conventionally placed amacrine cells. Out of a total of 21 cells in the cat's ganglion cell layer injected with Procion dye, Nelson *et al.* (*12*) found three cells that had a combination of features suggesting that they were amacrines. Our studies confirm findings (*13*) that some of the cells of the ganglion cell layer fail to degenerate following optic nerve section. Vaney and Hughes (*14*) pointed out that fewer axons seem to be present in the rabbit's optic nerve than neurons in the ganglion cell layer. Studies in which the optic nerve is exposed to horseradish peroxidase, like the present experiment in which small transported molecules were used, show a population of neurons in the ganglion cell layer that do not become labeled by retrograde transport (*15*). Our experiments, in which acetylcholine serves as a cellular marker, allow an added conclusion: that at least some of the degeneration-resistant neurons are the same cells that cannot be backfilled. When all of this evidence is considered together, it seems quite certain that a substantial number of the cells of the ganglion cell layer are displaced amacrines and that many of these cells synthesize acetylcholine.

All of the cholinergic neurons of the rabbit retina would thus appear to be amacrine cells. We are impressed by the geometry with which the cholinergic cells are positioned. Essentially equal numbers are located on either side of the inner plexiform layer, and their processes arborize at roughly equal distances from the midplane of the plexiform layer. A prime feature of this symmetry is that the cell bodies of half of the amacrine cells involved have been "displaced" to the proximal side of the plexiform layer. It would be interesting to know the functional or developmental reasons for this arrangement.

SHAWN A. HAYDEN
Department of Surgery, Massachusetts General Hospital, Boston 02114
JOHN W. MILLS
Department of Anatomy, Harvard Medical School, and *Department of Medicine, Massachusetts General Hospital*
RICHARD M. MASLAND
Department of Physiology, Harvard Medical School, and *Department of Surgery, Massachusetts General Hospital*

References and Notes

1. C. O. Hebb, *Q. J. Exp. Physiol. Cogn. Med. Sci.* **40**, 176 (1955); C. W. Nichols and G. B. Koelle, *J. Comp. Neurol.* **133**, 1 (1968); R. H. Masland and C. J. Livingstone, *J. Neurophysiol.* **39**, 1210 (1976); R. H. Masland and A. Ames III, *ibid.*, p. 1220.
2. R. W. Baughman and C. W. Bader, *Brain Res.* **138**, 469 (1977).
3. R. H. Masland and J. W. Mills, *J. Cell Biol.* **83**, 159 (1979).
4 The cholinergic cells are too large to be the glia associated with optic nerve axons. In any case, we know of no reports of acetylcholine synthesis by glial cells of the central nervous system.
5. H. G. J. M. Kuypers, C. E. Catsman-Berrevoets, R. E. Padt, *Neurosci. Lett.* **6**, 127 (1977); D. van den Kooy, H. G. J. M. Kuypers, C. E. Catsman-Berrevoets, *Brain Res.* **158**, 189 (1978); H. G. J. M. Kuypers, M. Bentivoglio, D. van den Kooy, C. E. Catsman-Berrevoets, *Neurosci. Lett.* **12**, 1 (1979).
6. E. Frank, W. A. Harris, M. Kennedy, *J. Neurosci. Methods* **2**, 183 (1980).
7. The medium was a bicarbonate-buffered salt solution resembling cerebrospinal fluid in its electrolytes and containing 10 m*M* glucose equilibrated with 95 percent O_2 and 5 percent CO_2. Extensive precautions were taken to prevent exposure of the tissue to toxic substances. Retinas maintained in this way retain protein synthesis, morphological integrity, acetylcholine metabolism, and functional activity as judged by ganglion cell receptive fields for at least 8 hours. A. Ames III, J. M. Parks, F. B. Nesbett, *J. Neurochem.* **184**, 215 (1976); H. Webster and A. Ames III, *J. Cell Biol.* **26**, 885 (1965); R. H. Masland and C. J. Livingstone, in (*1*); R. H. Masland and A. Ames III, in (*1*).
8. J. G. Hildebrand, D. L. Barker, E. Herbert, E. H. Kravitz, *J. Neurobiol.* **2**, 231 (1971).
9. As the transporting time increases, the population of faintly labeled cells becomes increasingly visible in whole mounts. Kuypers *et al.* (*5*) have observed a similar secondary labeling in the brain. On the basis of the appearance of the cells' fluorescence, they believe that the cells are glia labeled by diffusion from nearby neurons. In the retina it is clear that adjacent neurons are labeled as well. This is most striking in the mouse retina where the distance over which transport must occur is shorter and the labeling of neurons brighter than in the rabbit. In the mouse, brightly fluorescent displaced ganglion cells are observed; they are often surrounded by faint halos of other inner nuclear layer cells, and these are far too numerous to be glia. The issue is in any case not central to the present experiments since the faintly labeled cells cannot be observed after processing of the tissue into plastic.
10. See review by E. F. Domino, in *Chemical Modification of Brain Function,* H. F. Sabelli, Ed. (Raven, New York, 1973), p. 95.
11. V. H. Perry, *Proc. R. Soc. London* **204**, 363 (1979).
12. R. Nelson, E. V. Famiglietti, H. Kolb, *J. Neurophysiol.* **42**, 472 (1978).
13. J. T. Eayrs, *Br. J. Ophthalmol.* **36**, 453 (1952).
14. D. I. Vaney and A. Hughes, *J. Comp. Neurol.* **170**, 241 (1976); D. I. Vaney, *ibid.* **189**, 215 (1980).
15. A. H. Bunt, R. D. Lund, J. S. Lund, *Brain Res.* **73**, 215 (1974); U. C. Dräger and J. Olson, *J. Comp. Neurol.* **191**, 383 (1980).
16. The sections shown in Fig. 1 were exposed for 2 weeks, long enough to give a dense accumulation of grains over the cholinergic cells. Sections exposed for short times were also studied to ensure against the possibility that silver grains could obscure the fluorescense of cells.
17. We thank C. Liu and M. Whalen for technical assistance. Supported by NIH grants EY 01075 and HLO 6664, by a Research Career Development Award to R.H.M., and by a King Trust award to J.W.M.

29 February 1980; revised 19 May 1980

Pollen Tube Growth Rates in *Zea mays*: Implications for Genetic Improvement of Crops

Abstract. *Speed of pollen tube growth is positively correlated with the quality of the resultant sporophytic generation. Therefore, gametophytic competition may be an important adaptive mechanism. Furthermore, pollen tube growth rates may be used to predict the quality of F_1 crosses in crop species.*

Plant breeders evaluate inbred lines according to how they perform in different hybrid combinations. The mean performance of each line (the mean performance of the F_1's in crosses with other lines) is indicative of its general combining ability (GCA). The performance of a particular cross may deviate from the average of the two parental lines; this deviation is the specific combining ability (SCA) of that cross (*1*). We consider that pollen tubes within a style, like parental genomes in an F_1 hybrid, represent an interacting combination that can be analyzed in terms of SCA's and GCA's. Pollen GCA, for example, is the mean growth rate of one pollen tube type in several style types, and stylar GCA is the mean growth rate of pollen tubes in that style.

We tested for correlations between pollen GCA and classical GCA and between pollen-style SCA and classical SCA. Positive correlations would imply useful applications, such as a rapid test for combining ability, and would support the hypothesis that pollen tube competition may be of importance in adaptive processes (*2*).

Eleven lines of corn (*Zea mays* L. Poaceae) were used in this study. All are standard inbred lines employed in the production of commercial hybrid corn. Pollen tube growth rates of pollen from five of these lines (WF9, C123, M14, B37, and H3025) were determined in the styles of five of the other lines (W23, 33-16, Sil-91, B73, and B14). This was accomplished by comparing the pollen tube growth rate from each pollen source with that of a standard inbred pollen source, W22. Fertilizations by the standard resulted in red aleurone, while those by the other five pollen sources produced colorless aleurone. Because aleurone color is expressed in the kernel, it allows immediate determination of which kernels resulted from fertilization by tester (W22) gametes and which did not.

Determination of the relative growth rates of pollen tubes is based on the observation (*3*) that, if a mixture of two pollen types is applied to stigmas, and if these types, tagged by genetic markers,

0036-8075/80/1024-0437$00.50/0

Table 1. Regression coefficients of sporophytic traits of the F_1 progeny on competitive ability of parental pollen. Correlation coefficients (in parentheses) incorporate results of ability analyses.

Item	Sporophytic traits				
	Weight of seedling dry matter	Weight of ear	Kernel weight per plant	Mean kernel weight	Kernels per row
Pollen tube growth rate ($b_{P/S}$) and quality of sporophyte	0.0335* (.394)	3.6756† (.233)	2.3148† (.216)	0.2502† (.345)	0.1672 (.106)
Pollen GCA and classical GCA	0.0396* (.572)	9.1138* (.734)	6.8061* (.823)	0.3660* (.852)	0.9548* (.918)
Pollen-style SCA and classical SCA	0.0126† (.204)	−2.2683 (−.109)	−2.0067 (−.138)	0.1179 (.165)	−0.4252 (−.237)

*Significant at $P < .05$. †Significant at $P < .01$.

penetrate the stylar tissues at different speeds, then the probability of the faster type reaching the ovules first is proportional to the style length. That is, the longer the style, the more ovules will be fertilized by gametes of faster pollen tubes. This is explained by random effects of the environment (*4*). For example, in corn, the receptive portion of the style extends for several centimeters. Thus not all the pollen tubes start at the same position; furthermore, some grains land with the germination pore near the stigma, others do not. When the styles are sufficiently long, however, the relative speed of pollen tube growth outweighs these random effects. Since the styles of basal kernels on an ear of corn are longer than the styles of apical kernels, the faster pollen type effects more fertilizations in the basal kernels. Thus a basipetal increase in frequency of fertilization by one pollen type is a measure of that pollen type's greater speed. If the composition of the resultant kernel mixture is determined at different points on the ear, the slope of a regression of percentage of one kernel type against distance from the apex will express the relative speeds of the two pollen types in that stylar type, assuming that both types are capable of reaching the basal ovules.

The ears produced with pollen mixtures were divided, starting at the apex, into five segments of equal length (six kernels per row), and the kernels were scored for aleurone color. There were about eight ears per combination. Uncolored kernels from the third segment were sown in the greenhouse for evaluation of seedling weight, and were also used in a field trial to evaluate mean kernel weight, number of kernels per row, and kernel weight per plant. Three plots of 20 plants were planted. Data were obtained for five ears per plot, each ear from a different plant.

Relative pollen tube growth rates of the five lines are expressed as variations of the proportion (P) of uncolored kernels from the apex to the base of the ear. Accordingly, it was estimated as the linear regression coefficient ($b_{P/S}$) of P on ear segments: a positive value indicates a competitive ability greater than that of W22, the standard. Because the dependent variable is a proportion referred to binomial data, the method of Armitage (*5*) was used for statistical fitting of the linear trend and to determine deviations from regression. However, since the number of observations was very large in all cases (about 1200 kernels per segment of each combination), the values for $b_{P/S}$ obtained with this method were practically the same ($r = .98$) as those obtained with unweighted analyses in which P was used as the dependent variable. Association between sporophytic traits of the progeny and gametophytic competition ability ($b_{P/S}$) of the male parental line was evaluated by means of regression analysis, with $b_{P/S}$ considered as a covariate.

Except for the number of kernels per row, all sporophytic qualities measured showed statistically significant relations to the growth rates of the pollen tubes that gave rise to the sporophytes (Table 1). Pollen GCA was significantly related to classical GCA for all five sporophytic traits measured. From the agronomic point of view, these data may be of particular importance. For example, they demonstrate that pollen tube growth rates allow us to predict 67.7 percent of the variation in kernel weight per hybrid plant. This ability could be used in performing preliminary screening of parental lines of hybrid crops.

Evidence is accumulating that a substantial number of genes that are expressed in the pollen (gametophyte) are also expressed in the sporophyte (*6*). Presumably, the rate of pollen tube growth and the sporophytic qualities measured in the present study could both be limited by such primary metabolic processes as glycolysis, respiration, and synthesis of cellulose or callose. Most of the enzymes involved in these sporophytic metabolic processes have been found in pollen (*7*). If such systems are controlled by the same loci in both phases of the life cycle, gametophytic selection could easily modify the quality of the resultant sporophytic generation.

For at least one sporophytic trait, seedling dry weight, there was a statistically significant relation between pollen-style SCA and classical SCA (Table 1). This finding not only agrees with reports that the growth rates of different pollen tube types change with stylar environment (*4*), it suggests that pollen-style interactions favor pollen tubes of genotypes that give rise to vigorous seedlings.

A hypothesis to this effect states that since the pollen tube is at least partially dependent on the style for its nourishment, normal pollen tube growth is the result of physiological complementation between the male gametophyte and the style (*8*, *9*). Specific support for this hypothesis was provided by Linskens and Pfahler (*10*), who found evidence for physiological complementation between pollen tubes and styles, a complementation of the type that could also account for the differential transmission of male traits observed in the material used.

ERCOLE OTTAVIANO
MIRELLA SARI-GORLA
Institute of Genetics, University of Milan, 20/30 Milan, Italy
DAVID L. MULCAHY
Department of Botany, University of Massachusetts, Amherst 01003

References and Notes

1. D. S. Falconer, *Quantitative Genetics* (Ronald, New York, 1960).
2. D. L. Mulcahy, *Science* **206**, 20 (1979).
3. D. F. Jones, *Selective Fertilization* (Univ. of Chicago Press, Chicago, 1928).
4. P. L. Pfahler, *Genetics* **57**, 513 (1967).
5. P. Armitage, *Biometrica* **2**, 375 (1955).
6. J. B. S. Haldane, *The Causes of Evolution* (Longmans, Green, London, 1932).
7. D. B. Dickenson, J. E. Hooper, M. Davis, *Biogenesis of Plant Cell Wall Polysaccharides* (Academic Press, New York, 1973), p. 29; J. L. Brewbaker, in *Pollen Development and Physiology*, J. Heslop-Harrison, Ed. (Butterworth, London, 1971), p. 156; E. Ottaviano and M. Sari-Gorla, *Monogr. Genet. Agrar.* **4**, 89 (1978).
8. D. L. Mulcahy, *Science* **171**, 1155 (1971).
9. J. Heslop-Harrison, *Annu. Rev. Plant Physiol.* **26**, 403 (1975).
10. H. F. Linskens and P. L. Pfahler, *Theor. Appl. Genet.* **50**, 173 (1977).
11. Supported by Centro Nazionale della Ricerca grant 7902430671152343 to E.O. and by USDA/SEA competitive grant 5901-0410-9-0365-0 to D.L.M.

25 February 1980; revised 16 July 1980