mature (*6*); in an outbreeding system, archegonia might be expected to develop first. The necks of the archegonia of most of the Polypodiaceae (s.l.) are curved inward, away from the apical notch of the gametophyte (*6*), thereby pointing their mouths toward the antheridia and rhizoids. This appears to be an adaptation to self-fertilization. If this were a cross-fertilizing system, selection should favor archegonial necks curved in the opposite direction.

Thus, homosporous vascular plants are apparently characterized by degrees of homozygosity that would be unattainable in a heterosporous plant in many generations of selfing. The maintenance of heterozygosity at individual gene loci is unlikely in these homosporous plants because of continuing exposure of sporophytes to selection in the homozygous condition. The obvious evolutionary diversity and successful adaptation to diverse environments of these plants, however, raise the question of the means by which genetic variability (evolutionary potential) is maintained and realized.

On the average, the Pteridophyta have much higher chromosome numbers than the seed plants. The mean gametic chromosome number of those angiosperms whose numbers had been counted before 1963 is 15.99 (*7*). The mean gametic chromosome number of the Pteridophyta as a whole is calculated to be 55.27, based upon lists compiled by Chiarugi (*8*) and Fabbri (*9*). The mean gametic chromosome numbers of the heterosporous and homosporous Pteridophyta are 13.62 and 57.05, respectively. The magnitude of the chromosome number seems to be correlated with the occurrence of homospory or heterospory. In this connection, the close agreement of the mean gametic chromosome number for the angiosperms (which are heterosporous) and the heterosporous Pteridophyta (n = 15.99 and n = 13.62, respectively) in contrast to the homosporous plants (n = 57.05) is notable.

Figure 1 depicts the distribution of the known gametic chromosome numbers of 50 heterosporous and 1166 homosporous species of Pteridophyta. For comparative purposes the percentage of the total sample falling in each class of gametic chromosome numbers is plotted. Of the homosporous plants, 96 percent have a gametic chromosome number greater than 27, whereas 90 percent of the heterosporous ones have a gametic chromosome number less than 28. Unlike the homosporous species, complete homozygote formation in the heterosporous plants is highly unlikely, and the lower incidence of polyploidy in the latter may be related to this.

Polyploidy increases gene redundancy and, if dominant alleles are present, any recessive alleles of the same genes elsewhere in the genotype are kept from immediate exposure to selection even when the plants are homozygous at each locus. Because polyploidy increases the dosage of genes beyond the disomic condition, genes can mutate and take on new functions without depriving the organism of essential processes which are maintained by their former homologs.

This potential for genetic variation could gradually become expressed in the phenotype of the polyploid or could be released for recombination by occasional hybridization of gametophytes which possess duplicated genes that have mutated in different ways. The resulting sporophytes would be heterozygous and give rise to recombinant progeny, some of which might be adaptive. Intergenomic crossing-over is another possible mechanism for release of this kind of genetic variation.

Polyploidization may provide further selective advantages in the homosporous Pteridophyta by the production of immediate genotypic changes (especially where allopolyploids are produced). These new genotypes may be advantageous by reason of heterotic effects produced by intergenomic heterozygosity or the genetic change may directly result in a specific adaptation.

Thus polyploidization is a means by which homosporous Pteridophyta may maintain, create, and release genetic variability in spite of the production of homozygous sporophytes.

EDWARD J. KLEKOWSKI, JR.
HERBERT G. BAKER
Botany Department,
University of California, Berkeley

References and Notes

1. I. Manton, *Problems of Cytology and Evolution in the Pteridophyta* (Cambridge Univ. Press, Cambridge, 1950); W. H. Wagner, Jr., *Amer. Fern J.* **53**, 1 (1963).
2. D. Wilkie, *Heredity London* **10**, 247 (1956).
3. The experiments reported here were started at University of California at Los Angeles by E. J. Klekowski with the help and advice of H. Lewis and B. C. Parker.
4. B. C. Parker and H. C. Bold, *Amer. J. Bot.* **48**, 185 (1961). Modified B-solution was supplied by B. C. Parker.
5. A. J. Eames, *Morphology of Vascular Plants —Lower Groups* (McGraw-Hill, New York, 1936), p. 171.
6. L. R. Atkinson and A. G. Stokey, *Phytomorphology* **14**, 51 (1964).
7. V. Grant, *The Origin of Adaptations* (Columbia Univ. Press, New York, 1963), p. 486.
8. A. Chiarugi, *Caryologia* **13**, 27 (1960).
9. F. Fabbri, *ibid.* **16**, 237 (1963).

16 March 1966

Fungicide Selective for Basidiomycetes

Abstract. *Concentrations of 2,3-dihydro-5-carboxanilido-6-methyl-1,4-oxathiin lower than 8 parts per million prevented mycelial growth of a number of Basidiomycetes. By contrast, mycelial growth of various other fungi—Phycomycetes, Ascomycetes, and Deuteromycetes—was 50 percent inhibited only by concentrations of 32 ppm or higher. Two exceptions to this pattern of selective fungitoxicity were found: an isolate of* Rhizoctonia solani *was not as sensitive as other Basidiomycetes, and the deuteromycete* Verticillium albo-atrum *was inhibited by lower concentrations than affected other fungi in this group. Spore germination of two Basidiomycetes,* Uromyces phaseoli *and* Ustilago nuda, *was inhibited 95 percent or more at 10 ppm.*

The compound 2,3-dihydro-5-carboxanilido-6-methyl-1,4-oxathiin (oxathiin) has recently proved effective in chemotherapy of bean rust and of loose smut of barley (*1*). Tests in this laboratory, however, indicated that the compound is relatively nontoxic to three ascomycetous fungi. This difference suggested selective toxicity to Basidiomycetes. To explore this apparent

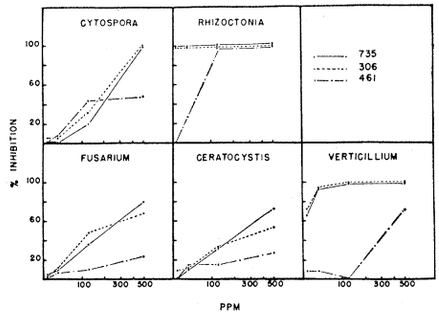

Fig. 1. Inhibition of growth of five fungi by 2,3-dihydro-5-carboxanilido-6-methyl-1,4-oxathiin (*735*) and two of its isomers: 2,3-dihydro-5-(*N-m*-tolyl) carboxanilido-6-methyl-1,4-oxathiin (*306*) and 2,3-dihydro-5-carboxanilido-6-methyl-1,4-oxathiin-4,4-dioxide (*461*).

selectivity, the toxicities of this compound and two of its analogs—2,3-dihydro-5-carboxanilido-6-methyl-1,4-oxathiin-4,4-dioxide (dioxide analog) and 2,3-dihydro-5(*N-m*-tolyl)carboxanilido-6-methyl-1,4-oxathiin (tolyl analog)—to fungi of the four taxonomic classes were determined.

A dilution series of the test compound was prepared in acetone, and 0.1 ml of such a solution was added to 10 ml of molten potato dextrose–agar in each growth tube to give concentrations of 0, 8, 32, 125, or 500 ppm; the agar was buffered at *p*H 5.3 with 0.1*M* potassium acid phthalate. For *Cytospora kunzei*, the medium described by French and Helton was used (*2*). Growth tubes were inoculated by transfer of 4-day-old mycelial discs of the test fungi to the sterile agar surface. Mycelium was allowed to grow for 2 to 14 days at room temperature, depending on the growth rate of the particular fungus. Each treatment was replicated three times.

Figure 1 shows that oxathiin and its tolyl analog have very similar patterns of fungitoxicity, which is greater in vitro than that of the dioxide analog. Being the most fungitoxic of the three, oxathiin alone was used in the remaining experiments. Figure 2 shows the lower fungitoxicity of oxathiin to Phycomycetes, Ascomycetes, and Deuteromycetes than to Basidiomycetes; the only exceptions were the deuteromycete *Verticillium albo-atrum* and the basidiomycete *Rhizoctonia solani* (isolated from aster). Both isolates of *R. solani* (one from aster, one from potato), under phase microscopy, proved to have similar dolipore septa (*3*).

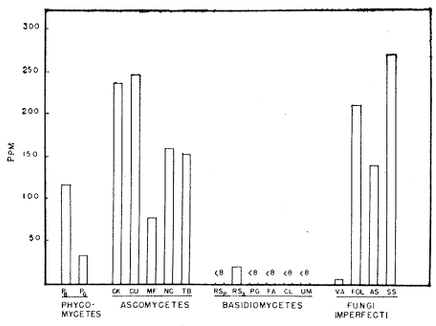

Fig. 2. Concentrations (ppm) of 2,3-dihydro-5-carboxanilido-6-methyl-1,4-oxathiin required for 50-percent inhibition of growth of various fungi. P_B, *Pythium* sp. isolated from bean; P_G, *Pythium* sp. isolated from geranium; *CK, Cytospora kunzii; CU, Ceratocystis ulmi; MF, Monilinia fructicola; NC, Nectria coccinia; TB, Thielaviopsis basicola;* RS_B, *Rhizoctonia solani* isolated from potato; RS_A, *Rhizoctonia solani* isolated from aster; *PG, Polyporus gigantea; FA, Fomes annosus; CL, Coprinus lagopus; UM, Ustilago maydis; VA, Verticillium albo-atrum; FOL, Fusarium oxysporum* f. *lycopersici; AS, Alternaria solani;* and *SS, Stemphylium sarcinaeforme.*

Inhibition of spore germination was determined by use of spores of two other Basidiomycetes: *Uromyces phaseoli* and *Ustilago nuda* (both obligate parasites). Spores of each fungus were exposed to oxathiin at concentrations of 10, 100, 1000, and 10,000 ppm. Only 22 percent of the bean-rust spores and 52 percent of the loose-smut spores germinated on the control slides. With both fungi all concentrations of oxathiin reduced the percentage of germination to from 1 to 4 percent; a few spores germinated even in saturated solutions containing many floating crystals of oxathiin.

Thus oxathiin is fungitoxic in vitro to Basidiomycetes and may be useful for controlling plant diseases caused by them.

L. V. EDGINGTON*
G. S. WALTON
P. M. MILLER
Department of Plant Pathology and Botany, Connecticut Agricultural Experiment Station, New Haven

References

1. B. von Schmeling and M. Kulka, *Science*, in press.
2. W. J. French and A. W. Helton, *Phytopathology* **52**, 610 (1962).
3. R. T. Moore and J. H. McClear, *Amer. J. Botany* **49**, 86 (1962).

* Present address: Department of Botany, University of Guelph, Guelph, Ontario.

1 May 1966

Electron-Microscopic Autoradiography of Rat Hypothalamus after Intraventricular H^3-Norepinephrine

Abstract. *Tritiated norepinephrine was injected into the lateral ventricles of rats, and its localization in the hypothalamus was determined by light and electron-microscopic autoradiography. Eighty percent of the autoradiographic grains were located over nerve endings and unmyelinated axons. Large, dense synaptic vesicles were present in most of the endings and axons with activity. Grains were rarely seen over myelinated axons, glia, or blood vessels.*

Parenterally administered tritiated norepinephrine (H^3-NE) has been localized in peripheral sympathetic axons of the rat pineal body by electron-microscopic autoradiography (*1*). Autoradiographic grains were located in or near unmyelinated axons containing granular ("dense-core") synaptic vesicles 400 to 500 Å in diameter. The dense vesicles have been correlated with the subcellular binding sites of peripheral norepinephrine (NE) as judged by morphological (*2, 3*), pharmacological (*4*), and fluorescent histochemical (*5*) experiments. Thus, there is good though not universal (*6*) agreement that NE-containing peripheral axons exhibit dense granular vesicles, which contain releasable NE (*4*).

We have used autoradiography to identify the structural correlate of norepinephrine binding sites within the brain. Evidence relating brain NE to the central dense granular vesicles has been mainly circumstantial. The dense granular vesicles of the central nervous system are generally 1.5 to 2.0 times as large as those described in the periphery (*7, 8*). Furthermore, the electron density of the dense "cores" is quite variable. Some investigators (*9*) have questioned the correlation between mono-amine fluorescence and the distribution of the dense vesicles in the central nervous system. Certainly, other biogenic amines present in nerve endings [obtained from density gradient fractionation of brain homogenates (*10*)] might interfere with either fluorescent or electron-microscopic histochemical methods (*11*).

Investigation of central NE binding sites is difficult because parenterally administered amine enters the brain poorly. It has been shown that tracer doses of H^3-NE injected into the lateral ventricle of the rat brain is readily taken up by the brain cells and metabolized similarly to endogenous NE (half-life of 3 to 4 hours) (*12*). The H^3-NE (but not the labeled catabolites) is found predominantly in the nerve ending fraction of brain homogenates. The labeled norepinephrine is, in addition, appropriately responsive to pharmacological agents affecting the stores of the endogenous amine. These results indicate that autoradiography following intraventricularly injected H^3-NE might provide direct evidence on the sites of uptake and binding of exogenous NE in the brain.