On the Origins of Polymorphisms

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POLYMORPHISM OF PHOTOPIGMENTS IN THE PLATYRRHINI

A polymorphism of cone pigments is present in more than one species of New World monkey (see, for example, Mollon et al., 1984; Bowmaker et al., 1984; Neitz et al., 1985). How might such polymorphisms arise and be maintained?

In his paper in this volume, Jacobs sets out the genetic model that is prompted by the behavioral, microspectrophotometric, and electrophysiological results for the squirrel monkey: Three alleles exist for a single locus on the X-chromosome; and heterozygous females come to have two types of cone in the middle- to long-wavelength spectral region because "Lyonization," or X-chromosome inactivation (Lyon, 1972; Gartler and Riggs, 1983), ensures that the different gene products are segregated in different cells. Can the polymorphism be explained by postulating a heterozygous advantage or, in other words, by postulating that the heterozygous genotype is biologically more fit than any of the homozygous types?

The most celebrated example of heterozygous advantage is that of sickle cell anemia, where the heterozygous carrier is more resistant to the malarial parasite than is the normal homozygote (Allison, 1964). An example that is closer to home is provided

by the polymorphism of the glucose-6-phosphate dehydrogenase (G6PD) gene, a gene that is known to lie on the q-arm of the X-chromosome, very close to the protan and deutan loci. In this case it is reported similarly that the GD^- alleles, that it, those that yield a deficiency of G6PD, are common only in populations exposed to malaria and that girls who are GD^+/GD^- heterozygotes have lower levels of parasitemia than do homozygotes (Luzatto and Battistuzzi, 1985). In the case of the squirrel monkeys, there is no doubt that the heterozygous females are at an advantage in laboratory tests. They are behaviorally able to make discriminations in the red-green range that are impossible for their dichromatic conspecifics (Jacobs, 1984; Jacobs, this volume). It remains a speculation to suppose that they are biologically more fit in their home environment, but it is a plausible one. Using color as a cue, the heterozygous monkey should be better able to find fruit or conspecifics in the dappled environment of a forest, for color vision is particularly useful not (as often claimed) in the rare case where an edge must be detected between equiluminant surfaces, but in the much more common case where luminance edges are masked by local variations in illumination. Furthermore, the heterozygous monkey should be better able to judge the ripeness of the fruit, since ripeness is often indicated only by delicate variations in hue.

How would the polymorphism arise in the first place? Consider a homogeneous dichromatic population and suppose that a rare mutant gene arises at the locus that specifies the single ancestral pigment in the red-green range. As long as the new allele is rare, it will almost invariably find itself paired with the wild-type allele when it is inherited by a female monkey. I have postulated that the heterozygous female is at an advantage and so the frequency of the new allele should rise. But the higher the frequency of the second allele, the more likely is it to be paired with itself and so be carried by homozygous animals that enjoy no advantage. Other factors being equal, the second alelle should reach an equilibrium frequency of 0.5. If, now, a third mutant allele enters the gene pool, it too will be at an advantage as long as it is rare, and its frequency should rise. A new equilibrium should be reached when each of the three alleles has a frequency of 0.33.

The heterozygous advantage postulated here is a particularly pure case. The advantage lies directly in the heterozygosity: It does not matter which two alelles are inherited, only that they are different ones.

An instructive feature is the role of Lyonization, or X-chromosome inactivation. Heterozygous advantage has often been thought to lie in the ability of the heterozygote to manufacture hybrid enzymes (Fincham, 1972). In the present case, however, it is crucial that the products of the two alleles are segregated in different cells; and Lyonization provides the mechanism. It is easy to imagine that analogous cases might occur in other physiological systems. If, for example, there were a polymorphic X-chromosome locus that specified a membrane receptor molecule, then an extra degree of differentiation might be achieved in the nervous system of the heterozygous female, because there would be two subtypes of neuron with different receptor properties. It is even conceivable that one of the reasons for X-chromosome inactivation is the segregation of polymorphic genes into different cells in the female.

POLYMORPHISM OF PHOTOPIGMENTS IN MAN

In the case of man, the common forms of color blindness have long been attributed to polymorphisms at the loci that specify the long- and middle-wavelength pigments (see Pokorny et al., 1979). Neitz and Jacobs (1986; see also Jacobs, this volume) have now postulated a more comprehensive polymorphism within the color-normal population. They report a striking bimodality of Rayleigh matches that is reminiscent of the bimodality reported 20 years ago by Waaler (1967; see Mollon, 1986).

Is it possible that the polymorphisms of human photopigments are also maintained by heterozygous advantage? Such an idea was advanced by Cruz-Coke and Varela (1966), who reported that carriers of color deficiency were more fecund than either mothers who were homozygous for normal color vision or mothers who were explicitly color blind. The subjects were patients in a Chilean maternity hospital. The mean number of progeny for normal mothers was 4.63, whereas the value for heterozygotes was 6.02.

Cruz-Coke and Varela (1966) give few details of their study and it certainly has one flaw. Each child of a carrier has a chance of only about 0.25 of being a color-deficient male, and thus, even a multiparous mother can remain silently heterozygous. The more children she bears, the more likely she is to reveal her heterozygosity. So some correlation between demonstrable heterozygosity and number of progeny must necessarily be expected. Nevertheless, the reported difference in fecundity was very large. And it is nowadays

even more plausible that the opsins of cone pigments might have a pleiotropic role in another physiological system connected with reproduction, for it is known that the opsins are members of a much larger family of homologous receptor molecules (Nathan et al., 1986a, 1986b; Dixon et al., 1986; Kubo et al., 1986). So I have recently been led to check whether there are any gross differences in fertility between normal and heterozygous mothers in a British population. This study was performed in collaboration with J. Watson and J. Ellis.

In a modern British population, any true difference in fertility is likely to be masked by the widespread use of contraception. Therefore, in addition to recording number of offspring, we recorded other potential indices of fertility: reported menstrual regularity, the interval between the date of marriage and the birth of the first child, the reported interval between ceasing contraception and becoming pregnant, and attendance at fertility clinics. Each control mother was a friend or neighbor of one of the heterozygotes. The homozygous and heterozygous mothers and their sons were tested on the Nagel anomaloscope, the Ishihara plates, and the OSCAR test (Estévez et al., 1983).

Figure 1 shows, separately for heterozygotes and normals, the interval from marriage to birth of the first child. Although the distribution for heterozygotes looks skewed to short intervals, a Mann-Whitney test shows no significant difference between the two distributions (U =525; z =1.46). None of our other fertility indices yielded significant differences. So, perhaps we should instead seek a heterozygous advantage more closely analogous to that postulated for trichromatic squirrel monkeys.

One of the purposes of our study of heterozygous women was to evaluate the OSCAR test as a rapid clinical means of identifying carriers of color deficiency. In this test, a surface is illuminated by red and green light-emitting diodes, which are flickered in counterphase. Rotation of a single control knob increases the depth of modulation of one light while it decreases the depth of modulation of the other. The observer is asked to find the setting at which the flicker is minimally visible. We had expected heterozygotes to give abnormal settings, because it is tests of spectral luminosity, rather than tests of color matching, that have classically indicated heterozygosity (Crone, 1959; see also Pokorny and Smith, this volume). Figure 2 shows the settings of heterozygous and normal mothers on the OSCAR test, each plotted against the setting of

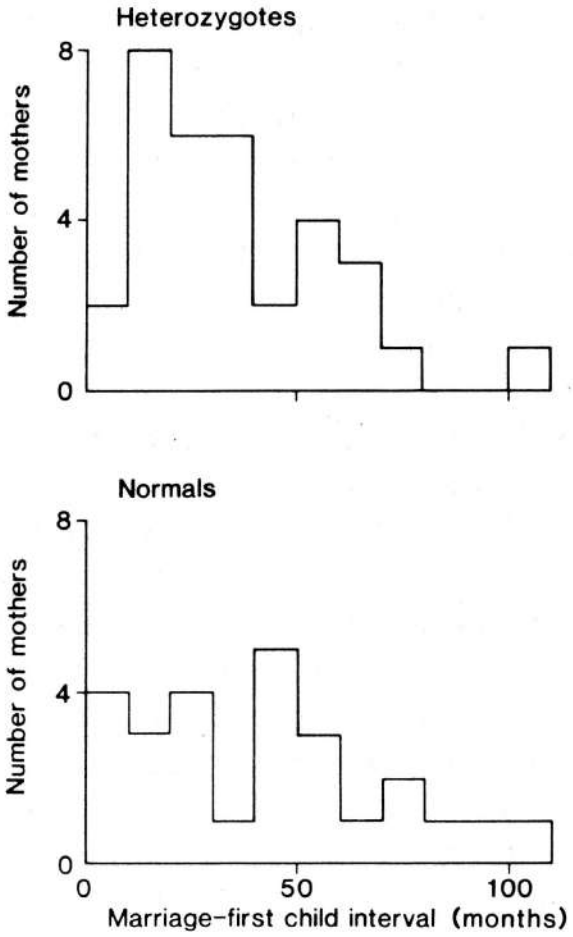

FIGURE 1 Histograms showing the interval (in months) between marriage and the birth of the first child for heterozygous mothers and for putatively homozygous mothers.

her own son. There was a highly significant correlation between mothers and sons ($r = 0.809$; $t = 9.92$; $P < 0.001$). As groups, the deutan and protan carriers were clearly distinct from the normals, but the test does not identify every individual heterozygote.

To explain why carriers of color deficiency have abnormal spectral luminosity curves, X-chromosome inactivation must again be considered. In any given cone in the retina of the heterozygote, only the paternal or only the maternal X-chromosome is expressed. In the case, say, of a carrier of a protan defect, on average half of her long-wavelength cones will resemble those of her deficient son, in that they will fail to manufacture a long-wavelength pigment or will manufacture one that is less sensitive to long wavelengths. The reduction in the number of normal long-wavelength cones would be expected to express itself in a lowered psychophysical sensitivity at long wavelengths (see Pokorny and Smith, this volume).

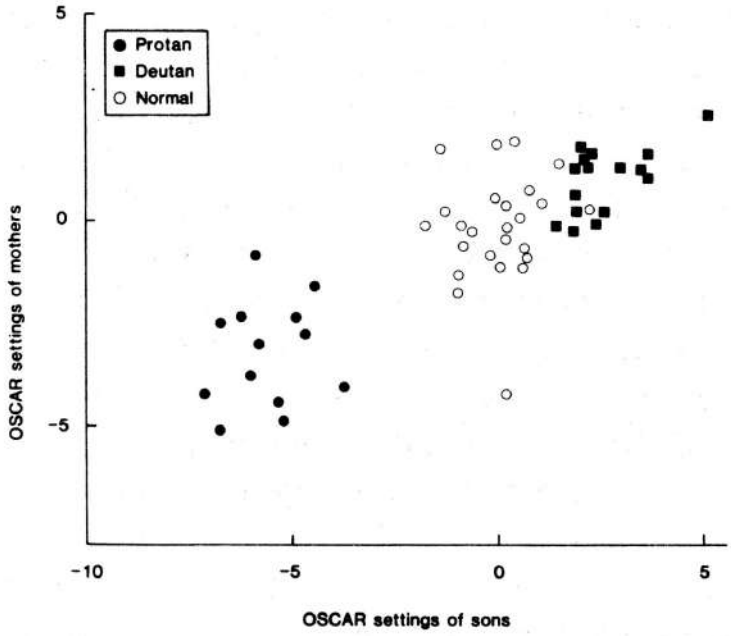

FIGURE 2 The mean OSCAR settings of heterozygous and homozygous women plotted as a function of the settings of their sons. The ordinates are expressed as scale readings, which are in arbitrary units, but are linearly related to the depths of modulation of the two lights.

The results of Figure 2 and their explanation by Lyonization might be taken to confirm the common view that classical heterozygotes simply share a little in the disability of their sons. Consider the majority of heterozygotes, however: those who are carriers of anomalous trichromacy. In the retinas of these women there are likely to be four classes of cone with four different spectral sensitivities: the three normal types and the anomalous receptor that their son inherits. There should also be four classes of cone in the retinas of those putative heterozygotes identified by Waaler (1967) and Neitz and Jacobs (1986). In the squirrel monkey, a basically dichromatic species, the heterozygotes become trichromatic as a result of X-chromosome inactivation. In the foveola of such monkeys, cones expressing the two alleles are completely intermingled so as to produce a fine mosaic (see Figure 8 in Mollon et al., 1984); and apparently, the nervous system is plastic enough to be able to exploit the differentiation among the cones so as to achieve behavioral trichromacy. Now the human species is basically trichromatic, but it is thought that there are many women who have four kinds of cone in their retina. It is logically possible that these women are tetrachromatic. If they are there is the additional possibility that their advantage maintains the polymorphism in the human species.

But would we not long ago have recognized our tetrachromatic conspecifics? The answer is that human trichromacy is never tested in the way that dichromacy is established in experiments on animals, that is, by determining the spectral mixtures that a subject cannot discriminate (cf. Cornsweet, 1970, p. 218). If a woman makes a trichromatic match for a male experimenter—or if she sets for him a two-variable Rayleigh match in the red-green spectral region—she may comment that the match is merely the best she can achieve; the experimenter will accept her match and is unlikely to offer her an additional primary.

The traditional idea that heterozygotes merely share in the disability of their sons is supported by reports that they exhibit slightly more errors than normal on pseudoisochromatic plates and the Farnsworth-Munsell test, and that their Rayleigh matches are somewhat sloppy (see Verriest, 1972, for references). But all of these tests are designed by trichromats for trichromats. Two printed colors that are near metamers for trichromats may vary along a dimension that is very salient for a tetrachromat, and so her performance on plate or arrangement tests might well be impaired. With regard to the Rayleigh match, suppose that we were mostly dichromats but that there were a few trichromats in our midst. We probably would have invented a clinical color-matching test in which a red and a blue light were mixed to match a white standard. Might we not scorn the trichromats who were unable to make a precise match between the purple and the white?

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REFERENCES

Allison, A.C.
1964 Polymorphism and natural selection in human populations. *Cold Spring Harbor Symposium on Quantitative Biology* 29:137–149.

Bowmaker, J.K., J.D. Mollon, and D. Travis
1984 Variation in the photopigments of the common marmoset. *Journal of Physiology* 353:25P.

Cornsweet, T N.
1970 *Visual Perception* New York: Academic Press.

Crone, R.A.
1959 Spectral sensitivity in color-defective subjects and heterozygous carriers. *American Journal of Ophthalmology* 48:231–238.

Cruz-Coke, R., and A. Varela
1966 Inheritance of alcoholism. Its association with colour-blindness. *Lancet* ii:1282–1284.

Dixon, R.A.F.
1986 Cloning of the gene and cDNA for mammalian β-adrenergic receptor and homology with rhodopsin. *Nature* 321:75–79.

Estévez, O., H. Spekreijse, J.T.W. van Dalen, and H.F.E. Verduyn Lunel
1983 The Oscar color vision test: Theory and evaluation (Objective Screening of Color Anomalies and Reductions). *American Journal of Optometry and Physiological Optics* 60:892–901.

Fincham, J.R.S.
1972 Heterozygous advantage as a likely general basis for enzyme polymorphisms. *Heredity* 28:387–391.

Gartler, S.M., and A.D. Riggs
1983 Mammalian X-chromosome inactivation. *Annual Review of Genetics* 17:155–190.

Jacobs, G.H.
1984 Within-species variations in visual capacity among squirrel monkeys (*Saimiri sciureus*): Color vision. *Vision Research* 24:1267–1277.

Kubo, T.
1986 Cloning, sequencing and expression of complementary DNA encoding the muscarinic acetylcholine receptor. *Nature* 323:411–416.

Luzatto, L., and G. Battistuzzi
1985 Glucose-6-phosphate dehydrogenase. Pp. 217–329 in H. Harris and K. Hirschhorn, eds., *Advances in Human Genetics 14*. New York: Plenum.

Lyon, M.
1972 Mechanisms and evolutionary origins of variable X-chromosome activity in mammals. *Proceedings of the Royal Society of London B* 187:243–268.

Mollon, J.D.
1986 Questions of sex and colour. *Nature* 323:578–579.

Mollon, J.D., J.K. Bowmaker, and G.H. Jacobs
1984 Variations in colour vision in a New World primate can be explained by polymorphism of retinal photopigments. *Proceedings of the Royal Society of London B* 222:373–399.

Nathans, J., T.P., Piantanida, R.L. Eddy, T.B. Shows, and D.S. Hogness
1986a Molecular genetics of inherited variation in human color vision. *Science* 232:203–210.

Nathans, J., D. Thomas, and D.S. Hogness
1986b Molecular genetics of human color vision: the genes encoding blue, green and red pigments. *Science* 232:193–202.

Neitz, J., and G.H. Jacobs
1986 Polymorphism of the long-wavelength cone in normal human vision. *Nature* 323:623–625.

Neitz, J., G.H. Jacobs, and M. Crognale
1985 Polymorphism of color vision in a Callitrichid monkey. *Investigative Ophthalmology and Visual Science* 26(Suppl.):185.