Abstracts of Papers Accepted for Presentation at the Sixty-Third Annual Meeting of The American Phytopathological Society, Philadelphia, Pennsylvania, 15-19 August 1971

Several aspects of the ecology and pathology of Fusarium oxysporum f. sp. cepae in organic soils. G. S. Abawi (Cornell Univ., Ithaca, N. Y.). A direct correlation was found between inoculum density of Fusarium oxysporum f. sp. cepae and damping-off of onion seedlings growing in organic soil artificially infested with the fungus under controlled environmental conditions (26 C-day and 21 Cnight; 16 hr fluorescent light/day at 2,000 ft-c; 60-70% relative humidity). A population of 5 × 10⁴ or more propagules/g oven-dry soil was needed before any significant development of disease could be detected in field soil; 100 propagules caused extensive disease development in sterilized soil. Seedling damping-off increased from 10 to 32 C. Conidia added to field soil exhibited a low percentage of germination, and germ tubes formed were either lysed or converted to chlamydospores. Population of the fungus decreased in the absence of onion and increased in its presence. Roots of Oxalis corniculata were heavily infected by the fungus when the plant was grown in artificially infested organic soils.

Some effects of pH and nitrogen source on growth of Physalospora obtusa in culture. J. J. Albert & C. R. Drake (Va. Polytech. Inst., Blacksburg). Physalospora obtusa was cultured in liquid synthetic media containing KNO3, NH4NO3, (NH4)2SO4, and DL-aspartic acid as nitrogen sources, and sucrose as the carbon source, at initial pH values of 3.5, 5.5, 7.5, and 9.0. The medium also contained KH₂PO₄, 1.0 g; MgSO₄ · 7 H₂O, 0.5 g; Fe(NO₃)₃ · 9 H₂O, 0.2 mg; $MnSO_4 \cdot 4$ H₂O, 0.1 mg; thiamine, 0.1 mg; and biotin, 0.005 mg. Standing cultures were incubated at 28 C and harvested at 4-day intervals for 24 days, and the mycelium was dried and weighed. At 16 days, dry weights in mg at was thed and weighed. At 10 days, dry weights in fing at initial pH values of 3.5, 5.5, 7.5, and 9.0 were as follows, respectively: KNO₃, 207, 238, 232, and 249; NH₄NO₃, 63, 82, 157, and 225; (NH₄)₂SO₄, 30, 51, 108, and 193; aspartic acid, 133, 153, 151, and 152. There was some evidence that pH of the medium affected growth, independent of the nitrogen source. Nitrate nitrogen statistically supported better growth than organic (aspartic acid) or ammonium nitrogen at each harvest date. Analysis was determined using Duncan's multiple range test for significance at the 5% level. The optimum initial pH value for growth was 9.0 for all nitrogen sources except aspartic acid, where growth was similar at initial values of pH 5.5, 7.5, and 9.0. Growth was extremely sparse when nitrogen was omitted from the medium.

Pathogenicity of Cylindrocladium theae and C. sco-Pathogenicity of Cytinaroctaanim theae and C. sco-parium to roots and leaves of azalea. S. A. Alfieri, Jr., R. G. Linderman, R. H. Morrison, & E. K. Sobers (Fla. Dep. Agr., Gainesville, ARS, USDA, Beltsville, Md., Univ. Minn., St. Paul, Univ. Ga., Coastal Plain Sta., Tifton). Cylindrocladium theae (Calonectria theae) was isolated from roots, stems, and leaves of several cultivars of greenhouse azaleas. Its pathogenicity was compared with C. scoparium to roots and leaves of three greenhouse and three hardy cultivars of azalea. No mortality occurred among plants grown for 2 months in soil artificially infested with comparable amounts of blended cultures of each species. However, significant root discoloration was noted in two of six cultivars grown in soil infested with C. theae, and the fungus was reisolated from all but one cultivar. Two of six cultivars grown in soil infested with C. scoparium showed reduced and discolored root systems; three cultivars exhibited discolored roots only; and one cultivar had apparently healthy roots from which the fungus could not be isolated. The leaves of all cultivars showed some degree of susceptibility to both species. Symptoms varied from a few irregularly circular, purplishblack lesions, 1-2 mm in diam, to those that covered a large portion of the leaf surface, with slight-to-heavy defoliation.

In general, C. scoparium was more virulent to leaves and roots than was C. theae.

Nuclear phenomena preceding basidiospore formation by monobasidiospore isolates of Lenzites trabea. T. L. AM-BURGEY (USDA Forest Serv., Gulfport, Miss.). Some monobasidiospore isolates of Lenzites trabea have the capacity to form fruiting bodies and viable basidiospores in culture. This study was initiated to examine this phenomenon cy-tologically. Most of the probasidia produced by monobasidiospore isolates do not form sterigmata (in the usual sense) and spores, but continue to grow by either apical or lateral proliferations. In mature basidia produced by monobasidiospore isolates, the nuclear cycle is the same as that in dikaryotic isolates. A fusion nucleus is formed which then divides to form haploid nuclei. These then migrate through the sterigmata and into the developing spores. The nuclei usually divide mitotically as the spores mature. In basidia produced by dikaryotic isolates, two nuclei infrequently enter a sterigma and presumably both enter the developing spore. If the two nuclei entering a sterigma contained the same sexual incompatibility factor (i.e., A_1 and A_1), the spore subsequently containing them, although heterokaryotic, would give rise to simple-septate mycelium (assuming that dissimilar sexual incompatibility factors are necessary for clamp-connection formation). Isolates derived from such spores may be the monobasidiospore isolates which have the capacity to fruit in culture.

Ultrastructural determination of the infection process of Macrophomina phaseoli in Glycine max. V. D. Ammon, T. D. Wylle, & M. F. Brown (Univ. Mo., Columbia). Surface-sterilized seed of the soybean cultivars Amsoy and Adelphia were grown in steam-sterilized vermiculite for 2-42 days. In addition, seed from the cultivars Amsoy and Calland were germinated in petri dish cultures of Macrophomina phaseoli. The root systems of the plants grown in pots were exposed to the charcoal rot fungus by injecting into the rhizosphere 50 ml of an actively growing shake culture. After incubating for 1-10 days, selected roots were fixed and embedded in paraffin and plastic in preparation for light and electron-microscopic examination. Roots of all cultivars of soybean seedlings tested became infected within ca. 3 days as determined by direct observation. Fungal invasion of root tissue occurred intercellularly and intracellularly. Penetration of soybean root tissue was accompanied by changes in the organization of the cytoplasm, disruption of membranes, and breakdown of cell walls. The disruption of cellular and tissue integrity is believed to occur by the production of pectolytic enzymes in association with mechanical pressure.

Heterokaryon formation in Thanatephorus cucumeris ("Praticola" type). N. A. Anderson, H. M. Stretton, & N. T. Flentje (Univ. Minn., St. Paul, Waite Inst., Glen Osmond, S. Australia). Field isolates of Thanatephorus cucumeris ("Praticola" type) produce single basidiospore progeny which segregate as if a single factor (H factor) controlled heterokaryon formation. When monokaryons of different H factors are paired, a tuft of hyphae forms where the colonies join. Thiamin and nitrate auxotrophs were paired with wild-type monokaryons of different H factors. Hyphal tip colonies established from the tuft mycelium produced basidiospores containing the parental factors, indicating that the tuft hyphae were heterokaryotic. Five single-spore isolates which failed to form tufts with either parental H factor were found upon sporulation to contain both H factors. Intrafactor recombination producing nonparental H factors occurred at a rate of 1.8% (sample 420) from a USA isolate. H factors were obtained

from 13 field isolates from Australia, Britain, Canada, and the USA. Heterokaryons were synthesized between monokaryons from the four countries.

Races of Fusarium oxysporum f. sp. pisi. G. M. Armstrong & Joanne K. Armstrong (Ga. Exp. Sta., Experiment). During the past 5 years, 25 cultivars (CVS) of Pisum sativum were inoculated with 19 isolates of f. sp. pisi from five countries, but not all CVS were inoculated with all isolates. Three CVS were uniformily susceptible; the others were resistant or susceptible to different isolates. There were four groups of isolates based on pre-vious identifications by the donors; 8 of race 1, 9 of race 2, one of race 4, and one of race 5. Differences in pathogenicity of the isolates showed that races 1 and 2 are not well defined. Six of eight isolates in the race 1 group are retained in race 1 due to similarities in pathogenicity on six CVS. Two isolates differed from these and from each other and are recognized as two new races. Four of the nine isolates in the race 2 group, as well as the race 4 isolate, are similar on seven CVS and are considered as race 2. Two somewhat divergent isolates were retained with these in race 2. Of the three remaining isolates of the race 2 group, one is the same as the recently described race 5. The other two constitute a new race. Thus, six races are recognized in the collection. Races 3 and 3A were unobtainable.

Efficacy of triarimol (EL-273) against Venturia inaequalis and Podosphaera leucotricha. W. R. ARNOLD, D. H. LADE, & C. D. CHRISTENSEN (Eli Lilly and Co., Newark, Del., Plainwell, Mich., & Cazenovia, N.Y.). Triarimol [α-(2,4-dichlorophenyl)-α-phenyl-5-pyrimidinemethanol] was evaluated for the control of apple scab and powdery mildew on Rome Beauty, McIntosh, Red Delicious, and Jonathan apple trees in experiments conducted in Pennsylvania, New York, and Michigan. Triarimol was applied every 7, 10, or 14 days through petal-fall, followed by a 14-day schedule for the remainder of the season with high-pressure hand guns and commercial speed sprayers. Triarimol was also applied on a 7-day schedule for the first three applications, followed by a 14-day schedule for the remainder of the season. Application rates ranged from 30-80 ppm. In areas with less than 10% scab incidence on fruit of control trees, 40 ppm applied on the 14-day schedule for the entire season provided good-toexcellent powdery mildew control, and excellent foliar and fruit scab control. In all experiments, good-to-excellent mildew control and excellent scab control were provided by 40 ppm applied on the 7- and 10-day spray schedules through petal-fall, followed by applications every 14 days. Similarly, excellent control of these diseases was obtained by applying 40 ppm on the 7-day schedule for the first three applications, followed by applications made every 14 days. Fruit quality was not adversely affected by triarimol.

Aggressiveness in Puccinia recondita tritici. M. ASLAM & L. E. Browder (ARS, USDA, Kans. State Univ., Manhattan). The relationship of aggressiveness to pathogenicity in Puccinia recondita was studied in three cultures: 66-763 (virulent at 8 out of 12 loci studied), UN01-68A (virulent only at the Plr10 locus), and UN01-68B (avirulent at all loci studied). Survival in mixtures, relative infectivity, and uredospore production were used as criteria for aggressiveness. Three composites of two cultures each were grown through several generations on Bison wheat, which has no known genes for resistance. Frequency of each culture in each generation was determined by assaying on a differential set of near-isogenic lines. UN01-68B in composite with UN01-68A and with 66-763 increased from 43% and 54% to 70% and 94%, respectively, after 6 generations on Bison; UN01-68A in mixture with 66-763 increased from 54% to 90%. A high positive correlation was found between number of generations on Bison and the frequency of the cultures with less virulence. Predomi-

nance among cultures was unaffected by temperature or photoperiod during growth. UN01-68B produced significantly more uredospores per pustule than either of the other cultures. The cultures also differed in infectivity on Bison, with the most avirulent culture being significantly more infective.

The effect of oxycarboxin on the bean rust disease caused by Uromyces phaseoli, J. J. Bates & B. G. Tweedy (Univ. Mo., Columbia). Healthy and diseased plants and the fungus, Uromyces phaseoli, were treated with various concentrations of oxycarboxin. Germination and respiration studies of uredospores were carried out using 10, 50, and 100 µg/ml of oxycarboxin with samples taken at hourly intervals. 14C-glucose was used to study the effect of oxycarboxin on the diseased and healthy plant. Radioautography was used to study the movement of the fungicide in the diseased plant. Results indicated that the fungicide delays spore germination but does not inhibit it. Germ tube extension and respiration were greatly inhibited. Oxycarboxin had no effect upon growth or respiration rate of the bean plant at concentrations which inhibited disease development. Translocation from the roots to the leaves was found to occur in 1 hr. The concentration of labeled material increased rapidly around the site of infection. After an incubation period of 72 hr, the concentration in the pustule was ca. 50 times that found in the noninfected parts of the plant.

Proteins from cultured cells of Xanthi-nc tobacco inoculated with tobacco mosaic virus. R. N. BEACHY & H. H. MURAKISHI (Mich. State Univ., E. Lansing). Cell suspensions of Nicotiana tabacum 'Xanthi-nc' tissue culture were inoculated with 100 µg tobacco mosaic virus (TMV)/ml, washed with fresh medium to remove excess inoculum, and incubated on agar medium under diffuse light. Local lesions were observed on inoculated cell cultures after 40 hr. At 24-hr intervals, inoculated and noninoculated control cells were harvested, and cell extracts were prepared by homogenization and centrifugation at low (15,000 g) and high (144,000 g) speeds. The supernatant from the highspeed centrifugation was subjected to electrophoresis on 7 and 10% polyacrylamide disc gels, stained for protein in amido black, and scanned in a Gilford densitometer. On a fresh wt basis, more proteins were extracted from lesionbearing inoculated tissue than from control tissues. One new protein band, characterized as TMV coat protein, was detected from extracts of infected cells. Intensities of several normal host protein bands diminished, whereas others increased following infection. Data indicate that protein changes can be detected in virus-infected cells from tissue cultures more easily than in virus-infected leaves.

Effects of fluometuron and prometryne on Rhizoctonia solani in soil. H. WAYNE BEAM & E. A. CURL (Auburn Univ., Auburn, Ala.). Technical grade fluometuron and prometryne [2,4-bis(isopropylamino)-6-methylmercapto-striazine] were tested in flasks with previously autoclaved sandy loam soil for effect on growth response of Rhizoctonia solani. The herbicides were applied in a nutrient solution to provide concentrations ranging from 1 to 40 µg/g of soil. The cultures were incubated at 27 C, and analyses were made at intervals of 2, 3, 4, and 6 days to determine soil-enzyme activity and nutrient uptake. In herbicide-free soil, there was a general increase in β-galactosidase activity during the first 3 days after inoculation with the pathogen; activity changed little thereafter. Enzyme activity for the 40-µg fluometuron treatment increased rapidly, but was far less at all sampling times than for the control or other treatments. Enzyme activity was higher in the 1-µg fluometuron treatment than in the control. Values for other treatments (5 and 10 µg) were intermediate. In prometryne-treated soil, the 40-ug treatment reduced β-galactosidase activity during the period between 2 and 4 days, whereas treatments of 1, 5, and 10 µg enhanced activity. Little change in soil pH occurred in any

treatments, but ion uptake was suppressed by the higher concentrations of both herbicides.

Production of cellulase, phosphatidase, polygalacturonase, and pectate lyase by prototrophic avirulent and partially revertant mutants of Erwinia carotovora. L. Beraha, B. A. BILLETER, & E. D. GARBER (ARS, USDA, Univ. Chicago, Chicago, Ill.). Avirulent and partially virulent mutants of Erwinia carotovora were produced after 5- to 12-min exposure of a 105/ml shaken suspension of 24-hr-old cells in 250 mg/1 N-methyl-N'-nitro-N-nitrosoguanidine in PO4 buffer at pH 7.0. The enzyme profiles of five prototrophic, independently obtained, avirulent mutants (AV) were compared with those of four partially revertant strains (PRV), the fully virulent revertant (RV), and wild-type (WT) strains. In supplemented growth media containing the inducers polygalacturonic acid, carboxymethycellulose, purified soybean lecithin, or lima bean extract, all AV strains produced low levels of cellulase, variable amounts of polygalacturonase and pectate lyase, and no phosphatidase. When compared to AV, those PRV strains producing the most cellulase and polygalacturonase were intermediate in virulence. Like the AV strains, all PRV strains tested failed to produce phosphatidase. By comparison, the virulent WT and RV strains obtained from the AV had consistently higher unit and RV strains obtained from the AV had consistently higher unit and RV strains obtained from the AV had consistently higher the large and the strains of the s sistently higher unit values of polygalacturonase, pectate lyase, cellulase, and phosphatidase. The manipulation of enzyme production and the resulting effects on virulence implicate these enzymes directly in the production of soft

Numbers of trapped Cercospora apii spores determined by length of temperature, favorable moisture periods, and disease incidence, R. D. Berger (Univ. Fla., Belle Glade). Spore trap monitoring in large Florida celery fields for over 1,250 "trap-days" revealed that numbers of trapped Cercospora apii spores were greatly reduced when the following conditions were not met: 12 hr/day of humidities near 100% above 15 C (blight favorable hours = BFH). The numbers of spores trapped when the BFH exceeded 12 hr/day were directly associated with the amount of disease observed in the field; the highest numbers (400-700 spores/day) occurred when disease expression was maximal. Low disease incidence, small leaf areas (young plants), morning cloudiness, and high daytime humidities were the most important factors giving low spore numbers (0-100 spores/day) when seemingly blight favorable conditions existed. Except for those occasions when mechanical operations (harvesting, tillivating, weeding, etc.) detached spores, significant spore numbers (over 50 spores/day) did not occur when weather unfavorable for blight prevailed. The daily number of trapped spores provided a valid estimate of daily "blight pressure", and to some extent estimated the amount of disease in the field. The spore numbers also provided the growers with a basis to establish satisfactory spray schedules and a means to determine the effectiveness of particular spray programs.

Nutrition and metabolism of the haustorial mycoparasite Tieghemiomyces parasiticus in axenic culture. F. L. BINDER & H. L. BARNETT (W. Va. Univ., Morgantown). Tieghemiomyces parasiticus, a haustorial mycoparasite, makes no growth on hexoses, pentoses, or trichloroacetic acid intermediates as carbon sources. Excellent axenic growth occurs in liquid media with glycerol. Cell-free extracts of glycerol-grown cultures contain all enzymes of the Embden-Meyerhof and hexose monophosphate pathways. In the presence of Tween 80, glucose is used as a carbon source. The parasite grows readily only when the level of casein hydrolysate is high (10-40 g/liter). Fractionation of casein hydrolysate showed that the parasite had an absolute requirement for cysteine or methionine, and is highly dependent upon valine and leucine for axenic growth. Removal of the aromatic amino acids, those derived from the aspartate family, and histidine did not affect axenic growth. The parasite requires thiamine and

biotin for growth. The inability of certain haustorial parasites to grow in axenic culture may be the result of multiple deficiencies of specific amino acids and vitamins, as well as problems associated with the uptake of specific nutrients.

Hosts and pathogenesis of a new Heterodera sp. on grasses. W. Birchfield (USDA, ARS, La. State Univ., Baton Rouge). A new species of Heterodera induced pathological symptoms on grasses different from those previously described for other members of this genus. Rice, Johnson grass, and barnyard grass were found to be hosts of this nematode, but no other grasses tested, including sugarcane, rye grass, winter wheat, nor any of several dicotyledonous plants, were parasitized. Larvae penetrated the epidermis and traveled intercellularly to the vascular cylinder, where they positioned themselves with the head toward the main root turned from the root tips. They penetrated the pericycle and fed in the phloem between the xylem vessels. No giant cells were formed as reported for other species of Heterodera, but enlargement of phloem near the mouthpart was evident. Cell walls near the feeding site were thickened, as evidenced by heavier stain absorption. Galling of roots and top symptoms were not observed on host plants. Vulvular secretions were not evident in egg laying. Swollen females erupted the cortex and epidermis and deposited a few eggs outside, but most were retained. Males did not penetrate to the vascular area, but remained in the cortex near the epidermis.

Residue monitoring: a system for timing spray schedules. H. Blazquez (Univ. Fla., Immokalee). A system for timing spray schedules was developed by relating the decay rate of fungicide leaf residues with their ability to inhibit spore germination in a bioassay test. Foliar residues of Difolatan 4F (cis-N-1,1,2,2-tetrachloroethyl thio-4-cyclohexene-1,2-dicarboximide) sprayed at the rate of 3 pints/100 gal of water were washed with benzene from tomato leaves beginning 3 hr after spraying and every 24 hr thereafter until complete breakdown occurred. Difolatan residue analysis was done by thin-layer chromatography. Neely's bioassay test was used to determine the per cent germination of Alternaria solani spores with daily concentrations of fungicide residue. Difolatan residues varied from 400 to 0 ppm at 3 hr and 7 days, respectively. Spore germination in the bioassay test increased 6 to 100% from 3 to 7 days after spraying. Concentrations above 100 ppm were fungicidal, while those above 75 ppm were fungistatic. It was possible to estimate the number of days of effective protection by knowing the Difolatan decay rate and foliar residue. These results suggest that this analytical bioassay method may be used to accurately determine the proper timing of fungicidal sprays for disease control.

Winter wheat as a reservoir for maize dwarf mosaic virus. C. W. BOOTHROYD & C. P. ROMAINE (Cornell Univ., Ithaca, N.Y.). Winter wheat (Yorkstar) with mosaic was found near a corn field (Pa 290) in which a few plants showed symptoms of maize dwarf mosaic (MDM). Corn seedlings (NY3 × D50) mechanically inoculated with leaf juice of diseased corn and of diseased wheat from this field showed mosaic symptoms. The isolates from corn and from wheat were transferred mechanically to corn (NY3 × D50) and to sorghum (Atlas), but not to wheat (Yorkstar). Each of the two isolates was identified serologically as MDMV-A. When aphids (Rhopalosiphum maidis) were placed on corn leaves mechanically inoculated with the wheat isolate, and subsequently on healthy corn and wheat seedlings, severe mosaic developed in the corn but only a mild mottle in the wheat. Mechanical inoculation of corn with leaf samples from these corn and wheat leaves, however, resulted in an expression of pronounced mosaic symptoms. In another test, Macrosiphum avenae, Myzus persicae, R. maidis, R. padi, and Schizaphis graminum were placed on corn leaves infected with the wheat isolate, then on corn and wheat seedlings. In 3

weeks, no symptoms developed on wheat, but mosaic symptoms appeared in corn exposed to all aphid species. The most severe leaf mosaic symptoms occurred in seedlings exposed to *Schizaphis graminum*, the least severe in those exposed to *M. avenae*.

The experimental infection of Solanum tuberosum by tomato ringspot virus. J. S. BOYLE (Pa. State Univ., University Park). Ten 6-week-old greenhouse-grown potato seedlings were mechanically inoculated with an isolate of tomato ringspot virus (TomRSV). Initial symptoms of infection, characterized by inconspicuous and sparse chlorotic and necrotic spotting, were observed on rubbed leaflets of three plants 6 days after inoculation. The virus was later recovered from terminal tissue from two of the plants, indicating that systemic infection had occurred. Tubers from these plants were saved and stored at ca. 40 F until they sprouted. Three tuber generations now have been produced from these plants with several unexpected results. The symptoms of chronic TomRSV infection in potato are strikingly different from any previously encountered syndrome in this plant. Many tubers formed on TomRSV-infected plants do not sprout, thus eliminating the virus from future generations. Plants that develop from tubers are characterized by a striking chlorosis restricted to the tips and margins of leaflets. As the plants grow, symptoms fade from the old leaves but continue to develop in the young expanding leaflets. Frequently, plants will continue to grow indeterminantly with large numbers of aerial tubers.

Purification and properties of the viruslike particles of Penicillium chrysogenum. R. F. Bozarth & H. A. Wood (Boyce Thompson Inst., Yonkers, N.Y.). The viruslike particles (VLP's) of Penicillium chrysogenum (ATCC No. 9480) were purified by extraction with chloroform and 0.1 m, pH 7.0, potassium phosphate buffer, differential centrifugation, sucrose density-gradient centrifugation, and sucrose density-gradient electrophoresis. In addition to the major component which sediments at 150 S, there were additional sedimenting components at 81, 101, and 212 S. All components had spherical VLP's 40 mµ in diameter and ultraviolet spectra typical of nucleoproteins. Nucleic acid extracts of purified VLP's made by the single-phase phenol-SDS procedure and assayed by polyacrylamide gel electrophoresis contained three classes of double-stranded RNA with molecular wt of 2.18, 1.99, and 1.89 × 106 daltons. Antiserum to the VLP's of P. chrysogenum reacted specifically with its homologous antigen and also with the VLP's of P. brevi-compactum. On the basis of serology and similar morphology, the VLP's of P. chrysogenum are considered to be related to those of P. brevi-compactum.

Screening red clover introductions for resistance to Stemphylium sarcinaeforme. S. W. Braverman USDA, N.Y. State Agr. Exp. Sta., Geneva). At the Northeast Regional Plant Introduction Station, Geneva, N.Y., 550 red clover (Trifolium pratense) introductions and 23 standard cultivars were grown in the greenhouse and evaluated for resistance to Stemphylium sarcinaeforme. Seedlings were inoculated using a mixture of two cultures isolated from highly susceptible red clover accessions, increased on potato-dextrose agar, comminuted in a blender, and subsequently applied under 70 psi on 3- to 4-inchtall 5-week-old seedlings. Inoculated plants were incubated under 100% RH at 20-24 C for 48 hr, then removed to a greenhouse bench. Plants were rated for amount and prevalence of infection 10 days after inoculation. Plant Introductions 235845, 235851, and 235858 were rated as highly resistant, 50 were rated as moderately resistant, while 64 varied from slightly resistant to slightly susceptible. Wisconsin Mildew Resistant, N1-17-1-64 (FC 39493), Common Indiana Medium (FC 39379), and Medium Red (FC 40232) were slightly resistant. The remaining 20 standard cultivars were moderately to highly susceptible to the pathogen.

The use of the computer in analysis of Puccinia recondita pathogenicity data. L. E. Browder (ARS, USDA, Kans. State Univ., Manhattan). The basic model of the genefor-gene relationship permits four experimental designs; one of these is to hold host-genotype for resistance and environment constant and study parasite pathogenicity. This design, used on a parasite sample, can be used to study pathogenicity of parasite populations in time and space. The infection-type data obtained from such experiments usually are so extensive that only the simplest analyses can be made by manual methods. The capacity of the computer was used to make detailed analyses of survey data of Puccinia recondita on Triticum aestivum. A parasite-culture X host-line infection-type matrix was put into core-storage along with appropriate indices of information about host and parasite entities used. Specific arrays of data could be associated with specific host-lines or parasite-cultures by subscripting techniques. The arrays could be compared in any combination. Programs have been developed to produce race summaries, to tabulate virulence frequencies to each host line used, to tabulate associated pathogenicities to two or to four host lines in all combinations, to summarize pathogenicity according to similarity to "type" cultures, and to sort data on collections by collectors' names.

Studies on the activity of triarimol (EL-273) against certain powdery mildew fungi. I. F. Brown, Jr. & H. R. HALL (Eli Lilly & Co., Greenfield, Ind.). Greenhouse studies have demonstrated the curative activity of triarimol against Erysiphe graminis f. sp. tritici and E. polygoni. When conidia of E. polygoni were placed on bean leaves sprayed with 1, 10, or 50 ppm of triarimol, no difference in germination or germ tube development was observed. Bean or wheat plants show no macroscopic signs or symptoms of disease when sprayed with 5-10 ppm. Triarimol at 10 ppm was applied to foliage of bean and wheat plants 0-4 days before or after inoculation. Regardless of the time of application prior to inoculation, or for 1 hr thereafter, the conidia of both fungi germinate and haustoria develop. However, no additional fungal growth occurs after the formation of haustoria in treated plants. However, application of triarimol 24, 48, 72, or 96 hr after inoculation arrests further development of these fungi. No additional haustoria develop from superficial mycelium. When triarimol is applied to soil at 0.5 mg/500 g soil, no macroscopic symptoms of disease can be observed on wheat or bean plants. Microscopic examination of treated plants shows that haustoria form, but no further fungal development occurs.

Germination of Fomes igniarius var. populinus basidiospores on aspen pith. T. S. Brown, Jr., & W. Merrill (Pa. State Univ., Univ. Park). Controversy exists as to whether Fomes igniarius var. populinus (F) penetrates through branch stubs. Some workers have reported that germination of F occurred only after 7 days' incubation in fresh wounds, and that F occurred in branch stubs only as a lateral outgrowth of an internal decay column. Following our observations that sometimes the pith of branch stubs appeared decayed whereas the remaining wood appeared only discolored, we placed spore suspensions of F onto microtome sections freshly cut from several Populus tremuloides branch stubs. Percentage germination (based on 200 spores/replicate section) reached 24% in 72 hr on pith and adjacent annual rings. Greatest germination appeared to be directly on the pith itself, but the small size of the pith made it impossible to accurately separate germination on the pith from that on the adjacent annual ring. Germination was never greater than 2.5% on any other material, and usually was 0.0% on the surfaces of the stub and fresh sapwood. Germination usually was greatest on pith cut from the distal end of the branch stub, and least on pith cut from close to the zone of discoloration usually present in the bases of the stubs. This suggests that the pith of branch stubs may be an avenue of entrance for F. igniarius var. populinus.

Influence of gamma radiation on vessel development and spore movement in American elm. R. CAMPANA (Univ. Maine, Orono). Conidia of Ceratocystis ulmi were inoculated in early June 1970 into 80 elm stems, 40 growing under chronic gamma radiation, and 40 growing in a nonirradiated control field. Irradiated elms had been exposed to 20 r/day for 2 years. Inocula at dosages of 1×10^6 or 8×10^6 spores/ml were introduced by knife incision through suspension droplets into xylem. After 24 hr, each inoculated stem was severed 30 cm below inoculation point, and cut into 6-mm-long sections, which were incubated aseptically on water agar in plates at room temperature. Presence or absence of the fungus was noted in the following 2 weeks. Cross sections of stems were studied histologically for vessel development. Most new vessels in irradiated stems were either lacking or immature. New vessels in all nonirradiated trees were mature. Distance of spore movement was significantly greater (0.01) in non-irradiated elms. Radiation delayed development of vessels and prevented vertical passage of spores. The data supplement other evidence of resistance in irradiated elms.

Accumulation of phenols and phytoalexins in hypocotyls of bean infected with Fusarium solani f. phaseoli. C. O. N. CARDOSO & M. O. GARRAWAY (Ohio State Univ., Columbus). Hypocotyls of Phaseolus vulgaris inoculated with F. solani f. phaseoli (FSP) and of healthy plants were extracted with ethanol at various intervals. The phenol concentration (Folin-Denis; 4-methylcatechol equivalents) of extracts from diseased hypocotyls increased with time to a maximum of 1.63 mg/plant 15 days after inoculation. The phenol concentration of a petroleum ether fraction (PEF) of the extracts reached 57.5 µg/plant at 12 days and remained constant thereafter. Healthy hypocotyls contained no detectable PEF phenols. Growth of F. solani f. cucurbitae (FSC), but not of FSP, was inhibited on PDA containing 5.8 µg/ml PEF phenols. Similarly, spore germination and germ tube elongation of FSC, but not of FSP, were inhibited in water agar containing PEF. Separation of PEF on silica-gel plates with pentane-etheracetic acid revealed that two of the phenols produced red colors with FeCls. The one at $R_{\rm F}$ 0.35 differed from phaseollin by having a UV abs max only at 280 nm. The other, with $R_{\rm F}$ 0.65, has a UV spectrum similar to phaseollin (max at 315 and 280, 315:280 ratio of 0.237). Both phenols selectively inhibited the nonpathogenic FSC. Data suggest that at least two phenolic antimicrobial substances, probably phytoalexins, accumulate in FSP-infected bean hypocotyls.

Environmental contamination by airborne fluorides in Montana. C. E. CARLSON & J. E. DEWEY (U.S. Forest Serv., Missoula, Ma.). Gaseous and particulate fluoride effluents from an aluminum reduction plant in western Montana have caused considerable environmental damage over a large geographical area. Herbaceous plants, shrubs, and trees showed foliar burn correlated with excessive fluoride accumulations. Conifer needles showed definite histological responses to high fluoride levels, including protoplasmic and nuclear hypertrophy of phloem parenchyma. High levels of fluoride were found in cambial feeding insects, indicating that fluorides may be translocated within the vascular system of conifers. The occurrence of elevated fluorides in insects indicates accumulation through the food chain of the ecosystem. Foliar necrosis due to fluorides was found in Glacier National Park, representing an unwanted intrusion by technology of man into one of the few remaining truly pristine habitats of the world.

Induced resistance in alfalfa to Corynebacterium insidiosum by prior treatment with avirulent cells. R. B. CARROLL & F. L. LUKEZIC (Pa. State Univ., Univ. Park). Resistance to virulent isolates of Corynebacterium insidio-

sum was induced in four susceptible alfalfa cultivars by leaf infiltration with dilute suspensions of avirulent mutants from the same original single-celled culture. Challenge tests showed that protection occurred as early as 12 hr and was complete by 24 hr. Challenge tests on companion leaflets indicated that the protection factor(s) is not translocated or light-dependent. Protection also occurred when roots of seedlings grown under sterile conditions were pre-inoculated with avirulent bacteria. Tests on small root sections of greenhouse and gnotobiotically grown alfalfa, maintained in a moist chamber, gave the same results as those obtained with infiltrated leaves. Protection in leaves and in root sections was also induced by avirulent cells that had been Formalin-treated, sonicated, or sonicated and then filtered through a 0.45-µ Millipore membrane. Protection could not be elicited by cell-free filtrates of nonsonicated avirulent cells or by virulent cells treated in the same manner as the avirulent. Protection was not induced by infective or noninfective isolates of Rhizobium meliloti, or by other isolates of unidentified bacteria isolated from alfalfa roots.

Evaluation of granular formulations of systemic nematicides for the control of Pratylenchus penetrans on potatoes. R. C. Cetas (Cornell Univ., Long Island Vegetable Research Farm, Riverhead, N. Y.). Preplant broadcast appli-Dasanit [0,0-D]ethyl 0-(p-[methylsulfinyl])cations phosphorothioate] 6.7 kg/ha) and of Nemacur [Ethyl 4-(methylthio)-m-tolyl isopropylphosphoramidate] 6.7 kg active/ha, and pre-emergence sidedresses of aldicarb, 290 and 319 mg/m of row, carbofuran, 319 and 580 mg/m of row, and Nemacur (290 and 580 mg active/m of row) reduced nematode populations and increased yields of the Superior cultivar of potato in soils infested with Pratylenchus penetrans and Verticillium dahliae. Pre-emergence sidedresses of methomyl, 484 and 580 mg/m of row, lowered nematode populations but failed to increase yields consistently. Pre-emergence sidedresses of Dasanit, 290 and 580 mg/m of row, gave inconsistent results, and those of Di-Syston [0,0-Diethyl S-(2-[ethylthio] ethyl) phosphorodithioate], 290 mg active/m of row, did not control P. penetrans or increase yields. Aldicarb, 290 mg/m of row, and carbofuran, 290 mg/m of row, were more effective when applied before than after emergence. Pre-emergence sidedresses of aldicarb gave the best control of P. penetrans and highest yields (20 to 70% over control).

A soluble antigen related to infection with apple chlorotic Lafayette, Ind.). Virus-specific antisera reacting in ring tests to titers of \(\frac{1}{4} - \frac{1}{128} \) were made using preparations of apple chlorotic leaf spot virus (CLSV) purified from Chenopodium quinoa by a method previously shown to give highly purified virus proportions. Head in ging tests give highly purified virus preparations. Used in ring tests at dilutions of up to 1/8, these antisera also detected a virus-related soluble antigen in supernatant fluids from infected C. quinoa extracts subjected to prolonged ultracentrifugation (15 hr at 27,000 g in a Spinco No. 30 rotor). In gel-diffusion tests, this soluble antigen was detected in crude extracts from infected *C. quinoa*. Furthermore, gel diffusion tests showed that this antigen was also present in virus preparations, but was distinguishable from another virus-specific antigen that was not present in preparations of the soluble antigen. The soluble antigen was concentrated by precipitating in 50% ammonium sulfate solution and resuspending in buffer. It remained antigenic after storage at 4 C for 1 week or at -20 C for at least 2 weeks. In gel diffusion and ring tests, no such antigen was detected in crude extracts from healthy C. quinoa, or in control preparations concentrated from healthy C. quinoa or from C. quinoa plants infected with several other viruses.

Germination of uredospores of Puccinia recondita inhibited by blue, red, and far-red light. Ho-Shii Chang & L. Calpouzos (Univ. Minn., St. Paul). A crude action spectrum was obtained for light inhibition of uredospore ger-

mination in the wheat leaf-rust pathogen (race UN-2). The illumination system consisted of a slide projector, water-cooling bath, interference filters (half-band width, 4 or 10 nm), and a sharp cut-off filter when testing the red and far-red wavelengths. Spores were exposed to 100% relative humidity overnight, then placed on water agar and illuminated for 2 hr at 8,000 erg/cm² per sec, and 100 spores were observed for germination. The agar plate was kept at 20-21 C. Inhibition of 97% or greater (as compared to germination of spores in darkness) occurred at wavelengths of 400, 419, 651, 699, 710, 720, and 750 nm. Moderate inhibition occurred at 390, 425, and 603 nm. Little or no inhibition occurred at 452, 493, and 552 nm. A response to far-red radiation is unusual in fungi.

Graminella nigrifrons as a vector of corn stunt agent. M. M. Choudhury & E. Rosenkranz (Miss. State Univ., ARS, USDA, State College, Miss.). Graminella nigrifrons was established as a vector of both the Ohio corn stunt agent (CSA-OH) and the Mississippi corn stunt agent (CSA-MS). This leafhopper recovered CSA-MS from corn plants inoculated with CSA-MS by the more efficient vector, Dalbulus maidis. The latent period of CSA-MS in G. nigrifrons was 15-18 days. The shortest incubation periods for CSA-OH and CSA-MS in corn infected by viruliferous G. nigrifrons were 11 and 15 days, respectively. After a 14day acquisition access followed by 14 days of transmission feeding, the efficiency of transmission of CSA-OH by G. nigrifrons was 34%, that of CSA-MS, 3%. Female leafhoppers appeared to transmit CSA-OH more efficiently than their male counterparts. Some individual G. nigrifrons were found to transmit both CSA-OH and CSA-MS to the same test plants after feeding on source plants that exhibited symptoms of only Ohio corn stunt (CS-OH) or only Mississippi corn stunt (CS-MS). Such doubly infected test plants consistently developed symptoms first of CS-OH and then of CS-MS. These studies also revealed the natural occurrence in field corn of both CSA-OH and CSA-MS at State College, Miss. Moreover, noninfective G. nigrifrons acquired both CSA-MS and CSA-OH upon feeding on the same naturally infected corn plants.

Influence of fertilizer treatments and cropping sequence on populations of spiral nematodes. R. J. Collins & R. RODRIGUEZ-KABANA (Auburn Univ., Auburn, Ala.). A 2year study on the effect of fertilizer treatments on populations of spiral nematodes was made in plots within a 10year-old fertility experiment. Plots studied were under the following rotation sequence: corn; winter wheat; soy-beans; fallow; cotton; and, in some plots, winter legume as green manure. Fertilizer treatments varied from a complete formulation (N, P, K, lime, minor elements) to treatments deficient in one or more components. Populations of spiral nematodes (predominantly Helicotylenchus dihystera) were highest in corn, followed by cotton and soybeans in decreasing order. Spiral nematodes occurred in high numbers only in soil and roots from plots receiving all major elements, lime, and a winter legume; the addition of minor elements did not affect spiral populations. In all crops, application of N in the form of a winter legume resulted in higher spiral populations than when inorganic N was used. With the exception of cotton, numbers of spiral nematodes were higher in plots deficient in P than those lacking K; difference between those treatments was not evident in cotton. Omission of lime restricted spiral populations in all crops.

Exchange of materials between corn tissues and Helminthosporium carbonum as determined by electron microprobe and conductivity techniques. J. C. Comstock & R. P. Scheffer (Mich. State Univ., E. Lansing). Many fungal spores lose nutrients when germination starts; external supplies are required. The microprobe was used to monitor content of elements in Helminthosporium carbonum (HC)-conidia on corn leaves. Magnesium in conidia at time of inoculation gave 1,200 cpm under our conditions. Conidia

lost Mg (-30 to -50%) on susceptible leaves for 16-24 hr; the original Mg level was then regained by 36-48 hr. Conidia lost more Mg on resistant than on susceptible leaves, and no recovery was evident. Conidia containing Rb lost equal amounts (80%) to both resistant and susceptible Rb-free leaves. Conidia lost equal amounts of S on resistant and susceptible leaves during 36 hr. Helminthosporium victoriae (oat pathogen) conidia on corn lost Mg, Rb, and S in amounts comparable to those of HC on resistant corn. Susceptible leaves inoculated with HC-conidia (10-20/mm² leaf surface) lost more electrolytes (23 µmhos) than did control leaves (4-5 µmhos) by 24 hr. Such materials are available to the fungus. Resistant leaves had no increase in electrolyte loss 32 hr after inoculation (10 conidia/mm²). Helminthosporium victoriae did not induce electrolyte loss from corn leaves. Exchanges between host and pathogen occur early, and differ in resistant and susceptible tissue.

Interaction of Verticillium albo-atrum and Pratylenchus penetrans under controlled inoculum densities. J. J. Con-ROY, R. J. GREEN, JR., & J. M. FERRIS (Purdue Univ., Lafayette, Ind.). The influence of Pratylenchus penetrans on incidence of infection of Verticillium albo-atrum on tomato under controlled inoculum densities was considered. Infection by V. albo-atrum on tomato was correlated with controlled inoculum densities. Infection incidence of 100% occurred at the inoculum density of 200 propagules/g soil, and infection levels were progressively lower at inoculum densities of 100, 75, 50, and 25 propagules/g. Consistent increases in infection were found at all inoculum densities in the presence of the nematode. The number of nematodes extracted from roots of tomato plants infected with V. albo-atrum was significantly lower than the number extracted from roots with the nematode alone. The role of the nematode in increasing susceptibility of tomato to V. albo-atrum was considered, using a split root technique. Slightly higher levels of infection were found when the fungus and the nematode were placed on opposite sides of the split root system as compared to the fungus alone on one side, suggesting a host physiological response to the nematode which makes the plant more susceptible to infection by V. albo-atrum.

Influence of nutrition and total nonstructural carbohydrate content on Helminthosporium sativum-incited leaf spot of Kentucky bluegrasses. H. B. Couch & L. D. Moore (Va. Polytech. Inst. & State Univ., Blacksburg). Thirteen Kentucky bluegrass cultivars were grown in expanded shale and supplied with three levels of nitrogen nutrition (N), respectively; standard Hoagland's solution (1.0 N) and Hoagland's solution modified to contain 0.1 × N or 3.0 × N. The plants were inoculated with an aqueous spore suspension of Helminthosporium sativum. Analyses were made of foliage collected prior to inoculation for total and reducing carbohydrates and fructose. Anheuser, Geary, Pennstar, and Belturf were moderately to highly resistant, and Delta, Merion, Fylkin, and Newport were moderately to highly susceptible at the three nitrogen levels. The highest levels of susceptibility occurred at the 3.0 X N nutritional regime. Cougar, Winsor, Park, BA6124, and Kenblue were highly resistant only at the $0.1 \times N$ nutritional regime. There was no correlation between susceptibility and the concentration of either total and reducing carbohydrates or fructose. These studies indicate that while susceptibility of Kentucky bluegrasses to H. sativum is generally increased under high nitrogen nutrition, the total range of susceptibility of a given cultivar is more fully described when grown under a broad spectrum of nutritional regimes, and is independent of the total nonstructural carbohydrate content of the tissue.

Development of multiple disease-resistant fresh market tomato varieties adapted for machine harvest. J. P. CRILL, D. S. Burgis, & J. W. Strobel (Univ. Fla., Bradenton, Gainesville). In Florida it has been desirable to exploit

genetically controlled disease resistances, as the limiting factor in tomato (Lycopersicon esculentum) production is plant disease. Tomato pathogens in Florida controlled by the use of resistant or tolerant cultivars include races 1, 2, 3, 4, and 6 of Cladosporium fulvum (leaf mold), Alternaria solani (early blight), Verticillium albo-atrum (Verticillium wilt), Fusarium oxysporum lycopersici (races 1 and 2 of Fusarium wilt), Stemphylium solani (gray leaf spot), and five strains of tobacco mosaic virus. Resistance to fruit pox, gold fleck, blossom-end rot, crease-stem, graywall, and yellowtop diseases is also genetically controlled. Genes for resistance to these diseases, along with the horticultural characters necessary for a machine-harvest freshmarket tomato, have been incorporated into a single genotype. Release of this variety should reduce production costs considerably and revolutionize the fresh-market tomato industry, as it can be harvested mechanically and possesses more disease resistance genes than does any previous cultivar. Estimated minimum number of disease resistance genes in this variety is 21. Genes for resistance to 12 more fungal, bacterial, viral, insect, and nematode pests have been identified and are being incorporated into this genotype.

The role of the basidial stage in the life cycle of Typhula idahoensis. B. M. Cunfer (Wash. State Univ., Pullman). Fertile sporophores of Typhula idahoensis, the major snow mold pathogen of winter wheat and barley in Washington, are usually found from late October until snowfall under field conditions. Sclerotia stored dry during the summer were placed outdoors at intervals during fall. Mature sporophores began to produce basidiospores in early November, and continued to produce spores until snowfall. Basidiospores were placed on 2% water agar on glass slides for 24, 56, and 72 hr at 1, 5, 10, 15, and 20 C in the dark. Germination occurred at all temperatures, but 10 C was most favorable for germ tube elongation. At 20 C, germination was lowest and germ tubes were short and abnormal. Winter wheat and barley were inoculated with basidiospores by suspending sporophores over plants in pots enclosed in polyethylene bags. Pots were kept at 5 C in the dark or in a greenhouse at 10 C for 3-20 days. After inoculation, plants were covered with moist cotton and incubated at 1 C for at least 90 days. Typhula idahoensis was reisolated from about 8% of the inoculated plants. Disease symptoms and typical sclerotia developed on the plants. Failure to obtain a high level of infection may be due to the inoculation technique or to an inherent low-inoculum potential of basidiospores.

Mechanics of infection of soybean seedlings by overwintered and secondary Pseudomonas glycinea. G. C. DAFT & C. Leben (Ohio Agr. Res. Devel. Cent., Wooster). Seeds were planted with abrasives (sand or Carborundum) and diseased leaf litter in growth room tests. Abrasives increased the number of cotyledon lesions on the emerged seedlings. In three tests, cotyledons bore 1.8-2.3 times more lesions than control cotyledons. Lesion area was also increased. Most lesions were on the proximal cotyledon end, which usually is the advancing end, as the cotyledon penetrates the soil. The proximal area most diseased was the "trough", the depression in which the radicle rests during dormancy. Apparently the trough was injured by abrasives caught between the swelling, growing radicle and the cotyledon. Lesions appeared rarely on unifoliolate leaves; none occurred on trifoliolates. Tests were made in which seedlings (1-5/pot) with lesioned cotyledons, unifoliolates, or first trifoliolate leaves were subjected to simulated windrain storms. New lesions were initiated on leaves above the organ diseased prior to the storm. These findings suggest that pre-emergence infection of cotyledons is the initial event in the increase of overwintered inoculum, and that secondary infection on seedling leaves occurs during windrain storms.

Germination of Penicillium digitatum spores as affected by solutions of volatile components of citrus fruits. P. L. DAVIS & J. J. SMOOT (ARS, USDA, Orlando, Fla.). Volatile compounds known to occur in mature citrus fruits were evaluated for their effects on spore germination of Penicillium digitatum, a major citrus fruit-rotting fungus. The compounds tested were obtained from commercial sources and included aldehydes, alcohols, esters, and terpenes. Spores from 2-week-old fruit cultures were added to liquid synthetic medium at about pH 4 containing solutions of the test materials. The aldehydes inhibited germination, whereas the esters, terpenes, and alcohols had no effect on germination. In quantitative tests with short-chain aldehydes, the inhibitory effect increased with chain length from C5 to C8. The minimum concentrations in mmole/ liter producing 50% or more inhibition were 1.0 for valeraldehyde, 0.2 for hexanal, 0.1 for heptanal, and 0.06 for octanal

Surface stain on sapwood of partially wetted longleaf pine logs. R. C. De Groot & F. J. Czabator (USDA Forest Serv., Gulfport, Miss.). A continuous spray of water over logs in storage can protect wood from decay and staining fungi. The determination of the proper volume of water needed is critical in the establishment of water sprinkling systems. Three patterns of biologic activity were observed under the bark of longleaf pine logs that were partially immersed under a continuous spray of water for 6 weeks. In portions of logs in which the bark was continuously and adequately wet, the sapwood remained bright. Where the volume of water was inadequate, orangered stain developed on the surface of the wood. That part of the log which remained dry except for occasional exposure to rain was rapidly colonized by deuteromycetes, blue stain fungi, decay fungi, and bark-inhabiting insects, but the orange-red stain did not develop there. The red stain spread over the surface of the xylem tissues, but did not penetrate the sapwood. Several bacteria and yeasts were associated with the stain, but their relationship to the discoloration has not been defined. It is suggested that freshly cut longleaf pine logs can be used to monitor the effectiveness of water sprinkling systems.

Resistance to rust in red clover. S. DIACHUN & L. HENson (Univ. Ky., Lexington). Rust, caused by Uromyces trifollii var. fallens, is common on red clover under natural conditions in late summer in Kentucky. Several hundred seedlings and detached leaves of several hundred plants were inoculated with suspensions of uredospores of an isolate of U. trifollii var. fallens from red clover. Inoculated leaves and plants were incubated 14 days in growth chambers. A rust-resistant plant in a population of Lakeland red clover was selected in 1965 and is maintained as a clone, designated KyCL75. In F₁ populations from crosses between KyCL75 and several rust susceptible clones, resistant and susceptible plants occurred in approximately equal numbers. In F1 populations from crosses between susceptible clones, all plants were susceptible. These results indicate that resistance in KyCL75 is inherited as a single dominant factor, and that clone KyCL75 is heterozygous for this factor.

Evidence for possible genomic masking between two unrelated plant viruses. J. A. Dodds & R. I. Hamilton (McGill Univ., Montreal, Quebec, Can.). Mixtures of purified barley stripe mosaic virus (BSMV) and tobacco mosaic virus (TMV) were prepared in two ways. The first involved purifying the nucleoprotein from barley plants doubly infected with the two viruses. The second involved mixing the two viruses in vitro so that the product resembled the mixture purified from the doubly infected plants. Both mixtures were repeatedly incubated with TMV antibody until no precipitate was obtained. The BSMV which remained from the natural mixture contained TMV infectivity, most of which was removed by incubation with BSMV antibody. The BSMV which remained from the in vitro mixture contained a much smaller amount of TMV infectivity which was not removed by BSMV antibody, but which was removed by TMV antibody. The production of genomically masked virus particles with the coat protein of BSMV and the nucleic acid of TMV in doubly infected plants would explain these results.

Implications of inoculum dispersal pattern, meteorological data, and serological comparisons to epidemiology of Scle-roderris lagerbergii. C. Dorworth & B. Mathieson (Can. Forestry Serv., Ontario Region Insect Pathol. Res. Inst., Sault Ste. Marie, Ontario). Scleroderris lagerbergii is extremely damaging to pine on cool moist sites in Europe, and in plantations on hot sandy plains in central Ontario and adjacent areas east and west. Epiphytotics in North America intensify most rapidly in geographical depressions and on plains, this effect usually being attributed to predisposition of the suscepts to infection by frost damage. Temperature differences in a red pine plantation, however, were too slight to account for a 100% reduction in survival in the depressions as compared with adjacent areas. Depressions received 1 to 4 times as much inoculum as adjacent areas, depending upon their capacity to serve as air drainage troughs or basins. Serological comparisons among five isolates of S. lagerbergii from central and northern Ontario indicated that all were closely related and four were identical. Scleroderris lagerbergii in Ontario is interpreted as a relatively uniform clone which escaped from the region south of Lake Superior, where it was largely contained in small areas where conditions were optimum for infection, and subsequent local spread was minimal. It is now as far north as Lat. 52 degrees, where cool moist weather and continuous high-suscept density exist, and there the pattern of the epiphytotic should more closely resemble that described from Europe.

Toxic Fusaria isolated from moldy sweet potatoes involved in an epizootic of atypical interstitial pneumonia in cattle. B. Doupnik, Jr., O. H. Jones, Jr., & J. C. Peck-HAM (Univ. Ga. Coastal Plain Exp. Sta. and Diagnostic and Research Lab., Tifton). Several isolates of Fusarium moniliforme, F. roseum, and F. solani were obtained from moldy sweet potatoes involved in an epizootic of atypical interstitial pneumonia (AIP) and screened for toxicity to chicks. The isolates were separately grown on moist, autoclaved corn for 2 weeks, dried, ground, and mixed with protein supplement (6:4, w/w), then fed to day-old Bab-cock B-300 cockerels for 2 weeks. Of 15 isolates screened, one of F. moniliforme and two of F. solani caused weight reductions of 20% or more when compared to control chicks and one F. solani isolate was lethal. Clinical signs and lesions of AIP were reproduced in a steer and a cow by giving orally a homogenate of viable sweet potatoes which had been sliced, inoculated with the lethal isolate of F. solani, and incubated for 10 days. An ether extract from a similar homogenate produced AIP when given orally to a cow. Although this disease has been known for over 40 years, this is the first reported success of its reproduction in cattle under controlled conditions using a known organ-

The periderm, a factor limiting apple scab development. R. DRAKE & G. M. SHEAR (Va. Polytechnic Institute, Va. State Univ., Blacksburg). A periderm was found to be associated with apple scab (Venturia inaequalis) infection of apple fruit, fruit pedicels, and leaf petioles, but not leaf lamina. The periderm usually was initiated in a row of cells immediately beneath the epidermis, where infection had occurred. Periderm development was more extensive in fruit and fruit pedicels than in leaf petioles. The activity of the periderm seemed to be related to the rapidity of hyphal proliferation. During early stages of infection and when the environment was favorable for rapid hyphal mat formation between the epidermal cells and the cuticle, the periderm was initiated, but there was only a minor morphological change in the host tissue. The most extensively developed periderms were associated with the older areas of infection. In the older infections, cell division in the periderm frequently continued until 12 to 14 rows of additional host cells were produced. In the case of fruit pedicels, the extra rows of phellem cells to the outside frequently turned the pedicel 15 to 30 degrees from its long axis, resulting in a deformed, weakened structure. The heavy, suberized phellem cells of the periderm inhibited radial movement of the pathogen, but had no effect on lateral proliferation of the advancing hypha.

Ultrastructure of soybean leaves affected by systemic toxemia, J. Dueck & R. J. Zeyen (Univ. Minn., St. Paul). Systemic toxemia in young trifoliolate leaves of soybean (Glycine max) is caused by toxin(s) produced in older leaves infected by the bacterial pathogen, Pseudomonas glycinea. Affected leaves can recover from symptoms. Ultrastructural changes in leaf mesophyll affected by toxemia, and of chlorotic areas adjacent to lesions, were observed in young leaves 8 days after inoculation. Recovered and comparable healthy tissue were sampled 18 days after inoculation. Mesophyll cells in young healthy tissue had large vacuoles and chloroplasts with well developed grana and abundant starch grains. Systemically affected leaves had small cells with dense cytoplasm, small vacuoles, and chloroplasts with few lamellae and little starch accumulation. Cells of recovered leaves had large vacuoles, normal chloroplasts with well-developed grana, and extreme starch accumulation similar to comparable healthy cells. The toxin(s) did not cause a disruption of cell organelles, but delayed mesophyll cell maturation.

Water relations of tomato plants infected with Fusarium oxysporum f. sp. lycopersici. J. M. DUNIWAY (Univ. Cal., Davis). Determinations of leaf water and solute potentials showed that wilting of infected plants was due to water stress. When the diffusive resistance of leaves to water vapor loss was measured as a function of decreasing leaf water potential, the diffusive resistance in infected plants was as high as, or higher than, the resistance in healthy plants at a given leaf water potential. A leaf disc experiment showed that the disease caused an inhibition of stomatal opening which was independent of disease effects on water potential. Measurements of water flow through excised root systems indicated that infection did not increase the resistance of roots to water flow. When water was forced through stem segments, the resistance of infected segments decreased markedly as water flow occurred, and accurate estimation of xylem resistance in infected plants was impossible by this technique. Steady-state measurements of transpiration and leaf water potential indicated that resistance to water flow in the petioles of infected plants approached infinity as wilting occurred, and it was concluded that a high resistance to water flow in xylem is the sole cause of the wilting which characterizes the Fusarium-wilt disease of tomato.

Survival of Corynebacterium michiganense in soil as free cells and in infected tomato tissue. E. ECHANDI (N. C. State Univ., Raleigh). Corynebacterium michiganense could not be recovered by planting tomato seedlings in soil collected from around the base of diseased tomato plants at the end of the growing season. However, tomato seedlings transplanted immediately into soil artificially infested with 2×10^7 cells/g of soil developed canker, while those transplanted 2 weeks later remained healthy. Moreover, C. michiganense was not recovered from centrifuged suspensions of soil artificially infested with 2×10^7 cells/g after 1 week at 25 C, nor from dialyzing bags containing the organism and buried in the soil under the same conditions. However, C. michiganense was recovered from naturally infected tomato stems, roots, leaves, fruit skins, and seeds which overwintered on the soil surface, and from undecomposed woody tissue buried in the soil. Tomato seedlings transplanted immediately into soil infested with surface plant debris became infected, but seedlings transplanted 2 weeks later remained healthy. When small pieces (ca. 0.5 × 10 mm) of infected, overwintered woody tissue from buried plants were placed on healthy tomato leaflets, 80% of the inoculated leaflets became infected. Corynebacterium michiganense was short-lived as free cells in the soil, and only survived overwintering in undecomposed plant tissues.

Evaluation of imines of sec-butylamine for control of Penicillium decay of oranges. J. W. Eckert, M. L. Rahm, & R. J. Stewart (Univ. Cal., Riverside). Imines synthesized by reaction of sec-butylamine (SBA) with 2-pentanone, 2-heptanone, 2-octanone, 2-undecanone, benzalde-hyde, furfural, and vanillin, respectively, were evaluated as vapor-phase treatments to prevent decay of Valencia oranges. At dosages equivalent to $0.1\,\mathrm{g}~\mathrm{SBA/m^3}$ air, the imines were more effective than SBA, but the fruit were injured by direct contact with 2-octanone, 2-undecanone, benzaldehyde, furfural, or their imines. The imines of 2pentanone and 2-undecanone (aliphatic imines) hydrolyzed rapidly in the gaseous state in moist air, whereas the imines of furfural and benzaldehyde (aromatic imines) were stable under these conditions. The latter imines hydrolyzed on moist paper, and the rate of hydrolysis was greater below pH 7. Hydrolysis of the aromatic imines increased the pH of unbuffered paper from 5 to 9.5 within 2 hr, and 80-100% of the SBA from the imines was recovered from the atmosphere over a 5-day period. The aldehydes and ketones at concentrations equivalent to the imines had no effect upon fruit decay. The superiority of the aliphatic imines over SBA in reducing decay must be due to the sustained evolution of gaseous SBA, but for the aromatic imines, the relative contribution of imine and SBA to disease control is difficult to assess.

The association of Fusarium moniliforme with the crown rot complex of asparagus. R. M. Endo & E. C. Burk-Holder (Univ. Cal., Riverside). Asparagus plantings in southern California are frequently unproductive and shortlived. Inland plantings and fields cut for 3 or 4 months are more severely affected than coastal plantings and fields cut for 1 or 2 months. Vegetative spears from affected plants decrease in number and size; reproductive stalks are yellow, stunted, and reduced in vigor; and fern stalks die in various stages of elongation without vascular discoloration. A dry, brown crown rot was observed on all plants in the field. Severity of crown rot appeared inversely related to the vigor and size of affected plants. Fusarium moniliforme was isolated from 93% of the crowns, F. oxysporum from 22%, and Penicillium martensii from 6%. Over 80 singlespore cultures of F. moniliforme isolated from affected crowns from 5 counties were pathogenic to Fusarium wiltresistant asparagus seedlings, UC 72 and 66; reddish-brown lesions were produced on the crown, stem bases, and storage and feeder roots. Removal of a portion of the spears or storage roots significantly increased the severity of crown rot. The fungus also was isolated from commercial lots of seeds, from asparagus debris, and from volunteer asparagus seedlings. Fusarium moniliforme is considered an important pathogenic component of the crown rot complex.

The pathogenicity of Helminthosporium maydis and Helminthosporium spp. on carnation, snapdragon, and geranium. A. W. ENGELHARD (Univ. Fla., Bradenton). Helmin-thosporium maydis race T and race O from corn, and Helminthosporium spp. isolates obtained from a petal and a leaf of Chrysanthemum morifolium and from a leaf of Maranta leuconeura, were pathogenic on geranium leaves and on the flowers of carnation, snapdragon, and geranium but not on the leaves of the azalea, Rhododendron obtusum 'Erie'. Infection on the flowers of each host was evident after incubation for 24 hr at high humidity. All five isolates induced similar symptoms on the same host. Tan necrotic spots up to 2 mm in diameter developed on snapdragon flowers. Larger lesions formed as the spots coalesced. General collapse of the snapdragon flowers occurred within 3 days. The symptoms were similar to those induced by Botrytis cinerea in the same experiment. Irregular, tancolored lesions, varying from the size of a pin point to 4 mm in length, developed on the petals of carnation flowers. Initially, lesions 1-2 mm in diameter developed on geranium flowers, followed rapidly by necrosis of large

areas of the petal tissue. Irregularly shaped, sunken lesions up to 8 mm in diameter, with smooth gray centers, developed on geranium leaves.

Stalk rot in corn influenced by Texas male sterile cytoplasm. A. H. EPSTEIN, J. E. REYNOLDS, & W. P. DIETZ (Iowa State Univ., Ames). Isogenic lines of hybrid corn (A619 × A632) containing normal (N) cytoplasm and Texas male sterile (Tms) cytoplasm were compared for incidence and severity of foliar disease and stalk rot. Stalk rot severity was measured in terms of stalk-breaking strength, or "moment". This value was determined by exerting a lateral force at the fifth node sufficient to break the stalk, and measuring the distance from the 5th node to the break. The product of the force in kg by the distance in cm is the "stalk-breaking moment". Foliar diseases such as yellow leaf blight (*Phyllosticta zeae*) and southern corn leaf blight (Helminthosporium maydis race T) were more severe on plants with Tms cytoplasm. The first five leaves of Tms plants were dead due to *Phyllosticta* sp. infection by 18 June; little further development occurred. Helminthosporium maydis infections were numerous on all leaves of Tms plants by 26 August; plants appeared dead by 1 September. Stalk rot in terms of stalk breaking moment was more severe in Tms plants. The average breaking moment was 102.5 for Tms plants and 278.2 for N plants.

Vertical spore concentrations of three wheat pathogens above a wheat field. M. G. Eversmeyer, C. L. Kramer, & J. R. Burleigh (Kans. State Univ., ARS, USDA, Manhattan). Kramer-Collins samplers were used to measure concentrations of Erysiphe graminis conida and Puccinia graminis and P. recondita urediospores within the wheat canopy, 1, 3, and 6 m above ground level. Although considerable daily and hourly variations were observed in spore concentrations, circadian patterns with distinct peaks occurred. Time of maximum spore concentrations varied with meteorological factors present during a particular hour or day. Changes in wind velocity or turbulence accounted for most of the peaks observed in hourly spore concentrations. Higher wind velocities were required to dislodge spores from wet plants than from dry plants. Higher wind velocities were required to disperse spores produced on leaves and stems lower in the canopy. Peaks in spore concentrations generally occurred after plant surfaces dried in midmorning; they occurred later on days when plant surfaces remained wet longer. Peaks in spore concentration occurred as wind velocities and turbulence increased prior to a rain shower or passage of a cold front, and not as a result of the impact of rain droplets on sporebearing tissue. Profiles of spore concentrations at different heights distinguished between endogenous, exogenous, and mixed inoculum sources.

A selective medium for assay of Colletotrichum coccodes in soil. J. D. FARLEY (Ohio Agr. Res. Devel. Cen., Wooster). A selective medium was developed which successfully assayed Colletotrichum coccodes in nonsterilized soil artificially infested with conidia or sclerotia at populations as low as 100 propagules/g. The medium consisted of the following ingredients/liter: 10 g polygalacturonic acid; 1.5 g KH₂PO₄; 4 g K₂HPO₄; 25 ml soil extract; 17 g Difco agar; and the following antimicrobial agents added after autoclaving: 0.1 g pentachloronitrobenzene (PCNB); 0.1 g benomyl; 50 mg streptomycin sulfate; and 50 mg chloroamphenicol. The pH was adjusted to 5.0 prior to autoclaving, and plates were incubated at 26 C. Colletotrichum coccodes colonies produced abundant sclerotia and brown mycelium within 10 days on the medium, and could easily be differentiated from other fungi when examined on a white background. In addition to restricting spread and development of other fungi on the medium, PCNB also enhanced sclerotial production of *C. coccodes*. The medium was used to isolate C. coccodes from overwintered tomato fruit, and shows promise of direct isolation of the pathogen from field soil.

Antibiotic production by Pseudomonas morsprunorum. D. M. FILIPEK & D. POWELL (Univ. Ill., Urbana). Pseudomonas morsprunorum, a pathogen of stone fruit trees, produces an antibiotic in peptone-containing liquid or solid media. This constitutes the first report of antibiotic activity by this bacterium. The antibiotic is readily produced in King A & B media, but not in potato-dextrose agar, nutrient agar, and synthetic media. The production of the antibiotic is greatly influenced by peptone source, amount, and lot. Modified King B medium (modifications: no agar, pH 7.0, and 12 g/liter of Neopeptone [Difco]) gives excellent yield. The antibiotic is dialyzable, passes readily through cellophane, and is heat stable in 0.12 M, pH 6.8, Tris-HCl after autoclaving for 15 min at 121 C and 15 psi. Extraction studies show that it is soluble in water, pyridine, and aqueous solutions (50% v/v) of ethanol, acetone, and pyridine. It is insoluble in ethanol, acetone, petroleum ether, ethyl acetate, benzene, and chloroform. Part of this information is based on results using crude solutions or lyophilized extracts. The antibiotic appears to be primarily antifungal. Geotrichum candidum is very sensitive to it.

Possible xylem translocation of pea streak virus. R. E. FORD & S. W. ROSSO (Iowa State Univ., Ames). Pea streak virus (PSV), Oregon isolate, produces in peas necrotic local lesions, necrosis of veins and veinlets, and savoy of youngest leaves. Infected Perfected Wales peas were fixed in glutaraldyhyde, postfixed in osmium tetroxide, embedded in Epon, sectioned, stained with uranyl acetate and lead citrate, and observed with an Hitachi HU 11C electron microscope. Large aggregates of rod-shaped particles were found in parenchyma cells. These aggregates were also found in immature vessel elements that were developing secondary wall thickenings. Particles of PSV were seen after cytoplasmic contents had disintegrated. This evidence suggests that PSV is quite stable in vivo and that PSV may move via the xylem or the phloem.

Rhizoctonia blight of water hyacinth. T. E. FREEMAN & F. W. ZETTLER (Univ. Fla., Gainesville). During 1970, surveys were conducted in search of phytopathogens exhibiting potential for biological control of noxious aquatic weeds. During one of these surveys in the Canal Zone of Panama, a blight of anchoring hyacinth (Eichhornia azurea) incited by Rhizoctonia solani was noted along the Rio Chagres. An isolate of the fungus from this source proved to be extremely pathogenic on water hyacinth (E. crassipes). Inoculation of water hyacinth with mycelial fragments caused a severe blighting of the immersed portion of the plant which frequently resulted in death of the entire plant. Disease development was favored by temper-atures of 22-27 C with a marked decrease in severity at 32 C. This decrease was not correlated either with growth of the parasite in vitro or with its pathogenicity on cu-cumber. Symptoms incited by this fungus and its host range were similar to that of R. solani isolated from kenaf in Florida; however, the Panama isolate was more pathogenic on water hyacinth. Sclerotia of this fungus maintained their viability without loss of virulence after being submerged for 9 months in lake water.

Germination of uredospores of wheat stem rust pretreated with volatile stimulators. R. C. French & M. D. Gallimore (Plant Sci. Lab., Ft. Detrick, Frederick, Md.) Many diverse types of chemicals, including nonanal isolated from uredospores, have previously been reported to stimulate the germination of uredospores floated on water. Pretreatment in Conway diffusion cells of dry spores with high concentration of vapors of polar stimulators (10 µ-liters n-nonanal or n-nonanol/4.0 ml water in the annulus) in the presence of water vapor, reduced germination to zero and spore damage was apparent. In the absence of water vapor, pretreatment with stimulator vapor had no effect. Hydration or pretreatment of spores with low concentrations (0.1 µliter nonanal or 0.01 µliter nonanol/4.0 ml water) stimulated germination above that of the controls. Nonane, one of several nonpolar hydrocarbon stimulators.

neither reduced germination nor damaged the spores at the rate of 10 µliters/4.0 ml water. Nonanol at 0.5 to 10 µliters/4.0 ml water stimulated germination when applied to spores floating on water. When the same concentrations were applied as 2-hr pretreatments to dry spores, subsequent germination on water was virtually zero.

Stimulation of Phytophthora cactorum by fatty acids and metabolism of oleic acid-1-14C. R. E. Gain, F. L. Binder, & C. D. Bisogno (Marshall Univ., Huntington, W. Va., W. Va. Univ., Morgantown). Oleic, linoleic, and linolenic acids (5 mg/liter) stimulated growth of Phytophthora cactorum when added individually to a basal glucose-asparagine medium. Five days' incubation with oleic acid in 25 ml medium at 20 C yielded a 5- to 7-fold increase in dry weight over the controls. Similar increases were observed with linoleic and linolenic acids. Saturated fatty acids such as lauric, myristic, palmitic, and stearic were also stimulatory, but to a lesser degree. When cholesterol (10 mg/liter) was added to the basal medium, the individual saturated fatty acids caused a slight growth increase over the cholesterol controls, whereas the unsaturated fatty acids resulted in an approximate doubling of the dry weight after 5 days. This additive stimulation of growth by cholesterol and the unsaturated fatty acids lessened with incubation time until maximum dry weights of test cultures were similar to those of the cholesterol controls after 10 to 12 days. *Phytophthora cactorum* was incubated with oleic acid-1-14C (2 µc, 5 mg/1) in a basal medium without cholesterol for 8 days in flasks equipped with CO2 traps, then extracted with chloroform and analyzed for transformation products. Less than 3% of the oleic acid was metabolized to CO2, ca. 27% of the recovered activity was unchanged oleic acid, and ca. 70% was esterified to unidentified compounds.

Changes in soil and the death of woody ornamentals associated with leaking natural gas. J. H. GARNER (N.C. State Univ., Raleigh). This study was made to identify changes in the biotic and mineral composition of the soil associated with leakage of natural gas and the deterioration and death of trees and woody ornamentals. In soil contaminated with natural gas, (i) the soil atmosphere was anaerobic and large numbers of hydrocarbon-utilizing and sulfur-reducing bacteria were present; (ii) the pH range was 5.5-8.5; (iii) there was an increase in manganese and organic matter; (iv) sulfides of hydrogen and iron and the associated "sour-gas" smell were present; (v) there was a decrease in numbers of nematodes, actinomycetes, and fungi when compared to control soils. The changes indicated by the study are consistent with those noted by petroleum microbiologists when studying the effects of leaking natural gas in the soil. The changes occur due to the adaptation of the microbial ecosystem to a new environment. Sulfides and excesses of manganese are detrimental to plant growth.

Chemical evidence that the short particle of tobacco rattle virus specifies coat protein. S. A. GHABRIAL & R. M. LISTER (Purdue Univ., Lafayette, Ind.). Biological evidence strongly suggests that the essential genome of to-bacco rattle virus (TRV) is divided between the ribonucleic acids of "long" and "short" tubular nucleoprotein particles, which have complementary functions. Thus, infectivity tests indicate that long-particle RNA controls viral replication, and serological and surface charge studies suggest that the coat protein is specified by short-particle RNA. More rigorous chemical evidence has now been obtained supporting the latter conclusion. Amino acid analyses were performed on the protein component of two biologically distinctive strains of TRV, TRV-Y and TRV-Z, and also on the protein of an artificial hybrid, "TRV-Y/Z" produced in mixed infections using long particles of TRV-Z and short particles of TRV-Y. Protein preparations were obtained by dialyzing purified virus for 24 hr against 1.5 M calcium chloride. Results showed that the protein of TRV-Y and that of TRV-Z differed markedly in amino acid composition, especially in the content of alanine, aspartic acid, glutamic acid, glycine, and serine. The amino acid composition of the hybrid TRV-Y/Z was the same as that of TRV-Y, the strain from which the short particles used were derived. In all cases, the protein subunit had a min molecular wt of ca. 20,000.

A single high-rate application of Difolatan for the control of apple scab. J. D. GILPATRICK, M. SZKOLNIK, & S. D. Gibbs (N.Y. State Agr. Exp. Sta., Geneva). A single foliar spray of Difolatan 4 Fl (captafol), at the high rate of 5 quarts/100 gal (350 gal/acre), to large, bearing trees at the green tip to half-inch green flower stages gave excellent control of apple scab beyond petal fall. This spray replaced and gave as good control as 5 weekly precover sprays of captan 50 WP, 2 lb./100 gal. Control through the rest of the 1969 season was obtained when Difolatan was followed by cover sprays of captan; but in 1970 these were not required. Gas-spectrophotometric analyses of residues on the surface of trees sprayed with Difolatan in 1969 and 1970 revealed levels of 300-700 ppm on the bark of branches until after bloom, declining to 3-40 ppm by harvest time. Residues of 350-700 ppm were found on cluster leaves, 60-100 ppm on flowers, and 35-75 ppm on terminal leaves during bloom, even though only partially exposed, if at all, to Difolatan at the time of application. Much lower residues were found on immature fruit, and none on ripe fruit. The success of this promising new method of scab control is due mostly to the persistence of Difolatan on the trees and its redistribution by rains so that sufficient protective levels of the chemical are present on susceptible tissues during bloom, a critical stage for infections.

Ribonucleic acid polymerase from Rhizopus stolonifer. C. Gong & J. L. Van Etten (Univ. Nebr., Lincoln). Extracts of germinated spores of Rhizopus stolonifer were separated into three active fractions (I, II, and III) of DNA-dependent RNA polymerase upon a diethylaminoethyl cellulose column. Similar preparations from ungerminated spores contained only two active fractions corresponding to fractions I and III. Fraction III from both spore states exhibited identical characteristics, whereas fraction I from dormant spores required conditions for optimal activity slightly different from those for fraction I of germinated spores. All fractions required four ribonucleoside triphosphates, a divalent cation, and template DNA for activity, and all were inhibited by actinomycin D but not by rifamycin. The individual fractions differed in the optimal Mn++ or Mg++ ion concentrations. However, optimal activities were obtained with Mn++ and denatured DNA for all fractions.

Re-evaluation of the role of NH₃ as the cause of the hypersensitive reaction. R. N. Goodman (Univ. Mo., Columbia). Contrary to published reports from my laboratory, recent experiments have disclosed that NH₃ accumulation and increased pH in tissues undergoing bacterially induced (10⁸ cells/ml) hypersensitive reaction (HR) are not causally related to the resultant permeability changes and membrane damage. In previously published experiments, we studied HR induced by incompatible bacteria (Pseudomonas pisi and others) in tobacco leaves that were kept in moist chambers. In this system, desiccation of tissue and other symptoms of HR were delayed more than 48 hr, which provided more time to study the otherwise rapid death of inoculated tissue. Data are presented from experiments with the same system that reveal a threefold increase in electrolyte leakage; electron micrographs disclose extensive membrane damage 6 hr after inoculation. These occur 18 hr before the observation of either a pH rise or an accumulation of NH₃.

Transpiration of bur oaks during oak wilt pathogenesis. G. F. Gregory (USDA Forest Serv., Delaware, Ohio). Transpiration (in a growth chamber) was determined for pairs of 2-year-old bur oak ramets after inoculation of

one member of each pair with Ceratocystis fagacearum. The other served as a control. All plants were weighed daily to determine transpiration loss. These pairs of ramets were harvested periodically, and the two members were compared for flow rate of sterile water through a stem section of each plant, water content of leaves and stem (by fresh and dry weight measurements), and ash content of the leaves and stem. Transpiration held steady, then declined rather rapidly at varying time intervals after inoculation. The period from inoculation to start of transpiration decline was computed as a per cent of the period from inoculation to first leaf symptoms. When averaged for plants showing this decline, this was 57%. Flow rate of water through stem sections was nearly always markedly reduced by the time leaf symptoms appeared. Symptomless inoculated plants that showed transpiration decline also had reduced waterflow capacity. Water content of the leaves of symptomless plants was not significantly different from their corresponding noninoculated ramets, whereas that of plants with leaves showing symptoms was reduced. No significant difference occurred in ash content.

Induction and photoreversal of peroxidase in leaves of Phaseolus vulgaris 'Pinto III'. H. HABECK & C. R. CURTIS (Univ. Md., College Park). Detached Pinto bean leaves 9 to 13 days old were placed in a moist chamber 6 inches from two germicidal lamps (G15T8, principal wavelength 2537A), irradiated for 0, 15, 30, 60, and 120 min, then incubated in darkness at 25 C for 24 hr. Two leaf discs (12-mm diam) from each of four leaves were homogenized in 10 ml of cold 0.1 m phosphate buffer, pH 5.8. The homogenate was filtered and the filtrate centrifuged for 30 min at 30,000 g at 2-4 C. The supernatant was assayed for peroxidase activity using a spectrophotometric guaiacel- $\rm H_2O_2$ method. Peroxidase activity was increased in extracts from all far UV (2537A)-irradiated leaves. The peroxidase stimulation was confined only to the irradiated areas, and did not occur in nonirradiated half-leaves or other unexposed portions of leaves. Peroxidase stimulation due to far UV irradiation of leaves was reversed by sub-sequent exposure to near UV (GE F15T8-BLB, principal wavelength 3650A); the degree of reversal was related to the length of exposure to near UV irradiation. Near UV irradiation alone failed to stimulate peroxidase activity. Since considerable increases in oxidative enzymes are known to occur during infection in certain plant diseases, these results indicate a means of changing peroxidase activity in host cells without infection.

The nature of polypeptide, polyamine, and protein molecules which promote phytoalexin production. L. A. HADWIGER (Wash. State Univ., Pullman). Many components released by plant pathogens and other microorganisms stimulate phytoalexin production in plants. Some of these components are basic polypeptides. The synthetic polypeptide poly-L-lysine (1 mg/ml) enhanced phenylalanine ammonia lyase (PAL) activity > 6-fold, and initiated the de novo synthesis of > 100 μ g/g pisatin in pea pod tissue within 24 hr after application. Synthetic polypeptides (containing lysine, ornithine, or arginine) effective in inducing PAL and pisatin contained 14 to 100 amino acid residues. Optimal induction was obtained with peptides containing 14-50 residues. Basic polypeptides with less than four or more than 700 residues and 20 other synthetic polypeptides not containing basic amino acids were ineffective as PAL inducers. Spermine, putrescine, cadaverine, protamine, and several other basic compounds also induced PAL and pisatin formation in peas. These and many other phytoalexin-inducing compounds have in common an affinity for DNA.

Some characteristics of pectic lyases produced by Pseudomonas fluorescens, a causal agent of "pink eye" disease of potato tubers. S. S. Hagar & G. A. McIntyre (Univ. Maine, Orono). Potato tuber slices were inoculated with Pseudomonas fluorescens. After 120-hr incubation at 20 C, the tissue was extracted. The extract was centrifuged, filtered,

dialyzed, and assayed for pectic enzyme activity. No pectin methylesterase activity was observed in extracts; however, there was lyase activity. The extract was placed on a pH 9.4 DEAE-cellulose column and eluted by a gradient of pH 7.6 Tris buffer (0.01-0.1 m). Two pectic lyase systems (peaks 1 and 2) were eluted. Peak 1 was more active than peak 2. Optimum pH range of activity was 8.5-9.5. Activity of both peaks was enhanced by Ca²⁺ (0.001 m). Both peaks were inactivated when held above 50 C for 1 hr. Substrate preference was Na-polypectate > pectin N.F. > polygalacturonic acid. Thin-layer chromatography of hydrolytic products showed that unsaturated uronides and pectic fragments were present after 2-hr hydrolysis, At 96-hr hydrolysis, only unsaturated uronides were observed. Molecular weight determinations by Sephadex G-200 filtration were ca. 18,000 (peak 1) and 22,500 (peak 2). Both enzyme systems appear to be endopectic lyases.

Egg hatching of Meloidogyne incognita in the neutral carbohydrate fraction of root exudates of gnotobiotically grown Dupuits alfalfa. R. A. HAMLEN, J. R. BLOOM, & F. L. LUKEZIC (Pa. State Univ., Univ. Park). Due to the implication by previous works of carbohydrates in hatching of Meloidogyne incognita, we studied egg-hatching success in soil sterilized by gamma irradiation and wetted with root exudates from plants in different stages of development and subjected to various levels of clipping. Carbohydrate components of the exudates were identified by gas chromatography-mass spectrometry. Correlation values suggested the involvement of glucose with hatch; however, subsequent testing of glucose alone did not confirm this association. Percentage of hatch also was positively correlated with concentrations of inositol, sucrose, and three unidentified compounds in the exudates. Greater hatch occurred in exudates from seedling alfalfa than from mature, nonflowering plants, and more occurred in exudates from flowering plants than from nonflowering plants. Hatch was significantly higher in samples from lightly clipped alfalfa than from severely clipped plants. However, hatch in root exudates was never significantly different than in distilled water. The results indicate that the total neutral carbohydrate fraction and component carbohydrates considered in this study were not effective hatching stimulants of M. incognita under these experimental conditions.

Cell permeability changes in sunflower caused by infection by Sclerotinia sclerotiorum. J. G. HANCOCK (Univ. Cal., Berkeley). Influx and efflux of water (bulk flow), urea (diffusion), and electrolyte leakage are less for sunflower (Helianthus annuus) hypocotyl sections from above lesions caused by Sclerotinia sclerotiorum than for those from healthy plants. Urea uptake by sections from above lesions is reduced (squash, tomato) or unchanged (bean) in other hosts after Sclerotinia infection, and it is reduced in diseased sunflower tissues even during brief incubation periods. Efflux of urea from sunflower hypocotyl sections is biphasic, suggesting diffusion from two compartments (cytoplasm and vacuoles). Urea efflux from cytoplasm in diseased tissues ($t_{1/2} = 59 \,\mathrm{min}$) is faster than from vacuoles of cells in healthy or diseased hypocotyl tissues ($t_{1/2} \cong$ 250 min), but slower than that from cytoplasm of healthy ones $(t_{1/2} = 38 \text{ min})$. Increased resistance to diffusion of urea in host cells above lesions apparently resides in the plasmalemma. In contrast to intact plants, electrolyte leakage is greater from tissues adjacent to lesions than from comparable noninoculated plant materials when excised sunflower shoots or celery petioles are used. Increased permeability stimulated by infection of excised organs is more reminiscent of senescent processes than disease or wound responses.

Toxicity of aqueous extracts from peas infected with Aspergillus ruber to healthy excised pea embryonic axes. G. E. Harman (N.Y. State Agr. Exp. Sta., Geneva). Aspergillus ruber (NRRL 52) is highly pathogenic to pea seed. Germination of surface-sterilized inoculated seed stored at

30 C and 87% relative humidity for 14 weeks was reduced from 98 to 30-35%, whereas germination of noninoculated peas was about 90%. Peas infected with A. ruber for 14 weeks, similar peas which had been placed under conditions favoring germination for 6 days, moistened, auto-claved peas infected with A. ruber for 7 days, or uninfected controls were comminuted in distilled water. The resulting mixtures were centrifuged, and the supernant liquids passed through sterile HA Millipore filters. The sterile liquids were mixed with a nutrient salts-sucrose agar medium and excised, healthy, surface-sterilized, embryonic pea axes placed on the medium. Extracts from infected autoclaved peas induced marked necrosis, prevented normal increases in fresh weight and elongation of the radicle, prevented the formation of root hairs, and caused extreme brittleness of the embryonic axes. Extracts from infected, living peas caused similar, though much less severe, effects. Extracts from uninfected peas had little effect on excised embryonic axes. Toxicity of the extracts was not lost on autoclaving for 15 min.

Electron microscopy of shoestring and stunt diseased highbush blueberry. J. X. Hartmann, G. R. Hooper, & J. E. Bath (Mich. State Univ., E. Lansing). Viruslike particles 28 to 30 nm diam were found in ultrathin sections of leaf and root tissues from highbush blueberry affected by shoestring disease. Leaf epidermal, palisade, spongy mesophyll cells, and xylem tissue contained characteristic particles. Crystalline arrays and large masses of particles were particularly evident in root xylem tissue, Particles hexagonal in outline, 26-28 nm diam, were partially purified from diseased leaves. No such particles were detected in healthy blueberry tissues. Blueberry stunt, another disease of unknown etiology (Chen in 1971 associated a mycoplasmalike organism with the disease) was studied concurrently with shoestring. In ultrathin sections of phloem sieve-tube elements from diseased Concord and Coville cultivars, mycoplasmalike organisms were found. Spherical, elongate, and irregular forms, as evidenced by serial section methods, were present in leaf and fruit pedicel tissue. A distinct external unit membrane, presumed ribosomes, and fibrillar nucleic acid were characteristic of these structures. No mycoplasmalike structures were observed in healthy blueberry plants of the same cultivars.

Polar lipids of fungi. J. W. Hendrix & G. Rouser (Univ. Ky., Lexington, City of Hope Med. Center, Duarte, Cal.). Polar lipids of eight fungi from all three fungal classes were determined, and phospholipids were deter-mined quantitatively. Lipids found in all fungi were cardiolipin, phosphatidylcholine, phosphatidylethanolamine, and phosphatidic acid. Phosphatidylinositol was found in all fungi except one yeast. Phosphatidylserine was found in all fungi except Pythium and Phytophthora; in these fungi, an unidentified compound which may contain serine was detected, but the compound did not behave chromatographically like phosphatidylserine. Various fungi contained at least three and as many as 10 unknown lipids. Pythium and Phytophthora contained more unknown lipids, both number and amount, than the other fungi. Pythium and Phytophthora were similar in their lipids, whereas the higher Phycomycete, Phycomyces blakesleeanus, resembled the Ascomycetes (Neurospora crassa, Saccaromyces fragilus, and S. pastorianus) and Basidiomycetes (Schizophyllum commune and Polyporus versicolor) more than the Pythiaceae. The unknown phospholipids in the Pythiaceae accounted for about one-third of the lipid phosphorus. Reproducing cultures of Pythium periplocum contained much more of two highly polar unknown lipids than nonreproducing cultures.

Heat therapy for elimination of seed-borne Phoma betae and other microbial contaminants from sugarbeet seed. L. J. Herr (Ohio Agr. Res. Devel. Cent., Wooster). A silver nitrate seed surface-sterilization treatment controlled contaminants on commercial sugarbeet seeds but failed to control the black root pathogen, Phoma betae,

present in seeds of some breeding lines. Seed-borne Phoma must be eliminated for gnotobiotic host-pathogen black root investigations. Phoma-contaminated seeds were treated in a water bath at 60 or 65 C for varying time intervals on the same day or on 2 consecutive days (seed dried at 28 C overnight). Treatment at 65 C reduced germination 60-100%, depending on length of treatment. Seeds treated on a single day, whether for one or two intervals, commonly were contaminated with spore-forming bacteria. Commercial monogerm seed was more sensitive to length of treatment at 60 C than was seed from breeding lines (25% germination vs. 80%, respectively, at 15 min). Treatment of three breeding line seed lots (monogerm and multigerm) for 10 min at 60 C on consecutive days failed to eliminate all *Phoma*; whereas a 15-min treatment interval did so with a 20% or lower reduction in germination. Variation in heat tolerance was not correlated with differences in average seed wt. The heat tolerance of different cultivars and breeding lines must be ascertained by

Callose deposition and abnormal secondary wall thickening as possible restrictive factors for virus spread. C. HIRUKI & J. C. TU (Univ. Alberta, Edmonton, Can.). Potato virus M (PVM)-incited local lesions on leaves of Red Kidney bean were investigated at the three developmental infection phases represented by nonnecrotic, seminecrotic, and necrotic cells from the peripheral zone toward the lesion center. Thickening, due to callose deposition, was detected on the inner wall of nonnecrotic cells immediately adjacent to seminecrotic ones by fluorescent microscopy 3 to 4 days after inoculation. The ultrastructure of such thickening was simpler than that of "abnormal secondary wall thickening", due to deposition of paramural bodies, in seminecrotic cells. The callose deposition was not detected in necrotic cells. PVM particles were observed in seminecrotic cells but not in necrotic cells. Temporary and partial blocking of PVM spread by callose deposition may occur at the early stage of individual cell infection. However, callose alone may not be the determining factor in restricting virus spread, since PVM lesions continued to expand for a few days even after a band of callose was deposited around seminecrotic cells. The movement of PVM appeared to be ultimately restricted by more complex "abnormal secondary wall thickening" which prevailed in the seminecrotic zone.

The relationship of light to growth and sporulation of Botrytis cinerea on potato-dextrose agar. R. E. HITE (ARS, USDA, Pa. State Univ., University Park). Certain isolates of Botrytis cinerea sporulate profusely when grown on potato-dextrose agar at 21 C and exposed to continuous near-UV radiation. The same isolates grown under continuous darkness produce only sclerotia. General Electric cool-white fluorescent lamps emit sufficient near-UV radiation to stimulate sporulation. Sporulation is accelerated by exposure to General Electric BLB fluorescent black light lamps which emit a continuous spectrum between 300 and 400 nm with 97% of the radiation in this region. When near-UV is filtered from the lamp source, colony growth consists of a fluffy, aerial, vegetative mycelium with no sclerotia and essentially no sporulation. Sporulation varied in relation to the near-UV transmission of specific filters. Sclerotia formed under dark conditions are also stimulated to produce asexual spores when exposed to either cool-white fluorescent or BLB fluorescent black light lamps, the latter being the more effective. When near-UV is filtered from the source, the sclerotia resume growth through the production of vegetative mycelium. The sporulation response requires very low levels of near-UV radiation under long-term exposure.

The ultrastructure of Aphanomyces euteiches during differentiation of primary asexual spores. H. C. Hoch & J. E. Mitchell (Univ. Wis., Madison). Ultrastructural changes in vegetative hyphae of Aphanomyces euteiches just prior to differentiation of primary asexual spores included the

segmentation of the central vacuole by cytoplasmic strands, the enlargement and striation of electron-dense vesicular inclusions, and the movement of nuclei from a peripheral position to one near the longitudinal axis of the hyphae. Differentiation of primary spores occurred within minutes, and began with the migration of cytoplasm toward each nucleus and the withdrawal of the plasmalemma from the hyphal wall. Simultaneously, the central vacuole evaginated and emptied its contents into the space developed between the plasmalemma and the hyphal wall. The tonoplast and the plasmalemma jointly formed the plasma membrane enveloping the primary spores. Plasmalemma-somes, initially consisting of tightly compacted tubules, became increasingly disorganized during differentiation. The irregularly shaped spores, generally connected in succession by strands of membrane-bound protoplasm, were entirely passive as the hyphal contents were rapidly extruded through a rupture in the sporangium apex. No flagella were observed in any spores at this stage. Within minutes after extrusion, wall material was excreted to complete the formation of the primary spore cyst.

Effect of different soil constituents on uptake of two benzimidazole compounds by American elm seedlings. W. K. HOCK & L. R. SCHREIBER (USDA, ARS, Delaware, Ohio). Benomyl and thiabendazole (TBZ) fungicides, applied as drenches to 4-month-old container-grown American elm seedlings, were absorbed and translocated throughout the plants. Leaf, wood, and bark sections were placed in petri dishes containing potato-dextrose agar seeded with either Ceratocystis ulmi (for benomyl bioassay) or Penicillium sp. (for TBZ bioassay). The diameter of the zone of inhibition around each tissue section was measured to determine the relative concentration of fungitoxicant present in each seedling. The zones of inhibition around tissues from plants grown in sand and treated with benomyl were about 2 times larger than from plants grown in treated soil, and about 3-4 times larger than from plants grown in a potting mixture of soil:peat:perlite (1:2:2). When seedlings were grown in their respective media for 90 days, elms grown in treated sand still contained more fungitoxicant than those in either treated soil or the mixture. Whenever peat was included in various media, uptake of both benomyl and TBZ was inhibited significantly. Plants grown in perlite and sand always contained the most fungitoxicant. Uptake of benomyl by seedlings was greater from heat-sterilized soil than from nonsterile soil.

Water solubility of a volatile factor inducing soil fungistasis. T. S. Hora, Maureen Romine, & R. Baker (Colo. State Univ., Ft. Collins). Earlier workers have experienced difficulty in extracting a fungistatic principle from soil. Extraction of this inhibitor was attempted, taking into account the volatile nature of the factor as previously reported. Germination of conidia of common soil fungi was significantly inhibited by air which had passed through a column of soil. This inhibitory influence of air, however, was lost upon passage through water, indicating water solubility of the volatile inhibitor. To demonstrate the activity of the fungistatic principle in aqueous solution, air washed in concentrated sulfuric acid was passed through 1 kg of clay loam soil (pH 8.6) packed in a 1,000-cc capacity burette at 25 C, and bubbled into 2 ml of water at 3 C for 24 hr with a flow rate of 6-8 cc/min. Conidia of eight of ten soil fungi showed a 20-65% decrease in germination when suspended in 0.1-ml aliquots of the water extract and placed on water agar discs for 24 hr. These observations suggest that the volatile fungistatic factor is soluble and extractable in water.

The effect of temperature on symptom expression of chrysanthemum inoculated with chlorotic mottle virus. R. K. Horst & S. P. Kryczynski (Cornell Univ., Ithaca, N.Y.). Symptoms expressed by Chrysanthemum morifolium 'Deep Ridge', subsequent to inoculations made by tissue implantation with chrysanthemum chlorotic mottle virus (ChCMV), are influenced by temperature. Experiments

were performed under controlled environmental conditions so that light and moisture were nonvariables and temperature was constant at 18, 24, and 30 C. Symptom expression was evaluated by determining numbers of leaves and lateral shoots developing symptoms after inoculation, and by length of time for symptoms to appear. Symptom expression, as determined by all three means of evaluation, was optimum at 24 C. Delay in symptom development was more pronounced at 18 than at 30 C. Mean temperatures resulting from a diurnal temperature program yielded results similar to those from corresponding constant temperatures. Recovery of ChCMV (by tissue implantation procedures) from the experimental plants was independent of temperature treatments.

The effect of triarimol on oxidation of glucose and acetate to carbon dioxide and on spore germination of Aspergillus niger and Cladosporium cucumerinum. L. D. House-WORTH, E. W. BRUNTON, & B. G. TWEEDY (Univ. Mo., Columbia). C-1, C-3, C-4, C-6, and uniformly labeled glucose and C-6 labeled gluconate were used to determine the effect of triarimol on the respiratory pathways of germinating spores of Aspergillus niger and Cladosporium cucumerinum. Triarimol did not affect the quantity of 14CO2 evolved when the organisms were incubated with any of the labeled substrates, indicating that the fungicide has no effect on the rate or participation of pathways for glucose catabolism. To determine the effect of triarimol on Kreb's cycle, C-1 and C-2 labeled acetate were administered, and triarimol did not alter the rate of 14CO2 evolution from labeled acetate. Fractionation of A. niger spores previously incubated with C-2 acetate showed that triarimol had no effect on the quantity of the methyl moiety incorporated into various organic fractions. These studies suggest that triarimol has no effect upon respiratory pathways during the initial stages of spore germination. Spore germination studies showed that triarimol had little or no effect on the initial processes of both organisms, but germ tube elongation of C. cucumerinum and A. niger was arrested at 0.5 μg/ml and 20.0 μg/ml, respectively.

Chemical limitation of discoloration and decay associated with wounds in red maple and yellow birch. D. R. HOUSTON (USDA, Northeastern Forest Exp. Sta., Hamden, Conn.). Chemicals, including fungicides, antibiotics, glue, and paraformaldehyde, were placed immediately into increment borer wounds made in 65 red maple and 35 yellow birch trees in New Hampshire. Three years later, the trees were dissected, the vertical extent of the discoloration and decay associated with the wounds was measured, and microorganisms were isolated. Discoloration in both species was reduced significantly by chemicals. Red maple wounds treated with glue, and yellow birch wounds treated with glue or Methocel, had the least discoloration. In red maple, the most extensive discoloration and decay, except for controls, occurred in trees treated with paraformaldehyde. Decay in this species was least when wounds were treated with Acti-dione. Microorganisms isolated from discolored and decayed tissues were typical inhabitants of woundassociated defects, and included bacteria, non-Hymenomycetes, and Hymenomycetes. Graphium sp. was common in both tree species.

Cassia obtusifolia, a possible reservoir for inoculum of Colletotrichum fragariae. C. M. Howard (Univ. Fla. Agr. Res. Center, Dover). Strawberry anthracnose caused by Colletotrichum fragariae has been a serious disease in Florida strawberry nurseries for many years. Since 1968, anthracnose has severely limited production in Florida of the popular but extremely susceptible cultivar, Tioga. Colletotrichum fragariae has not previously been reported attacking plants of any genus other than Fragaria and Duchesnea. In 1970, 3- to 6-inch chance seedlings of Cassia obtusifolia in strawberry seedling flats were found to be infected by a Colletotrichum species. Infection by a Colletotrichum species of C. obtusifolia seedlings up to 18 inches high in and around a strawberry seedling nursery were

found later. Typical anthracnose lesions developed on runners and petioles of potted strawberry plants inoculated with spore suspensions from isolates obtained from both Cassia sources. Acervuli with black setae developed on C. obtusifolia seedlings inoculated with isolates from the weed or from strawberry plants. Cassia obtusifolia is widely distributed in west central Florida throughout the year, and may be the principal source of primary inoculum of C. fragariae.

The role of bacteria in inoculated seedlings of sugar maple, Acer saccharum. R. HOWLAND (Univ. N.H., Durham). Sugar maple seedlings were inoculated with several selected species of bacteria representing the genera Bacillus and Pseudomonas. Subsequent isolation of the inoculated species proved successful in the majority of cases. Naturally introduced gram-negative organisms have also been isolated frequently from the same inoculated seedlings. Wood sections taken from the nonwounded control trees were free of bacteria. Several species representative of both the Aerobacter-Klebsiella and Pseudomonas groups have been identified. Cultural and histological studies indicate that some of the gram-negative bacteria that develop naturally in wounded sugar maple trees may be of importance in predisposing the tissues to further colonization by numerous other organisms higher than bacteria.

Peak concentrations of potato yellow dwarf viruses in Nicotiana rustica revealed by infectivity assay on vector cell monolayers. H. T. Hsu & L. M. Black (Univ. Ill., Urbana). The concentrations of two varieties of potato yellow dwarf virus (PYDV) in the stem portion bearing the acutely infected leaves of Nicotiana rustica plants, grown at 30 C, were determined by infectivity assay on their respective vector cell monolayers. The variety SYDV (sanguinolenta yellow dwarf virus) was assayed at pH 5.93 on AS2 cells, whereas variety CYDV (constricta yellow dwarf virus) was assayed at pH 5.25 on AC20 cells. Both viruses attained a peak concentration 10 days after the first appearance of systemic symptoms. The relative virus concentrations of each variety were calculated as percentages of its peak concentration. About 30% of SYDV infectivity was lost by the 12th day and 40% was lost by the 14th day; thereafter the infectivity remained almost constant until the last measurement on the 24th day. About 70% of the peak infectivity of CYDV was lost by the 12th day, and more than 80% was lost by the 14th and 16th days. Purification to obtain highly infectious PYDV free from microorganisms was best achieved by harvesting infected plant materials at the peak virus concentration. Purified viral inocula can be stored at -80 C for more than 1 year without detectable loss of infectivity.

35S transfer from host to parasite during primary infection of barley by Erysiphe graminis f. sp. hordei. S. C. HSU & A. H. ELLINGBOE (Mich. State Univ., E. Lansing). Six-day-old barley plants with different Ml genes (Mla, Mlg, Mlk, Mlp, or mlx) were inoculated with two isolates of Erysiphe graminis f. sp. hordei with genotypes Pa Pg Pk Pp and Pa pg Pk Pp. The percentage of the applied conidia that infected the host cells and produced secondary hyphae at least 10 µ in length was recorded at 2-hr intervals between 16 and 30 hr after inoculation. At various hours after inoculation, leaves carrying the several different M1 genes were allowed to take up 35SO4- for 5 hr. The portion of the parasite on the surface of the leaf was removed from the leaf with parlodion, and the $^{35}\mathrm{S}$ in the parlodion strips was determined by scintillationcounting. The parasite/host genotypes which had a high efficiency of infection (Pa Pg Pk Pp/mla mlg mlk mlp and Pa pg Pk Pp/mla Mlg mlk mlp) also had high rates of ³⁵S transfer from host to parasite. The incompatible parasite/host genotypes (Pa/Mla, Pg/Mlg, Pk/Mlk, or Pp/ Mlp) had lower efficiency of infection and a reduced rate of 35S transfer from host to parasite, as compared to compatible combination.

Properties of chloroplast structural proteins isolated from tobacco leaves undergoing bacterially induced hypersensitive reaction. J. S. Huang & R. N. Goodman (Univ. Mo., Columbia). Cloroplasts were isolated from tobacco leaves, by homogenization and differential centrifugation, 20 min, 3 hr, and 6 hr after infiltration with 108 cells/ml of Erwinia amylovora. The hypersensitive reaction (HR) is normally visible at 6-7 hr after infiltration. Isolated chloroplasts were sonicated and centrifuged to obtain membrane and lamellar fragments. Structural proteins (SP) were isolated from the fragments, using acetone. The resultant acetone powder was dissolved in 0.002 M Tris buffer (pH 8.5) containing sodium dodecyl sulfate and urea. The protein fraction collected was that precipitated with 12-16% (NH₄)₂SO₄. All SP thus prepared showed UV absorption spectrum maxima at 280 nm and minima near 250 nm. They were heterogeneous, however, and were resolved as several protein bands by electrophoresis. The SP prepared from leaves 6 hr after infiltration (during the "developing" stage of HR) when HR symptoms were apparent had a lower OD₂₅₀/OD₂₅₀ ratio, reduced phospholipid-binding capacity, and lower solubility than SP isolated from the leaves 3 hr after infiltration (during the "incubation" period). These differences in criteria of SP were even greater between the 3-hr sample and that taken 20 min after infiltration (during the "induction" period). It would seem, therefore, that SP denatures progressively as a consequence of HR.

Factors affecting production of microsclerotia by species of Cylindrocladium. B. B. HUNTER & H. L. BARNETT (West Va. Univ., Morgantown). Cylindrocladium scoparium, C. floridanum, and C. ilicicola were tested for production of microsclerotia. Effects of temperature and light (40 w General Electric cool-white fluorescent lamps, 125 ft-c) were tested on glucose-casein hydrolysate agar. A range of 24-28 C was optimum, and light had no apparent effect on numbers of microsclerotia. Effects of concentration and ratios of carbon to nitrogen were tested at 25 C on glucosepotassium nitrate agar medium and on sand supplemented with the same nutrients, minus agar. Similar results in agar and sand cultures were obtained with C. scoparium. The amount of mycelium decreased, but the number of microsclerotia increased (15 mg and 360 mg/culture, respectively) with widening carbon:nitrogen ratios (1:1 to 100:1). Casein hydrolysate and L-tyrosine provided carbon as well as nitrogen for microsclerotial production, but L-glutamic acid was a poor source of carbon. Microsclerotia were recovered from nursery soil to which 2 g of glucose were added to 25 g of soil and incubated in the laboratory for 12 days, but were not recovered from soil without added glucose.

Peroxidase activity in wheat leaves infected by Puccinia recondita. L. B. Johnson & B. A. Cunningham (Kansas State Univ., Manhattan). Peroxidase activity was determined in primary leaves of healthy and inoculated nearisogenic wheat lines differing in susceptibility to isolate UNI-68B of the leaf rust pathogen, Puccinia recondita. Activity was similar in healthy leaves of both lines, and increased with leaf age. In inoculated Thatcher, which develops a high infection type (4), peroxidase activities at 2-9 days were 20-48% higher than in healthy tissue. In LR10(TC), which developed a low infection type (X) in these experiments, peroxidase had increased up to 109% over healthy controls after 9 days. Disease development did not affect buffer-soluble protein (trichloroacetic acidprecipitable) in either line. Peroxidases from healthy and inoculated LR10(TC) were separated on a Sephadex G-100 column into two distinct molecular wt groups. Although total peroxidase activity was greater in infected tissues, the ratio of activities in these two molecular wt groups did not differ significantly between 9-day rusted LR10(TC) and healthy tissue. The low molecular wt group is composed of peroxidase isoenzymes with an average molecular wt of near 35,000. The higher molecular wt fraction has at least

twice this molecular wt. The physiologic significance of these two molecular sizes is unknown.

Telephones as computer terminals for plant disease information. A. L. Jones & S. B. Harsh (Mich. State Univ., E. Lansing). An automated method for handling questions on fungicide compatibility, disease diagnosis, and calibration of spray equipment has been produced using 12-button telephones as computer input-output terminals in combination with an IBM 7772 audio response unit capable of giving a verbal reply to digital input. Codes used frequently for identifying the user are entered using a "Call-a-matic" device or prepunched plastic cards and a card-dialer option. Names of fungicides and disease symptoms are entered in coded form, using the push-button keyboard. Data requests by the program and output from the analysis are returned over an auxiliary speaker or hand receiver. The programs, written in FORTRAN IV, allow the operator to alter the input values to obtain custom replies. Field tests indicate that the operating procedures are simple enough for use by Cooperative Extension workers. Because low cost, multipurpose telephones and standard communication lines are used, the voice answerback system is potentially available to a large number of remotely located users of disease in-formation. Push-button "pads" connected in parallel with rotary dial telephones can be used in areas without pushbutton telephones.

Interaction of five soil-borne pathogens of tomato. J. P. JONES & A. J. OVERMAN (Univ. Fla. IFAS Agr. Res. Education Center, Bradenton). Tomato seedlings of a breeding line resistant to Fusarium oxysporum f. sp. lycopersici (Fol), race 1, but susceptible to race 2, were grown in methyl bromide and steam-sterilized, composted, Leon fine sand artificially infested with Fol races 1 and 2, Verticillium albo-atrum, Tylenchorhynchus capitatus, and Meloidogyne incognita, using all possible combinations of these five pathogens. Race 1 interfered with pathogenesis by race 2 at 22 and 25 C. In 14 of 16 race 2 combinations containing race 1, disease incidence was significantly less when compared to identical combinations without race 1. Tylenchorhynchus capitatus did not affect Fusarium or Verticil-lium wilt development at 22 or 25 C. Meloidogyne incognita did not affect the development of Verticillium wilt at either temperature, but increased the incidence of Fusarium wilt incited by race 2 at 22 C.

Effect of glucose and amino acids on growth and sporulation of Fusarium oxysporum f. sp. lycopersici, race 2. J. P. Jones & S. S. Woltz (Univ. Fla., Bradenton). Race 2 was cultured 7 days in pH 6.0 liquid media containing in addition to a basal inorganic nutrient solution, 0.0, 0.3, and 1.2% glucose alone and in combination with 1,250 ppm of each of the following amino acids: L-asparagine, L-aspartic acid, L-cysteine, L-glutamic acid, L-proline, and L-tyrosine. Growth increased as the glucose concentration increased. All amino acids except cysteine supported some growth in the absence of glucose. Tyrosine increased growth the most at each glucose level, although it furnished less nitrogen than any other amino acid utilized. Few microconidia were found in the basal medium (no glucose or amino acids). Microconidial numbers increased as the glucose concentration increased. Several amino acids increased microconidial sporulation in the absence of glucose, but none did so at the 0.3% glucose level. Proline and glutamic acid increased microconidial production at the 1.2% glucose level. No macroconidia formed in the basal medium, but production was increased by increasing glucose concentrations. Asparagine inhibited macroconidial formation, whereas proline, aspartic acid, and glutamic acid increased macroconidial numbers 2 to 3 times. However, the effect of amino acids on macroconidial formation was highly dependent on the glucose concentration.

Inhibition of the polysaccharide-degrading ability of Fusarium oxysporum f. sp. lycopersici culture fluids by a protein isolated from tomato stem cell walls. T. M. Jones,

A. J. Anderson, & P. Albersheim (Univ. Colo., Boulder). When cultured in liquid medium containing 1% cell walls (w/v) isolated from tomato stems as the sole carbon source, Fusarium oxysporum secretes a variety of polysaccharide-degrading enzymes. These include an endopolygalacturonase, a cellulase, α - and β -D-galactosidases, an α -L-arabinosidase, and a β -4-xylosidase. The enzyme activities appear in the culture fluid in a definite order: the polygalacturonase is first to reach maximal activity, followed by the glycosidases, with cellulase activity reaching a peak after the other enzyme activities have diminished. The ability of culture fluid enzyme mixtures to degrade cell walls isolated from tomato stems has been demonstrated by determining the nature of the material solubilized during treatment of the walls with enzyme mixtures, and by examining the composition of the cell walls prior to and following enzyme treatment. A protein extracted from tomato stem cell walls by treatment with 0.5 m potassium phosphate buffer, pH 7.1, has been observed to inhibit the action of the polygalacturonase and to prevent cell wall degradation by culture fluid enzyme mixtures. Similar polygalacturonase inhibitors have been obtained from Phaseolus vulgaris stems and from suspension-cultured Acer pseudoplatanus cells.

Production of chlamydospores by Phytophthora palmivora. Janice Y. Kadooka & W. H. Ko (Univ. Hawaii, Honolulu). Only thin-walled chlamydospores were produced by Phytophthora palmivora, using the submergence and low temperature method described by Tsao for P. parasitica. However, observations of naturally infected papaya fruits showed that as many as 90% of the chlamydospores had thick walls. Similar results were obtained by culturing the organisms for 4 weeks at 22 C in a liquid medium containing 50% papaya juice and 0.2% CaCOa. Only thin-walled chlamydospores were observed in the 1st week of growth in the papaya medium, moderately thick-walled chlamydospores in the 2nd week, and mostly thick-walled chlamydospores in the 4th week. The percentage germination of thick- and thin-walled chlamydospores on water agar was 85 and 92%, respectively. On V-8 juice agar, thick-walled chlamydospores did not germinate, whereas 80% of the thin-walled chlamydospores germinated.

Histology of onion leaves infected with Pseudomonas cepacia. S. O. Каwамото & J. W. Lorbeer (Cornell Univ., Ithaca, N.Y.). When stab-inoculated into the base of onion leaf blades, Pseudomonas cepacia moved into the leaf sheaths through the intercellular spaces. Although large bacterial masses occurred, separate small groups of bacteria frequently were formed in the intercellular spaces of both blade and sheath. Such distribution was observed when the lesions were periodically sprinkled with water. Except for the xylem vessels and epidermal cells, expanding bacterial masses in the intercellular spaces crushed the surrounding cells. In the blade, the large parenchyma cells were the first to be crushed by the bacterial masses; the smaller, more closely packed parenchymatous cells near the periphery of the blade were crushed later. Tissues composed of these cells eventually were macerated. In the sheath, bacteria were widespread among the loosely organized parenchyma cells adjacent to the adaxial epidermis. Bacterial masses and scattered bacteria in close association with host cell walls were commonly observed in this area. Bacteria in the intercellular spaces of the closely packed parenchyma cells beneath the abaxial epidermis usually were restricted to dense, compact masses.

Nature of aerial strands of Erwinia amylovora. H. L. Keil & T. van der Zwet (ARS, USDA, Beltsville, Md.). Erwinia amylovora produced abundant aerial strands on incollated Bartlett plants sprayed with 1% commercial spray oil. Morphology of the strands was studied under a scanning electron microscope. Strands were easily distinguished from natural host trichomes. They were brittle, and broke when crushed, resulting in irregularly fractured

ends which showed proportionately more matrix than bacteria. Strands, when dissolved in water and vacuum-evaporated, formed a network of adhesive material which connected the bacterial cells. When the weblike network was placed in ethyl alcohol, this material was dissolved, exposing individual cells. Smooth strands were most common, had about the same diameter throughout their length, and appeared to have a uniform quantity of matrix. We believe that this matrix was extruded at a constant pressure. Beaded strands appeared to be formed by irregular pressure. Oil apparently plugs the natural openings of the plant, thus creating internal pressure. As the bacterial matrix is forced out it solidifies upon exposure to air, and the strand increases in length as more matrix is added basipetally. Since bacteria in these strands consistently proved virulent, aerial strands in nature may play an important role in the epidemiology of fire blight.

The electrotactic response of zoospores of several species of Phytophthora fungi. K. L. KHEW & G. A. ZENTMYER (Univ. Cal., Riverside). Electrotaxis was studied with five species of Phytophthora: P. cinnamomi, P. palmivora, P. capsici, P. cactorum, and P. citrophthora. In deionized $\rm H_2O$ at 0.1-0.5 μa d-c. (< 1.2 v/cm), zoospores of all five species exhibited an excitation and increased swimming velocity as they approached the anode. This was usually followed by an active-oriented attraction and accumulation. As intensity of current increased, a repulsion zone appeared at the anode while a progressive immobilization of zoospores occurred at the cathode. The zoospores at the cathode exhibited a decreased swimming velocity, rotation, cessation of motion, and bursting. Studies were also made in solutions of various mono- and disaccharides and metabolic inhibitors in an attempt to interfere with the basic pattern of electrotactic response. None of these completely altered or inhibited electrotaxis without also affecting motility. Microelectrophoresis studies of swimming and pre-encysted zoospores indicated that both were negatively charged. Electrokinetic force could contribute in part to electrotaxis. Both chemical and electrical stimuli can induce a change in the membrane potential of zoospores which might serve as a signal to initiate a respective chemotactic or electrotactic response.

Pythium stem blight of beans. S. H. Kim & J. G. KANTZES (Univ. Md., College Park, Md.). Association of Pythium spp. with bean stem blight was observed in Maryland and Delaware during 1968, 1969, and 1970. Pythium ultimum was isolated from diseased bean (Phaseolus vulgaris) plants in the field during June, July, and September; P. aphanidermatum was isolated only during the warmer months of July and August. Both species, however, were isolated from the soil at all times of the year. Infection and the development of symptoms required the presence of water droplets containing the organism on susceptible tissues. When water droplets containing zoospores or mycelium evaporated within 8 hr of inoculation, disease development at 23 C was prevented. Maintaining water droplets on buds and leaves for 2 days in a moist chamber, or 5 days of continuous sprinkler irrigation in the field, permitted full disease development on Tendercrop bean plants inoculated with zoospores of P. aphanidermatum. Laboratory and greenhouse studies on P. aphanidermatum revealed that zoospores from oospores were as pathogenic as zoospores from sporangia. Zoospores were released from 1-year-old dry oospores in water. Thus, zoospores may be vital to the development of stem blight of beans caused by P. aphanidermatum.

Resistance of Xanthomonas dieffenbachiae isolates to streptomycin. J. F. Knauss (Univ. Fla. Agr. Res. Center, Apopka). Bacterial leaf spot and tip burn of Philodendron oxycardium incited by Xanthomonas dieffenbachiae is the most important foliar disease of foliage plants grown in Florida. Foliage grower applications of 200 to 400 ppm streptomycin under summer conditions at 4- to 7-day intervals for disease control were found ineffective, and ap-

peared to intensify disease development. Direct isolation of the pathogen from infected leaves taken from streptomycinsprayed plants, from foliage nurseries, and also from experimentally inoculated but nonsprayed plants onto potatodextrose agar containing streptomycin (100 to 1,000 ppm) revealed vigorous growth of the pathogen only from sprayed leaves. Growth occurred at all streptomycin concentrations. In isolations from nonsprayed leaves, only slight growth occurred at 100 ppm streptomycin with none at higher concentrations. Inoculations with streptomycin resistant isolates to *P. oxycardium* sprayed with 600 ppm streptomycin resulted in more severe disease development than did similar inoculations made with streptomycinsensitive isolates.

A selective medium for determining population of Rhizoctonia solani in soil. W. H. Ko & Frances K. Hora (Univ. Hawaii, Honolulu). A medium containing 1 g K₂HPO₄, 0.5 g MgSO₄·7H₂O, 0.5 g KCl, 10 mg FeSO₄·7H₂O, 0.2 g NaNO₂, 0.4 g gallic acid, 90 mg Dexon [sodium p-(dimethylamino) benzenediazosulfonate], 50 mg chloramphenicol, 50 mg streptomycin sulfate, and 20 g agar in 1 liter distilled water enhanced the growth of Rhizoctonia solani from soil and suppressed the development of undesired microorganisms. Recovery of R. solani from natural soil inoculated with sclerotia was 90-100% with this selective medium. For determining R. solani population in naturally infested soil, 1g of soil was evenly distributed in 10 clumps on a plate of selective medium and observed microscopically for *R. solani* mycelium after 24- and 48-hr incubation. The population of R. solani in natural soil ranged from 1 to 9 propagules/10 g dried soil. Soils inoculated with the same amounts of R. solani sclerotia were capable of causing pre-emergence damping-off of beet. Low populations of R. solani in soil may account for the unsuccessful application of the dilution plate method to this fungus. The combination of carbon and nitrogen sources selective for R. solani and inhibitors of undesired microorganisms is the basis for the selective medium.

The effect of H2O2 under acid conditions on strength, swelling, alkali solubility, reducing capacity, and weight loss of cellulose. J. W. Koenigs (Forestry Sci. Lab., Research Triangle Park, N.C.). In earlier studies, brown and white rot basidiomycetes produced extracellular H2O2. Formation of H₂O₂ by both groups suggested that H₂O₂ might be involved in the decomposition of cellulose. In vitro, H2O2 swelled cotton-fiber cellulose and increased slightly its alkali solubility and reducing capacity. H2O2 affected neither strength nor the reducing capacity or weight loss of cellulose subsequently treated with cellulase. In the presence of iron, weight loss increased with $\rm H_2O_2$ concentrations. tration, but strength loss increased much more rapidly, especially at low concentrations of H2O2; swelling, alkali solubility, and reducing capacity increased in a more linear relation to H2O2 concentration than strength loss. These results suggest that H2O2 produced by microbial enzymes may swell cellulose, and that previously unconsidered mechanisms could be involved in cellulolysis. H2O2-swollen cellulose may be a suitable substrate for assessing the role of certain cellulase components.

Effect of low temperature on germination of teliospores of Cronartium ribicola. R. G. Krebill (USDA Forest Serv., Logan, Utah). Along the southern limits of its known range in the Rocky Mountains, blister rust is most frequently encountered in white pines at elevations below 8,000 ft. This study was made to determine if low temperatures such as might occur in pine stands at higher elevations, inhibit the spread of the rust by depressing basidial sporulation. Telia from inoculated Ribes lacustre kept in growth chambers at 18 C days and 13 C nights were removed from leaves and subjected in petri plate moist chambers to constant temperature at 3-C intervals from —3 to 15 C for periods of 3, 6, 12, 24, 48, and 72 hr. Basidiospores were cast from telia by 12 hr at 15 C, by 24 hr at 6 C, and by 48 hr at 3 C. No basidiospores were

cast at 0 and -3 C. Casting rates were reduced at low temperatures; after 72 hr, total basidiospores cast per mm of telium averaged from a few hundred at 3 C to several thousand at 15 C. These results suggest that low temperatures might interact with a scarcity of long, moist periods, and greatly reduce the inoculum potential of Cronartium ribicola in high-elevation white pine forests of the central Rocky Mountains.

Some properties of chrysanthemum chlorotic mottle virus. S. P. KRYCZYNSKI, R. K. HORST, & A. W. DIMOCK (Cornell Univ., Ithaca, N.Y.). Current studies confirm that chrysanthemum chlorotic mottle is caused by a virus. Since the causal virus differs from any previously recognized chrysanthemum virus, and no evidence of relationship to other common viruses has been found, the name chrysanthemum chlorotic mottle virus (ChCMV) is proposed. A technique which assures a good, consistent infection level by mechanical (sap) inoculation has been developed. The top, completely yellow leaves from systemically infected plants of a clone of the chrysanthemum cultivar Deep Ridge were used as the virus source. Chilled glassware and cold distilled water were used for sap extraction. Succulent young leaves of healthy Deep Ridge plants were inoculated. Typical symptoms appeared in 14-20 days. When distilled water was replaced by a 1% suspension of bentonite, virus transmission was even more consistent and the incubation period shorter. No virus symptoms have appeared on any of 15 other plant species, representing 6 families, inoculated by the above technique. These included the most common virus indicator plants, some of them Compositae. ChCMV has never been recovered either from inoculated or from newly developed leaves of any of these plants.

Triarimol, (EL-273), a new fungicide for tart cherry disease control. D. H. LADE & C. D. CHRISTENSEN (Eli Lilly & Co., Plainwell, Mich., Cazenovia, N.Y.). Triarimol $[\alpha-(2,4-\text{dichlorophenyl})-\alpha-\text{phenyl}-5-\text{pyrimidinemethanol}]$ was applied to foliage of tart cherries with high-pressure hand guns on commercial schedules at 20-80 ppm in several replicated experiments in Michigan and New York. Harvested fruit was subjected to standard fruit quality determinations. Dodine at 300 ppm and Difolatan [N-(1,1,2, 2-Tetrachloroethyl) sulfenyl-cis-4-cyclohexene-1,2-dicarboximide] at 900 ppm were reference compounds. At harvest, triarimol at 30 ppm provided 98-99% control of cherry leaf spot (Coccomyces hiemalis) and 97% powdery mildew (Podosphaera oxyacanthae) control. Ninety to 100% brown rot (Monilinia fructicola) control was provided by 30-40 ppm. Dodine and Difolatan provided leaf spot and brown rot control, but were ineffective for the control of powdery mildew. Triarimol ranked between dodine and Difolatan for the prevention of defoliation from late season leaf spot invasion. Yield, ripening rate, terminal growth, fruit, and lateral bud set were not affected by triarimol. Fruit quality as determined by color, character, freedom from defects, freedom from surface pits, grade, U.S. No. 1, size, pH, and per cent sugar were also unaffected by triarimol.

Genetic evidence for pleiotrophic factors involved in the tumor-inducing ability of Agrobacterium tumefaciens. R. A. LANGLEY & C. I. KADO (Univ. Cal., Davis). N-methyl-N'-nitro-N-nitrosoguanidine was used to mutagenize cells of A. tumefaciens 1D135. Mutants auxotrophic for various amino acids, resistant to neomycin, kanamycin, and rifampicin, were tested for tumor-inducing ability. A number of histidine, leucine, and tryptophan auxotrophs were avirulent on sunflower. Addition of the nutritional requirement to the inoculum (1 mg/ml of the amino acid) did not restore virulence. Spontaneous revertants of histidine-avirulent auxotrophs simultaneously regained virulence. Neomycin- and kanamycin-resistant mutants were highly virulent, indicating that alterations in the 30S ribosomal proteins has no direct effect on virulence. Rifampicin-resistant mutants were also as virulent as the parental wild-type strain, suggesting that changes in the structural gene of

DNA-dependent RNA polymerase also are not directly involved in promoting virulence. These results indicate that the tumor-inducing ability is not due to a single gene product, but requires an alteration of the physiology of the bacterium.

Growth responses of strains of Botrytis cinerea tolerant and susceptible to 2,6-dichloro-4-nitroaniline. R. K. LANкоw (Univ. Calif., Davis). In liquid media, growth (dry weight) of DCNA-tolerant strains of *B. cinerea* was not affected by 2,6-dichloro-4-nitroaniline (DCNA) exceeding 2 g/liter. On solid media, radial growth and hyphal tip extension of tolerant strains were stimulated 60% by 0.1 g DCNA/liter. Spores of the tolerant strains produced germ tubes normally on solid and liquid media containing 2 g DCNA/liter. In DCNA-susceptible strains, dry weight increase in liquid media and radial and hyphal tip growth on solid media were inhibited more than 90% by 5 X 10⁻⁵ M DCNA. At this concentration, germination occurred, but 70-80% of the germ tubes burst and further development of intact germ tubes was inhibited. When crystals of DCNA were placed near hyphal tips of susceptible strains on solid media, the growth rate decreased within 5-10 min. Within 15-20 min, hyphal tips developed narrow extensions or became distorted. In some hyphal tips, the protoplasm became granular and was then discharged through an apparent rupture in the wall of the subapical region. When DCNA was replaced with 10 mg cycloheximide/liter, growth of both DCNA-tolerant and susceptible strains was inhibited; bursting and abnormal development of hyphal tips did not occur. DCNA may affect some aspect of cell wall metabolism in DCNA-sus-ceptible strains of *Botrytis cinera*. The mechanism of tolerance to DCNA is unknown.

Sequential development of pinwheel inclusions induced by a potato Y group virus. R. H. LAWSON & S. S. HEARON (ARS, USDA, Beltsville, Md.). A developmental sequence of pinwheel and bundle inclusions induced by sweetpotato russet crack virus (RCV) was studied in aphidinoculated leaves sampled at several time intervals. Bundle inclusions perpendicular to the cell wall were first observed after 5 days, and were separating from the wall after 7 days. Loop and circle configurations were occasionally observed in a 7-day infection. After 10 days, more bundles were free in the cytoplasm, and loop and circle configurations were common. An increased number of loops and circles was correlated with fewer intact pinwheels. Bands of membrane aggregates first observed in a 10-day infection were common in plants infected 2 months. Bundle and pinwheel development in a 7-day systemic infection appeared similar to a 5-day local infection, and a 10-day systemic infection appeared similar to a 7-day local infection. A single sampling of inoculated and systemically infected tissues does not reveal all stages of pinwheel and bundle development induced by RCV.

A Fusarium-induced root rot of staked tomatoes. J. V. Leary & R. M. Endo (Univ. Cal., Riverside). A root and crown rot disease of staked tomatoes caused by Fusarium oxysporum different from the classical vascular wilt has been observed in southern California. Plants in the vegetative stage are stunted and chlorotic, but not wilted. Following fruit set, leaves of severely affected plants may become interveinally necrotic or the plant may wilt and die. Mildly affected plants are only slightly stunted, but yield is reduced. The entire root system is affected, exhibiting a dry, brown rot of the cortex and xylem. The fungus enters the feeder roots and progresses slowly into the tap and lateral roots, primarily by means of intracellular hyphae through the cortex and secondarily through the xylem. The xylem is frequently discolored several cm in advance of the mycelium. Stem cankers may develop at or above the soil line. The fungus is rarely recovered from the stem more than 3 inches above the soil line either in field or inoculated plants. The entire range of symptoms has been reproduced in greenhouse-grown plants. Fusarium

wilt-susceptible and -resistant varieties (I gene) are both susceptible to this strain of Fusarium.

Enhancement of diagnostic symptoms of potato spindle tuber virus by manganese. C. R. Lee & R. P. Singh (Can. Dep. Agr., Fredericton, New Brunswick). The tomato cultivar, Allerfruheste-Freiland, has been used successfully as an indicator host for the mild strain of the potato spindle tuber virus (MPSTV). Recently, some inconsistency in symptom expression was encountered. Manganese nutrition was suspected. Tomato plants were grown in sand cultures containing various levels of Mn, Fe, or Zn. Increasing Mn from 0 to 9 μg/ml drastically increased the characteristic veinal necrosis symptoms with MPSTV, as well as with severe strain. At 9 μg/ml Mn, increasing iron up to 10 μg/ml decreased the necrosis. Zinc had no effect on the necrosis. Other tomato cultivars (Bonny Best, Michigan-Ohio, Rutgers, and Sheyenne) previously reported symptomless to MPSTV developed necrosis when grown in sand cultures receiving 9 μg/ml Mn.

Study on interaction of soybean mosaic and bean pod mottle viruses in soybean. YIH-SHYONG LEE & J. P. Ross (N.C. State Univ., USDA, Raleigh, N.C.). Inoculation of soybeans with soybean mosaic virus (SMV), followed 1 week later by bean pod mottle virus (BPMV), often produced top necrosis. Factors affecting this reaction and associated cellular changes were studied. Conditions favoring top necrosis included diurnally fluctuating (27 C-21 C) rather than constant temperatures, use of inoculum containing high SMV titers, and inoculation of young rather than old plants. SMV titers in young leaves and stem apices were highest 3 weeks after SMV inoculation when determined by serological and infectivity dilution end points and by virus particle counts. By the last method, SMV titers were twice as high in doubly as in singly infected plants. Neither light microscope observations for inclusion bodies nor electron-microscope observations for pinwheel, circular, and bundle inclusions revealed any consistent differences in number or size of the structures between SMV and SMV + BPMV-infected plants. BPMVlike particles and SMV inclusions were found in the same cells. Starch granules were more numerous and larger in healthy than in singly or doubly infected plants. Lipid globules in chloroplasts adjacent to necrotic tissue in doubly infected plants were larger than those in more remote tissues; lipid globules were rarely observed in healthy plants.

Inhibition of F-2 (zearalenone) biosynthesis and perithecia production in Fusarium roseum 'Graminearum'. LIEBERMAN, J. C. WOLF, H. R. G. RAO, & P. K. HAREIN (Univ. Minn., St. Paul). Dichlorvos (2,2-dichlorovinyl dimethyl phosphate) was applied at 10²-10⁻⁴ µg to 1-cm discs of Coons' medium agar seeded with spores of F. roseum 'Graminearum'. The carrier solvent was 10 uliters diethyl ether-dimethyl sulfoxide (97:3, v/v). Ten days after seeding, control discs had perithecia, whereas discs treated with 10 µg dichlorvos had none. Cultures of the fungus on moist, autoclaved rice grown at 25 C for 1 week and held at 12 C for 3 weeks yielded 150 ppm F-2 (zearalenone), whereas if the medium contained 20 ppm dichlorvos, no F-2 could be detected. When 5-g slices of a culture of Fusarium on moist, autoclaved rice were sprayed with 2.5 ml diethyl ether containing 1.8 mg dichlorvos, 1-14C-acetate was incorporated into F-2 only 50% as rapidly as in slices sprayed with ether alone. Vegetative growth of the fungus was not affected by the toxicant. These results suggest that production of perithecia may depend on F-2 in strains of F. roseum 'Graminearum' which normally produce the sexual stage.

Influence of transmissible diseases on toxin production in Helminthosporium maydis. G. D. LINDBERG (La. State Univ., Baton Rouge). Almost all isolates of Helminthosporium maydis obtained from diseased corn in Louisiana in 1970 were diseased. The agent of disease was readily transmitted to apparently healthy H. maydis (HM-T) by

hyphal contact inoculation with diseased H. maydis (DHM-T); symptoms of diseased colonies were lysed cells and arrested growth. HM-T frequently changed to diseased (DHM-T), as if spontaneously. The toxin produced by HM-T and DHM-T was > 10 times more inhibitory to the roots of Fla-200A corn (T-cytoplasm) than was the toxin produced by a diseased isolate of Hooker's race O of H. maydis. The diseased isolate of Hooker's race O produced a toxin 5 to 10 times more active against roots of La-9211 corn (regular cytoplasm) than the HM-T and DHM-T toxin. The agent of disease of DHM-T was not transmitted to Hooker's race O. Isolate BlspHM occurred as a variant sector in a colony of HM-T, and was obtained also from Hooker's race O. BlspHM was immune to infection by the agent of disease of DHM-T, and produced no toxin. The agent of a diseased isolate of BlspHM (D-BlspHM) was transmitted to BlspHM but not to HM-T. D-BlspHM produced toxin, and toxin activity was induced in BlspHM by transmission of the agent of disease. A close relationship was observed, therefore, in strains of H. maydis, to susceptibility to a given agent of disease in the fungus and to specific toxin activity.

Presence of an antifungal compound in peanut cotyledons. D. L. LINDSEY & R. B. TURNER (New Mexico State Univ., Las Cruces). Surface-sterilized seeds without testa from freshly harvested peanuts were found to inhibit the growth of Aspergillus flavus and Trichoderma viride on an agar medium. No inhibition of these fungi by seeds with intact testa, by testa from freshly harvested peanuts, or by seeds without testa from dried peanuts was observed. A material inhibitory to A. flavus was extracted with acetone from the cotyledons of freshly harvested peanut seeds kept for 3 days in sterile petri dishes containing 10 ml sterile distilled water. The inhibitor was further purified by extraction of the dried acetone extract with peroxide-free dry diethyl ether. The ether-soluble fraction was chromatographed on Silica Gel G thin-layer plates. An active zone was detected at R_F 0.4 using benzene:1,4-dioxane (8:2) as the developing solvent. Partition extraction has indicated the active material to be acidic.

Chemical composition of the oospore-oogonium wall complex of Phytophthora megasperma var. sojae. Eleanor LIPPMAN, D. C. ERWIN, & S. BARTNICKI-GARCIA (Univ. Cal., Riverside). A homothallic strain of Phytophthora megasperma var. sojae (P405) produced abundant oospores on clarified V-8 juice agar in the dark at 25 C. These oospores (enclosed in their oogonium walls) were separated from the mycelium by homogenizing the entire culture and washed by centrifugation. The cells were broken in a Teflon Potter-Elvehjem tissue grinder, and the walls separated by differential centrifugation. The final wall fraction was free of cytoplasmic contamination and contained oospore walls, oogonial walls, and probably some unrecognizable fragments of antheridial walls. The bulk of these walls was composed of alkali-insoluble glucose polymers (78%), the balance consisting of protein (10.6%), readily extractable lipid (5.7%), and bound lipid (5.6%). Only 7% of the wall dry weight was cellulose; the susceptibility of the walls to digestion of exo-β-1,3-glucanases or a mixture of endo-β-1,3- and endo-β-1,4-glucanase revealed that the remainder of the wall glucan consisted of β-1,3-linked polymers similar to those found in hyphal walls of Oomy-

Survival of Helminthosporium maydis, race T, in Georgia. R. H. LITTRELL & D. R. SUMNER. (Univ. Ga. Coastal Plain Exp. Sta., Tifton). Infectivity of southern leaf blight-damaged field corn residue was studied under natural and artificial conditions. Replicated field plots were disk-harrowed, rotary-chopped, or plowed. Leaf residues were also placed in wire cages in the field and in cloth bags in an open shed. Samples were collected 15 January-15 March 1971. Residue was separated into stalk and leaf-shuck tissue, washed in water and filtered and centrifuged, and total Helminthosporium conidia/ml were counted. The suspen-

sion was sprayed on cultivars containing both Texas male sterile (T) and normal (N) cytoplasm. Lesions were counted, and selected lesions placed in moist chambers to determine conidial production. Numbers of lesions produced from 1g of residue declined from 13 to 0.4 in tissue in field plots during the winter, but remained at 24-40/g in leaf tissue in cages and 55-97/g in leaf tissue in cloth bags. Filtrates from residue samples from nine counties collected in late February caused 0.2-13.8 lesions/g. Only occasional typical lesions were noted on N plants, and those lesions produced only one-half as many conidia as lesions on T plants. Infectivity was greatest from leaf and shuck residue, although more total Helminthosporium conidia were observed in filtrates from stalk tissue. No H. turcicum or H. carbonum conidia were observed.

Helminthosporium maydis and temperature, light, and humidity. R. J. Lukens, P. E. Waggoner, & J. G. Horsfall (Conn. Agr. Exp. Station, New Haven). Conidiophores of the T-strain form most abundantly at 18 to 20 C on washed hyphal fragments suspended in 0.02 M phosphate buffer (pH 6.3) and placed on filter paper. No conidiophores form at 10 and 35 C. Two to four times as many form in light as in darkness. Sporulation of conidiophores is inhibited by light at 23 C and warmer. Conidial formation is max at 18 to 23 C in darkness and 18 C in light, whereas no conidia form at 30 C even in darkness. Conidia appear after 4 to 7 hr of darkness at 23 C. Four to 5 days after inoculation of maize leaves, conidia form within 24 hr after wetting the lesions at 23 C. Spores germinate imperfectly, and do not form appressoria on glass slides at 94% RH. In water, spores germinate about 50% in only 3 hr at 23 or 35 C. At 23 C, appressorial formation is 50% after 6 to 9 hr, but appressoria are rare after 9 hr at 35 C. Keeping leaves wet for 3 hr after inoculation permits some infection, but max infection follows 24 hr of wetness. Lesion enlargement is optimum at 30 C, and proceeds about the same whether leaf surfaces are wet or

Histopathology of infection of bean hypocotyls by Sclerotinia sclerotiorum. R. D. LUMSDEN & ROBERTA L. DOW (ARS, USDA, Beltsville, Md.). Dome-shaped infection cushions formed on bean hypocotyls within a few hours after inoculation with S. sclerotiorum-infested oats. Ingress into host tissue occurred by multiple infection pegs that formed by dichotomous branching of hyphae within the infection cushion next to host cuticle. The pegs forcibly penetrated the cuticle and formed large, swollen, granular infection hyphae beneath the cuticle. Some infection hyphae penetrated the epidermis and cortical tissue directly, whereas others radiated from the infection site continuing beneath the cuticle. The subcuticular hyphae advanced ahead of the cortical hyphae and moved more rapidly up the stem than across. After 24 to 48 hr of incubation, the advancing subcuticular hyphae appeared to move into the cortex where further advancement up the stem occurred. Movement of infection hyphae through cortical tissue was always intercellular; however, branches of small hyphae from the inflated infection hyphae soon ramified throughout the invaded tissue, frequently in an intracellular manner. In older portions of the lesion, the small hyphae (before invading phloem and xylem tissues) formed structures often resembling infection cushions. These small ramifying hyphae probably are of secondary importance in pathogenesis, as compared to the inflated infection hyphae.

Datura stramonium, a local lesion host for certain isolates of cauliflower mosaic virus. M. Lung & T. P. Pirone (Univ. Ky., Lexington). Necrotic local lesions were produced on leaves of Datura stramonium inoculated with the Campbell, CM1841, KK cabbage, and New York 8153 isolates of cauliflower mosaic virus (CIMV). Lesions developed 10-15 days after inoculation, and lesion numbers were directly proportional to virus concentration. No lesions or other symptoms were produced on D. stramonium inoculated with the cabbage B isolate. Electron-microscopic

examination of cells within lesions induced by the Campbell isolate revealed the presence of inclusion bodies in the cytoplasm. These contained compactly aggregated virus particles, and resembled the inclusions found in cells of mustard (*Brassica pervirdis* 'Tendergreen') systemically infected with CIMV. The morphology of the particles was typical of CIMV. Typical CIMV symptoms were produced on mustard inoculated with extracts from local lesions produced on *D. stramonium* leaves inoculated with purified CIMV. *Datura stramonium* is thus the second reported host of CIMV outside the Cruciferae, and the only reported local lesion host for this virus.

Fusarium wilt of susceptible and resistant tomato isolines: host colonization. M. E. MACE & J. A. VEECH (ARS, USDA, Beltsville, Md.). The effects of the site of inoculation and inoculum concentration on disease development were investigated in susceptible and resistant host isolines. No killing, no stunting, but a nearly constant amount of vascular browning were observed in the resistant host inoculated with 5×10^6 , 5×10^4 , 5×10^3 , and 5×10^2 spores/ml. In contrast, the susceptible host exhibited progressive sensitivity to increased inoculum concentration. In both host cultivars, the amount of mycelial colonization of the hosts decreased as the distance from the point of inoculation increased, even though there was nearly systemic distribution of fungus spores through the axis xylary system. The site of inoculation greatly affected the extent to which the host was colonized by the pathogen. Both the susceptible and resistant hosts showed increased resistance to colonization as the site of inoculation moved up the plant axis. It was concluded that two morphologically distinct zones, tissue above the cotyledonary node and tissue below the cotyledonary node, should be considered separately in comparative physiological or biochemical studies of susceptibility and resistance to Fusarium wilt of tomato.

Germination self-inhibitor of sunflower and snapdragon rust uredospores. V. Macko, R. C. Staples, & J. A. A. Renwick (Boyce Thompson Inst., Yonkers, N.Y.). The endogenous germination self-inhibitor of Puccinia helianthi and P. antirrhini uredospores has been identified as methyl 3,4-dimethoxycinnamate. Purification involved extraction with water, partition into ether, molecular distillation, and thin-layer (TLC) and gas chromatography (GLC). Analyses of the highly purified natural compound by infrared and mass spectrometry showed that it was identical to synthetic methyl 3,4-dimethoxycinnamate. In addition, both natural and synthetic compounds had identical R_F values in two different TLC systems, and identical retention times in two different GLC systems. The ED₅₀ of the natural and synthetic inhibitor for spores of both rust species was 5 × 10⁻³ µg/ml.

Properties of DNA bacteriophages from Agrobacterium tumefaciens. R. J. Manasse & R. C. Staples (Boyce Thompson Inst., Yonkers, N.Y.). DNA bacteriophages from related pathogenic and nonpathogenic strains of Agrobacterium tumefaciens (IIBV7 and IIBNV6, respectively) were isolated. The phage isolates examined, two from each bacterial strain, were shown to be composed of hexagonal heads with long flexible tails containing a set of six appendages. Phage isolate LIIBV7-1 had a sedimentation constant (S_{20,w}) of 596, a molecular wt of 72 × 10⁶ daltons, and a buoyant density of 1.5110. The adsorption rate constant is 7.1 × 10⁻¹⁰ cm² sec⁻¹, and the eclipse and latent periods are 60-75 and 135-150 min, respectively. Similar data on both the physical and biological properties of three other phage isolates have been obtained.

Artifact pseudocolonies on solid agar media inoculated with aster yellows or healthy plant material. K. Maramorosch, H. Hirumi, & B. Playsic-Banjac (Boyce Thompson Inst. Plant Res., Yonkers, N.Y.). Attempts to culture the mycoplasmalike presumptive agents of aster

yellows and corn stunt in liquid or solid media have not yet been successful. Commercial mycoplasma media containing serum agar were inoculated with phloem tissue and phloem sap from aster yellows-diseased Nicotiana rustica plants. Controls consisted of media inoculated with phloem tissue and sap from healthy plants, and of non-inoculated agar serum. Nonliving artifacts, resembling pseudocolonies described by Laidlaw (1925) and Brown et al. (1940), mimicking mycoplasma colonies in time of appearance and in size developed in all plates. Careful microscopic observation showed that these artifacts did not have the round granular "fried egg" or the vacuolated morphology of mycoplasma colonies. The pseudocolonies "grew" and could be "subcultured", but did not consist of mycoplasmalike bodies, as was ascertained by electron microscopy. Pseudocolonies are probably crystalline formations of calcium and magnesium soaps.

The application of an electronic cell counter to the study of growth of insect cell monolayers. G. Martinez-Lopez & L. M. Black (Univ. Ill., Urbana). The growth of Agallia constricta cell monolayers (AC20 cell line) was studied by using the Coulter Cell Counter at various times to count the number of AC20 cells present in cultures under the influence of several factors. The insect cells were grown in Chiu & Black's modification of Schneider's medium. A satisfactory seeding density for a 4-day interval between subcultures was between 2.5 and 3.0 × 106 cells for each flask with a 25 cm2 area for cell attachment. The optimal temperature for cell growth was estimated to be between 28 and 29 C. No differences in growth were detected at any temperatures from 27 to 30 C, but at 24 C the rate of growth was less, and at 33 C, cells died. A 10% concentration of fetal bovine serum was as good or better than 20%, and permitted a considerable economy in preparing the medium. Studies on the effect of infection by wound tumor virus (WTV) on the growth of AC20 cells provided definite evidence for cytopathic viral action. This effect was clearly observed after cells were subcultured, since the attachment of WTV-infected cells was about 25 to 30% less than that of cells free of WTV.

Effect of carboxin on mitochondrial activities. D. E. MATHRE (Montana State Univ., Bozeman). Carboxin was tested for its effect on the activities of mitochondria from several fungi, pinto beans, and rat liver. Succinate oxidation by mitochondria from the sensitive fungus, Ustilago maydis, was inhibited by low concentrations of carboxin, the K1 (inhibition constant) being 0.32 µm. The inhibition was noncompetitive. Succinate oxidation was also inhibited in the mitochondria from other sources, but not to such an extent as in those from U. maydis. Carboxin had little effect on the oxidation of reduced nicotinamide adenine dinucleotide. Carboxin did not appear to uncouple oxidative phosphorylation in the presence of succinate in tightly coupled rat liver mitochondria, but did decrease the respiratory control ratio. It is believed that carboxin inhibits mitochondrial respiration at or close to the site of succinate oxidation, and does not greatly affect the remaining portion of the electron transport system or the coupled phosphorylation reactions.

Influence of growth media on ultrastructure of hyphal tips of Sclerotinia sclerotiorum. D. P. Maxwell & P. H. Williams (Univ. Wis., Madison). The possible functional role of vesicles and crystal-containing microbodies (CB) in the production of oxalate, endopolygalacturonase (PG), or cellulase (Cx) by Sclerotinia sclerotiorum was investigated. The fungus was grown on mineral salts-yeast extract medium with glucose, glucose-succinate, sodium polypectate, or carboxymethylcellulose (CMC) as the carbon source. Relative Cx production on these media was < 0.5, 3.4, 179, and 384, respectively; while relative PG production was < 0.5, < 1, 211, and 23, respectively. Oxalate accumulation was 1.1, 1,600, 851, and 641 µg/mg dry wt of hyphae, respectively. No unique organelle was detected in tips grown on media inducing high PG or Cx produc-

tion. More CB's (about 3 times) were present in tips grown on polypectate and CMC than in tips grown on the other media. More membrane-bound vesicles (0.09-0.18 μ diameter) were associated with tips grown on the glucose-succinate medium than with tips grown on the other media. These results do not support a functional role for CB's in either PG, Cx, or oxalate production, but they do show that the growth medium can influence the ultrastructure of hyphal tips.

Relationship of moisture and light to ascospore discharge by Mycosphaerella ligulicola. R. E. McCov & A. W. DIMOCK (Cornell Univ., Ithaca, N.Y.). Ascospore discharge by Mycosphaerella ligulicola on diseased Chrysanthemum morifolium tissue was measured under controlled environmental conditions by means of a spore trap placed within a dew chamber. Moisture and, in some cases, light were found to be the controlling factors regulating violent discharge. Liquid water was required for discharge in all cases. Ascospore discharge occurred within 30 min after the sprinkling of dry perithecium-bearing tissue with water. When the tissue was wetted by dew, ca. 2 to 2.5 hr of condensation was required before discharge began. The rate of condensation of dew was 4.5 mg/cm² per hr, indicating a requirement for ca. 0.1 mm of moisture accumulation to initiate discharge. With 3 isolates (from California and New York), discharge occurred only in darkness; switching on lights (combination incandescent & cool-white fluorescent, 1,750 µw/cm2) prevented or stopped discharge even after a dew period which provided the requisite moisture had occurred in darkness. Ascospore discharge by a fourth isolate was not inhibited by light.

The distribution and fate of southern bean mosaic virus in the bean seed. J. G. McDonald & R. I. Hamilton (McGill Univ., Montreal, Quebec, Can.). Southern bean mosaic virus (SBMV) was recovered from immature embryos and seedcoats harvested from SBMV-infected Phaseolus vulgaris 'Logan'. However, when the embryo and seedcoats were treated to remove surface contamination, infectivity was eliminated from the embryos, but not from the seedcoats. Crude extracts of immature infected seedcoats were analyzed by sucrose density-gradient centrifugation, revealing a component, C2, sedimenting slightly faster than the SBMV monomer, C1. With drying of these seedcoats, the relative amount of C₂ increased, while that of C₁ declined. A third component, C₃, sedimenting faster than C2, was also observed in these preparations. These components were not observed in noninoculated controls, and they reacted positively with SBMV antibody. The OD 260:280 nm ratios of both C2 and C₃ (1.2-1.3) were considerably lower than that for SBMV monomer (1.55). With maturation of infected seedcoats, only a peak corresponding to C2 was observed, suggesting that the maturation process caused a drastic alteration in the morphology of SBMV.

Effect of soil porosity, moisture, and temperature on diffusion of 1,3-Dichloropropene nematicides. M. V. Mc-Kenry & I. J. Thomason (Univ. Cal., Riverside). Using techniques for direct chemical analyses (gas chromatography), the diffusion pattern of cis- and trans-1,3-Dichloropropene and 1,2-Dichloropropane was monitored under field and laboratory conditions. The three components of Telone diffuse at independent rates in soil. Increasing the soil moisture slowed the rate of diffusion. The diffusion patterns in a dry clay loam or sandy loam were similar. As moisture was increased, the rate of diffusion was decreased, depending upon the total volume of continuous soil air spaces. In soils with very little air space, the gas did not diffuse until pore spaces were sufficiently drained. In a very dry soil, adsorption to soil particles resulted in a complete lack of diffusion. Increases in temperature from 5 to 25 C resulted in a faster rate of diffusion, but also increased the rate of degradation of the 1,3-Dichloropropenes. In a clay loam soil, the maximum concentrations of trans-1,3-Dichloropropene were achieved 6 inches from the point of injection in 1.5 days when dry, 2.5 days when moist, and 9 days when soil was wet.

An electrophoretic examination of selected isoenzymes in stubborn diseased citrus. C. W. Meister, R. M. Allen, & R. L. Caldwell (Univ. Ariz., Tucson). Enzymes from citrus leaves are being examined to find a simple, inexpensive, and rapid method for diagnosing stubborn disease. Leaves of healthy or diseased 5-year-old citrus trees were homogenized in Tris buffer, pH 7.5, and clarified by centrifugation. Protein was precipitated with trichloroacetic acid and measured by the Lowry method. Equal quantities of protein were layered on 8% or 5-15% gradient polyacrylamide gel columns and subjected to electrophoresis with Tris glycine buffer, pH 8.3. Twenty-eight enzymes have been identified by using specific reaction stains. Isoenzymes extracted from leaves of healthy plants are being compared with those from similar leaves of plants suffering from stubborn or psorosis diseases, drought, or mineral deficiencies. Preliminary results indicate that leaves from stubborn diseased plants have higher activities of peroxidase, leucine amino peptidase, and shikimate dehydro-genase than those from healthy plants. Lower activities of catalase and malate dehydrogenase have been consistently noted in stubborn-diseased plants.

Epiphytic pseudomonads on soybean in the field. T. W. MEW & B. W. KENNEDY (Univ. Minn., St. Paul). A modified Kado's medium, selective for Pseudomonas, was used to detect populations of bacteria on leaves of three fieldgrown cultivars of soybean (Glycine max). By use of UV irradiation and leaf prints, we were able to demonstrate that the pseudomonads were in fact epiphytes, and that more saprophytic pseudomonads were on leaves early in the season than late. In the greenhouse, Phaseolus vulgaris 'Bush Blue Lake' was readily infected with isolates of epiphytic pseudomonads pathogenic to soybeans, and showed more or less typical water-soaked lesions, but there were no symptoms of systemic infection. As indicated by five major characters of Pseudomonas, it was evident that the pathogenic epiphytes were among the Group-I pseudomonads. It is concluded that epiphytic pseudomonads pathogenic to soybeans compose a unique group, but vary with the season and with genotype of the host

Pathogenicity of Heterodera virginiae to Va. 312 darkfired tobacco. L. I. MILLER (Va. Polytech. Inst. & State Univ., Blacksburg). The pathogenic effect of the horsenettle cyst nematode (Heterodera virginiae) to Va. 312 dark-fired tobacco was evaluated in microplots (metal cylinders 35 cm in diameter imbedded in soil to a depth of 90 cm and filled with methyl bromide-fumigated Woodstown fine sandy-loam soil). Cysts were mixed with the soil of six plots so that the infested soil contained an average of 30 eggs and/or larvae per g of air-dried soil. Six other plots were not infested. Single tobacco seedlings were transplanted to each microplot the 2nd week of May. Plants in the noninfested plots were harvested the 3rd week of August when they were judged to be of maximum quality and yield. Plants in infested plots were harvested the 3rd week of September to obtain the maximum yield. Average diameter at the base of the stalk from noninfested plots was 37 mm, and 21 mm from infested plots; dry wt of roots from noninfested plots was 46 g, and 20 g from infested plots; and dry wt of all aboveground parts was 205 g from noninfested plots and 76 g from infested plots. Soil samples collected the 4th week of September from infested plots averaged 98 eggs and/or larvae per g airdried soil.

Some physical properties of elm mosaic virus. H. E. Moline, G. L. McDaniel, & D. E. Mayhew (Iowa State Univ., Ames). A virus isolated from Ulmus americana (Moline elm) in Iowa is serologically related to the type strain of elm mosaic virus (EMV). Shadowed, negatively stained, and fixed tissue preparations were examined by

electron microscopy. The virus is a small polyhedron. It was associated with necrotic ringspots in inoculated leaves of Chenopodium quinoa. Infectivity is associated with EMV which consists of particles of three densities, as determined by analytical ultracentrifugation: top, middle, and bottom components with $S_{20,w}$ values of 45, 65, and 92, respectively. The middle component made up more than 60% of the total volume of purified preparations. The bouyant density of this component is $1.3326 \, \text{g/cm}^3$. The isolate has a dilution end point of 10^{-4} - 10^{-5} from systemically infected C. quinoa leaves. The longevity in vitro of EMV is 4 days in neutral 0.01 M phosphate buffer; it is inactivated by heating for 10 min at 62 C but not at 60 C. The Iowa EMV isolate does not seem to differ markedly from others previously described.

Preparation and storage of phytopathogenic bacteria in liquid nitrogen. L. W. Moore & R. V. Carlson (Oregon State Univ., Corvallis). Liquid nitrogen was evaluated as a method of storage for species of Agrobacterium, Erwinia, Pseudomonas, and Xanthomonas. Bacteria in exponential growth were harvested (108 to 109 colony-forming units/ ml) from a liquid shake-culture consisting of 4% extract, 20% dextrose, 0.4% peptone, and 0.5% (NH₄)₂SO₄ at pH 7. This medium plus 1.5% agar was used for viability counts (spread-plate method) before and after freezing. The harvested bacteria were frozen directly in their growth medium, or centrifuged and the pellet resuspended for freezing in the following media: fresh medium, fresh medium containing 1% dimethyl sulfoxide, 10% skim milk, or distilled water. Glass ampules containing 0.5-ml aliquots of a bacterial suspension were heat-sealed and plunged into liquid nitrogen. Recovery of viable bacteria was periodically determined over a 2-month period by thawing the ampule contents for 12 sec at 40 ± 1 C, diluting, and plating. Optimum recovery from a given freezing medium varied among the nine species of the four genera; recovery percentages were within a range of 65 to 118% of the initial viable counts.

Proteolytic enzyme production by Verticillium alboatrum. H. W. Mussell & Blanche Strouse (Boyce Thompson Inst., Yonkers, N.Y.). Twelve isolates of Verticillium albo-atrum from cotton produced extracellular proteolytic enzymes when grown using soluble or insoluble proteins as a sole carbon source. Proteolytic enzymes were not detected when glucose was used as a sole carbon source, but low levels of glucose with proteins enhanced enzyme production 3- to 6-fold over that induced by proteins alone. Neither amino acids nor the polypeptide glutathione induced proteolytic enzymes when used as the only carbon source or with glucose. Proteolytic activity had a pH optimum of 8.5, and was stable in solution for several months under aseptic conditions. Polygalacturonase activity was higher on media that induced proteolytic enzymes than on comparable media containing glucose. Cellulase activity, on the other hand, was uniformly low on glucose and protein-based media.

Etiology of Monochaetia mali in apple. R. NAIK & D. Powell (Univ. Ill., Urbana). Monochaetia mali has been known since 1900, but its etiology in apples has not been studied. In the current studies, the organism infected twigs through ovipositor wounds caused by cicada, and produced cankers. Infection occurred only through wounded tissue. Cankers were produced by inoculation of mature apple tree limbs. Inoculation of fruit produced lesions which were convex, light brown, and enlarged with age. Acervuli, which formed within 8-10 days after inoculation, produced characteristic conidia which were several-celled and dark, with hyaline-pointed end cells and a single apical appendage. Similar fruiting structures were found at the margins of limb cankers. The fungus grew well on lima bean agar. Optimum temperature for growth in vitro was between 25 and 30 C, and optimum pH was 6 with extremes of 3 to 9. Susceptibility of inoculated fruit was directly correlated with increase in tissue pH and warm

temperature of ca. 30 C. The incubation period was shorter in the field on mature fruit (4 days) than on 20-day younger fruit (5-7 days).

Pseudomonas glycinea inhibits germination of soybean seed. J. F. Nicholson & J. B. Sinclair (Univ. Ill., Urbana). A new role of Pseudomonas glycinea in soybean pathology was studied. Two isolates of an internally seedborne bacterium in soybean (Glycine max) inhibited the germination of Lee 68 and Amsoy soybean seed. In culture, one isolate had a smooth surface and margin, whereas the other had a rough surface and margin. Both isolates were identified as P. glycinea through a series of standard biochemical tests compared to stock cultures of P. tabaci and P. glycinea. Our isolates and the stock isolate of P. glycinea did not grow on Kado's selective medium D4 for Pseudomonads. When suspensions of three P. glycinea isolates were infiltrated by vacuum into sterilized Amsoy seed, germination was significantly inhibited by the roughmargined and stock culture isolates to 38 and 68%, respectively, whereas the smooth-margined isolate had normal germination (84%) compared with the control (88%). The bacterium was isolated from 17 seed lots of Lee 68 soybean collected from five states. The occurrence in individual seed lots was as high as 64% and as low as 3%.

The effect of \beta-sitosterol on the activity of certain enzymes of Phytophthora cactorum. C. Norman & R. A. CALDERONE (W. Va. Univ., Morgantown). β-Sitosterol (10 mg/liter) increased activity of aldolase, L-glutamic acid transaminase, and glucose-6-phosphate dehydrogenase of Phytophthora cactorum when added to cultures growing in a liquid glucose-asparagine medium at 25 C. The increase in activity generally occurred 48-72 hr after the addition of the sterol to 5-day-old cultures. Specific activities (units of activity/µg protein of cell-free extract) of both aldolase and transaminase was 1.6-1.7 times greater in cultures containing β-sitosterol than in the controls (no sterol). Likewise, the specific activity of glucose-6-phosphate dehydrogenase was 1.3-1.4 times greater than the control. The sterol-induced increases in enzyme activity paralleled increases in growth (mg dry wt) of the fungus over control cultures.

The influence of exogenous cytokinins on the development of the bacterial hypersensitive reaction. A. Novacky (Univ. Mo., Columbia). When tobacco leaves were treated with 5-10 μg/ml of cytokinins [kinetin, 6- (γ,γ-dimethylallylamino) purine or 6- benzylaminopurine] 48 hr prior to infiltration with 5 × 106 or 107 cells/ml of Pseudomonas pisi, development of the hypersensitive reaction (HR) was overcome. The initial appearance of HR was identical in leaves infiltrated with water or with cytokinins. In the former, the HR progressed rapidly from the initial shinyness and faint wrinkling of lower leaf surface to necrosis. In the latter, the initial reaction did not progress beyond the initial symptoms. Bacterial growth was not affected by cytokinins. Age of the treated leaves was critical in demonstrating the "cytokinin effect" on HR. Only fully expanded leaves showed this phenomenon. Extending the pretreatment period with cytokinin to 72 hr with an additional one or two applications of cytokinins prevented HR induction by 108 cells/ml. Other consequences of treatment of the tobacco with cytokinins were an increased resistance to water stress and a reduction in electrolyte leakage when compared with watertreated controls. Cytokinin-treated tissues also revealed lower peptidase activity. From these experiments we have postulated that cytokinins in some way preserve the integrity of vital plant cell membranes which are generally altered as a consequence of HR.

Remission of symptoms of pear decline in pear and peach X-disease in peach after treatment with a tetracycline. G. NYLAND (Univ. Cal., Davis). Recent reports indicate that mycoplasmalike organisms incite pear decline

and peach X-disease. One tree from each pair of 15 pairs of Comice and 15 pairs of Bartlett pear trees was treated with 3 liters of tetracycline hydrochloride solution (100 µg/ml) in May 1969. The solution was fed into the xylem of each tree by gravity via plastic tube connectors and plastic tubing attached to reservoirs suspended 3-4 feet above 4 equally spaced holes drilled into the trunk. All trees had shown symptoms of the leaf curl type of pear decline during each of the previous 2 seasons. Symptoms of leaf curl did not develop in 1970 in any of the 30 trees treated with tetracycline hydrochloride. Typical symptoms of leaf curl developed in 29 of 30 untreated trees. Four peach trees showing X-disease symptoms were treated with 2 liters of tetracycline hydrochloride solution (34 µg/ ml) fed into diseased branches in September 1969 plus 2 additional liters (1 µg/ml) fed in April 1970. Only untreated trees showed symptoms of X-disease in 1970. The treated ones remained symptomless.

Biological activity of conversion products of benomyl. J. M. Ogawa, Elaine Bose, B. T. Manji, E. R. White, & W. W. KILGORE (Univ. Cal., Davis). A mixture of 8 oz of 50% benomyl and Bordeaux mixture 10-10-100 (field dosages) formed a compound identified as an s-triazinobenzimidazole (STB). Technical benomyl placed in a solution adjusted to pH 12 with NaOH formed the same compound. The spectrum of activity of STB on 16 test fungi was the same as that of benomyl. For total suppression of Monilinia laxa on potato-dextrose agar, 10 to 50 ppm of STB were required, whereas for benomyl and MCB (methyl 2-benzimidazole carbamate), only 0.1 ppm was required. Using Botrytis cinerea as a test pathogen, 30 detached Drake almond blossoms/treatment were sprayed with each chemical, inoculated with a spore suspension, and incubated at 20 C and over 90% RH. At concentrations of 1 and 10 ppm, STB reduced infection by 10 and 90%; and benomyl, 23 and 80%, respectively. Seven single tree replications of Drake almond trees sprayed with field dosages for the control of M. laxa at the pink bud stage of bloom showed an average of 60, 23, 90, and 593 blighted shoots for STB, benomyl, benomyl plus Bordeaux mixture, and control, respectively. STB was used at the same concentration as benomyl.

Host specificity of Polyporus betulinus. R. L. Paine & W. MERRILL (Pa. State Univ., Univ. Park). Polyporus betulinus is restricted to Betula spp. in nature. Our previous studies of the mechanism of this host specificity showed that spores of the fungus germinated on host and nonhost species. Sapwood and heartwood of 40 trees each of Betula lenta, Picea abies, and Pinus strobus were inoculated with basidiospores or grain inoculum of P. betulinus. The fungus could not be recovered from Betula or Pinus 6 months later; it was recovered from P. abies after 6 months from four wounds on three trees inoculated with spores or grain inoculum, and after 1 year from one heartwood wound inoculated with spores. In the latter, the fungus was recovered from the margins of a pocket of yellowish-brown decay 8 × 3 cm in diameter. Identification of all isolates was confirmed by hyphal anastomoses with known isolates of P. betulinus. Host specificity of P. betulinus appears to be controlled by factors other than those acting to prevent spore germination and subsequent colonization. The multitude of competing microorganisms in wounds from which P. betulinus could not be recovered suggests that host specificity may be due partially to factors which control growth of these competitors.

Enhancement of Pseudomonas phaseolicola infection of bean by 3':5'-cyclic AMP. N. J. Panopoulos & M. N. Schroth (Univ. Cal., Berkeley). Infection of bean by Pseudomonas phaseolicola is regulated in a manner suggesting catabolite repression. Bacterial inocula (10⁵ cells/ml) were suspended in 2 to 5 mm solutions of glucose, glycerol, or other sugars, and infiltrated into the intercellular spaces of primary leaves of red kidney beans. The number of lesions was reduced by 50 to 60% in the pres-

ence of glucose or glycerol, compared to water and osmotic controls. Since catabolite repression of inducible enzyme formation in bacteria has been experimentally related to a lowering of the intracellular level of 3':5'-cyclic AMP, we examined whether infection was affected by this or other cyclic nucleotides. 3':5'-Cyclic AMP, 2':3'-cyclic AMP, and 3':5'-cyclic UMP at concentrations ranging from 2 to 5 mm (pH adjusted to 6.8-7.0 with NaOH) enhanced infection 2- to 5-fold over water controls. In contrast, ATP, the biochemical precursor of 3':5'-AMP, inhibited infection by 50 to 70% under these conditions.

Plant water stress and the development of Fusarium foot rot in wheat. R. I. PAPENDICK, R. J. COOK, & P. J. SHIP-TON (ARS, USDA, NATO Fellow, Pullman, Wash.). Foot rot of winter wheat (Triticum aestivum) caused by Fusarium culmorum is most severe in low rainfall areas in fields with high N fertility and early fall-seeded to semidwarf cultivars with high tillering capacity. Such conditions hasten extraction of the limited soil water and onset of plant water stress. Soil water depletion rates during May in field plots of Nugaines and Moro were generally greater with higher N (up to 200 lb./acre) and narrower row spacings (12 vs. 24 inches). Leaf water potentials measured by thermocouple psychrometry decreased steadily from to -27 bars in early May to -33 to -35 bars or less in mid-June. Potentials were generally lower, and onset of stress was earliest in plots with high N and narrow row spacings. Infection of crowns by F. culmorum was uniformily high in all treatments on 1 April, and remained latent through May, after which severe basal culm decay began occurring under greatest water stress. Five cultivars showed a positive correlation between onset of stress, onset of severe foot rot, and yield reduction, suggesting that disease control may be possible by breeding for cultivars having high water-use efficiency. A sixth variety that showed early stress had negligible foot rot and yielded well, suggesting that stress alone could not account for the yield reduction.

Exogenous carbon and nitrogen requirements for germination of conidia by Aspergillus flavus. T. Pass & G. J. Griffin (Va. Polytech. Inst., Va. State Univ., Blacksburg). In a phosphate-buffered (pH 5.7), inorganic salts solution (B), almost full dependence on exogenous carbon and partial dependence on exogenous nitrogen were found for complete germination of washed conidia by A. flavus over a range of conidial densities. At equivalent carbon, an amino-acid mixture supported higher germination than a sugar-organic acid-alcohol mixture plus NH4Cl; proline or alanine alone supported higher germination than several other single amino acids or single sugars plus NH4Cl. Glucose plus NH4Cl was the most stimulatory of the latter. Germination was more dependent on exogenous nitrogen as the conidial density increased from 103 to 105 conidia/ ml. At equivalent nitrogen, proline alone or an amino acid mixture supported higher germination than NH+4, NO-3 or several other single amino acids tested in B solution plus glucose. Percentage germination decreased as the conidial density decreased from 10^5 to 10^3 conidia/ml when glucose or glucose plus NH4Cl were supplied at constant amounts/conidium. However, little or no decrease in per-centage germination was observed in glucose plus an amino acid mixture, and only a slight decrease was observed in glucose plus peptone. Germination was best at 30-35 C, while a broad pH optimum of 3.0-8.0 was found.

Role of benzylisothiocyanate in the resistance of papaya fruit to postharvest fungal diseases. S. S. Patil, C. S. Tang, & J. E. Hunter (Univ. Hawaii, Honolulu, & Hilo). The level of benzylisothiocyanate (BITC), a compound ubiquitous in papaya fruit, decreases upon ripening of harvested fruit, and this decrease is inversely correlated with the incidence of postharvest diseases. In vitro studies on sensitivity of papaya pathogens to BITC showed that Rhizopus stolonifer and Phytophthora palmivora were completely inhibited at 40 ppm of BITC. At the same con-

centration, Stemphylium sp. and Phomopsis sp. were inhibited ca. 50%; and colonies of Colletotrichum gloeosporioides, Aschochyta cariacae, Botryodiplodia theobromae, and Fusarium solani were inhibited 30-40%. Control experiments were conducted on mature-green fruits which were wound-inoculated with R. stolonifer, incubated for 24-48 hr at 25 C, then sealed for 6 days in plastic boxes with paper discs impregnated with BITC. Two days after removal of papayas from BITC atmosphere, the diameters of rotted areas were measured. Rotting was reduced 50-60% at 60 µliters of BITC, and the papaya remained 50% greener than controls. Thus, BITC has potential for control of postharvest fruit rots and also as a retardant of papaya maturation.

Synthesis of tobacco mosaic virus nucleic acid in tobacco tissue culture. L. E. Pelcher & H. H. Murakishi (Mich. State Univ., E. Lansing). Suspension culture cells of Nicotiana tabacum 'Havana-38' were inoculated with 250 µg/ml of tobacco mosaic virus (TMV). The cells were then washed with fresh medium and incubated on agar. At 12-hr intervals after inoculation, cells were transferred to 2 ml fresh medium containing uridine-3H at a concentration of 25 μc/ml. Cells were allowed to incorporate uridine-3H for 12 hr. The cells then were harvested and washed, and the RNA was extracted by a modified SDS-phenol method. The extracted cellular and viral RNA then were separated by electrophoresis in 2.4% polyacrylamide gels. The gels were scanned at 260 nm, frozen in dry ice, and sliced into 1-mm sections. Radioactivity in each section was determined in a liquid scintillation counter. A discrete optical density peak corresponding to viral RNA could be detected as early as 36 hr postinoculation. The viral RNA OD peak increased until 96-120 hr, after which time little further increase could be detected. At 12-24 hr, uridine-3H incorporation into viral RNA could be detected. The rate of uridine-3H incorporation into viral RNA increased until 48-84 hr after which time the rate of incorporation of uridine-3H appeared to decrease throughout the 7-day period studied.

Effectiveness of benzimidazole and two benzimidazole derivative fungicides in reducing ozone injury to Phaseolus vulgaris 'Pinto 111'. M. Pellissier, N. L. Lacasse, & H. Cole, Jr. (Pa. State Univ., University Park). A hypothesis that protection from ozone injury on benomyl-treated plants was due to the benzimidazole moiety of benomyl was tested. Benzimidazole (Bd), benomyl (Bl), and another Bd-derivative fungicide, thiabendazole (Tb) [2-(4-Thiazolyl)-benzimidazole] were incorporated into a loam soil-peat-perlite (2-1-1) mixture. Equivalent amounts of each compound based on molecular weight were used at these Bd rates: 20, 40, and 80 µg/g (ppm) soil (dry soil wt basis). Plants grown in the treated soil mixture were fumigated under controlled conditions with 25 ppm O₃ for 4 hr. Amount of plant injury was evaluated 3 days later. Nontreated plants showed 50 to 60% leaf injury. Concentrations of Tb lower than 40 ppm did not give protection, but at 80 ppm a 50% reduction in injury occurred. Plants treated with Bl had 90% less injury than the nontreated plants at a concentration of 40 ppm, and complete protection was obtained at 80 ppm. A concentration of 40 ppm Bd provided 100% protection. Results indicate that the Bd moiety of each compound may be responsible for the protection from ozone injury observed in Bltreated plants.

A compound from onion roots inhibitory to Pyrenochaeta terrestris. F. L. PFLEGER & E. K. VAUGHAN (Oregon State Univ., Corvallis). A compound isolated from onion roots is fungistatic to the onion pathogen, Pyrenochaeta terrestris. An aqueous root extract from a highly resistant cultivar was passed through a Bio-Gel P-10 column, and 25 fractions, 10 ml each, were collected. Bioassay growth tests showed that inhibitory activity was confined to two fractions. Thin layer chromatography on Silica Gel-G, developed in methyl ethyl ketone:pyridine:water:glacial

acetic acid (70:15:15:2), indicated that the inhibitory compound had an $R_{\rm F}$ of .44, and reacted positively to ammoniacal silver nitrate, tetrazotized benzidine, and diazotized sulphanilic acid, thus suggesting a phenolic compound. Quantitative estimation of total phenols of several onion cultivars ranging from highly resistant to completely susceptible showed that resistant cultivars contained 12% more total phenols than susceptible ones. The inhibitor occurs in both infected and noninfected roots, indicating that invasion by the pathogen is not required for its production. The inhibitory effects of the compound are destroyed by heating and by acid hydrolysis.

A medium for the selective growth of Monilinia species. D. J. PHILLIPS (USDA, Fresno, Cal.). A medium containing 1,000 ppm PCNB (pentachloronitrobenzene), 20 g strained canned peaches, 100 ppm neomycin, and 500 ppm streptomycin was developed for the selective growth of Monilinia fructicola and M. laxa. While allowing rapid growth of Monilinia sp., the medium represses the growth of Mucorales and restricts other contaminating fungi and bacteria to discrete colonies. Counts of viable spores made with a hemocytometer corresponded to plate counts on the medium over a dilution range from 1:10 to 1:10,000 (1 =46,000 spores/ml). Counts of colonies on the medium served to measure the inoculum density when spores made up a very small portion of the total population. For example, a 6-month-old peach mummy had 25,000 viable spores in March 1971 when the total Monilinia spore population was estimated with the hemocytometer to be 85×10^6 . Washing freshly harvested peach, nectarine, and plum fruits in 1970 showed that the inoculum density ranged from 300 to 22,000 viable spores/fruit, which caused 1 to 50% decay after a simulated storage and marketing period of 24 hr at 2 C and 4 days at 25 C.

A disease of soybean caused by Neocosmospora vasinfecta. D. V. PHILLIPS (Univ. Ga., Ga. Exp. Sta., Experiment). Neocosmospora vasinfecta was isolated from several soybean (Glycine max) plants with discoloration in the pith and xylem of the stem. Soybean plants inoculated with N. vasinjecta developed internal stem discoloration, and the fungus was reisolated. Single ascospore isolates were used in subsequent pathogenicity tests. Fifteen soybean cultivars were susceptible. Symptom expression was greater at 27 C than at 21 or 15 C. Stem puncture inoculations resulted in a much higher percentage of diseased plants than did inoculation of injured roots. The length of internal stem browning in greenhouse-grown plants 4 to 7 weeks after inoculation did not exceed 13 cm. In fieldgrown plants inoculated in June and examined in October, internal browning extended a maximum of 47 cm above the inoculation point. Neocosmospora vasinfecta had little effect on plant height, and did not kill the soybean plant. Internal stem discoloration caused by N. vasinfecta in soybean plants is similar to that of brown stem rot caused by Cephalosporium gregatum, but N. vasinfecta hyphae are confined to the pith, whereas C. gregatum hyphae are found primarily in the xylem of young plants. Neocosmospora vasinfecta caused similar symptoms in cultivars of Phaseolus vulgaris and Vigna sinensis, but not in 11 other crop plants.

The differentiation between bacterial hypersensitive reaction and pathogenesis by the use of cycloheximide. Y. PINKAS & A. NOVACKY (Univ. Mo., Columbia). Injection of cycloheximide (70 µg/ml) into tobacco leaf tissue at or up to 3 hr after inoculation with Pseudomonas pisi (108 cells/ml) delayed the hypersensitive reaction (HR) for 18 hr. In vitro and in vivo experiments showed no inhibitory effect of cycloheximide (CHI) on bacterial multiplication. On the contrary, multiplication was higher in CHI-treated tissues. These data suggest that the delay caused by CHI is a result of its effect on the plant tissue; hence, HR is a plant response to the incompatible bacteria. An analogous delay of the HR symptoms was observed with Erwinia amylovora, another incompatible pathogen in to-

bacco. Since CHI did not delay symptoms in tobacco inoculated with the compatible pathogen *Pseudomonas tabaci*, it would appear that HR and pathogenesis are distinctly different phenomena. It was found that CHI strongly inhibited ¹⁴C leucine incorporation into tobacco leaf proteins, which suggests that HR is associated with plant protein synthesis. The labeled soluble proteins from bacterially injected tissue were separated by isoelectric focusing in polyacrylamide gel. The protein patterns were similar in controls and in tissues undergoing HR. However, the level of protein biosynthesis was increased in tissue undergoing HR. This increase is in large part due to plant biosynthesis

Effect of EDTA on transmission of purified cucumber mosaic virus by aphids. T. P. PIRONE (Univ. Ky., Lexington). Cucumber mosaic virus (CMV) (1 mg/ml in 5 mm borate buffer, pH 9), acquired by aphids (Myzus persicae) through a Parafilm membrane, was transmitted by the aphids to tobacco seedlings with an efficiency of 20-50%. Aphids did not transmit CMV when the virus suspensions contained 1 µmole/ml EDTA at pH 9. The addition of CaCl₂ (1 µmole/ml) to virus preparations which contained 1 µmole/ml EDTA restored aphid transmissibility of the virus. In some experiments, the presence of 0.1 µmole/ml CaCl2 in virus suspensions not pretreated with EDTA resulted in levels of aphid transmission which were higher than those obtained with preparations which did not contain CaCl₂. Infectivity of the virus, as measured by local lesion assays, was neither decreased by the presence of 1 µmole/ml EDTA, nor increased by the presence of 0.1 umole/ml CaCl2.

Characterization of barley stripe mosaic virus replicative form RNA. D. R. PRING (Univ. Neb., Lincoln). An apparent replicative form (RF) of barley stripe mosaic virus RNA was produced by in vivo 32P-labeling of the inoculated leaf one of Black Hulless Barley. The RF was resistant to deoxyribonuclease and to change in sedimentation coefficient upon formaldehyde treatment. The RF was resistant to ribonuclease in high salt conditions, susceptible to the enzyme in low salt conditions, and was alkali-labile. Estimated molecular wt of single-stranded viral RNA is about 8.9×10^5 , based on sedimentation after formaldehyde treatment. The sedimentation coefficient of the RF was 12.7 S, which gives a molecular wt of about 1.7×10^6 by Studiers formula. Buoyant density of the RF in Cs2SO4 was very close to 1.600, Maximum ³²P incorporation into RF occurred during a 72-96 hr postinoculation labeling period, or shortly before the maximum level of singlestranded RNA was apparent in the infected tissue.

Reduction of aflatoxin production in peanuts in the presence of dimethyl sulfoxide. G. W. RAMBO & G. A. BEAN (Univ. Md., College Park, Md.). The effect of dimethyl sulfoxide (DMSO) on the production of aflatoxin by Aspergillus flavus (ATCC 2221) in peanuts was determined. Fifty g of dry peanuts, cultivar Early Runner, were soaked for 30 min in distilled water containing 0.0, 0.6, 1.2, 2.5, 5.0, 10.0, or 20.0% DMSO. After soaking, they were autoclaved and inoculated with a spore suspension of A. flavus. The cultures were incubated for 7 days at 24 C, then extracted with 70% aqueous acetone and spotted on thin-layer chromatographic plates and developed in chloro-form-methanol (97:3) solvent. Developed plates were examined under UV irradiation, and relative amounts of aflatoxins were determined by amounts of florescence. The total amount of aflatoxins was determined spectrophotometrically at 365 nm and compared to aflatoxin standards. Aflatoxin production in cultures containing 0.6 and 1.2% DMSO was comparable to the controls. However, at concentrations of 2.5% and above, aflatoxin production decreased rapidly. At 20.0% DMSO, little or no fungal growth or aflatoxin was detected. These results were verified by a bioassay using chick embryos.

Tobacco etch virus tolerance in tomato. W. B. RAYMER (Campbell Inst. Agr. Res., Cinnaminson, N J.). A New

Jersey isolate of tobacco etch virus (TEV) was used to sap-inoculate 20 plants each of 288 tomato cultivars and P.I. lines. These accessions were evaluated for tolerance to TEV by means of a 0-5 rating system based on severity of mottling, rugosity, and leaf distortion. Only 5 of 125 Lycopersicon esculentum lines were considered highly tolerant, with ratings of 2 or less, whereas 9 of 17 L. hirsutum, 25 of 56 L. peruvianum, and 6 of 73 L. pimpinellifolium lines were in this category. All 7 lines of L. glandulosum and 1 each of L. cheesmanii f. minor and Solanum penellii were intolerant, with ratings of 3 or higher. No resistance to infection by sap-inoculation was found in these materials, as TEV was recovered from all symptom-less plants. P.I. 126955 remained symptomless for 4 weeks following inoculation, and was the most tolerant L. esculentum tested. P.I. 127827, 7 other L. hirsutum lines, and P.I. 247087 (L. peruvianum) also remained symptomless. Of the P.I. lines reported to be resistant to TEV in Florida, only P.I. 127827 was highly tolerant in these tests, with a 0 rating. P.I. lines 166989 and 183692 (L. esculentum) were given an intolerant rating of 4, and P.I. 134417 (L. hirsutum f. glabratum) was given an intermediate 3 rating.

An improved method for purification of wound tumor virus. D. V. R. REDDY & J. A. LESNAW (Univ. Ill., Urbana). The yield of wound tumor virus (WTV) purified by earlier methods was 2 to 3% of the virus contained in starting material. Moreover, the specific infectivity of purified virus was only 1% that of the initial virus. A simpler method of purifying WTV was developed using polyethylene glycol (PEG) 6000. Root tumors from sweet clover were ground in buffer first used for WTV by I. Kimura (0.1 m histidine and 0.01 M MgCl2, pH 7.0). After low-speed clarification, virus was precipitated using 0.3 M NaCl and 4% PEG. After standing 2 hr at 4 C, the precipitate was concentrated by low-speed centrifugation, and the virus resuspended in the buffer. After clarification by low-speed centrifugation, further purification was achieved by a single quasi-equilibrium zonal density-gradient centrifugation in sucrose. Infectivity was measured by inoculating mono-layers of vector cells (AC20) and counting the foci of infection after fluorescent antibody staining. The virus concentration in purified preparations was obtained from the optical density of their RNA, measured after its extraction with 1 N HCl. Purity of the virus was ascertained from determinations of protein/RNA ratios and of base ratios of the RNA. According to these criteria, no impurities were detected in the preparations. About 50% of the virus present in the starting tumor material could be recovered, and the purified virus retained about 30% of the original specific infectivity.

Induction of the soybean phytoalexin hydroxyphaseollin with fungicides. J. J. Reilly & W. L. Klarman (Univ. Maryland, College Park). Technical-grade fungicides were tested for their ability to induce hydroxyphaseollin (HP) in the soybean cultivar Harosoy 63. Cotyledons and hypocotyls were removed from 6-day-old soybean plants and incubated at 22 C in petri dishes each containing 7 ml of various concentrations of fungicides. Maneb, sodium dimethyldithiocarbamate dihydrate, nabam, Terrazole (5-ethoxy-3-trichloromethyl-1,2,4-thiadiazole), Mersolite-19 (phenyl mercuric salicylate), and benomyl induce HP as determined by bioassay, UV spectroscopy, and gas liquid chromatography. Zineb, ferbam, and Terrachlor (pentachloronitrobenzene) did not induce detectable amounts of HP. The amine degradation products of benomyl and maneb were potent inducers. In a subsequent study of six amines, isobutylamine was the most effective inducer of HP. The MBC (methyl 2-benzimidazolecarbamate) portion of the benomyl molecule is reported to be as fungitoxic as benomyl in vitro; however, MBC did not induce HP.

Oospore production, viability, and germination in relation to establishment of F_2 cultures of Phytophthora infestans. O. K. Ribeiro, M. E. Gallegly, & R. J. Young

(West Va. Univ., Morgantown). Four parental crosses, seven sibling crosses, and two backcrosses representing various combinations of races 1, 2, 3, and 4 of Phytophthora infestans were made on several different media. Differences in the number of viable oospores occurred which were consistent on the media used. The greatest genetic compatibility (35-58% failure to form mature oospores) was in a sib-cross of F1 cultures established from sister zoospores from the germ sporangium of a single oospore (inbreeding). The least compatibility (80-87% abortion) occurred in crosses of F1 cultures obtained from two different parental crosses (outbreeding). The original parental crosses had 16-30% abortion. The addition of 4 ppm gibberellic acid to a clarified lima bean V-8 juice medium supplemented with 10 mg/liter β-sitosterol and 100 μg/liter thiamine greatly increased the number, viability, and germination of oospores in all crosses. Viability was ascertained with 3-(4,5 dimethyl thiazolyl-2)-2,5 diphenyl tetrazolium bromide. Sporangia from germinated oospores of the F2 rarely released zoospores, and failed to establish mycelium on several media. Factors affecting the release and establishment of zoospores will have to be elucidated before further genetic studies can be undertaken.

Temperature and moisture requirements for infection of detached pineapple inflorescences by pink disease bacteria.
K. G. Rohrbach & Joan Pfeiffer (Pineapple Res. Inst., Wahiawa, Hawaii). Studies on pink disease of pineapple, caused by various strains of acetic acid bacteria (Acetomonas sp.), have been limited because the disease occurs sporadically and little is known about environmental conditions necessary for flower infections. To determine conditions required for flower infections, pineapple inflorescenses were detached and bacterial suspension was applied to the open flowers. The inflorescences were then subjected to various temperature and moisture regimes for time periods of up to 72 hr. Isolation of the bacterium in nectary gland and placental tissue was interpreted as positive infection, although a pathogenic relationship was not verified. At high humidity (polyethylene bag), 92% of the nectary glands were infected at 18 C for 48 hr, whereas only 47% occurred at 29 C. Infections decreased 4-fold at both temperatures at low humidity (no polyethylene bag). Only slight differences of infection occurred with an alternating 29-C day and 18-C night temperature when compared to a constant 18-C temperature (high humidity, 72 hr). Pathogen recovery from nectary glands was consistently higher than from placental tissue. When the length of time at high humidity was varied following inoculation, maximum infections occurred at 6 hr.

Johnson grass, an overwintering host of corn stunt agent. E. ROSENKRANZ (ARS, USDA, Miss. State Univ., State College). An epiphytotic of corn stunt (CS) occurred in the southern USA in the early 1960's, but the mode of overwintering of the corn stunt agent (CSA) is still unknown. A continued search for possible wild host plants of CSA resulted in the location, in 1970, of an apparently diseased Johnson grass shoot at State College, Miss., and a similar one at Clemson, S.C. Both young shoots, arising from rhizomes, exhibited leaf symptoms resembling those in corn experimentally infected with the Mississippi corn stunt agent (CSA-MS) in the greenhouse. These symptoms consisted of splitting of the leaf margin leading to laminar constrictions, marginal chlorosis, and occasional necrosis of the leaf apex; subsequent growth of the Johnson grass was symptomless. When noninfective Dalbulus maids and Graminella nigrifrons were exposed to the diseased Johnson grass clones, both leafhopper species became infective with CSA-MS. Transmission of CSA-MS from Johnson grass to corn was more efficient with D. maidis than with G. nigrifrons. The shortest latent period of these CSA-MS isolates in both vectors was 9 days, whereas their minimum incubation period in corn was 16 days. These findings contribute to an understanding of the disease cycle of CS and could help to explain the northern limits of its distribution in the USA.

Pathogenicity of Pseudomonas syringae on Phaseolus vulgaris as influenced by bacterial isolate source. S. M. SAAD & D. J. HAGEDORN (Univ. Wisc., Madison). Pathogenicity of 30 Pseudomonas syringae isolates from 10 hosts was compared on Tenderwhite bean in the greenhouse, using inoculum levels of 10³ and 10⁸ cells/ml. Bacterial suspensions were sprayed on both surfaces of the first trifoliate leaf. All bean isolates at both inoculum levels induced typical olive-green, water-soaked lesions within 3-4 days. These lesions soon became necrotic (dark brown), with marginal chlorosis. Isolates from pear, apple, sour and sweet cherry, lilac, peach, plum, walnut, and sorghum produced only tiny, dark necrotic flecks within 24 to 48 hr with inoculum containing 108 cells/ml. No macroscopic symptoms were obtained with the lower level of inoculum. Different isolates from the same host showed minor pathogenic differences. Growth characteristics of several isolates were studied in bean pods. In the compatible combination, the bacteria multiplied logarithmically for 4 days, inducing water-soaked lesions without brown necrosis. Logarithmic growth of incompatible isolates from pear, lilac, and sour cherry terminated after 2 days and induced brown, sunken necrotic lesions which fluoresced when exposed to UV light. These studies indicate a positive relationship of high pathogenicity to bean with *P. syringae* isolates from bean alone.

Remote sensing of southern corn leaf blight. G. R. Safir, G. Suits, & A. H. Ellingboe (Mich. State Univ., East Lansing, Univ. Mich., Ann Arbor). Corn plants of the hybrid W64A \times OH 43 with Normal "N" or Texas male sterile "T" cytoplasms were inoculated with Helminthosporium maydis race "T". The reflectance of leaves at wavelengths between 0.35 and 2.6 μ of inoculated and noninoculated plants was determined with a Beckman ratio-recording spectrophotometer. Greater reflectance, primarily at the chlorophyll and water-absorbing wavelengths, was obtained from leaf areas with lesions than from healthy leaves or portions of the inoculated leaves without lesions. A greater difference in reflectance in the water-absorbing wavelengths was observed if the inoculated corn plants had "T" cytoplasm than if the plants had "N" cytoplasm. The earliest detection of a difference in reflection between diseased and healthy tissues was ca. 36 hr after inoculation.

Identification, characterization, and transmissibility of a virus infecting St. Augustine grass in Florida. J. L. Saladini & F. W. Zettler (Univ. Fla., Gainesville). A virus of St. Augustine grass isolated in 1963 by E. H. Todd was identified as sugarcane mosaic virus strain E (SMV-E). It induced discrete localized lesions in Atlas sorghum, and a local and systemic necrosis followed by recovery in CP 31-294 and CP 31-588 sugarcane; it did not infect Johnson grass. Of 138 particles measured in leaf extracts, 89 were 675-782 nm long with a main maximum at 711 nm. Pinwheel, circle, bundle, and laminated aggregate inclusions occurred in diseased corn leaves. SMV-E was transmitted from corn to corn by Schizaphis graminum aphids in a stylet-borne manner. St. Augustine grass was more resistant than was corn to infection by mechanical inoculations to adaxial leaf surfaces. In trials using identical inocula, only 10 of 200 St. Augustine grass plants became infected as compared to 195 of 200 for corn. Once infected, however, symptoms persisted in St. Augustine grass plants, and SMV-E was always readily recovered by back assay to corn. Scanning electron micrographs revealed extensive wax deposits on adaxial but not on abaxial leaf surfaces of St. Augustine grass.

Protein content of tobacco roots following heat treatment and inoculation with Phytophthora parasitica var. nicotianae. G. E. SANDEN & L. D. MOORE (Va. Polytech. Inst., Va. State Univ., Blacksburg). Roots of intact plants of susceptible and resistant cultivars of flue-cured tobacco, Nicotiana tabacum, were subjected to the following treatments: (i) roots were heated by immersion in a water bath at 50 C for 1 min and inoculated; (ii) roots were heated only; (iii) roots were inoculated only; (iv) con-

trols were untreated. The root tissue was assayed immediately and 10 days after treatment for total protein content by the Lowry method. Extracts from the susceptible cultivar, Virginia Gold, which were inoculated only, heated only, or heated and inoculated, showed a decrease in total proteins as compared to extracts from roots which received no treatment. Extracts from the resistant cultivar, Coker 187, showed a decrease in total proteins from those roots which were heated only or inoculated only as compared with unheated checks, while roots which were both heated and inoculated showed an increase. The protein content of heated-only and inoculated-only roots was slightly higher for the resistant cultivar than for the susceptible cultivar. The low concentration of protein in roots of Virginia Gold, which were both heated and inoculated, may have been due to the degradation of the roots.

Impaired amino acid biosynthesis in phytopathogenic pseudomonads. D. C. Sands & M. L. Zucker (Conn. Agr. Exp. Sta., New Haven). Phytopathogenic fluorescent pseudomonads related to Pseudomonas syringae differ distinctly in amino acid metabolism from saprophytic members of the group. The pathogens cannot use a number of amino acids as a carbon source or for energy. The saprophytes can. A number of amino acids were toxic to 11 strains of pathogens but to no saprophytes. L-homoserine was toxic to all pathogens. Threonine, valine, alanine, glycine, and a combination of phenylalanine plus tyrosine were strongly inhibitory to one or more of the pathogens. In all such cases, the inhibitory effect was reversed by the presence of certain amino acids in the same biosynthetic pathway. Studies with mutants of saprophytes have shown that such toxicities and reversals are indicative of lesions in the regulation of the branched pathways of biosynthesis of amino acids. Perhaps these lesions result from selection, due to the high levels of amino acids in plants. These lesions may be linked to toxin production and host specificity factors.

Survival of Erwinia rubrifaciens in exudate on diseased walnut trees. N. W. SCHAAD (Univ. Cal., Davis). Erwinia rubrifaciens occurs in large numbers in a slimy substance which exudes through cracks and accumulates on the bark of infected walnut trees. To determine the longevity of the bacteria in dried gummy exudate, samples of exudate were collected at various time intervals after emission. After 62 days, numerous bacteria were still viable in exudate protected from further surface replenishment but left in its natural environment. For example, the original concentration of 1.4×10^9 viable cells/mg decreased only to 1.0×10^7 cells/mg after 41 days and to 8.6×10^6 after 62 days. When expressed as a survival rate, the data show that the half-life of the bacteria was 5.7 days for the first 41 days. When exudate was sampled after having been placed on a healthy tree or stored in the laboratory (room temperature), the results were somewhat different. For example, it took 5.6 days for half the bacteria to die after 48 days in the exudate placed at room temperature, but only 3.8 days for bacteria in exudate transferred to a healthy tree. It is concluded that E. rubrifaciens can survive for at least 123 days in exudate on the bark of diseased trees.

The influence of dew and atmospheric moisture periods on infection of slash pine seedlings by Cronartium fusiforme. R. A. SCHMIDT (Univ. Fla., Gainesville). Two-month-old seedlings of slash pine (Pinus elliottii var. elliottii) were exposed for 1-2 hr in a dew chamber beneath trays of oak leaves bearing germinating telia of Cronartium fusiforme. Inoculated seedlings (45/treatment) were subjected to the environmental regimes described below and planted in the nursery, where the percentage of seedlings with galls was determined after 9 months. Dew periods (x1) of 0, 1, 2, 4, 5, 6, 8, 10, 12, 16, and 24 hr resulted in disease percentages of 0, 0, 0, 0, 26, 16, 52, 29, 32, 47, and 71, respectively. Periods of 100% relative humidity (x2) of 2, 4, 6, 8, 10, 12, 16, and 24 hr resulted in disease percentages of 0, 3, 8, 67, 34, 40, 49, and 73,

respectively. Per cent disease (y) was related to postinoculation treatments x_1 and x_2 in a direct linear manner ($x_1 \le 4$, y=0; $x_1 \ge 5$, $y=15.71+2.04x_1$ and $x_2 \le 2$, y=0; $x_2 \ge 4$, $y=5.14+2.96x_2$). The significant correlation coefficients (r) were 0.672 and 0.751, respectively. Postinoculation drying periods (x_3) of 0, 1, 2, 4, 8, 12, 16, and 24 hr prior to incubation at favorable moisture conditions resulted in disease percentages of 62, 68, 46, 70, 31, 48, 60, and 51, respectively (y=58.0-0.408 x_3 , and r=-0.264).

Halo blight of mung bean incited by a new strain of Pseudomonas phaseolicola. A. F. SCHMITTHENNER, H. A. J. HOITINK, & M. E. KROETZ (Ohio Agr. Res. Devel. Cent., Wooster). Introduction of mung bean (Phaseolus aureus) has not been successful in Ohio because of severe losses from halo blight. The pathogen isolated was similar to both Pseudomonas phaseolicola and P. glycinea in cultural characteristics. Mung bean isolates caused water-soaking on mung bean, Red Kidney bean, Fordhook 242 lima bean, and Acme soybean, but not on other soybeans used to differentiate P. glycinea races. None of six P. phaseolicola isolates or six P. glycinea races caused water-soaking on mung bean. Since only the mung bean isolates are virulent to mung bean, it is proposed that they be named the mung bean strain of P. phaseolicola. The pathogen was introduced on seed. In experimental plots at 11 locations in Ohio initial infection occurred in plots of Berken, Jumbo, and Oriental cultivars from seed produced in Oklahoma. Plots from Brazil, Peru, or Thailand seed or plots from greenhouse-produced Berken seed had no initial infection. Halo blight eventually developed in most cultivars at all locations. However, at four locations, plots from greenhouse Berken seed isolated from a source of primary inoculum remained free of halo blight. At these four locations, Berken with halo blight (Oklahoma seed) yielded ca. 60% less than healthy Berken (greenhouse seed).

Multidense satellite of tobacco ringspot virus: a series of components with a uniform difference in density between each component. I. R. SCHNEIDER, R. HULL, & R. MARK-HAM (ARS, USDA, Beltsville, Md., John Innes Inst., Colney Lane, Norwich, England). The satellite (STRSV) tobacco ringspot virus (TRSV) ratio found in the inoculated plant was higher when the STRSV/TRSV ratio in the inoculum was increased. STRSV is composed of 11-14 homogeneous components ranging in buoyant density (BD) in CsCl from $\rho = 1.408$ through $\rho = 1.529$. Each component differs in density from its nearest neighbor by ca. $\rho = 0.009$. The BD's found in a STRSV population are not artifacts caused by CsCl, but correlate with the position of the particles sedimenting in a sucrose gradient. Two components of STRSV cannot be distinguished in BD from bottom component of TRSV; another component of STRSV cannot be distinguished in BD from middle component of TRSV. Since some TRSV is always present in STRSV infections, a satellite population has 11 components at the least, but may have as many as 14 distinct components. The data indicate that each STRSV particle of lowest BD has ca. 11 nucleic acid strands (molecular wt) of 86,000 daltons/strand). Each STRSV particle of next higher buoyant density has one more nucleic acid strand than its nearest neighbor of lower density. Each particle of highest density contains 24 nucleic acid strands.

Etiology of Cephalosporium gregatum in soybean. R. W. Schneider & J. B. Sinclair (Univ. Ill., Urbana). In greenhouse and field experiments, internal browning in soybean stems, caused by Cephalosporium gregatum, developed as greater extent in plants inoculated at a younger stage (6 weeks) than in those inoculated at later stages (8-12 weeks). These results suggest that if maximum symptom development is to occur in the field, the fungus must enter the roots at an early age. Evidence for conidial movement within the transpiration stream after root penetration suggests that this may be the principle mode of spread of C. gregatum in soybean plants. Cephalosporium gregatum spread to the tops (ninth node) of plants in the early pod-

filling stage within 1 and 2 days, respectively, when plants were grown in a fungal suspension or when the hypocotyls were dipped into a conidial suspension (25,000/ml). Though the fungus enters the roots, internal browning may be delayed by warm temperatures. There was a direct relation between exposure time of infected plants to cool temperatures (18-24 C) and extent of internal browning.

Invasion of rough rice in storage by strains of Aspergillus flavus. H. W. Schroeder & R. A. Boller (ARS, USDA, College Station, Tex.). Belle Patna rough rice was inoculated with spores of each of the three strains of Aspergillus flavus, separately and in all combinations. P-70-51i (I), green-spored, produces large quantities of aflatoxin; P-70-25f (II), green-spored, produces an orange fluorescent compound but no aflatoxin; and AF-2 (III), a white-spored mutant, produces little or no aflatoxin. In all treatments, invasion of kernels increased significantly only during the 1st week in storage in a relative humidity of 90% at 25 C. The white-spored mutant strain (III) failed to invade as readily as the two green-spored wild-type strains (I, II) when each strain was applied separately. Maximum toxins (33 ppb) were detected in rice inoculated with two non-producers of toxins (II, III). When mixed inoculum was used, all strains appeared equally capable of invading rice under the conditions of this experiment. The separate strains appeared to retain their identity and to mix physically rather than through asexual recombination of genetic materials.

Evidence for an infectious double-stranded RNA plant virus. J. S. Semancik & L. G. Weathers (Univ. Neb., Lincoln, Univ. Cal., Riverside). The replication of the free-RNA citrus exocortis virus (CEV) was determined by bioassay of phenol extracts from Gynura aurantiaca. Infectivity was max 40-60 days postinfection (p.i.), with 50% still recoverable 140 days p.i. When the nucleic acid was fractionated into 2 M LiCl precipiate, "DNA-windings", and supernatant, the supernatant fraction demonstrated the highest specific infectivity. However, the DNA fraction contained twice the total infectivity of the supernatant. Infectivity was reduced slightly when preparations were heated to 80-100 C for 10 min. Inactivation was linear from 100 to about 140 C. Concentrations of diethylpyrocarbonate which reduced the infectivity of single-stranded viral RNA by 95-100% inhibited CEV by only 40-70%. CEV infectivity was eluted from methylated albumin at 0.72 M NaCl as compared to DNA at 0.69 M and double-stranded RNA at 0.77 M. The infectivity distribution after Cs₂SO₄ equilibrium sedimentation conformed to a broad density range (1.55-1.64) and indicated the presence of single and double-stranded RNA species. These data suggest that the major infective unit of CEV is a free-RNA double-stranded structure.

Diseased leaves as a source of ascospores of Leptotrochila (Pseudopeziza) medicaginis for alfalfa inoculations. G. Semeniuk (S. Dak. State Univ., Brookings). Diseased leaves hand-picked from Medicago sativa plants in late June and early July in western South Dakota were airdried, then wetted and dried flat between blotting paper and secured in numbers of ca. 30 between 4- × 5-inch plastic window screens (screens 17 mesh/inch) sewed together with nylon thread. The leaf-carrying screens were placed (leaf adaxial sides up) between heavy galvanized wire wide-mesh screens and set outdoors in early August on open ground in the shade. Mature apothecia appeared by late August 1969 and late October 1970. Their number increased to ca. 5/leaflet (range 0-25) over an additional 2 weeks' exposure, and at that time the screened leaves were stored in a cool, dry place for use in greenhouse plant inoculations. Screened leaves were arranged on light-wt wide-mesh screens over plants, and the whole was enclosed for 3 days with a transparent plastic sheet. Seven hundred potted alfalfa plants in two 4- × 12-ft benches were inoculated the first time from 145 screens, first one bench, then the other; and a second time from 470 screens, both benches at once. The results suggest uniform ascospore discharge and yellow leafblotch development over the bench areas.

Uncoating of TMV-RNA in tobacco leaves treated with Carborundum or chloroform. J. G. Shaw (Univ. Ky., Lexington). Tobacco leaves were treated in three different ways prior to being rubbed with tobacco mosaic virus which was labeled with ¹⁴C in its protein moiety. Leaves were (i) rinsed with water and dusted with Carborundum; (ii) rinsed with chloroform followed by water; or (iii) rinsed with water only. Three min after inoculation, the leaves were rinsed with water and frozen. The amount of TMV-protein released from the virus was determined by centrifugal analysis of extracts of the leaves. Uncoating activity in leaves rinsed with chloroform was twice that in leaves dusted with Carborundum and up to 10 times that in leaves to which neither chloroform nor abrasive had been applied.

Successions of microorganisms and patterns of discoloration and decay after wounding in red oak and white oak. A. L. Shigo (USDA, Northeastern Forest Exp. Sta., Durham, N.H.). Trunks and roots of 23 mature red oak, Quercus rubra, and white oak, Q. alba, were dissected to determine the patterns of discolored and decayed (D & D) wood associated with 22-year-old basal fire wounds and mechanical wounds inflicted during subsequent salvage operations. The columns of D & D wood had advanced farthest along the sapwood-heartwood boundary present at the time of wounding. The D & D tissues associated with the wounds were confined to the tissues present when the wounds occurred. The heartwood cylinder constricted abruptly below the root collar. Heartwood formation was retarded about the wounds. Complex patterns of D & D wood occurred at the root-trunk transition zone. Isolations for microorganisms were made in a systematic way from columns of D & D wood in 19 trees. Bacteria and nonhymenomycetous fungi were isolated consistently from columns that contained only discolored wood, and from the discolored wood at the distal margins of columns that contained decay. Hymenocycetous fungi were isolated commonly from the tissues at the border of D & D wood. A large variety of microorganisms was isolated from the advanced decay. The results indicate that basic patterns of discoloration and decay and successions of microorganisms follow wounding in oaks.

The role of dew and temperature in the epidemiology of Botrytis leaf blight of onion. P. B. SHOEMAKER & J. W. LORBEER (Cornell Univ., Ithaca, N.Y.). Conidia of Botrytis squamosa (isolate 64a) were atomized onto leaves of onion plants (Allium cepa) which were then held for various periods in a dew chamber at 20 ± 1 C. Only plants whose leaves remained wet continuously for 6 or more hr developed an appreciable number of lesions when intervals of 0, 3, 6, 9, 12, and 18 hr were tested; lesion numbers were determined 40 hr after inoculation of the plants. There was a significant increase in lesion numbers for each interval beyond 6 hr. Necrosis (blighting), measured from the leaf tip, also increased significantly as the dew period was increased. When plants whose leaves remained continuously wet were kept 40 hr at constant temperatures of 7-30 C, lesions developed only in the range from 9 to 23 C, inclusive. Lesion numbers and amount of blighting were significantly greater on older outer leaves than on younger inner leaves. Leaf age and position may be important factors determining susceptibility. When conidia were brushed onto dry leaf surfaces, lesions developed only if the plants subsequently were kept wet. Germination of conidia was maximum at 15 C, and occurred over the range 12-27 C; growth in culture was maximum at 24 C and occurred from 9-31 C.

Distribution and metabolic fate of benomyl in dwarf pea plants. M. R. Siegel & A. J. Zabbia, Jr. (Univ. Ky., Lexington). ¹⁴C benomyl was added to Knop's solution in which 21-day-old peas (Pisum sativum 'Laxton Progress'), were growing. Plants were treated with benomyl for 4 days and harvested at 12-day intervals until senescence of the plants occurred (52 days after treatment). Neither 14C benomyl nor 14CO2 was recovered from the plants at any time. A fungitoxic derivative, 14C methyl 2-benzimidazolecarbamate (MBC), was present in large quantities in organic solvent extracts of the plants at all time periods. At senescence, 76% of the label was present in MBC and 7% in unknown water-soluble metabolites; 15% of the label was bound to the plant residue. When the plant residue was treated with NaOH, a portion of the bound label was present in 2-amino-benzimidazole. Distribution of the fungicide in plants was dependent on the age when harvested. The labeled products were translocated to the stems and leaves of the plant. Fifty-two days after treatment, 94% of the label was in nonroot portions (stems, leaves, and pods). However, only 12% of the total label was present in tissue formed after the plants were treated. These data suggest that long term chemotherapeutic effects could be dependent on the distribution, persistence, and concentration of MBC in plant tissue.

Effect of light on growth and sporulation of Colleto-trichum destructivum in culture. R. C. Sievert (ARS, USDA, Univ. Tenn., Greeneville). Cultures of Colletotrichum destructivum were grown on numerous solid media (generally 1.5% agar, w/v), at initial pH values from 4.0 to 10.0, and incubation temperatures from 15 to 30 C. Media varied in the carbohydrate and/or plant extract included and source of agar, in addition to pH and temperature. When a single medium was employed at different initial pH levels, growth (as measured by colony diameter) was faster, and conidial production was greater, for cultures incubated with a 16-hr light period (daylight fluorescent lamps) than for cultures incubated in the dark. For all media tested at incubation temperatures below 30 C, growth of cultures was more rapid and production of conidia was greater with a 16-hr photoperiod than in the dark. At 30 C, growth was more rapid and conidial production was greater on some media in the dark than in the light. Thus, a reversal in response occurred with the same media at 30 C as compared to lower temperatures.

Crown rust tolerance of Avena sativa-type oats derived from wild Avena sterilis. M. D. SIMONS (Iowa State Univ., Ames). To test the hypothesis that Avena sterilis indigenous to Israel has potentially valuable tolerance to crown rust (Puccinia coronata), three strains rated susceptible in reaction type were crossed with the highly susceptible oat cultivars Richland and Clinton. Cultivated-type rust-susceptible segregates selected from the resulting populations were evaluated for field tolerance in epiphytotics artificially initiated with a mixture of common crown rust races. Relative tolerance of segregates to infection was estimated by comparing the average wt of kernels from infected plants with rust-free controls. Data were adjusted for maturity differences among lines. Twenty of 23 lines from the Richland crosses were significantly more tolerant than Richland in 1969 and 1970. Similarly, 14 of 23 lines from the Clinton crosses were more tolerant than Clinton. No relationship was apparent under rust-free conditions between kernel weight and tolerance, but in yield the tolerant lines were uniformly inferior to their cultivated parents.

Additive effects of different types of ectomycorrhizae on growth of Douglas fir seedlings. W. A. Sinclair (Cornell Univ., Ithaca, N.Y.). The relationship of ectomycorrhizae to seedling wt increase was studied in a new Douglas fir (Pseudotsuga menziesii) nursery. Samples totaling 1,700 seedlings were lifted 20 weeks after seedling, and all ectomycorrhizae were classed as smooth (S) or tomentose (T) according to texture and color of their mantles. Colors of S and T mycorrhizae were, respectively, similar to and distinct from nonmycorrhizal roots. The seedlings were then grouped in four categories: 42% were nonmycorrhizal; 21% had only S mycorrhizae; 21% had only T mycorrhizae;

rhizae; 15% had both S and T types. The mean dry wt of trees in these groups differed significantly, and were 112, 144, 135, and 170 mg, respectively. The data are interpreted as evidence of stimulation of seedling growth by ectomycorrhizae, and of additive effects of different types of ectomycorrhizae on rate of seedling growth. Alternative interpretations not involving growth stimulation by ectomycorrhizae are that (i) large seedlings are more often mycorrhizal because their root systems are larger targets for colonization by symbiont fungi, or roots of fast-growing seedlings as substrates for growth of symbiont fungi; (ii) ectomycorrhizal seedlings may be protected by fungal symbionts against growth suppression by root pathogens.

Infectious low molecular weight ribonucleic acid from tomato infected with potato spindle tuber virus. R. P. Singh & M. C. Clark (Can. Dep. Agr., Fredericton, New Brunswick). Infectious low mol wt ribonucleic acid (RNA) from Sheyenne tomato tissues infected with potato spindle tuber virus was extracted and purified by precipitation with 3% aqueous cetyltrimethylammonium bromide followed by chromatography on diethylaminoethyl cellulose. Electrophoresis on 2.4% polyacrylamide gel columns and sedimentation on a 5-20% sucrose-density-gradient showed that the mol wt of purified RNA corresponded to 4-5 Svedberg units. Further purification by exonuclease digestion showed that the RNA was not completely resistant to attack by venom phosphodiesterase. Infectivity assays of the different fractions produced by purification were conducted on the local lesion host, Scopolia sinensis, and showed a 36-fold concentration from the crude sap to the DEAE-cellulose-treated fraction.

Effects of Alternaria alternata infection and other leaf injuries on subsequent growth of tobacco. L. J. Slana & J. R. Stavely (ARS, USDA, Beltsville, Md.). We reported that heavy infection by Alternaria alternata severely inhibits the expansion of the youngest inoculated leaves. To determine further effects on plant growth, we treated greenhouse-grown Coker 187 Hicks tobacco plants (having about 11 leaves) by inoculating with A. alternata or Colletotrichum destructivum, wounding, or removing the youngest leaves. Dimensions and numbers of leaves on these plants and controls were recorded after 0, 2, 4, and 6 weeks. At flowering, we recorded the date, plant height, and number of leaves per plant. All treatments inhibited expansion of the youngest treated leaves, but caused production of more posttreatment leaves than were produced on controls. The increase in leaf number due to Λ . alternata infection was proportional to infection intensity. The average increase in the number of leaves above the number on controls was 0.5 and 6 on lightly and heavily infected plants, respectively. Heavy A. alternata infection induced twice as many additional leaves as did leaf removal, and three times as many as severe wounding or C. destructivum infection. Stem elongation was inhibited in the youngest treated portion, but was not affected above this area. Severe A. alternata infection caused a reduction in the size of leaves produced above infected tissues, but other treatments had no effect.

Influence of nitrogen fertilizers on rhizosphere pH and take-all of wheat caused by Ophiobolus graminis. R. W. SMILEY & R. J. COOK (Wash. State Univ., USDA, ARS, Pullman, Wash.). Take-all of cereals and Ophiobolus-patch of turf can be controlled in the field by NH₄+-N, but not commonly by NO₃--N. Take-all control with (NH₄)₂SO₄ in greenhouse pots of Puyallup fine sandy loam (native pH 5.5) was negated by liming this soil to pH 7.7. Control was obtained with no N and with Ca(NO₃)₂ in Ritzville silt loam acidified to pH 5.7; but without acidification, only (NH₄)₂SO₄ was effective at the native pH 7.5. Disease was less at pH_b < 6.0 (bulk soil pH), but severity differed for each soil, and was only moderately correlated with pH_b. Disease ratings were highly correlated with pH_r (rhizosphere soil pH); and regardless of soil type, pH_b, N fer-

tility, and form of N applied, ratings were progressively less as pH_r decreased below 7.0. However, in fumigated soil (methyl bromide), the disease was uniformly severe at all values above pH_r 5.0. Similarly, in autoclaved soils, pathogen growth was prevented at pH < 4.9 and was optimal above 6.0 irrespective of the form of N. Apparently, the effect of pH_r on disease, induced by the form of N, is direct at pH_r < 5.0; possibly operating through soil microorganisms antagonistic to ectotrophic growth of O. graminis on the rhizoplane.

Isolation of substance II, an antifungal compound from Rhizoctonia solani-infected bean tissue. D. A. SMITH, H. D. VAN ETTEN, & D. F. BATEMAN (Cornell Univ., Ithaca, N.Y.). Phaseolus vulgaris hypocotyls infected with R. solani were triturated in 95% ethanol (1:4, w/v). The extract was filtered and centrifuged at 15,000 g for 10 min. Water (v/v) was added to the supernatant, and the ethanol removed in vacuo at 40 C. Phaseollin was extracted from the aqueous phase by partitioning twice with petroleum ether (1:4, v/v). Bioassays of the residual aqueous fraction revealed a second antifungal component. This fraction was extracted twice with CHCl3 (1:2, v/v), and the CHCl3 partitioned twice with 0.2 M Na2CO3 (1:1, v/v). The Na2CO3 fraction was adjusted to pH 6.0 with HCl, extracted twice with CHCl3 (1:1, v/v), and the CHCl3 fraction dried in vacuo at 40 C. The residue was dissolved in 95% ethanol and subjected to thin-layer chromatography on silica gel using benzene: p-dioxane: acetic acid (8:1:1). Area RF 0.25-0.37 was eluted and rechromatographed with CHCl3: methanol: acetic acid (8:1:1). Substance II has an RF of ca. 0.75 in the latter system, appears as a dull yellow-green spot under UV light in the presence of NH3 fumes, is phenolic, has a major UV absorption peak at 294 nm in ethanol, and undergoes a reversible hyerchromatic base shift of 36 nm. The ED50 for R. solani corresponds to an absorbance of 6.0 OD units/ml at 294 nm.

Aerobiology of peanut leaf spot fungi. D. H. SMITH & F. L. Crosby (Univ. Ga., Nat. Weather Serv., Experiment). Air was continuously sampled with a Hirst spore trap in a peanut (Arachis hypogaea 'Argentine') field during the growing seasons of 1969 and 1970. Cercospora arachidicola conidia and Leptosphaerulina crassiasca ascospores were present in the air during June, July, August, and September of both years. Cercosporidium personatum conidia were not trapped in either year. The concentration (spores per m3 of air per 24-hr period) of C. arachidicola conidia and L. crassiasca ascospores varied widely from day to day. The catches of C. arachidicola conidia and L. crassiasca ascospores increased rapidly with the drying of leaves in the morning and with the onset of rainfall. Although C. arachidicola conidia were trapped on 60 days in 1969 and 69 days in 1970, they were present in low numbers until the 2nd week of August. The peak concentration of C. arachidicola conidia (> 300 m3) occurred from 3-4 September 1970. Leptosphaerulina crassiasca ascospores were trapped on 71 days in 1969 and 79 days in 1970. Low numbers of L. crassiasca ascospores were present in the air until the level of peanut leaflet abscission reached 37% in late July of 1969 and early August of 1970. The peak concentration (> 1,300 m³) of L. crassiasca ascospores occurred from 23-24 August 1970. Very few phragmosporous L. crassiasca ascospores were trapped.

Trehalase activity in natural and artificially fungus-colonized soil. R. E. SMITH & R. RODRIGUEZ-KABANA (Auburn Univ., Auburn, Ala.). Trehalase activity (TA) was measured in natural soil and in soil inoculated with Sclerotium rolfsii (SR), Fusarium oxysporum f. sp. vasin-fectum (FOV), and Trichoderma viride (TV). In natural soil, incubated over a temperature range of 20-80 C, TA increased to maximum at 50 C, then declined. Acetate, propionate, citrate, succinate, maleate, and phosphate buffers (0.5 M) were used to determine maximal TA over a pH range of 3.5-7.5. Maximal TA occurred in propionate buffer at pH 4.95, and was at a high level in acetate buffer.

When pH was held constant at 5.5 (acetate), and ionic strength increased from 0 to 5 m, TA decreased. When trehalose concentration was varied in the assay mixture, maximal rates of increase in TA occurred between 0 and 2% (w/v). At constant volume, increases in amount of soil resulted in increased TA. There was a linear increase in TA with incubation time from 0 to 22 hr. Determination of TA in colonized soils was performed using 125-ml flasks containing 5 g soil, 5 ml 2% (w/v) trehalose, and 1 ml toluene. Soil pH values were 4.1, 5.3, and 6.4 for SR, FOV, and TV, respectively. During a 3-hr incubation period at 35 C, TA in soil colonized by the three fungi was in the following order: SR > FOV > TV.

Lead, mercury, and sodium contamination of urban trees. W. H. SMITH (Yale Univ., New Haven, Conn.). Acer platanoides planted in New Haven were analyzed for Pb and Na contamination. During the fall of 1970, branch samples were collected ca. 2 m aboveground from 50 trees located within 7 m of streets. Analysis was by atomic absorption spectrophotometry. Lead content of the growth of the previous growing season was as follows (µg/g dry wt basis, mean and se, unwashed and washed, respectively): intact branches 163 ± 13 , 167 ± 12 ; leaves $112 \pm$ 33, 96 \pm 20; twigs 86 \pm 20, 69 \pm 10. Sodium content was as follows: intact branches 374 ± 30 , 362 ± 35 ; leaves 469 \pm 183, 275 \pm 73; twigs 319 \pm 71, 347 \pm 57. The Pb concentrations are in excess of most reports of tree Pb levels. Since the concentrations of unwashed and washed samples were not significantly different, either the cations were located inside the tissues or the washing procedure was inadequate to remove surface deposits. Hg analyses revealed the following concentrations (µg/g) on single, unwashed samples: Acer saccharum leaves, 0.81; Ailanthus altissima leaves, 0.62; Platanus xacerifolia leaves, 0.73; twigs, 0.13; Picea abies intact branches, 0.22; Pinus nigra leaves, 0.17; twigs, 0.08; Quercus palustris leaves, 0.77; Taxus sp. intact branches, 0.84; and Tilia cordata leaves, 1.10.

Contamination of peaches and nectarines in packing houses. W. L. Smith, Jr., J. M. Wells, & R. W. Penney (ARS, USDA, Beltsville, Md.). Surveys indicate that peaches and nectarines are often essentially free of decayproducing organisms when they enter packing houses, but can be contaminated when exposed to unsanitary conditions in packing houses. Such contamination can cause a high percentage of the decay occurring during marketing. In these tests, most of the decay due to contamination was brown rot, caused by *Monilinia fructicola*. Chief sources of contamination were hydrocoolers and tanks for dumping bulk bins. After 3-9 days at shipping or room temperatures, fruit sampled from these areas had from 2 to 10 times as much brown rot as fruit sampled directly from the orchards. Additional contamination sometimes occurred during defuzzing. Suggestions for reducing contamination include (i) more frequent cleaning of hydrocooler and dump tanks; (ii) chlorination of cooling or dump tank water; (iii) hot water treatments (52 C for 1-2 min); and (iv) cooling of fruit in air rather than water.

Inheritance of leaf rust resistance in Red River 68, a semidwarf hard red spring wheat. G. D. Statler (N.D. State Univ., Fargo). Inheritance of seedling and adult plant resistance to wheat leaf rust incited by Puccinia recondita f. sp. tritici was investigated in Red River 68, a semidwarf hard red spring wheat resistant to prevalent races of wheat leaf rust. Red River 68 was crossed and backcrossed to the susceptible variety Thatcher for the genetic analysis. The F1, F2, and backcross progenies of the crosses were inoculated in the seedling and adult stages with a culture of race 15 which had been purified by three successive, singlepustule isolations. Seedling and adult resistance to physiologic race 15 of *P. recondita* was conditioned by a single dominant gene in Red River 68 as shown by three criteria. The F1 plants were all resistant to race 15; the 104 backcross plants segregated into a 1:1 ratio (P between .50 and .75) of resistant to susceptible; and the 308 F2 plants segregated to fit a 3:1 ratio (*P* between .50 and .75). The same gene was apparently operative in both seedling and adult stages, providing Red River 68 with resistance to wheat leaf rust.

Effect of Cercospora nicotianae infection on chemical constituents of cured tobacco leaf. J. R. STAVELY & J. F. CHAPLIN (ARS, USDA, Beltsville, Md., Oxford, N. C.). In 1970 we grew three burley, three Maryland, and four fluc-cured tobacco cultivars in the field in two blocks of six plants/each of five replicates per cultivar. One block was inoculated with *C. nicotianae*, the incitant of frogeye and green leaf spots. All inoculated cultivars were heavily infected, but the most frogeye occurred on burley, and the most green spot occurred on Maryland types. The second block remained disease-free. Leaves were harvested twice from equivalent positions on all plants. Half the harvested leaf from each replicate was air-cured; half was flue-cured. Analyses of all cured leaf showed that the disease caused respective decreases in total alkaloids of 24, 10, and 5% in the first and 36, 30, and 18% in the second harvest from burley, flue-cured, and Maryland types. Soluble phenols were reduced by the disease 83% in air-cured and 57% in flue-cured leaf. Reducing sugar content was decreased an average of 50% by the disease in all leaf that was fluecured. The greatest reduction occurred in flue-cured leaf of flue-cured cultivars, which had 2-4 times more sugar than the other types. Reducing sugar content and its reduction by disease was greater in the second than in the first harvest. Leaf spot caused a significant 3 and 9% increase in total nitrogen in the first and second harvest, respectively.

Identification of tenuazonic acid as an important toxic substance produced by Alternaria spp. J. A. Steele & C. J. Mirocha (Univ. Minn., St. Paul). Certain isolates of Alternaria spp., when grown on moist autoclaved corn or corn rice mixtures, produce substances toxic to rats, swine, and turkeys. One of these substances was previously isolated but not fully characterized. Fifty mg of the unknown toxin were obtained from moist autoclaved corn which had been inoculated with an Alternaria isolate known to produce large quantities of the toxin. This material was submitted for elemental analysis, flame emission spectroscopy, UV spectroscopy, and infrared spectroscopy. Chemical degradation studies were made by acid hydrolysis and treatment with sodium periodate. The spectral data and degradation products indicate that the toxin was the mixed potassium, sodium, and calcium salt of tenuazonic acid (3-pyrrolin-2-one, 3-acetyl-1-5 sec-butyl-4-hydroxy).

Dutch elm disease: control with soil-injected fungicides and surfactants. R. J. STIPES & DONNA R. ODERWALD (Va. Polytech. Inst., Va. State Univ., Blacksburg). The control of Dutch elm disease (DED) by soil-injected fungicides was investigated. Methyl 1-(butylcarbamoyl)-2-benzimidazolecarbamate (B), N-(trichloromethylthio)-4-cyclohexene-1, 2-dicarboximide (C), and thiabendazole [2-(4-Thiazolyl)benzimidazole] (T) suspensions alone or in 1.0% (v/v) Tween 80 (polyoxyethylene sorbitan monooleate) surfactant, were injected into the root zone of 3-year-old outplanted *Ulums americana* trees 14 days prior to artificial bole inoculation with *Ceratocystis ulmi*. Seven liters of a 6,000-ppm active ingredient suspension were administered to each of nine completely randomized elms/treatment. Mean visually estimated foliar symptoms in B-, T- and Ctreated and check trees were 1.0, 52, 78, and 68%, respectively. Tween 80, as an adjunct, further reduced symptoms ultimately by 11 and 8% over C and T alone, but C. ulmi was isolated in significantly higher frequencies in all Tween 80-treated trees. Bioassays of solvent-extracted leaves revealed the presence of B, C, and T 1 day after application. These findings suggest promise of controlling DED in outplantings with B by this method.

Dothiorella canker of dawn redwood (Metasequoia glyptostroboides). R. J. Stipes, F. S. Santamour, Jr., & R. C. Lambe (Va. Polytech. Inst., Va. State Univ., Blacksburg,

U.S. Nat. Arboretum, Washington, D.C.). We observed a new, destructive, perennial canker on 20-year-old dawn redwood (Metasequoia glyptostroboides) in the United States National Arboretum, Washington, D.C. Affected stems (2-20 cm diam) with wilting and necrotic leaves were ultimately girdled. Isolations from cankers consistently yielded a Dothiorella in pure culture. The colony on potato-glucose agar was first white, then turned olivaceous gray with a darkly pigmented agar. Koch's postulates were confirmed by inoculation of seedlings in the greenhouse. The fungus produced typical white rot lesions on apple (Malus sylvestris) fruits, and also cankers on stems of apple and white pine (Pinus strobus). The apple white rot fungus (Botryosphaeria dothidea = B. ribis), which we believe the pathogen to be, also produced similar cankers on Metasequoia stems. The syndrome on Metasequoia also included marked resin exudation and extensive discoloration of xylem and phloem tissues in advance of the visible canker lesion. Rapid blighting resulted commonly from xylem-inoculated stems. Neither pycnidia nor perithecia were observed on natural infections, whereas pycnidia were abundant on apple fruit and stem lesions.

Effects of fluoride on pollen germination and pollen tube growth. C. W. Sulzbach & Merrill R. Pack (Wash. State Univ., Pullman). Tomato plants continuously exposed in atmospheres having 5 µg or more of F/m³, as HF, produced smaller fruits or fruits with fewer seeds, or both, than similar control plants. Such treatment reduced pollen germination and subsequent pollen tube growth. Pistils of self- and manually pollinated flowers (the latter emasculated when flowers were fully open) were removed 2 to 10 days after pollination, stained with aniline blue, and observed under UV light. The amount of pollen on the stigma, per cent of grains which germinated, number of tubes growing to the base of the style, and number of tubes reaching ovules were determined. Pollen from HFexposed plants had lower germination percentages in vitro than pollen of the controls. A similar result was obtained when NaF was added to the germinating media. Less pro-nounced results were obtained in cross pollinations (both pollen of controls to HF exposed and the reciprocal), suggesting that the F affected both the male and female parts of the flower. Inhibition of germination and tube growth appears to be proportionately related to the concentration of the F; however, the degree of the F effect can be par-tially regulated by the amount of Ca within the nutrient solution or media.

Ecology of root rot of turnips grown for processing. D. R. Sumner (Univ. Ga. Coastal Plain Exp. Sta., Tifton). Turnip roots grown for processing (with greens) frequently are severely damaged by root rot in the Georgia Coastal Plain. Brown external discoloration is usually first observed near the top of the roots, and the disease progresses down the vascular tissue in the interior of the root. Both Colletotrichum higginsianum and Erwinia sp. were commonly isolated from infected roots. Roots were inoculated by wounding or nonwounding with both pathogens alone and together. Typical symptoms were produced only with C. higginsianum and C. higginsianum + Erwinia sp. The bacterium alone caused only an ashy gray-to-white soft rot with a distinctive putrid odor. In a field test, 71% of the roots were unacceptable for processing when the fungus + the bacterium were injected into the roots, as compared to 17% when the fungus alone was injected and 40% when the fungus was sprayed on the lower fo-liage and tops of the roots. When the fungus was sprayed on only upper leaves or when plants were not inoculated, only 15% of the roots were unacceptable. In greenhouse tests, soil moisture had no significant affect on root rot. Boron deficiency plus the pathogens significantly increased the percentage of unacceptable roots as compared to boron deficiency or the pathogens alone.

Etiology of crazy top of corn. M. H. Sun & A. J. Ullstrup (Purdue Univ., Lafayette, Ind.). Sclerophthora

macrospora has long been implicated as the incitant of crazy top of corn. However, proof of pathogenesis based on fulfillment of Koch's postulates has not been established. Artificial inoculation was achieved by the following technique: young "green island" leaf tissue of diseased seedlings growing from infected seeds was incubated in water at 24-26 C for 24 hr for production of sporangia. The sporangial suspension was then incubated at 16 C for 6-12 hr to stimulate sporangial germination and zoospore production. The motile zoospores were injected into coleoptiles of 2- to 3-day-old corn seedlings (Pioneer Hybrid 3306) with a syringe. The inoculated plants were incubated at 16 C for an additional 48 hr before transfer to the greenhouse (24-28 C). The first green island symptoms appeared on the fourth or fifth leaf about 4 weeks after inoculation. Thereafter, systemic symptom development was similar to that of the diseased plants growing from infected seeds. Sporangia were isolated from the artificially inoculated plants and successfully used for reinoculation. Infection has been as high as 70% with the method, and the technique is being used for testing susceptibility of corn genotypes.

Purification and characterization of a severe strain of peanut mottle virus. M. K. C. Sun & T. T. Hebert (N.C. State Univ., Raleigh). A severe disease of peanut (Arachis hypogaea) having chlorotic, sometimes necrotic, mosaic leaf symptoms is caused by a severe strain of peanut mottle virus (PMV-s). PMV-s infected only nine species of Leguminosae of 30 mechanically inoculated species in eight plant families. Yield reduction was 50% and 70% in Florigiant and NC₂ peanut in the field, respectively. Seed transmission was 0.001% in NC₂. In unbuffered sap from Pisum sativum 'Alaska Early', properties were: thermal inactivation between 60 and 65 C, dilution end point between 10-3 and 10-4, and longevity at room temperature 12 hr with a half life of 83 min. Virus was purified from Alaska Early peas by chloroform-butanol clarification, polyethylene glycol precipitation, and resuspension of the virus in 0.025 M Na₂HPO₄, pH 7.4, buffer containing 0.025 M sodium sulfite and either 0.1-0.6 M urea or guanidine-HCl. PMV-s particles are flexuous rods with a normal length of 740 nm. Virus inclusions were similar to members of the potato virus Y group. PMV-s was not serologically related to soybean mosaic, bean common mosaic, potato Y, or tobacco etch viruses.

Extractable phenols in clear, discolored, and decayed tissues of sugar maple and red maple. T. A. TATTAR (Univ. New Hampshire, Durham). Ethyl acetate extracts were prepared from hot water extracts of 10-g samples of clear, discolored, and decayed tissue of sugar maple, Acer saccharum, and red maple, A. rubrum. Extracts were run on 2-way cellulose thin-layer chromatography plates with butanol:acetic acid:water (6:1:2) and 7% acetic acid: 0.03% sodium acetate. Total phenols were determined by the Folin-Ciocalteu method on methanol extracts of clear red maple tissue. Gallic acid, its esters, and catechin were identified as the major phenols in clear tissue of both red maple and sugar maple. These phenols were absent from discolored and decayed tissue. An unidentified flavonoid appeared only in discolored tissues. Total phenols in clear red maple tissue were highest (10 mg/g) near the cambium, and decreased progressively toward the pith to 5 mg/g. The processes of discoloration and decay result in decreases in extractable phenols confined to clear unaffected tissue. Total phenols in clear tissue of red maple are highest in the most physiologically active tissues near the cambium. The low level of phenols in discolored wood may permit the growth of decay fungi unable to grow at the phenol concentration in clear tissue.

Synnemata induction in Ceratocystis ulmi. P. A. TAYLOR, E. B. SMALLEY, & F. M. STRONG (Univ. Wis., Madison). Wild-type isolates of Ceratocystis ulmi do not readily form synnemata (coremia) on common culture media unless such

media contain elm wood fragments or extracts of elm wood. Aqueous and methanolic extracts of elm wood, chromatographed on a polyamide column, yielded a "phenolic fraction" and a "water fraction" containing nutrients. The fungus grew well on 1% agarose gel containing the "water fraction", but did not produce synnemata unless the "phenolic fraction" was also present. Fructose in the "water fraction" was essential for synnemata production, but not for fungal growth. A phenol was isolated by successive chromatography on polyamide and silica gel columns. When added to the "water fraction"-agarose gel or to potato-dextrose agar (PDA), this phenol induced C. ulmi to form synnemata. Synnemata production in PDA was enhanced when the added glucose in PDA was replaced by fructose. The active phenol was identified as catechin (3,3', 4',5,7-pentahydroxyflavan) by UV and proton magnetic resonance spectroscopy, mass spectrometry, and cochromatography. Authentic D-(+)-catechin was identical to the isolated compound in both biological and chemical aspects.

A chloroneb-resistant mutant of Ustilago maydis. R. W. TILLMAN & H. D. SISLER (Univ. Md., College Park). A mutant of *Ustilago maydis* resistant to chloroneb was isolated from medium containing 8 µg/ml of toxicant. Growth of the mutant was not inhibited in medium containing 16 $\mu g/ml$ of chloroneb; whereas growth of the wild type was prevented by $8\,\mu g/ml$. There was no appreciable difference in the uptake of ring-labeled or methyl-labeled chloroneb-14C by the mutant or wild type. All radiolabel initially added to mutant cultures could be recovered after 24 hr as unaltered chloroneb. The chloroneb-resistant mutant was also resistant to 2,6-dichloro-4-nitroaniline, diphenyl, hexachlorobenzene, napthalene, p-dichlorbenzene, pentachloronitrobenzene, and sodium-o-phenylphenate. Wild type and mutant were equally sensitive to 2,4,6-trichlorophenol; 2,4,5,6-tetrachlorisophthalonitrile; o-benzyl-chlorophenol; methyl-2-benzimidazole carbamate; and 1,2,4trichloro-3,5-dinitrobenzene. Tolerance to chloroneb progeny from a cross of the resistant mutant to a sensitive wild type indicated single gene resistance.

The capacity of leaf extracts of tobacco to react with ozone. H. Tomlinson & S. Rich (Conn. Agr. Exp. Sta., New Haven). The capacity of leaf extracts of tobacco to react with ozone was tested by measuring the ability of these extracts to inhibit the peroxidation of methyl linolenate exposed to ozone. Methyl linolenate (60 µg) suspended in 3 ml neutral 0.025 M phosphate buffer plus 40 μg of sodium lauryl sulfate and 10% ethanol gave a linear increase in peroxidation when ozonated for 1-4 min by an air stream containing 6 ppm ozone bubbled through at the rate of 1 liter/min. Peroxidation was measured as the increase in malonyl dialdehyde (MDA). Cysteine (20, 40, and 60 µg), added before ozonation, inhibited MDA formation 45, 55, and 65%, respectively, when ozonated for 2min, and 10, 30, and 48% when ozonated for 4min. Juice expressed from a single frozen disc (3 cm-diameter) of tobacco leaf (Nicotiana tabacum) also inhibited formation of MDA when added before ozonation. After 2 min of ozonation, juice from an ozone-resistant and an ozonesusceptible cultivar inhibited the formation of MDA 52 and 55%, respectively; after 4 min of ozonation, MDA formation was inhibited 4% and 28%, respectively. Therefore the capacity of sap from the susceptible cultivar to react with ozone remained equivalent to 40 µg of cysteine, whereas the capacity of the resistant cultivar was depleted.

Processes of abnormal cell wall thickening in potato virus M-incited lesions in Red Kidney bean. J. C. Tu & C. Hiruki (Univ. Alberta, Edmonton, Can.). Abnormal secondary wall thickening due to deposition of paramural bodies was found in cells in the peripheral zone of potato virus M-incited lesions in Red Kidney bean. The initiation of the deposition in an affected cell was closely associated with increased cellular metabolic activities evidenced by increased numbers of ribosomes and mitochondria. Slight roughening of the plasmalemma was observed at this stage.

Later, the roughening extended into invagination of various degrees and, therefore, numerous invaginated periplasmic spaces were formed in the cell. Increase in numbers of dictyosomes and in deposition of dictyosome-related vesicles and microtubules occurred near or in the invaginated periplasmic spaces. Individual deposits of paramural bodies on the normal secondary wall became thickened, extended, and then coalesced with the adjacent depositions and finally coated the inner cell wall. The deposition initially resembled a loosely-packed spongelike matrix, later filled with fine fibrous materials, and eventually became very dense

Effect of streptomycin on development of fire blight in artificially injured pear fruit, T. VAN DER ZWET & H. L. Kell (PSRD, ARS, USDA, Beltsville, Md.). Immature Bartlett pear fruit (4-5 cm) in the orchard were injured by bruising with the head of a thumbtack or by puncturing with a needle or nail at intervals of 0, 1, or 2 days before or after inoculation. The fruit were inoculated by dipping for about 5 sec in an aqueous cell suspension (5 \times 10⁷ cells/ml) of Erwinia amylovora. At the time of injury, the fruit were dipped for 5 sec in a suspension of 100 ppm streptomycin. The antibiotic provided unsatisfactory control when applied at time of inoculation (0 days), regardless of the type of injury. Streptomycin applied 1 day before inoculation protected most of the punctured fruit against infection. Bruised fruit, with or without streptomycin, showed considerably more blight than did punctured fruit, regardless of the interval between injury and inoculation. In laboratory tests, when fruit were injured, inoculated, and dipped in streptomycin at the same time, 5 times more fruit blighted when skin rupture accompanied bruising than without rupture. Without streptomycin, all fruit blighted regardless of degree of bruising. Under the severe conditions of these tests, streptomycin was most effective in preventing development of fruit blight only when it was present at the time of injury.

Studies on the mode of action of phaseollin. H. D. VAN ETTEN & D. F. BATEMAN (Cornell Univ., Ithaca, N.Y.). Phaseollin at 9, 15, or 47 μg/ml inhibited the ability of Rhizoctonia solani to remove p-glucose-U-14C from a liquid medium (15 µg/ml is the min concentration needed to prevent an increase in dry wt of R. solani in shake culture). Microscopic examination of growing hyphae exposed to phaseollin revealed immediate (< 1.0-min) cessation of protoplasmic streaming, often accompanied by shrinkage of the protoplasts from the hyphal tips. Phaseollin (15 µg/ml) incubated with autoclaved or nonautoclaved R. solani mycelium was taken up rapidly by the mycelium during the first 10-15 min, after which the rate of uptake leveled off; after 1 hr ca. 30% of the phaseollin remained in solution. Biological degradation of phaseollin (15 µg/ml) was not detected 1 hr after exposure to live mycelium. When R. solani was exposed to phaseollin-14C (15 µg/ml) for 1 hr and then fractionated, most of the ¹⁴C (73%) was associated with the hyphal fraction removed by centrifugation at 500 g for 15 min. Phaseollin (23 µg/ml or above) rapidly lysed sheep red blood cells. The data support the hypothesis that the inhibitory action of phaseollin is associated with its ability to disrupt normal membrane func-

Comparative histopathology of the resistant and susceptible response of Pelargonium spp. to Xanthomonas pelargonii. S. H. Wainwright & P. E. Nelson (Pa. State Univ., University Park). The histology of Pelargonium spp. susceptible, moderately resistant, and resistant to Xanthomonas pelargonii was studied in inoculated and noninoculated plants. The most striking difference was the presence of tanninlike materials in the tissues of moderately resistant and resistant species, and their absence in susceptible species. The mode of spread of the pathogen was similar in all species, initially involving movement of the pathogen throughout the plant in xylem vessel elements and subsequent movement laterally into adjoining parenchyma cells.

The relative numbers of the pathogen and the numbers of fascicles initially invaded were low in resistant species and high in susceptible species. In susceptible species, bacterial pockets formed around affected primary xylem vessel elements, enlarging to encompass all xylem cells in the fascicle, then portions of the cambium, phloem, cortex, and epidermis. *Pelargonium* spp. responded to infection by proliferation of a ring of cells around affected portions of fascicles, with cells immediately inside this ring having a suberinlike material formed on their walls. This response was greatest in the most susceptible plants, and decreased as resistance increased.

Composition of the surface of the cotton boll in relation to spore germination and resistance to Diplodia gossypina infection. S.-Y. C. Wang & J. A. Pinckard (La. Agr. Exp. Sta., Baton Rouge). Diplodia gossypina attacks the bolls of many cultivars of cotton, being most destructive of very young bolls, less than 15 days of age, and bolls over 25 days old. The nature of the resistance of bolls of intermediate age, 15 to 25 days, was the purpose of this investigation. Surface washings of intermediate aged bolls and the nectarial fluids of the cultivar Deltapine 16 showed the presence of fructose, glucose, galactose, sucrose, raf-finose, and 2 unknowns which stimulated spore germination. Upon removal of boll waxes with hexane and analysis of the leachates, similar sugars were found, with the exception of raffinose. Maximal amounts of waxes and cutin acids were found on bolls of 17 and 20 days of age. These materials inhibited spore germination and mycelial growth, and may account for the observed resistance. Exopoly-galacturonase, one of the important cell-wall degrading enzymes produced by D. gossypina, was detected during spore germination, but only in the presence of boll leachates, suggesting that surface waxes and cutin are potential barriers to infection. A further study of the waxes and cutin of bolls of several cultivars may indicate materials useful for improved resistance.

Enhanced growth of Armillaria mellea on extracts from roots of defoliated sugar maple trees. P. M. WARGO (USDA Forest Serv., Northeastern Forest Exp. Sta., Hamden, Conn.). Defoliation of trees by insects appears to predispose them to attack by Armillaria mellea. Certain sugars, amino acids, and fatty acids that occur in roots stimulate growth of A. mellea in vitro. Defoliation can alter some of these chemical constituents, and these changes may be related to increased attack of defoliated trees by A. mellea. Root tissues from defoliated and nondefoliated sugar maple saplings were analyzed for changes in starch sugars, amino acids, and fatty acids. Defoliation caused a significant decrease in the starch content of whole roots, and a corresponding increase in glucose and fructose in the outer wood but not in the bark. Both total number and concentration of some amino acids increased in roots of defoliated trees, especially in the outer wood. Defoliation had no apparent effect on fatty acids, and concentrations were less than 0.0010% dry wt. Growth of A. mellea was significantly greater on outer wood extracts from defoliated trees, but not on bark extracts. It appears that defoliation causes chemical changes in the outer wood of roots that are favorable for the growth of Λ . mellea.

Influence of soil temperature on root rot of peach caused by Cylindrocladium floridanum. D. J. Weaver (ARS, USDA, Byron, Ga.). Six-week-old Elberta peach seedlings were transplanted into steam-sterilized soil, either noninfested or infested with microsclerotia of C. floridanum, and maintained in water baths at 15, 20, 25, and 30 C in the greenhouse. Six weeks later, roots were washed free of soil, rated for severity of rotting, dried, and weighed. Samples of infected roots from each inoculated treatment were subjected to routine isolation procedures. Cylindrocladium floridanum was readily isolated from roots of inoculated seedlings. Root rot was slight at 15 C, moderate at 20 C, and severe at 25 and 30 C. Generally, soil temperatures that favored root rot were less favorable for growth

of roots. The fungus was grown on potato-dextrose agar for 7 days at temperatures ranging from 9 to 36 C. Greatest colony diameters occurred at 27 C.

Studies on the relationship of the hoja blanca virus to the planthopper vector, Sogatodes oryzicola. A. J. Webber, Jr., V. D. Damstegt, & C. L. Graham (Plant Sci. Lab., Ft. Detrick, Frederick, Md.). The effects of the hoja blanca virus (HBV) of rice on Sogatodes oryzicola were studied through several generations with replicated single-pair matings and reciprocal crosses of transmitting and nontransmitting individuals in all combinations. Reduced fe-cundity, nymph viability, and adult longevity in the transmitting lines indicated that the virus adversely affected the vector. Both mean daily and total nymph production by viruliferous females was less than that by nonviruliferous females regardless of the virus-transmitting ability of the fertilizing male. Virus acquisition studies with nontransmitting insects from populations containing 8 and 64% transmitters (through transovarial passage of HBV) and from a virus-free population resulted in acquisition rates of 7.5, 6.0, and 7.0%, respectively. Deleterious effects of the virus on the vector and the low rate of virus acquisition may be compensating factors stabilizing the virus level in a randomly breeding population.

Influence of conditioning environment of Bermudagrass on sex differentiation of Meloidogyne graminis. A. J. Webber, Jr., & J. A. Fox (Va. Polytech. Inst., Va. State Univ., Blacksburg). Sex differentiation of Meloidogyne graminis was found to differ on Tifgreen Bermudagrass conditioned under different temperature and nutrient regimes. Bermudagrass sprigs from plants conditioned at 26 and 32 C or at a low and high nutrient level were rooted in distilled water. Rooted sprigs were inoculated with 150 larvae at 26 C and incubated at 26 or 32 C. Differences in sex ratios obtained at an incubation temperature of 26 C were not significant, but at an incubation temperature of 32 C the nematodes from sprigs conditioned at 26 C averaged 44% males, whereas those from sprigs conditioned at 32 C averaged 21% males. Nematodes from sprigs conditioned at a low nutrient level averaged 45% males, whereas those from sprigs conditioned at a high nutrient level averaged 16% males. The indirect influence of environment on sex differentiation may be the result of differences in physiological or biochemical factors of host origin.

Virulence repression in Rhizoctonia solani by 3-0-methyl glucose. A. R. Weinhold & T. Bowman (Univ. Cal., Berkeley). To investigate the influence of exogenous substrates on virulence in Rhizoctonia solani, 5-day-old cotton seedlings (Gossypium hirsutum 'Acala 4-42') were supported by quartz sand on glass plates and inoculated with mycelium grown in liquid culture. The inoculum was placed in contact with sand saturated with water or solutions of test materials. Previous studies have shown that an exogenous supply of glucose (sand saturated with a 10 g/liter solution) reduced disease severity and repressed the production of pectolytic enzymes by R. solani in vitro. When the sand was saturated with a solution of 3-0-methyl glucose (5 g/liter), disease development was completely inhibited. Growth of the pathogen was somewhat reduced in the presence of 3-0-methyl glucose (3-0-MG), but this was overcome by addition of a small amount of glucose (2.0 g/liter). The action of 3-0-MG differs from that of glucose. The pathogen readily grows on the surface of the cotton stem in both cases. With 3-0-MG, however, infection cushions did not form. The production of pectolytic enzymes in vitro is not repressed by 3-0-MG. These results suggest that 3-0-MG interferes with infection cushion formation, and indicates that pectolytic enzyme production is not the sole factor in virulence of R. solani.

Heated wax-emulsions with benomyl and 2,6-dichloro-4-nitroaniline for control of postharvest decay of peaches. J. M. Wells (ARS, USDA, Beltsville, Md.). Mean lesion

diameters of peaches, inoculated with Monilinia fructicola or Rhizopus stolonifer and treated with heated (52-C) or unheated (24-C) dips or sprays for 10, 20, or 30 sec with 225, 450, or 900 ppm 2,6-dichloro-4-nitroaniline (DCNA) suspended in a wax-emulsion, were significantly smaller than those on fruit similarly treated with DCNA suspended in water. Similar wax-benomyl treatments at 10, 33, or 100 ppm were also more effective than benomyl treatments alone. Heated DCNA treatments, whether as dips or sprays, with or without wax, were more effective than unheated treatments. Heat improved the effectiveness of all benomyl-dip or of 10-sec wax spray treatments. Means of per cent decay of noninoculated peaches and nectarines due to Monilinia and Rhizopus infections were significantly less when treated with a 10-sec heated wax spray containing 450 ppm DCNA and 100 ppm benomyl than with a 3-sec unheated spray.

Isolation of Alternaria alternata from Nicotiana species. R. E. WELTY & H. W. SPURR, JR. (USDA, N.C. State Univ., Raleigh). We determined the incidence of the brown spot organism, Alternaria alternata, on leaves of susceptible (C-298) and resistant (PD 121) cultivars of field-grown, fluecured tobacco, and on leaves of Nicotiana rustica, N. glutinosa, and N. sylvestris. Discs (9-mm diam) were cut from apparently healthy leaves, surface-disinfected in 1% NaOCI for 1 min, rinsed in tap water, and placed on either rose-bengal streptomycin or Czapek + 6% NaCl agars or both. No measurable host differences were noted as the fungus was recovered from 97 to 100% of the discs cultured regardless of host. In a study of the effect of leaf age on the frequency of isolation of the fungus, leaf discs were ran-domly cut from leaves of C-298 and PD 121 in the plant bed before and after the ferbam applications for blue mold control were discontinued, and at 10 weekly intervals after transplanting. The percentage of leaf discs from which A. alternata grew was 0 when ferbam was applied, 29% after it was discontinued, 64-94% for 3-week-old leaves, and 93-100% for 7-week-old leaves (mature). Thus, the frequency of isolation of A. alternata is high in tobacco leaves of all ages, except in those protected by selected chemicals.

Ultrastructural investigation of susceptible and resistant isolines of soybean inoculated with Xanthomonas. W. P. WERGIN & M. E. MACE (ARS, USDA, Beltsville, Md.). A fine structural study was made of the host-parasite inter-action between soybean (Glycine max) and Xanthomonas phaseoli var. sojensis. Fully expanded leaves of susceptible (Clark) and resistant (Clark 63) isolines of soybean were spray-inoculated with log-phase cultures of the bacterial pustule pathogen. One day after inoculation, examination of leaves from the two isolines revealed small colonies of bacteria appressed to the walls of the spongy parenchyma cells in the lower substomatal chambers. Regions of the wall bounded by the bacteria appeared to undergo gradual hydrolysis. Within 48 hr, progressive loosening and dispersal of wall microfibrils resulted in the rupture of several mesophyll cells. Three days after inoculation, structural differences between the isolines could be observed in the remaining mesophyll cells. In susceptible plants, the cells enlarged, pushed the lower epidermis outward, and formed a pustule. In the resistant plants, cellular expansion did not occur; however, cisternae of endoplasmic reticulum, pro-lific numbers of ribosomes, and an abundance of vesicles were present in the parietal layer of cytoplasm. Since these organelles are associated with protein synthesis and extracellular secretion, they may be related to a mechanism of disease resistance.

Lima beans resistant to Phytophthora phaseoli, strain C. R. E. Wester, V. J. Fisher, & V. L. Blount (ARS, USDA, Beltsville, Md., Univ. Del., Newark). Of 34 lima bean plant introductions from Central America that were tested in October 1970 at Beltsville for resistance to downy mildew, strain C, four showed complete resistance. These introductions were also resistant to downy mildew strains A and B. The others tended to be completely susceptible.

A few seedlings survived inoculation, but apparently these were escapes, as all progenies from them were completely susceptible to strain C. Two of the resistant introductions were white-seeded (P.I. 195342 and P.I. 197025); two were black-seeded (P.I. 195337 and P.I. 195345). The white-seeded introductions closely resembled small-seeded commercial varieties, and bloomed earlier than the black-seeded introductions. The white-seeded introductions were chosen as parents for crossing to Dover, U.S.D.A. line 1068, Del. line 255-S4, and U.S.D.A. lines 169, 269, and 369. F1 seedlings were screened and found resistant to all known strains of downy mildew (A, B, C). The F2 generation segregated in ratio of three resistant to one susceptible, indicating that resistance is controlled by a single dominant gene as reported earlier for strains A and B.

Host responses and ultrastructural changes in maize leaves infected with Colletotrichum graminicola. H. Wheeler, D. J. Politis, & A. S. Williams (Univ. Ky., Lexington). Young maize plants (10 days old) sprayed with a spore suspension (2 × 10⁵ spores/ml) from an isolate of Colletotrichum graminicola obtained from naturally infected maize developed visible lesions within 36 hr. and many were killed within 5 days. Older plants (15-20 days) usually survived, but lower leaves were killed and many lesions formed on upper leaves. All maize cultivars tested (14 commercial hybrids and 4 inbred lines) were susceptible, but some were damaged more than others. Appressoria formed within 5 hr, epidermal cells were invaded within 8 hr, and acervuli with setae developed within 24 hr after inoculation. Spores, germ tubes, and appressoria showed no unusual ultrastructural features. The earliest host response was an increase in electron density and a swelling of the cell wall beneath appressoria. As penetration progressed, many lomasomelike cell wall structures developed. The ability of this isolate to attack intact, nonwounded tissues and to kill young seedlings indicates that it is more pathogenic to maize than others of C. graminicola previously found in the USA.

Synergistic effect between Heterodera schachtii and Pythium ultimum on damping-off of sugarbeet vs. additive effect of H. schachtii and P. aphanidermatum. E. D. WHITNEY (ARS, USDA, Salinas, Cal.). Damping-off tests of hybrid sugarbeet inoculated with H. schachtii-P. ultimum and H. schachtii-P. aphanidermatum combinations were run simultaneously at various inoculum levels. The effect of the first combination on damping-off was synergistic but the second was additive. The synergistic effect was apparent at most inoculum levels and for both preand postemergence damping-off. In four tests, the presence of the nematode (10 larvae/g of soil) had increased damping-off by an average of 260% 10 days after planting. No reduction in seedling numbers resulted from the nematode alone. The mean number of infection centers per flat of beets was approximately the same whether the nematode was present or not. This suggested that the synergistic effect was due to increased rate of spread of the fungus in the soil around the infection centers when the nematode was present.

Inheritance of resistance to Xanthomonas campestris in cabbage. P. H. WILLIAMS & T. STAUB (Univ. Wis., Madison). Resistance in cabbage to black rot, caused by Xanthomonas campestris, was obtained from the Japanese cultivar, Early Fuji. When bacteria were inoculated through the hydathodes, resistance was expressed as either minute dark areas at the hydathodes or as nonexpanding light brown panels, 0.5-1 × 1-4 cm, bounded by the larger veins and surrounded by a dark brown rim. Lesions on susceptible plants were V-shaped and surrounded by an expanding chlorotic zone. Based on field screening of F1, F2, and backcross progenies, resistance was found to be controlled by one major recessive gene, r, the expression of which in the heterozygous condition was influenced by one recessive and one dominant modifier gene. Of over 350 cultivars and breeding lines screened for black rot resistance, none

contained the major r gene for resistance; however, some contained the modifier genes.

Comparison of peroxidase and o-diphenol oxidase in postharvest apple decay by Penicillium and Physalospora. D. M. WILSON & W. B. DARBY (Univ. Vt., Burlington). McIntosh apples inoculated with Penicillium sp. or Physalospora obtusa were used. Penicillium causes a light soft rot. As P. obtusa decay progresses, the skin is brown and the pulp light; later the skin turns black and the flesh darkens. Peroxidase, o-diphenol oxidase, and catalase activities were measured using skin and pulp extracts from healthy apples, decayed areas, uninvaded areas, and samples from the first 5 mm of firm tissue starting at the edge of visible change. Using guaiacol and H2O2 as substrates, peroxidase activity was low in decayed areas. Peroxidase activity in edge or uninvaded tissue and skin of both rots increased 2.5 times over controls. o-Diphenol oxidase activity using 3,4-dihydroxyphenylalanine as the substrate was undetectable in rotted areas. When P. obtusa-rotted apples were darkening, o-diphenol oxidase activity was again present. Inhibition of o-diphenol oxidase by low molecular wt substances was seen only in edge or macerated Penicillium-infected tissue. Catalase was absent in rotted tissues. Soluble protein decreased with P. obtusa only. These results indicate that enzymes in uninvaded tissue respond differently during pathogenesis. Inhibition of o-diphenol oxidase by Penicillium may contribute to the light color; the dark P. obtusa mycelia are responsible for most of the black color in black rot.

Relationship of basidiospore germination of Fomes igniarius to host specificity. J. E. Winch & P. Manion (State Univ. N. Y. Coll. Forest., Syracuse). Basidiospores collected from fruiting bodies of Fomes igniarius on aspen, birch, back and butters to be a specific production of the state of beech, and butternut were used to inoculate birch, beech, aspen, and maple in all possible combinations. Spores were introduced into holes drilled into the sapwood of living trees in the field. Spores also were placed on cellophane membranes in contact with freshly cut cross sections of stem pieces and were applied directly to cross sections of wood. In both laboratory tests, a short piece of 6-mm tygon tubing, closed at the upper end, was placed over the inoculation site forming a miniature chamber to maintain favorable relative humidity. Germination was determined after 1 week. In the field, germination occurred when spores from fruiting bodies on aspen were inoculated into aspen and failed with all other combinations. Spore suspensions placed on stem sections with membranes germinated in all combinations used, but an initial laboratory test using the spore source from aspen indicated that germination does not occur when spores are placed directly on the cross sections of the 4 hosts.

Electron microscopic studies on the effects of tetracycline HCl on the mycoplasmalike bodies in corn stunt and aster vellows-infected plants. B. S. Wolanski, M. Klein, & K. MARAMOROSCH (Boyce Thompson Inst. Plant Res., Yonkers, N.Y.). The effect of tetracycline HCl on the presumptive agents of aster yellows and corn stunt disease was studied by electron microscopy of thin sections of treated aster and corn plants. The effects of the antibiotics on the morphology of the mycoplasmalike bodies could be observed 24 hr after treatment, and were most pronounced after 48 hr. After 72 hr, most of the phloem tissue examined was almost free of mycoplasmalike bodies; any still present were in obvious stages of degradation. The changes in morphology of mycoplasmalike bodies resulting from the tetracycline HCl treatment included condensation of the granular and fibrous contents of the structures into sporelike masses. This was followed by complete emptying of the bodies and disruption of their membranes. Tetracycline treatment suppressed symptoms of the disease in asters for a period of 2 weeks, at the end of which time typical mycoplasmalike bodies could again be found in the phloem tissue.

Regulation of the perfect stage in Fusarium roseum 'Graminearum' by F-2 (zearalenone). J. C. Wolf & C. J. Mirocha (Univ. Minn., St. Paul). Fusarium roseum 'Graminearum' was seeded onto 1.5-cm discs of Coons' medium agar. F-2 in quantities ranging from 10^{-2} to 10^{-4} µg was delivered onto the medium in diethylether-dimethylsulfoxide (97:3, v/v). In general, amt from 10^{-4} to 10^{-2} µg enhanced the production of perithecia, whereas amt from 10^{-1} to 1 µg inhibited perithecial production. The period of effective inhibition of perithecial production was longer when amt of 10 µg and greater were used. The time of F-2 application to the developing colony was important. Greater amt of F-2 were needed to inhibit production of perithecia when introduced into the culture 1 day after seeding than 3 days after seeding.

Identification of ozone-type air pollution injury to vegetation in Philadelphia. F. A. Wood (Pa. State Univ., Univ. Park). Exposures of 35 species of broad-leaf woody plants to ozone (O3) during the past 5 years indicated that chlorotic-to-purple stippling of the upper leaf surface of middle-aged to-older leaves was the usual response. Recently established vegetation in Independence National Historical Park, Philadelphia, Pa., was examined 4 times during the summer of 1970. Purple stippling of the upper leaf surface was observed on American basswood, tree of heaven, and a Prunus sp. in July. In addition, ozone-type stippling was observed on Carolina silverbell, English oak, European mountain ash, flowering dogwood, grape, haw-thorn, mulberry, and rhododendron in August. Forty white ash seedlings were planted as O3 indicators at seven locations in the park. Stippling typical of O3 injury developed on these plants within 4 weeks after planting. Fluoridetype symptoms also were observed, and fluorine levels in some of the plants indicated a fluoride problem, but ozonetype stippling was the most common symptom. These results substantiate the philosophy that one should examine the relative sensitivity of an array of species in the laboratory and then survey for symptoms in the field rather than attempt to elucidate the specific cause of each symptom complex as it is encountered in the field.

Genetic complementation between the nucleoprotein components of the cowpea mosaic virus. H. A. Wood & J. B. Bancroft (Boyce Thompson Inst., Yonkers, N. Y., Purdue Univ., Lafayette, Indiana). The bottom (B) and middle (M) nucleoprotein components of cowpea mosaic virus strain Vu (wild isolate) interact, resulting in increased levels of infectivity. Two local lesion mutants, designated 3d and 7b, were isolated following nitrous acid treatment. The 7b mutant had an altered nucleoprotein component ratio. In systemic assays, homologous and heterologous mixtures of the wild and mutant B and M components were infective at a 100-fold lower concentration than the individual components in the mixtures. Mixed component (MC) isolates from the infectivity dilution-end points produced only one lesion type. The lesion types and component ratios of the MC isolates were generally the same as the isolate from which the B component in the mixture was taken. However, the MC isolates arising from 3d-B and 7b-M component mixtures had wild lesion types and component ratios. The nucleoprotein components from this MC isolate were mixed and assayed with the wild and mutant B and M components. The backcrossed-MC isolates had lesion types and component ratios which indicated the occurrence of genetic complementation, as the MC-B and -M components had the same properties as the original 3d-B and 7b-M components, respectively.

Use of dew point and ambient temperature differences to explain epidemic development of mushroom bacterial blotch. P. J. WUEST (Pa. State Univ., University Park). Bacterial blotch of the commercial mushroom is caused by Pseudomonas tolaasi. All commercially grown strains of Agaricus bisporus are susceptible to the disease. The pathogen is soil-borne and a normal component of casing (soil) microflora, so disease occurs when environmental condi-

tions permit. Disease occurrence and development were studied in two experiments in which PSU 310 was the suscept. Temperature variation in each forced-air mushroom-growing room did not exceed ± 2 F and relative humidity did not vary more than 4%. Mushrooms were harvested daily, scored for disease incidence, and weighed. Data from harvests were summarized weekly. Disease development was measured by calculating "r" values. Dew points were computed hourly from temperature and RH values. Disease development was compared with various dew point/ambient temperature (DP/AT) differentials. When compared with days of DP/AT differentials of 3 F or more for 15 hr or more, disease increased when only 2 and 3 days, respectively, occurred during two periods of 21 days each. Disease was constant when 16 such days occurred during a different 21-day period. Disease decreased when such days occurred every day during a 14-day period.

The lack of effect of pokeweed inhibitor on retention of tobacco mosaic virus by inoculated bean leaves. S. D. WYATT & J. G. Shaw (Univ. Ky., Lexington). Pinto bean leaves rubbed with ¹⁴C-labeled viral inhibitor from *Phy*tolacca americana retained from 5 to 15% of the label after thorough rinsing with water. The percentages varied among experiments, but did not vary with changes in concentration of inhibitor within the same experiment. The amount of inhibitor retained was not altered by the presence of up to 10 mg/ml of TMV in the inoculum. The amount of 32P-labeled TMV retained by an inoculated leaf after rinsing was not significantly affected by the addition of inhibitor to inocula. Nor did the presence of inhibitor affect the amount of nuclease-susceptible 32P-RNA found in extracts of leaves 10 min after inoculation with 32Plabeled TMV. Inhibition of infection, therefore, does not appear to depend on alteration of the amount of TMV retained by inoculated leaves or alteration of the amount of uncoated TMV-RNA in extracts of the leaves.

Selective stimulation of ion uptake by Helminthosporium carbonum toxin. O. C. Yoder & R. P. Scheffer (Mich. State Univ., E. Lansing). Susceptible corn roots from 4day-old plants were treated for 4 hr with the host-specific toxin of Helminthosporium carbonum (2 µg/ml), then placed in NO₃- solutions (0.04-10 mm) for 30 min. Control roots absorbed 200-300 nmoles NO₂-/g fresh wt/hr, whereas toxin-treated roots absorbed up to 3 times that amount. The same toxin concentration did not stimulate NO3- uptake by resistant roots. Increased NO3- accumulation appears to be caused by increased influx rather than decreased efflux or decreased NO3- reduction, because a 30-min desorption period gave slight but equal loss of NO3- from treated and control roots. NO3- uptake by toxin-treated and control roots was greatly reduced at 5 C. Na+, K+, Ca++, and Cl- were not required for toxin stimulated NO₅- uptake. Toxin increased Na+ absorption from 0.2- and 20-mm solutions by 50-60%. Toxin did not affect, or slightly inhibited, absorption of K+, SO₄--, and PO4-, and did not affect P incorporation into perchloric acid-soluble and insoluble organic materials. In no case did toxin affect ion efflux under the conditions used. Enhanced absorption of NO₃- and of Na+ was not required for toxicity because toxin affected susceptible tissues in the absence of either ion. Data indicate that toxin has selective effects on membrane properties, and does not generally derange the structure.

Production of tryptophol and indoleacetic acid in culture by Pythium debaryanum. K. Yoshii & D. J. Hagedorn (Univ. Wis., Madison). Pythium debaryanum produced 0.34 µg/ml indole-3-acetic acid (IAA) after 3 days of incubation in potato-dextrose broth (PDA). Addition of tryptophan to the medium invariably resulted in a significant increase, to 0.57 µg/ml in the production of IAA, suggesting that this fungus synthesizes IAA from tryptophan. Indoleacetamide and indoleacetonitrile added to the medium at 0.1 mm increased IAA production to 2.5 and 20.0

µg/ml, respectively, but tryptophol and tryptamine had little effect. Therefore, indoleacetamide and indoleacetonitrile may possibly be precursors for IAA synthesis by this organism. Pythium debaryanum also produced a neutral indole compound in PDA. The compound was extracted with ethyl acetate from the culture filtrate at pH 7.0, eluted with chloroform from silicic acid columns, and further purified by silicic acid partition chromatography with formate and a mixture of n-hexane and ethyl acetate as the stationary and mobile phases, respectively. It was identified as tryptophol by its melting point, chromatographic behavior, and ultraviolet and mass spectra.

Tobacco mosaic virus replication in suspensions of separated tobacco leaf cells. M. Zaitlin, A. O. Jackson, & R. I. B. FRANCKI (Univ. Ariz., Tucson). Cells were separated from tobacco mosaic virus (TMV)-infected tobacco leaves with the aid of a pectinase procedure 3-4 days after inoculation. Suspensions of cells were presented with 3Huridine. At various times after incubation in the light at 25 C, unlabeled virus was added as a carrier to facilitate the isolation of the virus from the cells and to determine the ³H-uridine incorporated into virus. By this means, virus was found to multiply at a constant rate for up to 45 hr of incubation (the longest time tested). Infected cells were also allowed to incorporate radioactive amino acids and uridine in order to detect virus-specific proteins and nucleic acids. Analysis by polyacrylamide gel electrophoresis revealed at least three high molecular wt virusrelated proteins in addition to the coat protein, and two viral-induced RNA species plus the viral RNA. One of these RNA species is of very high molecular wt (ca. 4×10^6) and is considered to be the TMV replicative form. The other is a lower molecular wt RNA $(2.5\text{--}3.0 \times 10^5)$ of unknown origin and function.

Growth and movement of Xanthomonas campestris in natural fluids in relation to infection of cabbage. B. G. ZOLLER & T. KOSUGE (Univ. Cal., Davis). Movement of Xanthomonas campestris in natural cabbage fluids was studied. Cells were grown in minimal medium (0.05% sucrose as carbon source) and resuspended (4 × 108 colony-forming units/ml) in condensed water collected in the field from cabbage leaves. Ends of capillary tubes (2.38 X $10^{-4}\,\mathrm{cm^2}$ in cross section) containing various field-collected samples of guttation fluid were immersed in the bacterial suspension. After 2-10 min at 24 C, up to 100 times as many bacteria had moved into capillary tubes containing field-collected cabbage guttation fluid than moved into capillary tubes containing the resuspending fluid. Conductivity bridge measurements revealed that the conductance of the guttation fluid was higher than that of the condensed water from the leaves. Thus, the number of bacteria moving into the guttation fluid appeared to depend, at least in part, on the conductance of the fluids. Similar results were obtained in identical experiments using salt solutions having the same conductances as the natural fluids. The bacteria had a generation time of 2.7 hr at 26 C in the condensed water leaf fluid, and 3.1 hr at 26 C in guttation fluid.