately), we constructed ordered contingency tables for association of the display characters male brightness, female brightness, and male song, with the incidence of each disease. First we constructed disease expectations per bird: $X_{ij} = P_{ij}/n_j$. Then to spread the X_{ij} and give a discrete table when distributed over display ratings, we used, in most cases, 10 X_{ij} rounded to the nearest integer, so that the table set out "expected cases per ten birds" (*30*). From the contingency tables, Goodman-Kruskal gammas were calculated as the coefficients of association (*31*). Our choice of this in preference to more familiar multiple regression methods followed from the very nonnormal distributions of the P_{ij}. For skewed distributions and for sparse entries in its tables, the Goodman-Kruskal method, on the null hypothesis of no association, leads robustly to a standard normal distribution of the following statistic: the gamma estimate divided by its standard error (*31*). This is what we tested.

The tests lead us to reject the null hypothesis of no association. The associations are much more commonly positive than negative and often are positive individually at significant levels (Fig. 1). Coefficients for female brightness are less positive and significant than those for male brightness and song. These conclusions apply broadly across localities and diseases, although diseases are erratic in the trends shown in the different localities.

Toxoplasma was exceptional in giving all negative coefficients in the one set recording it; two of these were significantly negative at the 5 percent level. Algonquin data (the most northerly set and by far the largest) gave the strongest evidence of association, while Cape Cod (second most northerly and third largest set) gave no significant coefficients at all. The negative results for *Toxoplasma* are unexpected, but significantly positive results would not be expected either; this parasite is an extreme generalist regarding its secondary hosts, including birds, and the feedback from any one host species to the *Toxoplasma* gene pool must be correspondingly slight and coevolutionary cycles consequently are unlikely (*32*). Values of gammas themselves are seldom high, 89 percent being below .4. That they are on the whole mildly positive and that numerous values are needed before their trend is established conforms to theory, for it would be most surprising if our first-selected subset from among all disease organisms, namely, the subset of the blood parasites, were to provide all relevant activators of the cycles that underlie sexual selection. In our results it is the high gammas (two reach almost .8) that are puzzling but at present they are easily regarded as fortuitous.

The positive association found for female brightness might follow from imperfect sex limitation of gene expressions only selected for in males. However, we prefer the idea that in monogamous birds sexual selection aiming at genes for health can be working in both sexes at once. As with other suggestions for sexual selection under monogamy, this precludes the very large fitness differences that can occur among males under polygyny and also precludes extremes of a runaway process (*17, 18*). Under any of these views sexually symmetrical ornament is expected to be relatively modest. This is so. However, there is also a widely reported phenomenon often involving bisexual ornament that seems explicable by our view and not by those alternatives where an advantage in natural selection merely primes sexual selection (*17, 18*), or where the whole process is imagined to work on arbitrary characters from the start (*19*). Both bisexual brightness and degree of sexual dimorphism tend to decline on islands (*33*). In our view this could happen because bird species escape from many of their parasites when they colonize islands.

Correlations of disease susceptibility (measured by X_{ij}) and the display characters (including body weight) indicated that in two of the data sets, log weight correlated more strongly with disease susceptibility than did any other character. These data sets (Alg and SCG) were also those that conformed best to our hypothesis for male display characters. Larger birds may provide a bigger target for the biting flies that carry protozoan diseases; or they may tolerate bites more because each bite is a relatively smaller subtraction of blood. Thus our result could be compatible with the predictions of classical sexual selection theory, since greater sexual selection and showiness are often connected with larger body size (*16*) and this is easily rationalized through the superiority of large males in fighting. Still, if size increase occurring for any reason leads to more disease (*34*), increased sexual selection through our suggested linkage is still expected and may entrain, among other things, further increase in size.

Other alternative explanations for the result could be offered. Evaluation of the many plausible and overlapping theories proposed about display and sexual selection, including our new one, is a daunting task and must be left to the future. For the present we conclude that eugenic sexual selection can work and may be common, and that our results hint at chronic disease as one agitator of the dynamic polymorphism that such selection requires.

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Opiate Receptor Distribution in the Cerebral Cortex of the Rhesus Monkey

Abstract. *The distribution of opiate receptors in the cerebral cortex of the rhesus monkey* (Macaca mulatta) *was determined by autoradiographic visualization of [^{3}H]naloxone binding to tissue sections. Naloxone was bound in relatively large amounts to the cortical laminae containing the cell bodies of output neurons, to a varying set of additional laminae in different cortical fields, to fields closer to more primitive types of cortex, and to polysensory cortical fields. From these laminar and areal variations in distribution, it appears that opiate receptors play a role in specific aspects of cortical function.*

Opiates are best known for their analgesic properties, but administration of these compounds results in a constellation of effects. Some of the effects suggest an influence on the cerebral cortex (*1, 2*). While opiate binding levels in broad cortical regions have been described (*3, 4*), no analysis has been made of binding in different cortical laminae or of changes in binding patterns that may occur at the boundaries of cortical fields. We present here an autoradiographic investigation of the laminar and regional distribution of opiate receptors in the cerebral cortex of the rhesus monkey (*5, 6*).

Autoradiographs of [^{3}H]naloxone binding to opiate receptors (*7*) were prepared following incubation of cryostat-sectioned, unfixed, slide-mounted tissue in 2.5 n*M* [^{3}H]naloxone (specific activity, 50 Ci/mmole) and 0.05*M* tris-HCl with 100 m*M* NaCl (*p*H 7.4) at 0°C (*8*). The sections were fixed in formaldehyde vapor after incubation, apposed to tritium-sensitive film, and exposed for 8 weeks at room temperature. The sections were subsequently stained with Thionine for cytoarchitectonic analysis. The ratio of total to nonspecific [^{3}H]naloxone binding was determined to be 9.1:1 by adding unlabeled etorphine (1 μM) to the incubation medium as a competitive blocking ligand. One hemisphere was sectioned in a frontal plane, the other in a parasagittal plane.

Several findings emerged from our analysis. The infragranular layers (*9*) showed relatively high levels of opiate receptor binding throughout most of the neocortex. For example, labeling was heavy in layer VI of the precentral motor cortex (Figs. 1a and 2b) and premotor cortex (not shown). The postcentral somatic sensory areas (Figs. 1a and 2b), the striate visual cortex (Fig. 1), and the peristriate visual cortex (Fig. 1) (*10*) displayed enriched binding in layers V and VI. The primary auditory cortex, the second somatic sensory cortex, and some visual fields (such as the inferotemporal cortex) had high binding levels in layer V, with lower levels in layer VI (Fig. 2a). Many additional fields, especially those in the superior temporal and lateral sulci, also had relatively high levels of binding in infragranular layers (Fig. 1a). Since laminae V and VI contain the cell bodies of the vast majority of corticofugal neurons (*11*), the prevalence of opiate receptors there suggests that opiates may play a role in the control of cortical output.

The influence of opiates on cortical output could be tested directly, since neurons in these layers are identifiable by antidromic activation of corticofugal fibers. It would be of interest to determine, for example, the effect of systemically or locally applied opioid compounds or antagonists on properties of the visual receptive fields of cells in layers V and VI in the monkey's striate visual cortex.

Although the infragranular layers showed relatively high levels of opiate binding in most cortical fields, overall laminar binding patterns varied in different cortical fields (*5*). For example, in the precentral motor cortex, layer I

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