

Abstracts of the 1971 Annual Meeting of the Northeastern Division of
The American Phytopathological Society

Pathogenicity of cedar apple rust from New York and other states. H. S. ALDWINCKLE (N. Y. State Agr. Exp. Sta., Geneva). One-year-old trees of eight apple cultivars selected for a range of susceptibility to cedar apple rust (*Gymnosporangium juniperi-virginianae*) were spray-inoculated with an aqueous suspension of basidiospores from moistened telial galls. After 48 hr at 100% RH and 18.5 C, trees were returned to the greenhouse. At least one sample from each state (Ark., Ga., Ind., Minn., N.H., N.Y., and Va.) produced pycnia on Rome Beauty, Jonathan, and Yellow Newton. One Ark. and two N.Y. samples produced nonsporulating orange lesions on Delicious; one of these N.Y. samples produced similar lesions on McIntosh and the other produced very sparsely sporulating pycnia on Cortland. These N.Y. and Ark. galls had yielded the highest frequency of germinable teliospores. No sample produced pycnia on Delicious, McIntosh, Melrose, or Arkansas Black. The highest number of aecia per lesion developed on Rome, followed by Jonathan and Yellow Newton. Aecia appeared on Rome and Jonathan with samples from all eight states, but Yellow Newton, which averaged less than two aecia per lesion, developed no aecium with several samples. No other cultivar showed aecia. Results were interpreted as differences between collections in teliospore germinability rather than in pathogenic races.

Fusarium species associated with corn stalk rot in Pennsylvania in 1970. J. E. AYERS & P. E. NELSON (The Pa. State Univ., University Park). Samples of rotted corn stalks from 133 fields in 28 counties of Pennsylvania were collected in the fall of 1970. Small pieces of stalk tissue were placed on Nash's Medium or potato-dextrose agar in petri dishes and placed under 40-w Ultra-Lux fluorescent lamps (Nu-Lite Division, El-Tronics, Inc., Warren, Pa.) operating on a 12-hr cycle at 21 to 23 C. *Diplodia zeae*, which is associated with corn stalk rot in several areas of the country, was not found in any sample. *Fusarium* spp. were transferred by means of single spores, to potato-dextrose agar plants, grown under the fluorescent lamps at 21 to 23 C and identified. Of the 237 isolates of *Fusarium* spp. obtained, 48% were *F. moniliforme*, 17% were *F. moniliforme* 'Subglutinans', and 12% were *F. roseum* 'Graminearum'. The remaining 23%, in order of occurrence, were *F. oxysporum*, *F. roseum* 'Gibbosum', *F. solani*, *F. tricinctum*, *F. episphaeria*, and several unidentified cultivars of *F. roseum*.

Sulfur dioxide - ozone synergism on eastern white pine. W. M. BANFIELD (Univ. Mass., Amherst). Typical field symptoms of white pine blight characterized by profuse spotting and death of needle ends have been consistently induced in repeated 12-hr daylight exposure periods to mixtures of sulfur dioxide and ozone. Ozone alone did not induce symptoms unless fumigation was continued at 18 pphm for 18 or more days. Sulfur dioxide alone induced symptoms when the concentration exceeded 7 pphm and exposure was continued for 3 or more days. All plants exposed to 18 pphm for 3-10 days developed spot or blight symptoms. In trials in which O₃ was held at 10 pphm, SO₂ induced blight on an occasional plant at 0.5 pphm, on half the plants at concentrations of 1.5 to 4 pphm, and on all plants at 5 pphm when exposure was continued for 1 to 12 days. Exposure for 4 days to ozone at 10 pphm followed by SO₂ at 5 pphm for 4 days after an interval of 9 days did not induce symptoms. Mild spot symptoms were induced on one of six plants when this ozone treatment followed at once after SO₂, and on four of ten plants when 12-hr daylight SO₂ and O₃ treatments were alternated over a period of 8 days. Four hundred sixty-nine susceptible plants grown in

charcoal-filtered greenhouses for 2 years prior to fumigation were used in this work. Five to ten plants were used in each trial.

Specificity of p-fluorophenylalanine for inhibiting sporulation of Ceratocystis ulmi. W. L. BIEHN (Conn. Agr. Exp. Sta., New Haven). Systemic invasion of the vascular system by wilt pathogens depends in part upon sporulation and transport of propagules in the host, according to current ideas. If so, then a compound that inhibits sporulation in the infected host should reduce the rate of spread of infection. In previous work, we found that DL-p-fluorophenylalanine (FPA) has chemotherapeutic activity against Fusarium wilt of tomato. Fungitoxicity studies suggested that FPA might have antispore activity. Therefore, studies were made on the effect of FPA on sporulation (conidia/ml) and growth (mg dry wt) of *Ceratocystis ulmi*. In shake culture, 5 and 10 ppm FPA inhibited sporulation of *C. ulmi* 91 and 95%, respectively. FPA at 450 ppm inhibited growth of *C. ulmi* 82% in shake culture and 68% in still culture. Thus, sporulation of *C. ulmi* is at least 90 times more sensitive to FPA than vegetative growth. In comparison, Teresan LSR (maneb, 80%, plus zinc) inhibited sporulation and growth of *C. ulmi* about equally. Sporulation of *C. ulmi* is optimum in shake culture, whereas vegetative growth is optimum in still culture. Therefore, the specificity of a compound for inhibiting sporulation of *C. ulmi* can be quantitatively measured by the ratio of its ED₅₀ for sporulation in shake culture to its ED₅₀ for growth (mg dry wt) in still culture.

Viruslike particles from a culture of Diplocarpon rosae. R. F. BOZARTH, H. A. WOOD, & A. GOENAGA (Boyce Thompson Inst., Yonkers, N.Y.). Viruslike particles (VLPs) were isolated from mycelium of *Diplocarpon rosae* Wolfe grown in shake culture for 8 days on Czapek-Dox medium supplemented with corn steep liquor. The mycelium was lyophilized, ground in a ball mill, and extracted with chloroform and 0.1 M, pH 7.0, potassium phosphate buffer. The VLPs contained in the aqueous phase were purified by two cycles of differential centrifugation, sucrose density-gradient centrifugation, and sucrose density-gradient electrophoresis at pH 8.5. After sucrose density-gradient centrifugation, the VLPs appeared multicomponent in nature with the fastest component sedimenting at about 110 S. Negatively stained preparations contained isometric particles ca. 34 nm in diam. A typical nucleoprotein ultraviolet absorbance spectrum was obtained. When tested against antiserum to the complex of viruses previously reported in *Penicillium stoloniferum* (ATCC 14586), the VLPs appeared to be related to the slow VLPs (PsV-s) of the *P. stoloniferum* VLP complex.

Phytoalexin production in Ginkgo biloba in relation to inhibition of fungal penetration. T. G. CHRISTENSEN & T. SPROSTON (Univ. Vt., Burlington). Three nonpathogens of *Ginkgo* (*Botrytis allii*, *Monilinia fructicola*, and *Stemphylium sarcinaeforme*) germinated and grew on the surface of living leaves, but did not penetrate the cuticle. Circular zones of small, clear globules were observed in the epidermis beneath many appressoria. Freeze-dried leaves were susceptible to penetration, and no globules were formed, suggesting that leaves must be metabolically active to be resistant. Leaf-surface wax coated on collodion membranes, and enzymatically isolated leaf cuticle, did not inhibit germination, growth, or penetration by the three fungi, suggesting that the cuticle is not a major factor in the resistance of the leaves. A fungal inhibitor was present in the acetone extract of detached, living leaves inoculated with *B.*

allii and incubated 10 days. This inhibitor was absent from detached, noninoculated leaves and from freeze-dried, inoculated leaves, indicating that it is a phytoalexin. In bioassays, the phytoalexin inhibited penetration of collodion membranes by *B. allii* but was inactive against germination and growth. Production of the phytoalexin was inversely correlated with leaf age.

Helical filaments associated with a mycoplasma-like organism in corn stunt-infected plants. R. E. DAVIS, J. F. WORLEY, R. F. WHITCOMB, T. ISHIZIMA, & R. L. STEERE (ARS, USDA, Beltsville, Md.). Helical mycoplasma-like filaments were seen, by phase contrast light microscopy, in juice expressed from leaves, stems, tassels, or roots of corn stunt (CS)-infected corn plants. The filaments, 140 to 200 nm in diam and about 3 to 10 μ in length, were sometimes dichotomously branched and often were attached to spherical bodies 400 to 600 nm in diam. A few filaments could be found about 2 to 4 days before symptoms appeared. Numbers increased with age of infection and, in leaves, were correlated with CS symptom severity. Tetracycline (but not penicillin) delayed development of symptoms and appearance of filaments. Freeze etching and thin-section electron microscopy of phloem and negative staining of juice revealed helical filaments presumed to be the same as those first seen by phase contrast. Each filament is bounded by a single unit membrane, and contains ribosome-like granules and strands presumed to be DNA. No helical filaments could be found in CS-free plants, even when CS-free leafhopper vectors were reared on the plants. Observation of helical filaments by phase contrast microscopy is diagnostic of CS infection in corn, and has been used to detect subclinical infections in inoculated plants that fail to develop symptoms. The evidence indicates that the helical filaments are formed by the presumed mycoplasma-like CS agent in plants.

Heterokaryon transfer of the "killer" factor in Ustilago maydis. P. R. DAY & S. L. ANAGNOSTAKIS (Conn. Agr. Exp. Sta., New Haven). Certain strains of *Ustilago maydis* produce a protein which diffuses through agar medium and which rapidly kills cells of sensitive strains. Killer cells contain an extranuclear factor (I) required for production of the lethal protein, and a second extranuclear factor (S) which appears to protect cells which carry it. Cells which contain neither factor are sensitive unless they carry a recessive nuclear gene for resistance. Although killer and sensitive strains will not mate to form a dikaryon on most media, they will do so on minimal agar medium containing 1% activated charcoal. When such dikaryons were isolated and allowed to dissociate into their haploid parental types, the strain which was formerly sensitive was found to be killer. The extranuclear nature of killer has thus been confirmed by heterokaryon transfer. Our colleagues (Bozarth & Wood, Boyce Thompson Institute) have found that killer cells contain viruslike particles which are not present in sensitive cells.

The water status of leaves of healthy and Fusarium-infected tomato plants. A. E. DIMOND & NEIL C. TURNER (Conn. Agr. Exp. Sta., New Haven). Stomatal resistance of the lower leaf surface was compared in tomato leaves from healthy and *Fusarium*-wilted plants, using a ventilated diffusion porometer. Water potential of leaf tissue and quantity of fluid extracted with increase in pressure were also measured, using a pressure chamber. The latter method potentially discriminates between mechanical vascular plugging and damaged membranes as a cause of wilting. Infection increased the stomatal resistance to transpiration, indicating that stomatal openings were narrower on leaves of

infected plants than in healthy leaves. The water potential in infected leaves did not increase in a consistent pattern as infection progressed. No evidence of membrane damage was found in leaves which were not yet wilting 7 or 9 days after inoculation. At a pressure of 250 psi, as much as 25 to 30% of the total water in a healthy leaf can be extracted from the cut end of the petiole. Characteristically, as symptoms of disease increase, the amount of water extracted from leaves becomes smaller. In heavily wilting infected leaves, so little water can be removed that effects on membranes are totally obscured.

The exposure of eight cultivars of chrysanthemum to peroxyacetyl nitrate. D. B. DRUMMOND & F. A. WOOD (Pa. State Univ., University Park). Several cultivars of chrysanthemum (*Chrysanthemum morifolium*) have been reported to demonstrate peroxyacetyl nitrate (PAN) symptoms in the eastern USA. Eight cultivars of chrysanthemum: Gaiety, Delight, Forty Niner, Golden Gate, Princess Ann, Streamer, Torch, and Tuneful were fumigated with PAN at concentrations ranging from 0.2 to 0.6 nliters/ml for 4 hr. The first five cultivars listed have demonstrated PAN symptoms in the eastern USA. During exposure, temperature was maintained at 24 C, relative humidity was 75%, and light intensity was 3,400 ft-c. More than 225 individuals were fumigated in the series of eight fumigations. Symptoms were not observed following any of the fumigations except for atypical symptoms on one plant fumigated at 0.6 nliters/ml. Pinto bean included in several fumigations exhibited typical PAN symptoms. This suggests that PAN type damage observed on chrysanthemums in the eastern USA may be due to some pollutant other than PAN, or a combination of pollutants which may or may not include PAN.

A selective medium for isolation of Botrytis squamosa. L. A. ELLERBROCK & J. W. LORBEER (Cornell Univ., Ithaca, N.Y.). Martin's rose bengal medium was amended with 20, 7, 2, 150, and 40 ppm, respectively, of pentachloronitrobenzene (PCNB), a 5.2:1 (w/w) mixture of ammoniates of [ethylenebis(dithiocarbamate)]-zinc and ethylenebis(dithiocarbamic acid) bimolecular and trimolecular cyclic anhydrosulfides and disulfides (Polyram), 2,6-dichloro-4-nitroaniline (Botran), streptomycin sulfate, and chlorotetracycline HCl (Aureomycin). This medium is satisfactory for the isolation of *Botrytis squamosa* from both soil (with dilution plates) and air (direct exposure of plates). Plates are incubated in the dark at 15-17 C, and in 4-5 days, *B. squamosa* colonies, which can be recognized by the characteristic dark-red pigmentation they develop on this medium, are readily counted by a viewing through the underside of the plate. Reaction to the medium was similar for all of 10 Orange County, N.Y., isolates. Recovery of *B. squamosa* from natural organic soil artificially infested with conidia ranged from 72-95%, and averaged 80%.

Purification and properties of russet ring virus of apple. NATALIE I. FENG & G. N. AGRIOS (Univ. Mass., Amherst). Russet ring virus (RRV) of apple was mechanically transmitted to the herbaceous hosts *Chenopodium amaranticolor* and *C. quinoa*. Retransmission of RRV from *C. amaranticolor* to *C. amaranticolor* produced local lesions followed by systemic infection. *Chenopodium* plants were most susceptible when inoculated at the four- and six-leaf stage following a 48-hr preinoculation darkness, and when kept at 24 C. The inoculum was prepared by grinding leaf tissue in four parts 0.01 M neutral phosphate buffer containing 0.02 M sodium diethyldithiocarbamate and 0.02 M cysteine-HCl. To this, Mg-bentonite was added at the rate

of 2 ml/5 g tissue. The mixture was centrifuged at 44,000 g for 15 min, and the supernatant was used as inoculum. RRV in crude *Chenopodium* sap has a dilution end point of 1:10³ to 1:10⁴ and a thermal inactivation point of about 50 C. In crude sap kept at 24 C, the virus loses all infectivity in about 6 hr, but is still infective after 2 months' storage in frozen leaf tissue and after 9 months in freeze-dried tissue powder. RRV was purified by differential high-low speed centrifugation and by sucrose density-gradient centrifugation. Electron microscopy of crude leaf sap and of purified preparations of RRV revealed filamentous particles about 920 nm long by 15-20 nm in diam.

Transmission of apple stem-grooving virus from squash to apple seedlings. R. M. GILMER & J. K. UYEMOTO (N.Y. State Agr. Exp. Sta., Geneva). Five to eight stem-grooving virus (SGV) isolates were transmitted mechanically to 8-day-old McIntosh apple seedlings from concentrated infected sap of squash (*Cucurbita maxima*). SGV isolates were initially transmitted from apple to *Chenopodium quinoa*, purified there by single-lesion selection, and multiplied 14-18 days in *C. maxima*. After concentration and partial purification by differential centrifugation, each SGV inoculum induced 100-200 lesions/leaf in cowpea (*Vigna sinensis*). These inocula were rubbed on corundum-dusted leaves and cotyledons of five McIntosh apple seedlings grown at 16-hr days and five other seedlings preconditioned by a 72-hr dark period. The rates of SGV transmissions varied from 0 to 60%, and were commonly higher in predarkened seedlings. No symptoms appeared in infected apple seedlings, but SGV was recovered from them 22-75 days after inoculation by indexing on *C. quinoa*.

Seed deterioration due to aging and to infection by Aspergillus ruber involves membrane damage. A. L. GRANETT & G. E. HARMAN (N.Y. State Agr. Exp. Sta., Geneva). Sterile peas (*Pisum sativum* 'Alaska') were inoculated with *Aspergillus ruber* and stored 14 weeks at 30 C and 21% moisture content (MC). Control seeds were stored similarly (aged control) or at 10 C and 6% MC (low-moisture control). After storage, inoculated peas were completely infected, and had 15% germination. Aged control seeds were sterile and had 80% germination, whereas low moisture control peas germinated 92%. Leachates from inoculated peas had higher conductivity and contained more Mg, K, and P than leachates from aged control peas. Leachates from low-moisture peas had still lower conductivity and contained fewer ions. Light microscopy of unstained hypocotyl tissue revealed dehydrated cytoplasm. When examined by electron microscopy, hypocotyl tissue had shrunken cytoplasm characterized by withdrawn plasmalemma. Greater shrinkage was evident in the tissue from aged control and infected seeds than in that from the low-moisture control seeds. Extreme shrinkage was accompanied by plasmodesmata rupture and plasmalemma breakage. These data suggest that infected seeds had greatest membrane injury with less damage occurring to aged control seeds. Membranes of low moisture control peas were comparatively undamaged.

Ultrastructure of ozonated pollens. B. H. HARRISON, W. A. FEDER, & F. SULLIVAN (Univ. Mass., Boston). Petunia and corn pollen, exposed to 10 to 50 ppm ozone for 3 hr in vivo and in vitro, show a reduction in germination. Chemical analysis showed significant increase in amino nitrogen proportional to the amount of ozonation, accompanied by decreased amounts of peptide-bound reducing sugars, both of which suggest cell wall damage. Ozonated, ungerminated pollen were fixed in glutaraldehyde-osmium tetroxide,

suspended in 1% agar blocks, and prepared for electron microscopy. Sections of the Epon-embedded blocks were stained with uranyl acetate and lead citrate. Upon examination, the exine layer of nonozonated petunia pollen was uniformly dense, with some strata in the nexine. The intine was fibrillar, but largely electron-transparent. Ozonated petunia pollen exhibited a change in the nexine 2 layer; the layer became electron-transparent and appeared completely unstained. Normal corn pollen had an electron-dense and evenly stained exine layer with channels running from inner to outer surface; these channels were filled with electron-opaque material. Ozonated corn pollen showed little or no change in staining characteristics, but the channels appeared empty, as they were electron-transparent. In both petunia and corn pollen plasma, membranes and starch granules appeared normal.

Distinction between sex and compatibility in Ceratocystis ulmi. F. W. HOLMES (Phytopathologisch Lab. "Willie Commelin Scholten", Univ. Mass., Amherst). Of 10 isolates of *Ceratocystis ulmi* collected in September 1970 from trees of *Ulmus hollandica* 'Belgica' afflicted with Dutch elm disease in various Dutch towns, seven were of compatibility type "B" and three were of type "A". Peeled, split, autoclaved elm twigs were dipped for 10 sec into a 3-day-old liquid shake-culture (in Tchernoff medium) of a "B" isolate and then, 3 days later, into a similar culture of an "A", and compared with similar twigs dipped first into "A" and 3 days later into "B". Abundant, fertile perithecia formed in all but seven of the 42 possible combinations: isolate No. 1 (an "A" from Doorwerth, The Netherlands) formed perithecia with each of the seven "B" isolates only when it was the second inoculum. It thus acted as a male, able to serve as effector but not as receptor. These tests were made with 3-month-old isolates. Reisolation from the same branch of the original tree in April 1971, yielded a culture that was again type "A" but was hermaphroditic rather than male. Passage of the male culture through elms yielded typical symptoms of Dutch elm disease but did not change its unisexual nature.

Large-scale aerial color photography - a tool for studying forest tree diseases. D. R. HOUSTON (Northeastern Forest Exp. Sta., Hamden, Conn.). Large-scale aerial color photography was tested as a tool for detecting symptoms, patterns of disease development over time, and site features contributing to disease development. The basal canker disease complex of white pine was used in the study. This disease develops when bark-cankering fungi invade the lower stems through wounds created by snow and ice or by ants. Infrared and true color 70-mm stereo photos taken of eight 0.2-acre diseased plots at scales of 1:792 to 1:1,584 in May and/or August for 5 years were compared to ground records. Visible, but not previsible, foliar symptoms were detected readily at both seasons by both films. Current mortality was shown best by August photography. Dead, bare trees and site features (that contribute to disease development) such as swales, depressions, hedgerows, rock piles, and ant mounds, were seen best on spring photos made before growth of herbaceous vegetation. Patterns of disease development over time associated with these site features could be detected readily with time-sequenced photography.

Degradation of bean cell walls by enzymes produced by Sclerotium rolfsii. T. M. JONES & D. F. BATEMAN (Cornell Univ., Ithaca, N.Y.). *Sclerotium rolfsii* produces a number of polysaccharide-degrading enzymes when grown on autoclaved bean hypocotyls, including a galactanase capable of releasing monomeric galactose from *Lupin* galactan. Isolated cell walls from 4-day-old red kidney bean hypocotyls were treated with a 0.1% solution of a lyophilized extract of a bean hypocotyl

culture of the fungus at pH 4.5; ca. 25% of the cell wall material was liberated as monomeric carbohydrate within 4 hr at 30 C. The major components released were arabinose, galactose, and galacturonic acid. Smaller amounts of rhamnose, xylose, and glucose were also released. Approximately 90% of the galactose present in 4-day-old bean cell walls was released by treatment with *S. rolfii* enzymes. Enzymes from *S. rolfii*-infected bean hypocotyls and enzymes produced by the fungus in culture degraded isolated bean cell walls in a similar manner. Furthermore, cell wall material isolated from the lesion areas of *S. rolfii*-infected bean hypocotyls contained substantially less galactose, arabinose, and galacturonic acid than wall material isolated from comparable healthy hypocotyls.

Factors affecting infection of onion leaves by Pseudomonas cepacia. S. O. KAWAMOTO & J. W. LORBEER (Cornell Univ., Ithaca, N.Y.). Different procedures were used to inoculate onion leaves with *Pseudomonas cepacia*. Symptoms resulted only if the leaves had been wounded. Spray inoculations following a number of different wounding procedures and stab inoculations resulted in small lesions. In the absence of free moisture, these lesions expanded very slowly. When water was supplied to the inoculation site, lesion expansion was rapid. When the developing lesion area was water-soaked, expansion was at a more rapid rate. Stab inoculations at the junction of the leaf blade and the leaf sheath (the leaf blade axil) was most successful in establishing the disease. The leaf blade axil appears to be most suitable as an infection court because water accumulates there. The rate of lesion expansion was directly proportional to the frequency of lesion wetting and to incubation temperature between 10 and 32 C.

Synergistic effect, on hybrid poplar, of sequential exposures to ozone and peroxyacetyl nitrate. L. W. KRESS (Pa. State Univ., University Park). More than five hundred 6-week-old hybrid poplar (*Populus maximowiczii* × *trichocarpa*) cuttings were exposed to 0.1 to 0.15 nliters/ml ozone (O₃) for 2-4 hr, and to 0.2 to 0.4 nliters/ml peroxyacetyl nitrate (PAN) for 4 hr in sequential exposures in a growth chamber maintained at 24 C, 75% relative humidity, 12-hr photoperiod, and 3,300 ft-c. There was a time delay of from 0 to 24 hr between exposures to each pollutant. Approximately half the trees were maintained at pre- and postexposure environmental conditions identical with the exposure conditions, while the remaining trees received pre- and postexposure treatments under greenhouse conditions. Control trees were subjected to the same environmental conditions as the test trees. Symptoms, evaluated 5 to 10 days after exposure, varied from light chlorotic stipple to total necrosis. A synergistic effect was noted when trees which had been maintained in the growth chamber were exposed to O₃ and then PAN the same day, or to PAN and O₃ on successive days. The severity indices of the O₃-exposed trees varied from 34 to 107; and of the PAN exposed trees, from 0 to 29, whereas those of trees exposed to both gases varied from 53 to 155. Exposures of trees maintained in the greenhouse yielded erratic results.

Preference for diseased roots by the clover root borer. K. T. LEATH & R. A. BYERS (U.S. Regional Pasture Res. Lab., University Park, Pa.). The ability of adult borers (*Hylastinus obscurus*), collected from field-grown plants, to select between diseased and healthy red clover roots was determined in walking bioassays. When caged with access to aqueous leachates from diseased and healthy red clover roots, 155 borers selected the leachate from the diseased roots, 18 selected the leachate from the healthy roots, and none

selected the distilled water control. In several tests, nearly all borers placed on pieces of healthy roots walked to and bored into pieces of diseased roots contained within the same test chamber. In reciprocal tests, no borers left pieces of diseased roots to move to healthy roots. Roots infected with *Colletotrichum trifolii* or *Fusarium roseum* were as attractive to the borers as were naturally diseased roots from the field or greenhouse. Leachates from diseased roots stimulated extensive feeding by the borers in nearly all tests; leachates from healthy roots stimulated feeding less frequently and less extensively than did leachates from diseased roots.

Phytohemagglutinins (lectins) and hemolysins in potato plants and their relation to virus infection. K. C. LIU & J. S. BOYLE (Pa. State Univ., University Park). Extracts from potato foliage or tubers agglutinate and may also hemolyze erythrocytes from various sources. Extracts from tubers of USDA 41956, 6-RF-1, Norchip, Wauseon, and 7-GO-28 among 12 test cultivars showed hemagglutinating activity of 1:64 or higher, and were not infectious to *Gomphrena globosa*. Extracts from other cultivars showed lower or no hemagglutinating activity, and were infectious to this plant. No changes in hemagglutinating or hemolytic activity of extracts from plants inoculated with TMV U-1, TMV-Yellow, PVX, or PVX plus TMV U-1 were observed. Extracts from younger leaves were distinctly higher in hemolytic activity than those from older leaves, and an inverse correlation between number of lesions on the indicator plants and hemolytic activity of these extracts was observed. Extracts from second generation plants infected with TMV-Yellow or PVX plus TMV U-1 almost lost their hemolytic activity, whereas those from plants infected with TMV U-1 or PVX alone retained the activity. Heat at 55 C enhanced infectivity of PVX extracts, but did not affect hemolytic activity of the extracts. And relative infectivity remained the same; e.g., fewer lesions from the younger leaves with the higher hemolytic titers, and more lesions from the older leaves with the lower hemolytic titers.

In vitro development of dodine tolerance in Venturia inaequalis. B. H. MAC NEILL & JANICE SCHOOLEY (Univ. Guelph, Guelph, Ont., Can.). When large populations of conidia of *Venturia inaequalis* were exposed to dodine in potato-sucrose agar at pH 8, a number of survivors were found which tolerated ca. 3 × the normally lethal dosage of the fungicide. These survivors were evaluated for their ability to maintain dodine tolerance after several successive transfers of mycelial discs to a medium lacking the selective agent. Of the 34 survivors tested in this way, seven exhibited a mutational change to permanent dodine tolerance without any other apparent change either in morphology or physiology. Tolerance also frequently developed when mycelial discs instead of conidia were cultured on dodine medium. In this latter instance, although the level of tolerance was equal to that exhibited by mutants recovered after exposure of conidia to the selective agent, this attribute was lost upon a single subculture to a nonselective medium. Adaptive tolerance in the colonies from mycelial discs, although reversible, appears to be no less effective than mutation as a mechanism of survival when the fungus is exposed to the selective pressure of dodine.

A histological study of Austrian pine needles injured by ambient air pollutants. J. M. MAIELLO, E. G. BRENNAN, & IDA A. LEONE (Rutgers Univ., New Brunswick, N.J.). A needle blight induced by air pollutants has been common among New Jersey Austrian pines. Blighted needles are characterized by orange-brown necrosis restricted to needle tips and progressing into adjacent tissues, resembling

semimature-tissue blight of white pine. Trees vary in susceptibility, but only physiologically active needles of the current year are damaged. A study of successive cross sections beginning near the tips of freshly blighted needles shows complete disorganization of both mesophyll and stele cells at the tip. Adjacent sections indicate injury to mesophyll cells with necrotic regions adjoining the substomatal chambers. Endodermis as well as transfusion cells of the stele are distorted. Sections below this point reveal less generalized damage which is restricted to the transfusion tissue. Frequently, mesophyll cells in these sections exhibit no signs of disorganization and appear healthy, contrary to results of white pine studies where comparable cross sections indicate damage confined to the mesophyll. Sections ca. 0.5 mm below the base of the blight show decreasing transfusion tissue injury and no injury to the mesophyll. Greenhouse fumigations with SO₂ and formaldehyde have produced injuries similar to those occurring in nature.

Respiration and organic acid content of virus-infected apple fruits. J. S. MAKARSKI & G. N. AGRIOS (Univ. Mass., Amherst). The respiration and organic acid content of apple fruits infected with either apple russet ring virus (RRV), which causes both foliar and fruit symptoms, or with apple mosaic virus (AMV), which induces only foliar symptoms, were determined. Fruits were collected at monthly intervals during two growing seasons from healthy and AMV or RRV-infected McIntosh trees. No differences in respiration were detected between the healthy and the diseased fruits. The effect of virus infection on the respiratory climacteric was determined by measuring the CO₂ evolution from whole mature fruits with an infrared CO₂ analyzer. Virus infection does not hasten or delay the onset of the climacteric, although in one season's data, there was an apparent virus-induced increase in the height of the climacteric. The organic acid content of freeze-dried samples prepared from healthy and virus-infected fruits was determined by ion-exchange column chromatography. Virus infection did not result in the appearance of any detectable new acids; the content of malic acid, one of the two major acids present, was slightly decreased by virus infection; however, there was no apparent change in quinic acid concentration in the infected fruits.

Influence of long-term low levels of ozone on the leaf surface mycoflora of Pinto bean plants. W. J. MANNING & P. M. PAPIA (Univ. Mass., Waltham). Pinto bean plants (*Phaseolus vulgaris* strain 111) were grown for 4, 7, 14, 21, or 28 days in greenhouses containing ambient air, charcoal-filtered air, or ozone at 6 pphm for 8 hr/day. Ozone was produced with a Welsbach generator and monitored with Mast meters. The upper and lower surfaces of the first set of simple true leaves were used to make leaf prints on acidified potato-dextrose agar plates (PDA) at each sampling period. Discs cut from these leaves were washed 10 times in sterile water and plated on PDA. Results with leaf prints showed that species of 25 genera of fungi were present in recognizable successions on all leaves. The number of fungi per cm² leaf tissue increased with leaf age for all leaves, with the greatest number occurring on 28-day-old leaves with accumulated ozone flecks. Differences between leaves by sources was more quantitative than qualitative, with the exception of *Aspergillus niger*, which was common only on the leaves of plants grown in ambient air. *Botrytis cinerea* was commonly found on plates printed with leaves that had ozone fleck. Isolates of *Candida*, *Cryptococcus*, and *Penicillium* were the most abundant fungi on all leaves. Similar results were obtained with plated washed leaf discs,

except that the number of fungal genera present was reduced from 25 to 11.

Influence of long-term low levels of ozone and benomyl on growth and nodulation of Pinto bean plants. W. J. MANNING, W. A. FEDER, & P. M. PAPIA (Univ. Mass., Waltham). Pinto bean plants (*Phaseolus vulgaris* strain 111) were grown for 20, 40, or 60 days in nonsterile sandy loam in greenhouses containing ambient air, charcoal-filtered air, or ozone at 6 pphm for 8 hr/day. Ozone was produced with a Welsbach generator and monitored with Mast meters. Benomyl treatments, consisting of a 0.5% seed soak, 0.5 and 2% seed slurries, and 25-, 50-, 75-, and 100-ppm soil amendments, were evaluated for protectant action against chronic ozone injury to Pinto bean plants. All control plants exposed to ozone showed reduction in the number, size, and weight of Rhizobium nodules, fresh and dry weights of roots and tops, plant height, number of leaves per plant, number and length of pods, and number, size, and weight of seeds when compared to control plants grown in filtered and ambient air. Growth and nodulation of all benomyl-treated plants were either similar to or lower in value than those achieved with control plants from any of the three greenhouses. Control plants grown in ambient air had the greatest top weights and pod and seed yield. Temporary protection against chronic ozone injury was obtained with benomyl soil amendments at 50 ppm and above. This was accompanied by varying degrees of leaf chlorosis and burning, plant stunting, and yield reduction.

The etiology of witches'-broom of Opuntia. K. MARAMOROSCH, M. KLEIN, & B. S. WOLANSKI (Boyce Thompson Inst., Yonkers, N.Y.). For the past decade, it has been known that *Opuntia tuna* could be transformed into *O. tuna monstrosa* by the grafting of scions from the ornamental onto the normal cactus. The graft transmissible agent was considered a virus, although direct evidence for viral etiology was lacking. When ultrathin sections of *O. tuna monstrosa* were examined by electron microscopy, mycoplasma-like bodies as well as rod-shaped viruslike particles were observed in phloem elements. To elucidate the etiology of the witches'-broom disease, cuttings from diseased plants were immersed for 3 hr in a 100-ppm solution of tetracycline HCl, potted in soil, supplied water-soluble nitrogen fertilizer, and maintained for 3 weeks at 35 C. New branches that developed afterwards in the greenhouse resembled normal *O. tuna*. Within 18 months, 75% of all treated plants reverted to the *monstrosa* type. Electron micrographs of cured branches revealed that mycoplasma-like bodies were absent, whereas rod-shaped particles were still present. Branches that reverted to the *monstrosa* type contained mycoplasma-like bodies as well as viruslike particles. This indicated that the witches'-broom disease of *O. tuna* was caused by the mycoplasma-like agent, and not by a sole or mycoplasma-combined virus infection.

Relative susceptibility of four species of Helminthosporium to fungicides. P. M. MILLER (Conn. Agr. Exp. Sta., New Haven). The susceptibility of *Helminthosporium maydis*, *H. turcicum*, *H. triseptatum*, and *H. vagans* to nine fungicides was tested in the laboratory on potato-dextrose agar in petri dishes. Control of *H. maydis* and *H. turcicum* was also tested on corn. Growth of all four species was inhibited at 10 ppm of maneb, thiram, ferbam, and ziram. *Helminthosporium triseptatum* was the most resistant of the four species to thiram, captan, daconil, carboxin, and Terrazole [5-ethoxy-3-(trichloromethyl)-1,2,4-thiadiazole]. *Helminthosporium maydis* was more resistant to carboxin, zineb, ziram, maneb, and daconil than *H. turcicum* or *H. vagans*. *Helminthosporium turcicum*

and *H. vagans* were equally susceptible to most fungicides, but *H. vagans* was more resistant to Terrazole than was *H. turcicum*. In greenhouse tests, Dyrene [2,4-dichloro-6-(*o*-chloroanilino-*s*-triazine)], maneb, and zineb gave the best control of *H. maydis* and *H. turcicum*. Carboxin, thiram, and ferbam washed easily from the leaves and gave poor control. Captan gave good control of *H. maydis* and *H. turcicum* on corn, but caused yellowing of the leaves under moist conditions.

The effect of aerated steam treatment of mushroom casing on recolonization by Verticillium malthousei. R. K. MOORE & P. J. WUEST (Pa. State Univ., University Park). Phialospore germination on buried water agar slides measured the capacity of *Verticillium malthousei* to initiate colonization on soil treated at various temperatures with aerated steam. Phialospores of 7-day-old *V. malthousei*, C-PW-707 isolates, were used in all experiments. Sterile slides coated with a 1.5% agar-spore suspension were placed in contact with soil previously treated at 60, 82, or 98 C for 30 min. Similar slides were placed in moist chambers to ascertain spore germination. Slides were incubated for 24 or 72 hr at 27 C in the dark. Slides were removed, fixed with lactophenol, and per cent germination was calculated. Phialospore germination decreased with decreasing treatment temperatures. The pathogenic potential of *V. malthousei* on *Agaricus bisporus* PSU 310 seeded into casing treated at 60 and 98 C was studied. The crop was grown at 17-19 C and 95% relative humidity in 10-inch plastic bulb pots. Soil was treated 3 days prior to casing. Phialospores were atomized 3 days before; 2 days after; or 12 days after casing. Mushrooms were harvested from three replicate pots for 13 days. On soil seeded before casing and treated at 98 C, sporocarp production was reduced by 90%. Sporocarp yields from other treatments were not affected, and disease incidence was very low.

Endopolygalacturonase: evidence for involvement in Verticillium wilt of cotton. H. MUSSELL. (Boyce Thompson Inst., Yonkers, N.Y.). Isolates of *Verticillium albo-atrum* from cotton produced two polygalacturonases, an exopolygalacturonase (exoPG), and an endopolygalacturonase (endoPG) when cultured on a glucose-salts medium. When assayed on cotton cuttings or leaves, the exoPG was not toxic, but the endoPG caused leaf chlorosis, necrosis, and desiccation identical to that seen in *Verticillium*-infected cotton plants. This endoPG was also toxic to potato tuber and cucumber pericarp cells in a standard bioassay. Symptom expression evoked by endoPG was greatly enhanced by the presence of 5 mM magnesium, although this ion had no effect on the enzymatic activity of endoPG in vitro. After pretreatment with the respective substrates, application of either glucose oxidase or amino acid oxidase to cotton leaves caused symptom appearance similar to that observed in *Verticillium*-infected cotton. The above results, coupled with the fact that a catalase pretreatment greatly reduced symptoms generated by the endoPG, suggest that one of the mechanisms of tissue damage operative in *Verticillium*-infected cotton leaves may involve the intramural generation of hydrogen peroxide.

How a slight decrease in susceptibility or in virulence can account for a great decrease in disease. R. R. NELSON (Pa. State Univ., University Park). Epidemiological research to monitor disease increase historically evaluates the impact of time and climatic parameters on various phases of the infection cycle. Such an approach can be misleading when it is assumed that different cultivars of a susceptible complex or different isolates of a virulent race will act as constants and respond similarly to various climatic regimes. Studies with

two isolates of race T of *Helminthosporium maydis* and two male-sterile, susceptible corn hybrids illustrate the point. After 72 hr, average lesion size on hybrid SX29 was 3.3 and 6.0 mm on plants exposed to 6 hr dew at 22 C followed by colonization at 25 and 28 C, respectively. Average lesion size on the less susceptible sterile hybrid Pioneer 3369A was 3.4 mm when plants were exposed to the same dew period and temperature and the same colonization period at 28 C. After 72 hr, average lesion size on SX29 was 6.1 and 4.5 mm when two different isolates were used and plants were exposed to 6 hr dew at 20 and colonization at 31 C. Decreases in disease by alteration of the host and the pathogen are comparable to those incurred by using less favorable dew and temperature regimes, and thus have comparable effects on epidemic development.

RNA polymerase activity in crown-gall tissues. R. M. NILES & M. S. MOUNT (Univ. Mass., Amherst). Groups of *Vicia faba* seedlings were either inoculated in the first internode with *Agrobacterium tumefaciens*, wounded with a sterile needle, or left untreated. After 21 days, chromatin from these internodes was extracted and purified. The DNA-dependent RNA polymerase associated with chromatin from crown-gall tissue produced 63% more RNA than did the same enzyme from wounded healthy tissue. An estimation of template availability was determined by saturating chromatin with *Escherichia coli* RNA polymerase. Crown-gall chromatin had 49-54% more available substrate than the controls. The results indicate that tumor chromatin has more genetic sites available for transcription, and it is hypothesized that it may also have more than one type of chromatin-bound RNA polymerase.

The adenine nucleotide status of ozonated Pinto bean foliage in relation to symptom development. EVA J. PELL & EILEEN BRENNAN (Rutgers Univ., New Brunswick, N.J.). Two-week-old Pinto bean plants were exposed to 0.25-0.30 ppm ozone for 3 hr. Primary leaves of control and treated tissue were harvested, weighed, and frozen immediately after ozonation when visible symptoms were not yet apparent. Adenosine triphosphate (ATP) content was analyzed by the luciferin-luciferase method. Adenosine diphosphate (ADP) and adenosine monophosphate (AMP) were converted to ATP with creatine phosphotransferase, and total adenine nucleotides were then analyzed by the luciferin-luciferase technique. Ozonated tissue had significantly higher concentrations of ATP and total adenylates than did control tissue. These increases do not appear to be the result of uncoupled phosphorylation, but rather of increased ATP synthesis. In order to determine whether this phenomenon persisted as the symptom developed, two kinetic studies were conducted. Tissue was harvested 0, 6, 21, and 72 hr after ozonation, and then analyzed for ATP and total adenylates. The symptom began to develop within 21 hr, becoming prominent in all samples within 72 hr. The increase in total adenylates in ozonated tissue persisted through the 21-hr period. ATP content of ozonated tissue was significantly higher than that of controls at the 0-, 6-, and 72-hr periods but not at 21 hr.

Electron microscopy of the xylem of ratoon stunted sugarcane. BILJANA PLAVSIC-BANJAC & K. MARAMOROSCH (Boyce Thompson Inst., Yonkers, N.Y.). Since no viruses, bacteria, or fungi have been isolated from ratoon-stunted sugarcane, the disease has usually been listed as one of unknown etiology. Young and old ratoon-stunted plants from plantations in Puerto Rico were obtained for electron-microscopic examination. Orange-reddish-colored portions of internodes were excised at Rio Piedras, P.R., and immediately immersed in cacodylate buffered 3%

glutaraldehyde. Further processing for electron microscopy was carried out at Yonkers, N.Y. Electron micrographs revealed the presence of pleomorphic bodies in the xylem, but not in the phloem, of old diseased canes. Young ratoon plants and healthy controls contained no pleomorphic bodies and appeared normal. The bodies resembled very small bacteria or mycoplasma-like bodies. Their association with ratoon-stunted sugarcane suggests their possible etiologic role in this disease.

Effects of some divalent cations on the macerating activity of two pectic lyases produced by Pseudomonas fluorescens. A. L. PRATT & G. A. MCINTYRE (Univ. Maine, Orono). Effects of several divalent cations on the macerating activity of two partially purified pectic lyases isolated from *Pseudomonas fluorescens* were studied, using potato discs 400 μ thick as a substrate. Rate of tissue maceration was inhibited at pH 8.5 by 0.001 M and 0.0001 M Ba^{+2} , Zn^{+2} , and Ca^{+2} . Macerating activity was reduced to a lesser degree by Mg^{+2} , Fe^{+2} , Ni^{+2} , Mn^{+2} , and Cu^{+2} at both 0.001-M and 0.0001-M concentrations. Studies of viscosity reduction using sodium polypectate (NaPP) as an artificial substrate at pH 8.5 showed that Ca^{+2} enhanced viscosity reduction. Results suggest that divalent cations inhibit tissue maceration, yet Ca^{+2} stimulates viscosity reduction of NaPP.

Effect of constant and fluctuating temperatures on in vitro growth of Ceratocystis species. P. E. REYNOLDS, W. H. SMITH, & K. F. JENSEN (Yale Univ., New Haven; U.S. Forest Service, Delaware, Ohio). The effect of constant and fluctuating temperature regimes on in vitro growth of various *Ceratocystis* species, including *C. adiposa*, *C. pluriannulata*, *C. minor*, *C. coerulea*, *C. pilifera*, and 14 isolates of *C. ulmi* was investigated. A constant temperature of 25 C seemed optimal for growth of most of the *Ceratocystis* members tested. There was considerable variation, however, in response to the various constant and fluctuating temperature regimes. Generally, fluctuating temperature reduced growth. Occasionally, a minor fluctuation would suppress growth, whereas subsequent larger fluctuations would enhance it. This phenomenon was termed "initial-negative" stimulation. No correlation between geographic source of an isolate or species and its reaction to temperature regime was observed.

Inhibition of foliar respiration in ozone-exposed Pinto bean plants. C. P. RIPALDI, EILEEN BRENNAN, & IDA A. LEONE (Rutgers Univ., New Brunswick, N.J.). A frequent and early symptom of ozone toxicity in Pinto bean (*Phaseolus vulgaris* 'Pinto') following fumigation (0.30-0.35 ppm O_3 /3-4 hr) is water-soaking of the foliage. After 24 hr, characteristic necrotic stipple or bleaching develops. We found a significant inhibition of respiration concurrent with the water-soaking (control 2.62 and O_3 1.89 $\mu\text{liters O}_2$ /mg dry wt per hr). This contrasts with the increased respiration rate that accompanies visible injury. Macdowall (1964) also reported an inhibition of respiration prior to the occurrence of necrosis, and subsequent stimulation with necrosis. Vacuum infiltration of healthy bean foliage with water also decreased respiration, but the inhibition was not of the magnitude found in ozonated tissue; nor was the external symptom the same under both conditions. Replacing ambient air with 100% O_2 in the Gilson respirometer flasks increased the rate of the ozonated bean foliage to that of the controls (control, 2.31 and O_3 , 1.64 $\mu\text{liters/mg dry wt per hr}$; control + 100% O_2 , 2.72 and O_3 + 100% O_2 , 2.40 $\mu\text{liters O}_2$ /mg dry wt per hr). These data support the hypothesis that O_2 diffusion was limited in the early stages of ozone damage.

An unexpected outbreak of lethal yellowing in coconut palms on Key Largo, Florida. D. A. ROBERTS, J. W. MILLER, C. P. SEYMOUR, J. H. KNOWLES, C. F. DOWLING, & W. H. PIERCE (Univ. Fla., Gainesville). The range of lethal yellowing of coconut palms in the United States has previously been restricted to Key West, Fla., and the adjacent Stock Island. Between September 1969 and April 1971, however, 13 coconut palms died from lethal yellowing in a planting of some 125 trees in Key Largo, ca. 100 miles northeast of Key West. Diseased trees showed symptoms typical of lethal yellowing, and the pathogen was transmitted from a diseased palm to 2 of 23 mechanically inoculated young coconut palms. The source and method of dissemination of the lethal yellowing pathogen to Key Largo are not known. The disease has not occurred in the Key West area since March 1968, and to suggest that trees there were sources of inoculum would presume an inordinately long latent period of 18 months. We believe the pathogen was carried from a Caribbean island to Key Largo by wind-borne arthropod vectors. The recent outbreak of lethal yellowing on Key Largo re-emphasizes the threat of this devastating disease to the thousands of ornamental coconut palms on the nearby mainland of the Florida "Gold Coast".

Relative abilities of pathogens and nonpathogens of alfalfa to induce production of and degrade medicarpin. T. SAKUMA & R. L. MILLAR (Cornell Univ., Ithaca, N.Y.). Three pathogens (*Stemphylium botryosum*, *S. sarcinaeforme*, and *Ascochyta imperfecta*) and eight nonpathogens were tested by the drop-diffusate method for their ability to induce production of medicarpin by excised alfalfa leaves. The ability of the same fungi to degrade medicarpin was determined in Czapek's medium. Medicarpin was assayed by spectrophotometric, chromatographic, and biological procedures. Except when live spores of *S. botryosum* and *A. imperfecta* were used, medicarpin was detected in tests with each of the fungi regardless of whether the material tested on leaves was a suspension of live spores, the supernatant from an autoclaved spore suspension, or the filtrate of a suspension of spores germinated in vitro. No medicarpin was detected in response to suspensions of live spores of *S. botryosum* and *A. imperfecta*, presumably because these fungi degraded medicarpin too rapidly. The three pathogens rapidly degraded medicarpin in vitro in essentially the same manner; the nonpathogens, except *Fusarium solani* f. *phaseoli*, either did not degrade medicarpin or degraded it to compounds unlike those determined for the pathogens. Data for *F. solani* f. *phaseoli*, though different in some details, reflected those obtained for the pathogens.

Effect of benomyl on the growth of several plant species. L. R. SCHREIBER & W. K. HOCK (USDA, ARS, Delaware, Ohio). Benomyl was mixed with air-dried soil (133 ppm, w/w) 10 months or immediately prior to planting seeds of corn, pea, bean, pepper, tomato, marigold, American elms, sycamore, or buckthorn. Untreated soil served as a check. After the plants were harvested, their height growth, and fresh and dry root and shoot weights were measured. The presence of a fungitoxicant was determined by placing stem and leaf tissue onto agar seeded with *Ceratocystis ulmi* conidia. The growth characteristics of corn, pea, and tomato were reduced little or not at all, whereas those of marigold and the woody plants were reduced over 80% in most cases. Growth responses and the bioassays from the two benomyl treatments indicated similar levels of chemical activity. These results show a substantial chemical residue in soil treated 10 months prior to seeding. The growth response to benomyl is related to other factors in addition to its concentration in the tissues, since the levels in corn and pea, equal to those in woody plants, did not reduce growth.

Evaluation of chloroneb for control of Philippine downy mildew of corn (Sclerospora philippinensis). O. E. SCHULTZ (Cornell Univ., Ithaca). Control of Philippine downy mildew by protectant fungicides is ineffective due to susceptibility of young seedlings, rapid expansion of leaves, and frequent heavy rain. Slurry and dip treatments of seed with chloroneb 65% WP (5 g/kg and 8 g/liter for 4 hr) reduced disease incidence for at least 12 days after planting, after which symptoms readily appeared. Preplant soil incorporation of 7.5% granules (100 kg/hectare) as well as in-furrow spraying and post-plant surface drenching with 65% WP (1.5 kg in 300 liters/hectare and 27.5 kg/54,000 liters/hectare) extended control to 2½-3 weeks after planting. Additional treatments included foliar applications combined with seed and soil treatments and overnight plant covering after emergence. The most effective combination was covering plants for 2-3 nights after emergence, spraying immediately after removal of covers with 65% WP (4.8 g/liter) and 2-3 additional applications during the next 12 days. Plants treated thus remained symptomless for ca. 4 weeks after emergence (period of extreme susceptibility), whereas all untreated plants were affected 18 days after emergence. This combination yielded a 7- to 11-fold increase in marketable ears. Placement of chloroneb-coated granules or seeds (rice or mung bean) into whorls was not as effective as were foliar sprays.

Relationship of cropping history and soil fumigation to the development of ectomycorrhizae in a new Douglas fir nursery. W. A. SINCLAIR (Cornell Univ., Ithaca, N.Y.). Quantitative aspects of development of ectomycorrhizae on Douglas fir (*Pseudotsuga menziesii*) seedlings in fumigated (450 liters/hectare of MIT, a mixture of 20% methylisothiocyanate and chlorinated C₃ hydrocarbons) and unfumigated soil in a 4-year-old nursery were studied. The soil had been used previously for production of corn and beans. Mean dry weights of 1-year-old seedlings in fumigated soil in Douglas fir monoculture for 4 years and 1 year were 0.43 and 0.18 g, respectively. The mean frequencies of tomentose ectomycorrhizae on roots of these trees were 58 and 21% dry weight of roots, respectively. Hartig nets were detected in 98 and 66% of sectioned roots of 1-year-old trees from fumigated soil in the 4-year-old and 1-year-old portions of the nursery, respectively. In unfumigated soil, 98 and 33% of the roots from the 4-year-old and 1-year-old areas, respectively, had Hartig nets. The data indicate that soil fumigation enhances ectomycorrhizal development, and that the second and subsequent Douglas fir seedling crops on a new nursery site grow and develop ectomycorrhizae more rapidly than the first crop.

Method for studying transcuticular movement of fungicides. Z. SOLEL & L. V. EDGINGTON (Univ. of Guelph, Guelph, Ont., Can.). The systemicity of fungicides is dependent upon transcuticular movement and subsequent translocation within the plant. A method for studying cuticular penetration was developed using cuticle isolated from apple leaves by either the pectinase or zinc chloride-HCl technique. Discs of cuticle were supported by thin aluminum foil rings and placed in contact with potato-dextrose agar seeded with spores of *Penicillium*. Equimolar amounts of test fungicides were then applied on the cuticle, and 24 hr later, the zones of inhibition of growth were measured and compared with known standards. The relative transcuticular movement of three benzimidazole fungicides was in the order of benomyl > methyl-2-benzimidazolecarbamate > Thiabendazole. The lower cuticle was more readily penetrated than was the upper cuticle. The method is relatively simple, and amenable to the study of spray adjuvants, droplet size, and other factors upon transcuticular movement. It

simulates the air:spray droplet:cuticle interfaces encountered in agricultural practices.

Serological and biological properties of two strains of potato virus X. J. C. STUDENROTH, R. C. MC CRUM, & F. E. MANZER (Univ. Maine, Orono). A mild strain of potato virus X (PVX) failed to produce symptoms on *Datura stramonium*, whereas a common strain caused distinct mottling. The common strain consistently exhibited higher serological and infectivity titers in crude sap extracts and purified preparations from *Nicotiana glutinosa* as revealed by microprecipitin tests and the local lesion host *Gomphrena globosa*. Preliminary evidence with Ouchterlony microslides and cross-absorbed antisera indicates that the two strains are serologically identical. Electron microscopy studies of ultrathin sections of infected *N. glutinosa* leaf tissues indicate that the mild strain of PVX occurs in fewer cells than the common strain, but both strains appear to behave identically in those cells where they are found. It is suggested that the differences in biological behavior between these two strains is not necessarily reflected in an antigenic disparity, and the role of cistrons other than those governing coat protein formation are considered as a possible explanation for the observed strain differences.

Prevention of Botrytis and Penicillium rots and scald of apples in storage with postharvest dip treatments. M. SZKOLNIK (N.Y. State Agr. Exp. Sta., Geneva). McIntosh apples were artificially wounded after harvest, dip-treated in fungicide suspensions for 30 sec, inoculated with *Botrytis* sp. or *Penicillium* sp. spores, and stored at 34 F in November. Rot control data taken in February revealed that Mertect 360W (Thiabendazole) at 12 oz/100 gal water, benomyl at 8 oz, and Pennwalt TD 1771 70W [1,2-bis(3-methoxycarbonyl-2-thioureido) benzene] at 1 lb. gave excellent protection against *Penicillium* rot. Mertect and TD 1771 gave good control of *Botrytis* rot, whereas benomyl gave fair-to-good control. Captan 50W at 2 lb., Difolatan 4 Flowable at 1 qt, Thynon 75W (dithianon) at 8 oz, Polyram 80W at 2 lb. and Eli Lilly EL-273 25W (triarimol) at 10.7 oz were not effective against these rots. In a similar dip test with Cortland apples, Mertect, benomyl, and TD 1771 gave excellent control of *Penicillium* rot, whereas captan was ineffective. The scald-preventing products "No Scald" [diphenylamine (DPA) 83% WP] at 2 lb. and "Stop Scald" (1,2-dihydro-6-ethoxy-2,2,4-trimethylquinoline, 70% liquid) at 3 pt. used as a dip treatment alone or with Mertect, benomyl, or captan effectively prevented fruit scald and did not change fungicidal effectiveness in rot control.

Benomyl controls Botrytis stem rot of tobacco seedlings. G. S. TAYLOR (Conn. Agr. Exp. Sta., Windsor). Stem rot (*Botrytis* sp.) can increase rapidly on susceptible strains of cigar tobacco when growers water seedbeds heavily to pull transplants. The dithiocarbamate sprays used against blue mold (*Peronospora tabacina*) give little or no control. In seedbeds at Windsor, in May and June 1969 and 1971, seedlings of susceptible tobacco (Connecticut Broadleaf) were sprayed weekly with benomyl and various dithiocarbamates. After the last spray (a) and 4 weeks later (b) naturally occurring stem rot was rated (1 = no disease, to 5 = stem girdled) on 10 random plants in each of three or four replications. In 1969, the ratings for benomyl at 0.75 lb. actual/100 gal at a and b times were 1.1 and 1.2, respectively, a significant difference from 2.0 and 3.5, respectively, for the average of seven dithiocarbamates. Levels of sugar in the stem sap did not differ significantly. In 1971, benomyl at 1 lb. actual/100 gal gave complete control (1.0) at both rating dates, either alone or in combination

with ferbam, maneb, metiram, or zineb. Without benomyl, average ratings were 1.5 and 2.5, respectively. There were no adverse effects upon plant growth. The long residual action suggests that benomyl would be an effective periodic addition to regular seedbed sprays for Connecticut tobacco.

The effect of carbon and nitrogen sources on polygalacturonate trans-eliminase production by Fusarium oxysporum f. vasinfectum. M. TIEN & R. CAPPELLINI (Rutgers Univ., New Brunswick, N.J.). The effect of 20 different carbon and nitrogen sources on growth and polygalacturonate trans-eliminase (PGTE) production of *F. oxysporum f. vasinfectum* was studied. When ammonium nitrate was used as the nitrogen source, xylose, glucose, galactose, mannitol, sucrose, and pectin were the most favorable carbon sources for growth. Of all the carbon sources tested, only pectin could induce the production of PGTE. When pectin was used as the carbon source, the organism utilized the different nitrogen sources under test almost equally well in respect to growth. However, the various nitrogen sources differed considerably in their influence on the amount and the time of maximum PGTE production. Urea, asparagine, and glutamine were the best nitrogen sources for PGTE production. When ammonium sulfate, glutamic acid, and lysine were used as substrates, maximum PGTE production occurred several days after the maximum growth of the fungus was attained. On the contrary, with glutamine and serine, maximum enzyme production preceded the period of maximum growth. In all other cases, PGTE was at its peak during the maximum growth period of this fungus.

Water balance and membrane damage studies with the pressure chamber technique. N. C. TURNER & A. E. DIMOND. (The Conn. Agr. Exp. Station, New Haven). The pressure chamber technique is widely used for studying the water balance of plant tissue. A detached leaf is quickly sealed into the chamber with the cut surface of the petiole or leaf protruding. Pressure from a cylinder of nitrogen gas is applied to the leaf until the meniscus of the xylem sap returns to the cut surface; this balancing pressure approximates the leaf water potential. Collection of the increments of fluid expressed by the step-wise application of further pressure gives the osmotic potential of the tissue, and can detect membrane breakdown. With this technique, we observed the destruction of osmotic integrity in susceptible but not in resistant oat leaves that had absorbed toxin of *Helminthosporium victoriae* (1.2 units/ml) for 3 hr. The toxin of *Fusicoccum amygdali* (10^{-5} M), sprayed onto peach leaves, accelerated dehydration but did not destroy the osmotic integrity of the cells. The technique could not detect the membrane damage known to occur in corn leaves injured by ozone or in leaves showing lesions of *Helminthosporium maydis*, probably because some of the cells in the test leaves were uninjured. Thus, the pressure chamber technique can detect membrane damage when most of the cells in a diseased leaf have been affected.

Isolation of tobacco necrosis virus from leaves or petals of apple, pear, and apricot. J. K. UYEMOTO & R. M. GILMER (N.Y. State Agr. Exp. Sta., Geneva). Isolation of tobacco necrosis virus (TNV) from crude petal or leaf sap of infected apple, pear, or apricot was erratic; such inoculum only occasionally incited 1-2 lesions/*Chenopodium quinoa* indicator. When apple leaf sap from two infected trees was concentrated and partially purified in two trials, these inocula incited an average of 38, 39, and 20 0.5 lesions/C. *quinoa* leaf, respectively. Several TNV isolates from apple and one from apricot incited mixed white (nonnecrotic) and red (necrotic) lesions on cowpea (*Vigna sinensis*). Selections

for white lesion type were maintained in cowpea at 28 C, but reverted to red types at 15 C. White lesion isolates were antigenically alike, and the one apple isolate tested activated satellite virus, type B, selectively. A second apple isolate that induced only red lesions differed antigenically from white lesion isolates and activated satellite virus, type C, selectively.

Ultrastructure of the feeding apparatus of Criconemoides curvatum. G.-Y. WEN & T. A. CHEN (Rutgers Univ., New Brunswick, N.J.). Electron microscopy of the feeding apparatus of *Criconemoides curvatum* shows that its stylet consists of tooth, shaft, and knobs. The tooth appears to be more cuticularized than the other parts, and forms the partial outer covering and the inner lining of the stylet. It can be separated from the shaft by treating an isolated stylet with 10% sodium hypochlorite. The stylet lumen opens subterminally on the ventral side of the tooth. The shaft begins at the basal knobs and extends toward the tip of the stylet. Six ducts within the shaft are connected with the cytoplasm outside the stylet, and extend from the base to the tip of the shaft. The telorhabdion consists of three knobs which are attached to three muscles extending anteriorly to the lip region. The stylet guiding apparatus is characterized by the presence of 12-15 longitudinal foldings along its inner wall. The esophageal glands are composed of numerous honeycomblike compartments. The dorsal esophageal gland cell is heterogenous in appearance, with tracheal-like canals connecting to its orifice, while the two subventral esophageal glands appear to be homogenous, with similar canals connected to the tri-radiate lumen at the base of the median bulb valve.

Evidence for ethylene injury in fusaric acid treated tomatoes. D. M. WILSON, B. ETHERTON, & R. JAGELS (Univ. Vt., Burlington). Absorption of calcium fusaric acid solutions (10^{-3} to 10^{-6} M, pH 7.0) by tomato cuttings (Bonny Best) was followed by leaf epinasty. To test for possible ethylene involvement in the fusaric acid response, etiolated pea seedlings were placed with treated cuttings, and the triple response of the peas was used as a bioassay for ethylene production. Fusaric acid-treated tomatoes were able to cause the triple response with an initial treatment of 10^{-3} , 10^{-4} , 10^{-5} , or 10^{-6} M. Tomatoes treated with picolinic acid did not show leaf epinasty or cause the triple response in peas. Picloram (4-amino-3,5,6-trichloropicolinic acid) caused tomato epinasty and caused the triple response in peas at concentrations from 10^{-3} to 10^{-10} M picloram. Picloram is known to stimulate ethylene production and to possess auxin activity. The effects of all three compounds on oat coleoptile elongation were compared. Picloram stimulated elongation with 10^{-3} to 10^{-7} M, while picolinic acid only slightly stimulated elongation at 10^{-4} to 10^{-6} M. Fusaric acid inhibited extension growth from 10^{-3} to 10^{-6} M. These results suggest that fusaric acid may stimulate ethylene production in tomatoes.

The response of 11 hybrid poplar clones to ozone. F. A. WOOD & J. B. COPPOLINO (Pa. State Univ., University Park). Individuals of 11 hybrid poplar clones were exposed to 0.25 nliters/ml O_3 for 4 hr at 24 C, 75% relative humidity, and 3,300 ft-c of light. Plants were maintained out of doors prior to and after the fumigations, and exposures were made at 3, 4, 6, 8, 10, 12, and 14 weeks after bud break. An interveinal, bifacial, dark brown-to-black necrosis was the most common symptom observed; brown-to-black pigmented stipples of the upper leaf surface were observed infrequently. The 11 clones could be classified into three groups on the basis of sensitivity. The least sensitive group yielded severity indices (SI) of 21 to 25; the moderately sensitive group, SI's from 48 to 62; and the most sensitive group, SI's of 88 to

135. The clones were most sensitive at 4 weeks after bud break; sensitivity declined rapidly at 8 weeks, increased during the 10th week, and declined thereafter. The relative sensitivity of clones also varied with time after bud break, and the middle to older age leaves were most sensitive.

A comparison of ribosomes from bean rust and an axenic wheat rust. Z. YANIV & R. C. STAPLES (Boyce Thompson Inst., Yonkers, N.Y.). Ribosomes from uredospores of the as-yet-uncultured bean rust fungus (*Uromyces phaseoli*) and the saprophytic Australian wheat rust fungus (*Puccinia graminis* f. sp. *tritici*, race 126-ANZ-6,7) were compared for transferase activity, capacity to bind polyuridylic acid, and capacity to incorporate leucine into ribosomes in vivo. During initiation of germination, ribosomes from the germ tubes of bean rust spores were at their peak of responsiveness to polyuridylic acid, and polyribosomes appeared. After 20 hr of germination, when elongation of the germ tube had nearly ceased, the responsiveness of the ribosomes to polyuridylic acid declined to one-tenth their original activity. Polyribosomes were still abundant. In contrast, ribosomes from uredospores of the Australian wheat rust fungus, which produce growing hyphae after several days on suitable media, remain fully active during germination.

Effect of source and test plants, feeding time, and aphid numbers on transmission of tobacco etch virus. M. YILMAZ & E. H. VARNEY (Rutgers Univ., New Brunswick, N.J.). Aphids, *Myzus persicae*, were cultured on *Capsicum annuum* 'California Wonder' and starved 80-100 min prior to acquisition feedings on source plants infected with tobacco etch virus (TEV). Aphids were equally effective as vectors after 30- and 60-sec acquisition feedings, but became less effective after 120 sec. Transmission was influenced by the combination of source and test plants selected. Aphids fed on infected California Wonder pepper transmitted to test plants (*Nicotiana tabacum* 'Samsun' and 'Samsun NN', California Wonder pepper, and *C. frutescens* 'Tabasco') with an efficiency of 20-37% as compared to 23-57% for aphids fed on infected Samsun NN tobacco. When Samsun NN was used as the source plant, aphids transmitted the virus to 17 out of 30 Samsun as compared to 11 out of 30 Samsun NN seedlings. A similar relationship was found in additional tests. Tabasco pepper was more susceptible than California Wonder to aphid inoculations. As expected, *Chenopodium quinoa*, a local-lesion host, was a poor source of virus. A single Samsun tobacco was infected in one experiment. One aphid was usually as efficient as two to five aphids in transmissions from good source plants.

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