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Population of Fusarium oxysporum f. sp. cepae in organic soils in New York. G. S. ABAWI & J. W. LORBEER (Cornell Univ., Ithaca, N.Y.). A selective agar medium developed for the assay of *Fusarium oxysporum f. sp. cepae* in New York organic soils consists of Martin's rose bengal medium supplemented with 2, 1, 100, and 100 ppm chlorotetracycline HCl (Aureomycin), bis(dimethylthiocarbamoyl) disulfide (thiram), *p*-dimethylaminobenzenediazo sodium sulfonate (Dexon), and pentachloronitrobenzene (PCNB), respectively. This medium also was satisfactory for the assay of *F. oxysporum* in the same soils. Only the sporodochial form of *F. oxysporum f. sp. cepae* was isolated from naturally infested soils. The fungus was not uniformly distributed within individual fields (top 15.2 cm) cropped to onion (*Allium cepa*), and was isolated to depths of 30.4-45.7 cm. The average population for individual fields (top 15.2 cm) ranged from 300 to 6,555 propagules/g of oven-dry soil. The highest populations of the fungus generally were found in fields with long histories of *Fusarium* basal rot. In several instances, however, high populations of the fungus were detected for fields with either light or no known outbreaks of *Fusarium* basal rot even when susceptible cultivars had been grown.

Onion white rot caused by Sclerotium cepivorum as affected by soil temperature, pH, and inoculum density. P. B. ADAMS & G. C. PAPAVIDAS (ARS, USDA, Beltsville, Md.). The optimum temp for white rot development on onion (cultivar Yellow Globe Danvers) was 15 C. Disease severity was less at 10 and 20 C than at 15 C. The optimum temp for germination of sclerotia of *Sclerotium cepivorum* in autoclaved soil was 20 C, with an optimum range from 15 to 25 C. Little germination occurred at 5, 10, and 30 C. More than 50% of the sclerotia germinated in autoclaved soil at several pH levels tested between 4.5 and 7.8. At pH 5.0 or below, less than 30% of the plants were infected; at pH 6.0 or higher, 90% of the plants were infected. Several isolates of *S. cepivorum* from Europe, Canada, and the USA were tested for virulence on Yellow Globe onion. Sclerotia were produced on Czapek-Dox agar (pH 5.2), separated by wet sieving, air-dried, and added to soil to establish known inoculum densities. Of 12 isolates tested at an inoculum density of 25 sclerotia/g of dry soil, 10 were considered highly virulent. As few as 5 sclerotia/g of dry soil were required to obtain visible infection. Twenty-five sclerotia/g of dry soil were required to obtain 50% infection. Fifty sclerotia/g resulted in more than 85% infected plants.

Correlation of counts of wound tumor virus particles with their RNA and protein content. M. E. AHMED & L. M. BLACK (Univ. Ill., Urbana). Particle counting has shown a linear correlation between different concn of wound tumor virus (WTV) particles and their RNA and protein contents. The RNA from 10^{12} WTV particles in 1 ml of solution has an OD of 1.0 with a 1-cm light path and $\lambda = 260$ m μ . Each 10^{12} virus particles contain 108 μ g protein. The percentages of RNA and protein were found to be 22% and 78%, respectively. The molecular wt of the virus calculated from particle counts and direct measurements of RNA and protein was 85.8×10^6 avograms, and that of the RNA was 19.9×10^6 avograms. Linear correlations were determined between different wt of WTV-protein and bovine serum albumin and OD, at $\lambda = 750$ m μ , which develop as a result of the protein reactions with the Folin phenol reagent.

Cytology of cabbage root hair penetration by Plasmodiophora brassicae. J. R. AIST & P. H. WILLIAMS (Univ. Wis., Madison). After the primary zoospore of *P. brassicae* attaches to a root hair wall, the flagella coil and the axonemes are retracted during a period of less than 1 min. The flagellar sheath is apparently not taken into the cytoplasm. During the next 2 hr, a bilayered cyst wall and an electron

opaque bullet-shaped rod (situated in an invagination of the cyst wall) are formed. A vacuole, associated with lipid inclusions, then develops within the cyst. Due presumably to pressure created within the expanding vacuole, the invagination of the cyst wall is evaginated to form a bulbous appressorium. The bullet-shaped rod punctures the root hair wall, and the parasite is quickly injected into the root hair. Total time required for appressorium formation and penetration is about 1 min. A callose-containing deposit is soon formed between the root hair wall and plasmalemma at most penetration sites following penetration. Young amoebae in root hairs have no lipid inclusions, and are surrounded by 7-layered envelopes.

Two new greenhouse tomato varieties resistant to all five Ohio strains of tomato mosaic virus. L. J. ALEXANDER & G. L. OAKES (Ohio Agr. Res. Dev. Ctr., Wooster). The gene *Tm-2a* for resistance to the five Ohio strains of tomato mosaic virus (TMV) has been transferred from a selection of the wild species, *Lycopersicon peruvianum*, P.I. 128650, to commercial cultivars. The female parent was a breeding line similar to Ohio W-R 29. Embryo culture was necessary to secure the cross and first outcross. After these first two crosses, a cross was made to Ohio W-R 7, followed by four backcrosses to it and an outcross to Ohio W-R 41. Ohio M-R 9 resulted from nine selections after the last outcross. Ohio M-R 12 resulted from one additional outcross to Ohio W-R 29. The new mosaic-resistant cultivars produce yields and high-quality fruit comparable to the present wilt-resistant commercial cultivars Ohio W-R 25 and Ohio W-R 29. Both cultivars are resistant to fruit cracking, tolerant to high soil manganese, and moderately susceptible to blotchy ripening. F_1 , F_2 , and F_3 data indicate that a close association exists between TMV resistance and blotchy ripening susceptibility.

A new disease of Zygocactus truncatus in Florida. S. A. ALFIERI, JR., & J. W. MILLER (Fla. Dep. Agr. Conserv. Service, Gainesville). A new and potentially serious disease has recently been observed affecting Christmas cactus (*Zygocactus truncatus*). The first symptoms on potted plants occur as a wilting of stems which droop slightly and exhibit a lifeless, dull gray-green color. Closer examination of the basal portion of the affected stems, usually at or just below the soil line, reveals a water-soaked but rather firm necrotic area with a faded reddish border. Infected roots are water-soaked, brown, soft, and necrotic. Repeated isolations from affected stems and roots, and results of controlled inoculations have shown that *Phytophthora parasitica* is the causal agent. Among four fungicides tested for disease control as a soil drench prior to inoculation, Dithane M-45 [coordination product of zinc ion and manganous ethylenebis (dithiocarbamate)] provided complete control; Dexon [*p*-(dimethylamino) benzenediazo sodium sulfonate], very good control; whereas Terrazole (5-ethoxy-3-trichloromethyl-1,4-thiadiazole) and Demosan (1,4-dichloro-2,5-dimethoxybenzene) provided the least effective disease control.

Accumulation of safynol in resistant and susceptible safflower infected with Phytophthora drechsleri. E. H. ALLEN & C. A. THOMAS (ARS, USDA, Beltsville, Md.). The first internode stems of 8-week-old Biggs (resistant) and Nebraska-10 (susceptible) safflowers (*Carthamus tinctorius*) were wound-inoculated with *Phytophthora drechsleri* and held at 30 C. Safynol (*trans*, *trans*-3,11-tridecadiene-5,7,9-tri-ene-1,2-diol), an antifungal polyacetylene, was extracted from infected stems and quantitated with the aid of thin-layer chromatography and ultraviolet absorbance at 269 nm ($\epsilon = 61,600$). The concn of safynol increased rapidly in the stems of Biggs in the first 48 hr after inoculation and was relatively constant during the next 48 hr. In the susceptible variety (N-10), the concn of safynol increased rapidly in the first 24 hr and decreased in the next 72 hr.

A concn of 30 µg/ml which completely inhibited the growth of the fungus in vitro was reached at the portal of infection in Biggs stems 48 hr after inoculation. The highest concn at the portal of infection for N-10 (17 µg/g fresh wt) was reached 24 hr after inoculation. Loss of water from infected N-10 stems was much greater than from infected Biggs stems.

Mechanism of action of thiabendazole. P. M. ALLEN & D. GOTTLIEB (Univ. Ill., Urbana). Thiabendazole prevented germ tube elongation and growth of *Penicillium atrovenetum* at 2 µg/ml, but growth inhibition was only 90% even at 10 µg/ml. Minor inhibition by thiabendazole occurred in the synthesis of protein, lipids, nucleic acids, and cell wall, and in the uptake of nutrients. Consumption of oxygen with exogenous glucose was inhibited at 20 µg/ml, but with endogenous required more than 100 µg/ml. Respiration by mitochondria was prevented completely. The fungicide did not inhibit cytochrome *c* oxidase, but did inhibit NADH and succinic oxidases and their cytochrome *c* reductases. It inhibited succinate dichlorophenolindophenol reductase at 2 µg/ml in the presence of antimycin A, and coenzyme Q at 15 µg/ml. The fungicide apparently inhibits respiration and growth by preventing electron transport in mitochondria at a site between the substrate and coenzyme Q.

Forms and races of Fusarium oxysporum causing wilts of cucurbits. G. M. ARMSTRONG & JOANNE K. ARMSTRONG (Univ. Ga., Ga. Exp. Sta., Experiment). Accounts of cross inoculations with *Fusarium oxysporum* f. sp. *cucumerinum*, *melonis*, and *niveum* report damping-off and some wilting, indicating a lack of host specificity. We inoculated 8 cultivars of cucumber, 39 of muskmelon, and 22 of watermelon. Isolates of *cucumerinum* were from Germany and the USA; *melonis* from Canada, Belgium, France, and the USA; and *niveum* from Japan and the USA. Variations in virulence for a specific host occurred, but each f. sp. showed selective pathogenicity for its host and is retained as a valid forma specialis. Isolates of *melonis* from Canada and the USA are different in specific pathogenicity on muskmelon from races 1, 2, and 3 from France, and constitute a new race. Formae speciales *luffae* and *lagenariae* from Japan were tested on gourds and other cucurbits, with *luffae* causing wilt of *Luffa* and a cultivar of muskmelon and *lagenariae* causing wilt of *Lagenaria* only. Fifty-two different plants, other than cucurbits, that are useful in the identification of other forms and races were nonsusceptible to f. sp. *cucumerinum*, *melonis*, and *niveum*. Fifty-two different wilt fusaria, other than those mentioned, were nonpathogenic on cultivars of cucumber, muskmelon, and watermelon susceptible to their respective forms.

Development of Helminthosporium victoriae in blends of resistant and susceptible isogenic oat lines. D. K. G. AYANRU (Iowa State Univ., Ames). Clintland-type isogenic oat lines, differing in resistance and susceptibility to *Helminthosporium victoriae*, were developed from crosses with Ascencao oats and studied in *H. victoriae*-infested soil in pure and mixed stands. In the greenhouse, the percentage of diseased seedlings was not proportional to that of susceptible plants in the blends; consistently, fewer susceptible plants became diseased in blends than in pure stands. In field plots, bundle wt, grain yield, and test wt increased more than did the increase in the proportion of resistant plants in the blends, as did the increase in the number of roots on greenhouse seedlings grown in root-observation boxes. Increase in *H. victoriae* spore population and the spread of the fungus in soil were greater in the rhizosphere of pure stands of susceptible plants than in the rhizosphere of blends in field and growth chamber experiments. The buffering of susceptible plants from *H. victoriae* infection by resistant plants in mixtures is implicated.

Fungi associated with cotton boll-rot in the Yazoo-Mississippi Delta. H. S. BAGGA (Delta Branch Exp. Sta., Stoneville, Miss.). Relative frequency and pathogenicity

of different fungi associated with the cotton boll-rot were determined. Surveys in 1966, 1967, and 1968 at Shaw and Stoneville, Mississippi, indicated that species of four genera accounted for 71.2% of the boll-rot: *Fusarium* spp. 35.6%; *Alternaria tenuis* 17.7%; *Aspergillus* spp. 9.6%; and *Diplodia gossypina* 8.3%. Twenty-six species responsible for 26.1% of the boll-rot were: *Ascochyta gossypii*, *Botryosphaeria berengeriana*, *B. ribis*, *Cercospora gossypina*, *Chaenophora cucurbitarum*, *Cladosporium herbarum*, *Curvularia* spp., *Glomerella gossypii*, *Helminthosporium* spp., *Macrophomina phaseoli*, *Myrothecium roridum*, *Nigrospora sphaerica*, *Nigrospora* sp., *Pellicularia filamentosa*, *Penicillium luteum*, *Penicillium* spp., *Pestalotia* sp., *Phoma* spp., *Phyalospora rhodina*, *Phytophthora parasitica*, *Phytophthora* sp., *Rhizopus stolonifer*, *Thielaviopsis basicola*, *Trichoderma viride*, *Trichothecium roseum*, and *Verticillium albo-atrum*. Unidentified fungi accounted for 2.7% of the boll-rot. *Glomerella gossypii*, often reported as a major cause of boll-rot, was isolated from only 1.3% of the samples collected at Shaw and 0.5% of the samples collected at Stoneville. All the fungi listed caused boll-rot when tested in the field.

In vitro production of pectolytic enzymes by Xanthomonas malvacearum. C. H. BALDWIN, JR. (Univ. Mo., Columbia). For the determination of in vitro pectolytic enzyme production by *Xanthomonas malvacearum*, the six selected races (1, 4, 10, 11, 13, and 14) were grown in shake culture on a defined medium containing (NH₄)₂SO₄, 0.5%; MgSO₄, 0.02%; KCl₂, 0.02%; KH₂PO₄, 0.64%; and Na₂HPO₄, 0.32%; or a modified Richard's solution (NH₄)₂SO₄, 0.5% plus the carbon source for 4 days at 28 C. The carbon sources were 1% glycerine, pectin, sodium polypectate, polygalacturonic acid, and galacturonic acid. Pectolytic enzyme production by *X. malvacearum* was determined by employing the cup-plate, viscosity reduction, and reducing group assays. The results of these assays showed that none of the races of *X. malvacearum* tested produced pectinesterase, polygalacturonase, pectate lyase, or pectin lyase on any of the carbon sources.

Synergism of oat blue dwarf virus and aster yellows in flax as observed by light and electron microscopy. E. E. BANTTARI & R. J. ZEYEN (Univ. Minn., St. Paul). Simultaneous infection of flax with oat blue dwarf virus (OBDV) and the aster yellows agent caused greater destruction in vascular tissues of stems and leaves than did either pathogen alone. Hypertrophy and hyperplasia of phloem fibers and hyperplasia of phloem elements caused stems to thicken markedly. Normal cortical parenchyma was displaced, and lacunae developed when hypertrophied fibers collapsed. Obliteration of phloem was prevalent, and some intact sieve tubes appeared occluded. Leakage of substances into cells adjacent to phloem was apparent in both leaves and stems. Infection of flax with OBDV alone caused hypertrophy and hyperplasia of phloem primarily in marginal veins of leaves, and was associated with the development of typical crinkle symptoms. The aster yellows agent alone in flax caused histological abnormalities similar to those already reported. Oat blue dwarf virus occurred mainly in phloem elements of leaves, and was frequently enclosed in saclike structures. Mycoplasma-like bodies were observed in phloem of aster yellows, or aster yellows and OBDV infected leaves and stems.

Histological investigations of the antagonistic interaction between Heterodera glycines and Rhizobium japonicum on soybean. K. R. BARKER & D. HUISINGH (N. C. State Univ., Raleigh). Some races of *Heterodera glycines* inhibit nodule development on soybean. Greenhouse and histological investigations were conducted to determine histological basis for this and to determine if the timing and/or sequence of inoculation of soybean with nematode (Hg) and *Rhizobium* (R) affect this interaction. Soybean plants were inoculated with 200 crushed cysts of Hg and/or 200 mg of commercial inoculum of R, encompassing the following treatments:

Hg + R added simultaneously ("0" time) to 1-week-old plants; Hg at "0" time + R at 2 days, 1 week, and 2 weeks; R at "0" time + Hg at 2 days, 1 week, and 2 weeks; R and Hg alone at above times; and noninoculated controls. Greatest inhibition of nodule development (93-100%) occurred with simultaneous inoculations. Other combination treatments gave 0 to 98% reduction of nodulation. Histological studies showed nodular tissues to be unsuitable for nematode development. Infection of the same tissues by both organisms produced hypersensitive reactions in the area around the nematode. Syncytia induced by Hg, when adjacent to developing nodules, usually failed to develop fully. Cells surrounding larvae which had penetrated nodules usually became necrotic. Although a few mature cysts developed on nodules, most infections of nodules by larvae failed to induce syncytia.

Inheritance of resistance to tomato anthracnose. T. H. BARSDALE (ARS, USDA, Beltsville, Md.). For 3 years, P.I. 272636 showed resistance to tomato anthracnose, caused by *Colletotrichum coccoides*, both in the field and in laboratory inoculation experiments. This line was crossed with the susceptible tomato cultivar, Roma. In 1969, plants of the two parents, their F_1 and F_2 , and backcrosses of the F_1 to both parents, were rated for resistance. Fruit from a single destructive harvest were counted as diseased or healthy, and counts converted to per cent anthracnose. Average natural infection for five replications for P.I. 272636, Roma, the F_1 , and the F_2 was 1, 55, 13, and 16%, respectively. The backcross to the P.I. line showed 3%, and that to Roma 35% anthracnose. The data for plants in the F_2 or in either backcross generation did not show segregation into resistant and susceptible groups, but were either normally distributed between the amounts of anthracnose shown by their parents, or skewed toward resistance. These data indicate that resistance in P.I. 272636 is multi-genic, and probably strong enough to be useful in a breeding program.

Differential stabilization of certain labile viruses. O. W. BARNETT & R. W. FULTON (Clemson Univ., Clemson, S. C.; Univ. Wis., Madison). Stabilization of isometric labile ring-spotting viruses has been compared in extracts of different hosts. The results showed that reducing agents stabilized many labile viruses while certain chelating agents stabilized others. To eliminate variance caused by different hosts, Tulare apple mosaic virus (TAMV) and Prunus necrotic ringspot virus (NRSV) were aged in sap of infected cucumber. A reducing agent, 2-mercaptoethanol (2-ME), stabilized TAMV infectivity for 4 hr. A chelating agent, sodium diethyldithiocarbamate (DIECA), inactivated TAMV. NRSV was stabilized by DIECA, but 2-ME had only a slight stabilizing effect. Cucumber mosaic virus (CMV) was stabilized better in cucumber sap by 2-ME than by DIECA, but when aged in tobacco sap, CMV was stabilized equally by DIECA and 2-ME. In a synthetic inactivating system composed of commercial tyrosinase and *p*-coumaric acid, TAMV was again stabilized by 2-ME but not by DIECA, and NRSV was better stabilized by DIECA than by 2-ME. Results with the synthetic inactivating system suggested that differences in properties of NRSV and TAMV caused the different response to DIECA and 2-ME. The different response to stabilizers by CMV aged in sap from cucumber and tobacco suggested that the inactivating systems of the two hosts are different.

Loss of galacturonate from bean hypocotyl cell walls during pathogenesis by Rhizoctonia solani and Sclerotium rolfsii. D. F. BATEMAN (Cornell Univ., Ithaca, N.Y.). Eight-day-old bean plants (*Phaseolus vulgaris*) were inoculated with *R. solani* and *S. rolfsii* and held in a greenhouse at 30 ± 3 C. Tissues infected with either pathogen and comparable tissue from noninoculated plants were harvested periodically during pathogenesis. Tissue samples were dried at 70 C for 24 hr and ground to pass a 60-mesh screen. Cell-wall samples (10 mg) from these tissues were treated

at pH 4.0 for 20 hr with a 0.8% solution (2.5 ml) of a dialyzed freeze-dried enzyme mixture produced by *S. rolfsii* when grown on autoclaved bean hypocotyls. Debris was removed from the cell wall digests by centrifugation. Galacturonate in 0.2-ml aliquots of the supernatants was determined enzymatically with uronic acid dehydrogenase. Cell walls from healthy plants contained 8.2 and 7% galacturonate after 8 and 11 days' growth, respectively. Cell walls of tissue infected with *R. solani* contained 1.7 and 1.4% galacturonate 48 and 96 hr, respectively, after inoculation. Hypocotyl tissue inoculated with *S. rolfsii* contained, respectively, 4.7, 1.9, and 0.3% galacturonate 24, 48, and 72 hr after inoculation. Depletion of cell wall galacturonate in the susceptible was associated with production of pectic enzymes by the pathogens during pathogenesis.

Local lesion and viral crystal formation in tissue culture cells of different species and cultivars of Nicotiana. R. N. BEACHY & H. H. MURAKISHI (Mich. State Univ., E. Lansing). Callus cells of tobacco mosaic virus- (TMV) susceptible (*N. rustica*, *N. suaveolens*, *N. tabacum* 'Havana-38', 'Maryland Mammoth', and 'White Burley'), hypersensitive (*N. glutinosa*, *N. tabacum* 'NN Burley' and 'NN Samsun'), and resistant (*N. tabacum* 'Ambalema') tobacco were grown on modified Murashige and Skoog (MS) agar medium. The callus (300-400 mg) in 3 ml of MS liquid medium were inoculated with 37.5 μ g of tobacco mosaic virus by mechanical dispersion with a Vortex mixer, washed with 15 ml liquid MS medium, and incubated on agar under 24-hr illumination (90 ft-c) at 21 C. Thirty-nine hr after inoculation, discrete necrotic areas appeared on NN Samsun cultures only, and were countable after 72 hr. Few (13 of 500 examined) of the living cells surrounding the necrotic areas contained viral crystals. Inclusion-bearing cells were most abundant (72% of cells contained 1 to 10 crystals) in susceptible but symptomless Havana-38 callus. Seven and 10 days after inoculation, virus titers were highest in susceptible tobacco callus based on local lesion assays on leaves of *N. tabacum* 'Xanthi-nc'. Titters of virus propagated in hypersensitive and resistant cells were considerably lower.

Rhizoctonia infestation in field soil following a long-term fertilization program. H. W. BEAM & E. A. CURL (Auburn Univ., Auburn, Ala.). *Rhizoctonia* was assessed in soils of 10 field plots which had been subjected for 10 years to fertilizer treatments ranging from a full complement of N, P, K, lime, and minor elements to treatments deficient in one or more of these constituents. All plots since 1967 had received the same 3-year rotation system of cotton, corn-wheat, and soybeans, with or without a winter legume included. *Rhizoctonia* was assessed by two known methods. Natural debris particles were wet-screened from the soils, serially washed, and plated on streptomycin-water agar. Also, autoclaved soybean-stem segments were buried in samples of plot soils for 4 days, recovered, serially washed, and plated as before. *Rhizoctonia* colonization was observed microscopically in 24 hr. High infestation of *Rhizoctonia* occurred in plots that had received N, P, and K plus lime and a winter legume in the rotation. These plots also produced highest seed-cotton yields, whereas low infestation and low yields were most evident in plots deficient in N, P, or lime. Levels of *Rhizoctonia* were higher in the fall following crop harvest than in spring prior to planting. When plot soils in greenhouse pots were planted with cotton, seedling emergence was generally low and root damage high in soils of high *Rhizoctonia* infestation.

Comparative evaluations of helicopter and ground equipment applications of fungicides to golf course fairways. K. M. BECKMAN, S. F. RICKARD, & J. F. NIEDEBALSKI (The Upjohn Co., Kalamazoo, Mich.). Biological, analytical, and time and motion evaluations were made to compare helicopter and ground application of cycloheximide fungicides to golf course fairways. Three treatments were replicated on three fairways for each mode of application. Residue deposits were collected and bioassayed from 10 glass plates

from each of two sampling sites on two replicated fairways/treatment per application. Disease control ratings of 0-5, where 0 equals no disease and 5 equals 100% disease, were made at similar sites. Preparation, mixing, travel, application, and clean-up times were recorded for each treatment. The primary advantage offered by the helicopter is precision application with increased speed. Additional features include min interference from adverse weather, no damage or compaction to fairways by heavy spray equipment, greater flexibility in the application schedule to permit normal operations of the golf course without interruption, and reduction in capital equipment investments. These studies indicate that helicopter fungicide applications to golf course fairways can reduce labor costs without sacrificing effective disease control.

Infection of peanut pods by Aspergillus flavus as affected by Meloidogyne arenaria and length of curing time. D. K. BELL, N. A. MINTON, & B. DOUPNIK, JR. (Univ. Ga. Coastal Plain Exp. Sta.; ARS, USDA, Tifton, Ga.). To determine the effects of *Meloidogyne arenaria* and curing time on pod infection by *Aspergillus flavus*, Argentine peanuts were grown in field microplots containing methyl bromide-treated soil inoculated with either *A. flavus*, *M. arenaria*, or *A. flavus* + *M. arenaria*. Noninoculated microplots served as controls. Each treatment was replicated 6 times. At maturity the plants were dug, the pods rated for root knot galling, and one-third of the pods harvested. The remaining pods were left attached to the plants and placed on a wire-mesh greenhouse bench to cure. Additional harvests were made after 5 and 12 days of curing. After each harvest, 25 two-seeded pods from each replicate were assayed for fungi. The remaining pods were dried for subsequent aflatoxin analyses. Pods from *M. arenaria*-inoculated plants were heavily galled, but the incidence of *A. flavus* and total fungi was not affected. The length of curing time also did not affect the incidence of *A. flavus* or total fungi. Only in pods from *A. flavus*-inoculated plants was the incidence of *A. flavus* increased. Aflatoxin contamination was not affected by any of the treatments. We concluded that *M. arenaria* damage to peanut pods did not affect *A. flavus* infection.

Heat-cure of sugarcane infected with sugarcane mosaic virus. G. T. A. BENDA (ARS, USDA, Houma, La.). Repeated hot-water treatments of single-bud cuttings of sugarcane stalks were used to cure sugarcane mosaic virus disease. Some cuttings infected with strains A or B were cured by 7-min treatments at daily intervals at temp of 54.8, 56.5, 57.3, and 57.3 C, successively. Some cuttings infected with strain H or I were cured by 10.5-min treatments at temp of 54.8 and 55.8 C, 1 day apart. Cured plants assayed negative for sugarcane mosaic virus were free of symptoms through two or more vegetative generations unless inoculated, and, when reinoculated with the strain from which heat-cured, developed typical symptoms. Cuttings which were not heat-treated remained infected. This is believed to be the first cure reported for sugarcane mosaic virus disease, and the first use of multiple treatments at different temp to obtain a cure of any virus disease.

Polyphenol oxidase activity in leaves of corn seedlings infected with maize dwarf mosaic virus. S. P. BENIWAL, R. T. GUDAUSKAS, & BRYAN TRUELOVE (Auburn Univ., Auburn, Ala.). Polyphenol oxidase activity in extracts from inoculated and subsequently emerging leaves of *Zea mays* 'HY × C103' seedlings was measured spectrophotometrically at intervals for 6 days after inoculation with maize dwarf mosaic virus (MDMV). Inoculations were made by the Carborundum-gauze pad technique with MDMV in sap from infected corn leaves; control plants were inoculated with sap from healthy leaves. No difference in polyphenol oxidase activity of MDMV-inoculated and control leaves was found at 0 and 24 hr after inoculation. Three days after inoculation, enzyme activity in MDMV-inoculated leaves was slightly higher than that of control leaves. A 20% increase in activity was measured in the first system-

ically-infected leaf (immediately above inoculated) 2 days after inoculation. By the 4th day, polyphenol oxidase activity in this leaf was 11% higher than the control, and declined to a level equal to that of the control by 6 days postinoculation. A similar pattern of activity was measured in the systemically infected fourth leaf.

Forecasting Helminthosporium turcicum attacks in Florida sweetcorn. R. D. BERGER (Univ. Fla., Belle Glade). *Helminthosporium turcicum* required at least 7 hr of humidities near 100% and temp above 15 C (blight favorable hr = BFH) for significant sporulation. Microscopic observation and spore-trapping data showed that *H. turcicum* spores were formed at night and released in the morning as the humidity rapidly decreased. Nearly 50% of the trapped spores were caught from 8 AM to noon; only 5% were caught from 8 PM to 8 AM. Cumulative records of daily BFH obtained from hygrothermograph charts provided an accurate method to predict the seasonal threat of blight. A daily average of 6.5 BFH or less resulted in little blight and not enough disease to justify fungicidal control. Seven-8 BFH/day required occasional sprays for control; light to moderate blight occurred in unsprayed fields. Eight or more BFH/day required a regular fungicide program to avoid serious losses. An average of more than 11 BFH/day often resulted in blight epiphytotic regardless of spray schedules. Spore trapping was valuable in determining the actual daily blight threat during all seasons, and proved especially useful during borderline conditions. For the past 3 seasons, growers successfully reduced fungicide applications without disease buildup following this forecasting method.

Effect of carbon sources on the production of polygalacturonase by Ceratocystis ulmi. W. L. BIEHN & A. E. DIMOND (Conn. Agr. Exp. Sta., New Haven). Studies were made of polygalacturonase (PG) production by *Ceratocystis ulmi* growing on two sources of pectin. The fungus was grown in shake culture on a pectin-asparagine-salts medium initially adjusted to pH 3.5 with lactic acid. The pectin source was added at 1% and consisted of either citrus pectin or a cell-wall fraction from green elm shoots. The elm preparation contained about 0.15% pectin on a dry wt basis. PG production per unit mycelial dry wt was more than 35 times greater on the elm preparation than on citrus pectin. Glucose (0.1 M) repressed PG synthesis 85 to 95% on the elm preparation and on citrus pectin. Galacturonic acid (0.05 M) reduced initial PG production at least 55% on the elm preparation. On citrus pectin, galacturonic acid reduced PG production about 65 to 80%. These studies indicate that PG production by *C. ulmi* is influenced by the nature of pectin as well as by the presence of sugars and sugar derivatives. Host pectin may be essential for the secretion of large amounts of PG by *C. ulmi*.

The biology of a new cyst nematode on grasses. W. BIRCHFIELD (USDA, ARS, La. State Univ., Baton Rouge). An undescribed species of cyst nematode in the genus *Heterodera* was found parasitizing barnyard grass, *Echinochloa colonum*. The pathogenicity, host preferences, and life cycle were studied. Larval penetration, growth, cyst development, and egg-laying were observed on grass seedlings germinated in infested soil under greenhouse conditions. Roots in toto were killed, fixed, and stained with lactophenol-acid fuchsin. Larvae penetrated roots and oriented along the vascular cylinder in 12 days. They began to enlarge and were fully formed in 18 days. The white, lemon-shaped cysts raised the epidermis and finally cracked through it, but no galling of the host tissues was observed. A few eggs were deposited on the epidermis after 24 days, but most of the eggs remained in the cysts. No gelatinous matrix was secreted by the parasite. Necrosis of host tissues was noticed in the feeding site, but no other symptoms were expressed. Roots were parasitized by large numbers in all stages of development, including secondary infection, after 64 days. A few brown cysts were found at that time. Mature females were observed on Johnson grass in 19

days. Several rice cultivars and sugarcane, cultivar Canal Point 44-101, were not parasitized.

Heritable wound tumors in several common bean varieties. J. BIRD, J. SÁNCHEZ, & K. MARAMOROSCH (Univ. Puerto Rico, Rio Piedras; Boyce Thompson Inst. Plant Res., Yonkers, N. Y.) Common bean (*Phaseolus vulgaris*) plants of the cultivars Criolla, Jamaica, Diablo, Dominicana, and Experimental No. 1208 consistently developed tumors 10 to 14 days following wounding by insect pins or by pruning and laceration with a razor blade. Chains of tumors sometimes developed on wounded plants, and occasionally tumors appeared a short distance from the inflicted wound. Injury resulted in tumor formation irrespective of whether the plants were grown in an open greenhouse or under insect-proof cages, in normal garden soil, or in sterilized soil. Plants grown from surface-sterilized seed also developed wound tumors when wounded with sterile needles. Attempts to isolate a causative agent such as a bacterium, a virus, or a fungus have failed. Electron microscopy of thin sections from neoplastic tissue failed to reveal the presence of abnormal cell constituents or particles that would resemble viruses. Apparently the pathological condition is genetically inherited and requires the stimulus of wounding.

Seed and seedling measurements of vigor for predicting postemergence damping-off and stands of cotton plants. L. S. BIRD, W. E. BATSON, JR., K. M. EL-ZIK, & R. F. LYNCH (Texas A&M Univ., College Station). Sets consisting of 22, 24, 25, 28, and 33 strains of *Gossypium hirsutum* were used. The strains in each set were known to represent ranges of differential ability to give stands of plants under field conditions. Some strains were common to each set. Seed lots for each strain were produced at the same location, none were deteriorated, they were acid-delinted, and protectant chemicals were not used. Data from replicated tests for each strain of the five sets included germination after 7 days at 18 C; oven-dry wt (ODW) of cotyledons and root-hypocotyl sections of 4-day-old seedlings; stands for three sets in field and two sets in outside tray plantings; and postemergence damping-off (DO) in one field and the two tray plantings. Within each set of strains, germination was positively correlated with emergence and DO where measured, and negatively correlated with final stands. High germination at 18 C and low cotyledon ODW are generally considered to indicate seed vigor. These results confirm earlier findings that vigor in this sense for nondeteriorated seed is undesirable with respect to DO and final stands. These data so suggest, and explain results where breeding for no germination of non-deteriorated seed at reduced temp aids in control of seedling disease.

Low volume application of vegetable fungicides by air. C. H. BLAZQUEZ & H. S. POTTER (Univ. Fla., South Fla. Field Lab., Immokalee). The low volume application by air of five fungicides on potatoes and watermelons was investigated during the spring of 1970. Aerial sprays of Dithane M-45 and Manzate 200 (both coordination products of zinc ion and manganous ethylenebis [dithiocarbamate]), Polyram (a mixture of 5.2 parts by wt of ammoniate of [ethylenebis(dithiocarbamate)] zinc with one part by wt ethylenebis[dithiocarbamic acid] bimolecular and trimolecular cyclic anhydrosulfides and disulfides), Difolatan 4 Flowable (*cis-N*-[1,1,2,2-tetrachloroethylthio]-4-cyclohexene-1,2-dicarboximide), and a combination of Nu-Film (beta-pinene polymer) and Manzate 200 were compared with conventional tractor-driven ground sprays for the control of late blight (*Phytophthora infestans*) and early blight (*Alternaria solani*) of potatoes, and of gummy stem blight (*Mycosphaerella citrullina*) and downy mildew (*Pseudoperonospora cubensis*) of watermelons. There was no significant difference with either crop in disease control or in yield between aerial and ground applications.

*Effects of nitrogen compounds in the growth medium on resistance of *Gossypium arboreum* 'Nanking' seedlings to*

anthracnose. K. BOLLENBACHER & N. D. FULTON (ARS, USDA, Univ. of Ark., Fayetteville). *Gossypium arboreum* 'Nanking' seedlings are highly resistant to *Colletotrichum gossypii*. Nanking seedlings were grown in a medium supplemented singly with L isomers of 23 amino acids, KNO_3 , $\text{NH}_4\text{H}_2\text{PO}_4$, $(\text{NH}_4)_2\text{SO}_4$, and NH_4NO_3 at a concn to supply 0.5 g of nitrogen/liter. Plants were inoculated by placing a spore suspension of *C. gossypii* on hypocotyls at point of contact with the substrate. Media supplemented with asparagine at 0.5 g of nitrogen/liter and peptone at 2,500 ppm induced complete susceptibility and were not toxic to noninoculated plants. Other amino acids either induced less disease or were phytotoxic. Except for those which were phytotoxic, inorganic nitrogen compounds did not break down resistance. Inoculated plants remained healthy in a medium supplemented with KNO_3 and glucose at rates furnishing nitrogen and carbon at the levels supplied by asparagine at 0.5 g of nitrogen/liter. Evidence is strong that the supplements predispose the seedlings rather than increase the virulence of the fungus. Asparagine was added to the medium by a method that allowed no contact between inoculum and plant growth medium, and the seedlings became completely susceptible.

*Interaction of some pesticides with *Sclerotium rolfsii*.* G. A. BOZARTH & B. G. TWEEDY (Univ. Mo., Columbia). The effect of 2-chloro-4-(ethylamino)-6-(isopropylamino)-s-triazine (atrazine), 1,1-dimethyl-3-(*a,a,a*-trifluoro-*m*-tolyl) urea (fluometuron), 3-(*p*-bromophenyl)-1-methoxy-1-methoxy-1-methylurea (metobromuron), *a,a,a*-trifluoro-2,6-dinitro-*N,N*-dipropyl-*p*-toluidine (trifluralin), and bis(dimethylthiocarbamoyl) disulfide (thiram) on mycelial growth and production of sclerotia by *S. rolfsii* was investigated. The fungus was grown in petri dishes on potato-dextrose agar containing the pesticides. At 50 ppm, thiram completely inhibited mycelial growth, fluometuron inhibited growth 66%, and the other herbicides inhibited growth 20 to 30%. All four herbicides inhibited production of sclerotia, with fluometuron being the most inhibitory (80% at 50 ppm). The wt (expressed as per cent of control) of sclerotia produced on media containing 50 ppm pesticide, however, was: fluometuron 222, atrazine 189, trifluralin 179, metobromuron 165, and thiram 109. The effect of fluometuron on some growth activities of *S. rolfsii* was also studied in liquid culture experiments. Concentrations of 25 ppm and greater inhibited mycelial dry wt, increased oxalic acid content, and altered polygalacturonase activity of the culture filtrates.

*Purification and properties of two viruses from *Penicillium stoloniferum*.* R. F. BOZARTH & H. A. WOOD (Boyce Thompson Inst., Yonkers, N.Y.). Cultures of *Penicillium stoloniferum* (ATCC 14586) contain nucleoprotein particles which are considered to be viral in nature on the basis of physical, chemical, and serological properties. Highly purified preparations obtained by extraction of mycelium with 0.1 M phosphate buffer, pH 7 and chloroform, differential centrifugation, and rate zonal sucrose density-gradient centrifugation contained two electrophoretic and serologically distinct species. The slow and fast electrophoretic species each contained four sedimentation components and spherical particles of about 34 m μ diam. Double-stranded RNA obtained by single-phase phenol extraction and analyzed by acrylamide gel electrophoresis contained fractions of 0.24, 0.86, 0.95, 1.02, and 1.11×10^6 daltons. Additional ribonucleic acid components with sedimentation coefficients between 3 and 6 Svedberg units in sucrose density-gradient were also observed.

Respiration and terminal oxidases of late blight-infected tomato leaves. J. A. BRENNEMAN & L. L. BLACK (La. State Univ., Baton Rouge). The respiratory rate of tomato (*Lycopersicon esculentum*) leaf tissue infected by *Phytophthora infestans* at the time of symptom production was approximately 50% greater than that of healthy leaf tissue. Comparisons were made of the in vitro activity of terminal oxidases in healthy and infected tissue at this stage. The

infected plants showed a 3-fold increase in the activity of polyphenol oxidase and peroxidase, a 2-fold increase in ascorbic acid oxidase, no difference in catalase and cytochrome oxidase, and a slight decrease in glycolic acid oxidase. The stimulated respiration in the infected tissue was not inhibited by treatment with inhibitors of copper-containing enzymes (diaca and thiourea), nor was it further enhanced by addition of substrates for these enzymes (catechol, chlorogenic acid, ascorbic acid). Although increased activity of the above enzymes suggests that stimulated respiration in infected tissue may be due to activation of a noncytochrome system, such a relationship could not be established by the methods used.

Protection from Rhizoctonia solani by pentachloronitrobenzene accumulated in plants. P. R. BRISTOW & J. KATAN (Mich. State Univ., E. Lansing). Bean seedlings, grown for 1 week in loam soil containing 25 µg pentachloronitrobenzene (PCNB) per cc, were inoculated with *Rhizoctonia solani* and replanted in steamed soil without PCNB or placed on moist paper towels in a humid growth chamber. At the time of inoculation, hypocotyls contained 4-7 µg PCNB/g fresh wt, a concn equal to or greater than that required to limit growth of the fungus in culture. Disease severity in plants containing PCNB was significantly less than in control plants. PCNB was also taken up in varying concn by the underground parts of several other plant species from soil containing ca. 17 µg PCNB/cc. PCNB was rapidly lost from underground parts, such as cotyledons, after their emergence from soil. More PCNB was taken up by plants from soils with low than with high organic matter contents. Peat was mixed with an equal volume of loam soil and amended with 10 µg PCNB/cc, and the mixture incubated for 1 week. Peat separated from the mixture contained ca. 10 times more PCNB than the remainder of the soil. Sorption of PCNB by soil organic matter apparently reduces the amount available for uptake by plants, and may correspondingly decrease protection from *R. solani*.

Vertical distribution and population fluctuations of three nematode species as correlated with soil temperature, moisture, and texture. B. B. BRODIE & B. H. QUATTLEBAUM (ARS, USDA, Univ. Ga. Coastal Plain Exp. Sta., Tifton). Soil samples, taken weekly for 12 months in sandy soil at 15-cm increments to a depth of 105 cm, were processed for nematodes. Percentage sand, silt, and clay was determined at each depth; moisture and temp were recorded weekly for each depth. Populations of *Belonolaimus longicaudatus*, *Pratylenchus brachyurus*, and *Trichodorus christiei* varied with sampling depths and time of year. At any time, only one species predominated at a given depth. *Belonolaimus longicaudatus* was found only in the top 30 cm (88% sand, 5% clay). Highest numbers occurred from June through September when soil temp 0-30 cm deep was above 20°C and soil moisture averaged 15-20% by volume. *P. brachyurus* was detected at all depths, but was most prevalent 45-90 cm deep (78-80% sand, 15-16% clay). Highest numbers occurred from December through June when the soil temp 45-90 cm deep was 15-20°C and soil moisture averaged 30%. *Trichodorus christiei* was detected at all depths, but was most prevalent 15-30 cm deep (87% sand, 7% clay). Highest numbers occurred from December through March when the soil temp 30 cm deep averaged 10-15°C and soil moisture was 25%. We conclude that soil texture determines vertical distribution of each species, and that temp and moisture determine population density within a given texture.

A histological study of the curative action of EL-273 against Venturia inaequalis. I. F. BROWN, JR., & H. R. HALL (Eli Lilly & Co., Greenfield, Ind.). Greenhouse and field experiments have previously demonstrated the ability of EL-273, α -(2,4-dichlorophenyl)- α -phenyl-5-pyrimidinethanol, to prevent symptom expression of the apple scab disease when the chemical is applied to apple foliage after infection has occurred. Histological techniques were

utilized to demonstrate the effect of EL-273 on the development of the scab pathogen within apple leaf tissue. Greenhouse-grown apple seedlings in the 7- to 8-leaf stage were inoculated with a conidial suspension of the scab fungus. Subsequently, at 1-day intervals, a single application of 25 ppm of EL-273 was applied to the inoculated seedlings. The last application was made 6 days after inoculation. When EL-273 was applied prior to inoculation, or 1 or 2 days after inoculation, the conidia germinated and germ tubes elongated, but no appressoria formed. An application of EL-273 after the fungus had penetrated the leaf arrested further development of stromatic hyphae. EL-273 applied after secondary stroma formed (6th day) prevented the formation of conidiophores. The curative action of EL-273 against the apple scab fungus results from its ability to arrest fungal growth within leaf tissue.

Ultrastructure of sclerotia of Sclerotium rolfsii. M. F. BROWN, D. A. KINDEN, & R. FAUP (Univ. Mo., Columbia). Morphological development of sclerotia of *Sclerotium rolfsii* can be followed by collecting specimens over the color range of white, yellow, tan, and brown. Such a collection was made to study the ultrastructure by means of transmission and scanning electron microscopy. The specimens were fixed in glutaraldehyde, postfixed in osmium tetroxide, and embedded in epoxy resin mixtures for transmission microscopy. Similar preparations were coated with gold for scanning microscopy. Differentiation within a sclerotium into rind, cortex, and medulla is initiated prior to the yellow stage and completed by the brown stage. In the white stage, all cells are multinucleate, possess numerous organelles, and are connected via dolipore septa. A zone of peripheral cells enlarges and the contents of the outermost cells degenerate. The residual walls of the outer dead cells stretch circumferentially as the sclerotial body enlarges to form flattened rind cells. Cortical cells enlarge and become tightly compressed, while medullary hyphae remain filamentous. Cytoplasmic degeneration of some outer cortical cells occurs during the yellow and tan stages. This process contributes to the rind layer. Remaining cortical and medullary cells are capable of germination, and do so by penetration of the outer dead cells.

Urediospore and grain yields from interacting crown rust races and commercial multiline oat cultivars. J. A. BROWNING, BLANCHE M. COURNOYER, D. JOWETT, & J. MELLON (Iowa State Univ., Ames). Early oat cultivars Multiline E68 and Multiline E69, midseason cultivars Multiline M68 and Multiline M69, and resistant and susceptible isogenic lines in each maturity class were grown in 50- \times 50-ft plots in two replications. Transplants heavily infected with races 216, 264, 290, and 326 were set in each plot. Rotorod spore samplers were exposed at the leeward edge of each plot 2 hr daily from 28 June to 15 July 1968. Accumulated spore counts produced sigmoid curves described by the logistic equation. Time of max spore release μ from multilines was 3-4 days later than from susceptible midseason line X122-12. Over twice as many spores were trapped at μ from X122-12 as from the multilines. A seasonal total of 5,600 spores/100 liters of air was harvested from X122-12. Two-thirds as many was harvested from the susceptible early line, while multilines yielded less than one-third as many as X122-12. Grain yield of X122-12 was 53% and of C237-89IV was 73%, and the multilines were 81 to 86% of their respective resistant checks. Grain and spore yields were highly negatively correlated ($r = -.90$). Thus, grain yield and time and quantity of spore yield indicated that commercially available multilines buffered against a crown rust epiphytotic much more severe than one likely to occur in a farmer's field.

Pectolytic enzymes involved in charcoal rot disease of soybean. W. E. BRYANT & T. D. WYLLIE (Univ. Mo., Columbia). Extraction in a 0.1-M potassium phosphate buffer (pH 7.2) containing 0.1 M sodium ascorbate and 1.0 M sodium chloride, and subsequent treatment with Polyclar AT (GAF Corp.) made possible the isolation of

pectolytic enzymes from soybean stem tissue infected with *Macrophomina phaseoli*. Extracts of diseased tissue and culture filtrates from the fungus grown on a sodium polyacetate medium contained exopolysaccharonase and calcium-dependent endopolygalacturonase *trans*-eliminase as determined using paper chromatography, increase in reducing power, increase in OD at 235 nm, and decrease in viscosity of reaction mixtures. At pH 5.0, infected tissue extract released reducing groups from polygalacturonic acid 12 times faster than did healthy tissue extract. *Trans*-eliminase activity was not detected in noninfected tissue extract. Extracts of infected tissues exhibited a greater pectin methyltransferase activity than did those from noninfected tissues. The demonstration of pectolytic enzymes in soybean tissue infected with *M. phaseoli* in conjunction with the demonstrated capacity of the fungus to produce pectolytic enzymes in culture suggests that such enzymes are involved in the development of charcoal rot disease of soybean.

Casein hydrolysates and peptones for artificial culture of Puccinia graminis f. sp. tritici. W. R. BUSHNELL & R. B. RAJENDREN (ARS, USDA, Univ. Minn., St. Paul). Culture nutrients from commercial sources were tested for ability to support growth of mycelial subcultures of *Puccinia graminis f. sp. tritici*, race 126-Anz-6,7. Each material was tested at 13 to 14 C at a concn of 0.1% (w/v) in media that also contained 3% glucose, Czapek's minerals, and 1.5% agar. Colony diam increased 45-81 μ /day in 16 successive 4-week trials with Evans' peptone. Similar rates of growth occurred in single trials with each of four other peptones and three acid hydrolysates of casein. With several enzymatic hydrolysates of casein, yeast extracts, and other peptones, growth rates were 28 to 65% of those with Evans' peptone. In tests over a range of concn with peptone (Evans') and casein hydrolysate (Difco Casamino Acids, tech.), max rates of dry wt increase (0.16-0.17 mg/day per colony) and colony expansion (85 to 99 μ /day) occurred with 0.7-1.0% casein hydrolysate without peptone. This was about 1.4 times the max yield obtained with peptone alone. But peptone (0.1-0.4%) combined with casein hydrolysate (0.3-0.5%) gave fairly rapid growth (0.15 mg/day per colony) with unusually large masses of spores. Both materials can be useful for rust culture, at least until specific nutrient requirements are determined.

Root inhibition of sugarcane seedpieces by Ceratocystis paradoxa. R. S. BYTHER & G. W. STEINER (Exp. Sta., Hawaiian Sugar Planters' Assoc., Honolulu). *Ceratocystis paradoxa* inhibited nodal rooting on sugarcane (*Saccharum sp.*) stalks incubated in plastic bags. The stalks were 60 cm long and consisted of five internodes. Inoculations which resulted in disease development in basal internodes inhibited root development at all nodes on the stalk. Root germination and growth was inhibited in noninfected single-node cane pieces suspended in flasks containing cultures of the fungus. Ethylene was detected under these conditions, but could not be detected in flasks containing only the fungus. Exogenously supplied ethylene caused a similar inhibition. Shoot inhibition is purportedly caused by ethyl acetate (EtAc) produced by the fungus. Production of EtAc by *C. paradoxa* was corroborated. High levels of EtAc inhibited rooting, and when root inhibition occurred, ethylene was detected. It is proposed that a volatile compound produced by the pathogen stimulates the plant to produce ethylene, which inhibits rooting. This inhibition is a possible factor reducing germination and vigor of seedpieces infected with *C. paradoxa*.

General resistance ("slow rusting") to Puccinia recondita f. sp. tritici in winter and spring wheats. R. M. CALDWELL, J. J. ROBERTS, & Z. EYAL (Purdue Univ., ARS, USDA, Lafayette, Ind.). The spread of leaf rust in apparently "slow rusting" winter wheats has been studied in plots separated by 38-ft buffer zones. Comparisons of pustule numbers on flag leaves were made at predetermined dates

and stations following inoculation of plot centers. Slow rusting cultivars as compared with the "fast rusting" cultivars Seneca and Sel. 45 were Vigo, LaPorte, Dual, Purkof, Bulgaria 88, and Fairfield. Vigo, grown on nearly 2 million acres in 1954, has remained free of severe leaf rust in pure stands in Indiana, although highly susceptible at maturity. It may become severely infected after senescence starts. Spring wheat cultivars have shown promising levels of slow rusting at presenescent stages under heavy inoculum. A pattern of infection, wherein early pustules occur mainly on the basal 10-25% of the blade, was found in Mentana and its derivatives Lerma 50 and 52. As senescence progresses, pustules appear on the distal portion. Such resistance to infection was observed in the cultivar Lerma Rojo 64 and its derivatives Inia 66 in 1970 in Mexico, where it gave significant protection in commercial fields. Blades of Ble Tendre and Menkemen also show the Mentana-type infection pattern, which appears to act similarly to the universal exclusion by the sheath and peduncle.

Preservation of single-cell isolates of Corynebacterium insidiosum in sterile soil without loss of virulence. R. B. CARROLL & F. L. LUKEZIC (Pa. State Univ.). Forty-five single-cell isolates of *Corynebacterium insidiosum* were maintained for 1 year at 4 and 21 C in a sterile 3:1:1 soil: peat:perlite mix (moistened with 1-ml water/5 g mix). Forty-two of the isolates kept at each temp persisted over this time interval and produced typical colonies when cultured on beef-lactose agar (BLA). No variants were detected. The effect of storage temp on virulence was determined by a random selection of eight virulent isolates tested on 6-week-old DuPuits alfalfa seedlings grown under sterile conditions. No change in virulence due to storage temp was detected. Parent isolates of the single-cell isolates persisted for 20 months in sterile soil kept at 4 C with no apparent change. In contrast, only 50% of the isolates could be recovered from sterile distilled water at the end of 1 year. These grew poorly on BLA and produced only a small amount of the typical blue pigment. This soil method of preserving single-cell isolates is more convenient and reliable than previously described methods, and should be useful for maintaining a large number of isolates for long periods without loss of virulence.

The dynamics of the permeation of Aphelenchus avenae by halo-organic nematicides and other substances. C. E. CASTRO, I. J. THOMASON, & M. BELD (Univ. Calif., Riverside). The rates of permeation in and out of *Aphelenchus avenae*, and the equilibrium distribution within the nematode and in the external solution, for organic halide nematicides and a variety of other substances, including hydrocarbons, ketones, esters, acids, alcohols, and salts, has been determined. The rates in (k_{in}) range from relatively fast for 1,3-dichloropropene (k_{in} 1.3 min^{-1}) to slow for *n*-butanol (k_{in} 0.003 min^{-1}). Equilibrium concn within the nematode range were from 16 to 0.7 times the external concn. The organic salt sodium propionate was barely detectable internally in the nematode after an exposure of 2 hr. The permeation in (k_{in} \sim 0.2 min^{-1}) and egress (k_{out} \sim 0.2 min^{-1}) for water and acetone are similar. In addition, the rate out is faster than all other substances studied, and the kinetics suggest that a different path of permeation exists for water and acetone.

Heterogeneity in the nucleic acid from tobacco streak virus. M. F. CLARK & R. M. LISTER (Purdue Univ., Lafayette, Indiana). Analysis of purified preparations of two strains of tobacco streak virus (TSV) by rate-zonal density gradient centrifugation revealed three sedimenting nucleoprotein species, designated "top", "middle", and "bottom" according to position. They occurred in various characteristic ratios according to strain, culture host, and mode of preparation. Ribonucleic acid extracted from preparations of the two strains using sodium lauryl sulphate, though usually considerably degraded, also contained

characteristic corresponding species, as identified by zonal density gradient centrifugation and by electrophoresis on polyacrylamide gels. Comparisons of the ultraviolet absorption profiles obtained in analyzing nucleoprotein and RNA preparations of each strain suggested that the RNA contained in middle component particles was heterogeneous. For one strain, this heterogeneity resulted in increased peak size for the ultraviolet absorption peak characterizing top component RNA in acrylamide gels. For the other, it resulted in increased peak size for one of two peaks characteristic of top component RNA. Analysis of RNA samples prepared from separated nucleoprotein components confirmed these results. Conceivably, the parallel between this property and the biological properties of top and middle nucleoprotein components of TSV is significant.

Relationship of fertilizer treatments to nematode populations. R. J. COLLINS & R. RODRIGUEZ-KABANA (Auburn Univ., Auburn, Ala.). The effect of fertilizer treatments on populations of free-living and plant-parasitic nematodes was studied in field plots planted to corn followed by winter wheat. Fertilizer treatments ranged from a complete complement (lime, N, P, K, + trace elements) to treatments deficient in one or more components. Each plot received the same treatment continuously for 10 years, and was subjected to the same rotation system. Soil samples for nematodes were taken postharvest to corn, preplanting to wheat, and at various other times during the growing season. Species of *Xiphinema*, *Meloidogyne*, and spiral nematodes occurred in highest numbers in the highest yielding plots which received P, K, and N; lower numbers occurred in plots deficient in one or more of the fertilizer components tested. Numbers of predacious nematodes (*Dorylaimus* sp., and *Mononchus* sp.) followed a similar pattern; numbers of these nematodes were very low in unfertilized plots. Numbers of lesion nematodes were highest in soils from limed plots that received incomplete fertilization and no winter legume. Plots with complete fertilizer combinations + winter legume had lowest soil populations of lesion nematodes. Stubby root nematodes (*Trichodorus* sp.) occurred in high numbers only in the fall in nonlimed plots after corn harvest.

The role of Helminthosporium carbonum toxin in corn leaf invasion. J. C. COMSTOCK & R. P. SCHEFFER (Michigan State Univ., E. Lansing). *Helminthosporium carbonum* (HC) conidia germinated and penetrated the cuticle of resistant, intermediate, and susceptible corn leaves by 12 hr after inoculation. Hyphal growth in susceptible leaves was extensive at 24 hr; conidiophores were evident at 48 hr. With 10 conidia/mm² leaf surface, fungal growth usually was confined to the first penetrated resistant cell; with 20 conidia/mm², several cells were invaded, but growth stopped by 48 hr. Invasion of tissues of intermediate corn was more extensive than of resistant corn, but less than of susceptible corn. The development of *H. victoriae* (HV), which is not pathogenic to corn, in susceptible, intermediate, and resistant corn leaves was similar to HC development in resistant corn. HC-toxin (0.2 µg/ml) added with HV spores had no effect on development of HV in susceptible or resistant leaves. Toxin at 2.0 µg/ml allowed HV to invade susceptible leaves, but did not aid development in resistant leaves. Toxin at 20-40 µg/ml allowed restricted growth of HV in resistant tissue, but hyphae were abnormal. Thus, toxin and toxin-sensitive tissues are required for successful colonization. Electrolyte leakage from HC-inoculated susceptible leaves was greater than from controls by 28 hr; leakage from inoculated resistant leaves did not differ from controls.

Effect of calcium on loss of electrolytes from pepper leaves inoculated with Xanthomonas vesicatoria. A. A. COOK & R. E. STALL (Univ. Fla., Gainesville). The influence of calcium on loss of electrolytes from inoculated leaves of *Capsicum annuum* cultivars hypersensitive and susceptible to race 2 (pepper strain) of *Xanthomonas vesicatoria* was determined. Maximum suppression of electro-

lyte loss from both types of leaves was obtained by adding 0.2 M calcium nitrate to the inoculum immediately prior to injection. Lower concn of calcium nitrate were progressively less effective, but no influence of calcium nitrate on viability of bacteria in vitro was detected. Comparable results were obtained when calcium nitrate in corresponding concn was injected into hypersensitive pepper leaves 16 hr prior to inoculation with bacteria. The chloride forms of calcium and strontium, but not magnesium, were equally effective as calcium nitrate. The effect of calcium on electrolyte loss from hypersensitive and susceptible leaf tissue induced by ammonia or by volatiles from bacterial cultures was less marked.

Crown rust epiphytology and heterogeneous oat cultivar-rust race populations. BLANCHE M. COURNOYER, J. A. BROWNING, D. JOWETT, & J. MELLON (Iowa State Univ., Ames). Near-isogenic oat lines were grown in pure and mixed stands in 50 × 50-ft plots and inoculated with several crown rust (*Puccinia coronata avenae*) races. Accumulated urediospore counts from Rotorod spore samplers exposed 2 hr daily outside the plots produced sigmoid growth curves. These were reasonably and consistently well described by the logistic equation. We derived and used the biologically meaningful parameters μ (the time of max spore release), M (the max spore release), and K (the final cumulative spore count at infinite time) to show epiphytotic differences in heterogeneous host and pathogen populations. Pathogen yields (final cumulative spore counts) were correlated negatively with host yields, and their regressions on percentage susceptibility showed that incorporation of more resistant plants significantly decreased numbers of spores trapped. Log_e pathogen yields were directly proportional to percentage of susceptibility in the host population. Spore and grain yield data from 96 large plots during 1966 to 1968 support the multiline theory that mechanical mixtures of near-isogenic lines buffer the host population against the rust population.

Effect of tobacco mosaic virus on fruit yield of tomato. J. P. CRILL, J. W. STROBEL, & B. VILLALON (Univ. Fla., Gulf Coast Exp. Sta., Bradenton; Univ. of Fla., Sub-Tropical Exp. Sta., Homestead). Tomato seedlings (*Lycopersicon esculentum* 'Walter') were transplanted into the field 4 weeks after seeding and inoculated with Alexander's strain 5 of tobacco mosaic virus (TMV) at 6, 7, 8, 9, and 10 weeks after seeding. A plot of noninoculated plants was included with the five plots of inoculated plants to make a total of six treatments in each of four replications. Yields were obtained from 10 plants in the center of each plot row. Sanitation procedures were practiced to avoid TMV contamination of noninoculated plots. At first harvest, the noninoculated plots, when assayed on local lesion tobacco, were 29, 36, 50, and 0% infected with TMV. Twenty-five plants were assayed at random from the inoculated plots; all contained TMV. Early infection reduced yields significantly more than later infections. Fruit yield progressively decreased from 1,140 to 778 bu/acre, and number of fruit per plant decreased from 42 to 32. Yield of culls did not increase in TMV-infected plots.

Genetic control of the gold fleck disease of tomato fruit. J. P. CRILL, J. W. STROBEL, & B. VILLALON (Univ. Fla., Gulf Coast Exp. Sta., Bradenton; Univ. Fla., Sub-Tropical Exp. Sta., Homestead). Pox and gold fleck are tomato diseases characterized by symptom expression on both ripe and green fruit. Symptoms of the two diseases are distinct, but gold fleck has previously been considered an early symptom of pox. Observations of fruit in several states and tomato-producing areas in the past year indicated pox could develop without fleck occurring previously; in many cases, fleck was the only symptom to occur. Gold fleck appears to be a genetically controlled disease distinct from pox. F₁ and F₂ ratios coupled with the study of F₃ families of fleck × no fleck crosses indicated that the genetic mechanism was more involved than a single gene, and expression appeared to be dominant.

Bacterial blight of soybeans in relation to wind-rain storms. G. C. DAFT & C. LEBEN (Ohio Agr. Res. Dev. Center, The Ohio State Univ., Wooster). During 1969, tearing of young leaf laminae occurred in the three major wind-rain (storm) periods. Prior to the first storm, a few lesions incited by *Pseudomonas glycinea* were on the lowest leaves. Two to 5 days after each storm, blight was in a horizontal stratum of leaves throughout the plots. Lesions were on the two youngest, unfolded leaves. Few or no lesions were on folded and partially-folded leaves, probably because they were not abraded by older foliage. (In greenhouse inoculations, young leaves were more susceptible than old ones.) During one long storm, when leaf damage was most severe, lesions were concd in abraded regions between main veins and along margins. Virtually no lesions were produced on leaves of any age following periods of rain without leaf-damaging winds. Evidence indicated that inoculum in amounts sufficient to incite lesions rarely was transferred more than 1 m. Surface water from lesioned leaflets, apparently-healthy leaflets, and buds immediately following storms was infective in greenhouse tests. After simulated storms in the field and greenhouse, symptom patterns were similar to those following natural storms. Foliage injury produced in wind-rain storms is indicated to be a major factor in blight initiation.

Enhancement of cowpea chlorotic mottle virus infectivity in vivo by 2-thiouracil or manganese sulfate. W. O. DAWSON & C. W. KUHN (Univ. Ga., Athens). Sap infectivity and specific infectivity of purified cowpea chlorotic mottle virus (CCMV) extracted from cowpea plants treated with toxic levels of 2-thiouracil (TU) or manganese sulfate (Mn) increased markedly as compared with controls. Infected plants were treated by immersing excised stems in 0.001 M TU or 0.01 M MnSO₄ or by spraying intact plants with 0.005 M TU or 0.05 M MnSO₄. This enhancement of virus infectivity by TU is in contrast to numerous reports indicating that virus nucleoprotein and infectivity decrease in plants treated with TU. Infectivity and virus nucleoprotein of tobacco mosaic virus in TU-treated tobacco was reduced 90%, whereas the infectivity of CCMV in cowpea treated with TU or Mn beginning at the time of inoculation increased 1.5 to 4 times (number of local lesions). When treatment was begun 20-30 days after inoculation, the infectivity increased 2 to 10 times. The increase in infectivity of CCMV in cowpea treated with TU occurred 8 to 14 days after treatment initiation. The Mn-induced infectivity increase began within 5 to 6 days, gradually reaching a max by 14 days of treatment. The virus nucleoprotein content of plants treated with TU or Mn sometimes increased, but never as dramatically nor as consistently as the infectivity increased.

The dikaryon of Ustilago maydis. P. R. DAY & S. L. ANAGNOSTAKIS (Conn. Agr. Exp. Sta., New Haven). Mating between compatible strains of *Ustilago maydis* occurs on a min medium containing 1.0% activated charcoal, and is revealed as zones of white aerial hyphae where cells of the two types come into contact. When the mated strains carry complementary auxotrophic mutants the mycelium is stable and shows slow growth. Subcultures to a complete medium show that the mycelium is heterokaryotic, forming sporidia of both parental types. Only the hyphal tip cells of the mycelium contain cytoplasm and nuclei. We have not observed clamp connections. We have shown that this heterokaryon may give rise to diploid cells. These are solopathogenic, and alternate between mycelial and sporidial growth on charcoal min medium. The diploid cells are more vigorous and soon overgrow the heterokaryotic mycelium.

Laboratory testing of tubers for multigenic resistance to late blight. K. L. DEAHL & M. E. GALLEGLY (W. Va. Univ., Morgantown). Using cultivars of potatoes with varying levels of multigenic resistance to *Phytophthora infestans*, bioassay techniques were developed to measure the amount of resistance in tubers. A positive correlation was found

between foliage and tuber resistance. This correlation was evidenced in the amount of infection observed when inoculations with race 1,2,3,4 were made after wound periderm formation in tuber discs was allowed to proceed for 24 to 48 hours at 20 C. Dehydrogenase activity measured by the reduction of triphenyltetrazolium chloride was greater in inoculated susceptible tissue than in resistant tissue, and decreased with an increase in levels of resistance. After 48 hr of wound periderm formation, the amount of chlorogenic acid and total phenols was much greater in the resistant tissue than in the susceptible tissue. Within a cultivar, the rate of phenol biosynthesis and the amount of accumulated polyphenols was greater in cortical tissue than in the parenchyma. Chromatographic analysis showed marked differences in the pattern of phenol synthesis initiated by wounding as compared to infection by *P. infestans* in resistant tissues.

Induction of phytoprecipitins in sunflower gall tissue in response to infection by Agrobacterium tumefaciens. J. E. DE VAY, R. J. ROMANI, ANNA M. MONADJEM, & MARILYN ETZLER (Univ. Calif., Davis). Serological comparisons of antigen preparations (AP) from *Agrobacterium tumefaciens* and from aseptic tissue cultures of crown gall and normal stem of *Helianthus annuus* 'Mammoth' confirmed a previous report that common antigens are shared by these tissues and the bacteria. Precipitin reactions involving only antigens were also observed. Three distinct precipitin bands appeared in agar gel diffusion tests between AP of gall tissue and virulent bacteria, whereas only two weak bands occurred between AP of normal tissue and virulent bacteria. Precipitin reactions between AP of avirulent bacteria and gall or normal tissues gave one or two weak bands. When AP from gall or normal tissues were titrated with AP from virulent bacteria, the nitrogen (N) and/or protein (P) contents of the precipitates reached a max and then declined as is characteristic of antigen-antibody reactions. Maximum reaction occurred when the ratio of bacterial N or P to gall or normal tissue N or P was in the range 7-10 to 1. The total N or P in the precipitate from the reaction of bacterial AP and gall AP was always greater than the reactions with normal tissue AP. These results indicate that a unique antibodylike substance which we call a phytoprecipitin is formed in sunflower tissue which has been transformed to the gall state by *A. tumefaciens*.

Circular ribonucleic acids in healthy and potato spindle tuber virus-infected tomato tissue. T. O. DIENER (Plant Virol. Lab., USDA, Beltsville, Md.). Potato spindle tuber virus (PSTV) ribonucleic acid (RNA) was insensitive to treatment with exonucleases. Infectivity was not materially affected when extracts containing PSTV-RNA were incubated with either snake venom phosphodiesterase or bovine spleen phosphodiesterase under conditions that completely inactivated tobacco mosaic virus RNA. Incubation of PSTV-RNA with a combination of exonuclease and alkaline phosphatase similarly had no significant effect on infectivity. Thus, PSTV-RNA is either a closed circular molecule or "masked" at both the 3' and 5' termini from attack by exonucleases. Exonuclease-resistant RNA was isolated by gel filtration (Sephadex G-100) of extracts extensively digested with exonuclease and alkaline phosphatase. The excluded, high molecular-wt RNA was concd and further fractionated by hydroxyapatite chromatography. Density-gradient centrifugation revealed a single component that sedimented at a rate of ca. 10 Svedberg units, and closely coincided with infectivity. Exonuclease-resistant RNA with similar properties could also be isolated from healthy leaves. Electron microscopy (in cooperation with Dr. T. Koller, Swiss Federal Institute of Technology) revealed circular RNA molecules in preparations from healthy and PSTV-infected tissue.

Cohabitation of Verticillium sp. and Sclerotinia lagerbergii in Pinus resinosa. C. E. DORWORTH (Can. Forest Service, Sault Ste. Marie, Ontario, Canada). Infection of young red and jack pine by *Sclerotinia lagerbergii* typi-

cally results in branch dieback, followed by stem canker formation and death of as many as 90% of the individuals in a plantation. A green discoloration of the wood and inner bark often accompanies infection, and is considered a reliable symptom of the disease. A species of *Verticillium* was isolated from the leading edges of several cankers which exhibited the green color symptom typically associated with infections by *S. lagerbergii*. Acetone extracts of the wood and from cultures of *Verticillium* and *S. lagerbergii* each contained a green pigment. The pigment was partially purified by successive washes with alkanes, followed by elution with cold diethyl ether from a silica gel column. An identical R_F was revealed for each pigment upon development on Eastman cellulose thin layers in nine different solvent systems, the more polar systems generally yielding the higher R_F values. This and other evidence indicate that the same pigment was extracted from each source. The presence of either a genetic or an ecologic connection between the two fungi is postulated.

The detrimental effect of rust on the water relations of bean. J. M. DUNIWAY & R. D. DURBIN (Univ. Wis., Madison). Bean plants with primary leaves infected by *Uromyces phaseoli* (60 colonies/cm²) became unusually susceptible to drought as sporulation occurred. Under the conditions used (1,500 ft-c, 27 C, and 60% relative humidity), such plants, including noninfected leaves, wilted at soil water potentials greater than -1 bar. More severe conditions, e.g., a 4-fold decrease in soil water potential, were required before healthy plants wilted. Determinations of leaf water potential and osmotic potential confirmed the observation that diseased plants suffered greater water and turgor losses under stress conditions. The determinations also showed that an osmotic alteration in the diseased leaf was not responsible for wilting. When the diffusive resistance of leaves to water vapor loss was measured as a function of decreasing leaf water content, the diffusive resistance of healthy leaves was found to increase markedly, while that of diseased leaves remained very low. We concluded that water loss through sporulating uredia was of such magnitude that stomatal closure in response to leaf water stress did not limit water loss. Apparently, this non-stomatal transpiration, together with the significant reduction in the root:shoot ratio which occurs in diseased plants, upsets the water economy of the diseased host under mild drought conditions.

Sources of resistance to stunt of beans incited by peanut stunt virus. E. ECHANDI & T. T. HEBERT (N. C. State Univ., Raleigh). Stunt of beans incited by peanut stunt virus (PSV) appeared in epiphytotic form in 1969 in North Carolina. No resistance was found in over 80 different commercial pole and snapbean varieties tested under greenhouse conditions. Susceptible plants showed epinasty, stunting, crinkling, and mottling of the true leaves. Apical necrosis was observed in a few varieties. The following species of *Phaseolus* were grown in the greenhouse and inoculated mechanically with bean and peanut isolates of PSV: *P. mungo*, *P. acutifolius*, *P. acutifolius* var. *latifolius*, *P. lunatus*, *P. angularis*, *P. aboriginus*, *P. bracteatus*, *P. dumosus*, *P. atropurpureus*, *P. aureus*, *P. calcaratus*, and *P. aconitifolius*. The last five species mentioned appeared to be resistant. Recovery of virus from the true leaves of resistant plants was unsuccessful. Two of three different accessions of *P. coccineus* inoculated with PSV were resistant. The virus was not recovered from the true leaves of resistant plants. Resistance to the western strain of PSV was also found in *P. dumosus* and *P. coccineus*.

*Accumulation and metabolism of aliphatic amines by hyphae of *Penicillium digitatum* in relation to growth inhibition.* J. W. ECKERT, M. L. RAHM, & B. BRETSCHNEIDER (Univ. Calif., Riverside). Twelve-hr-old hyphae of *Penicillium digitatum* were suspended in a growth medium at pH 4.5 containing 5 μ moles/ml of an aliphatic amine. After 4 hr, the μ moles amine/g dry hyphae and per cent inhibi-

tion of growth were, respectively: ethylamine, 10, 13%; *n*-propylamine, 17, 15%; *n*-butylamine, 27, 10%; 1-methylbutylamine, 72, 14%; isobutylamine, 32, 52%; (+) *sec*-butylamine (2-aminobutane), 149, 47%; (-) *sec*-butylamine, 194, 54%. In another experiment, hyphal mats deaminated in 4 hr 20-40% of the ethylamine, *n*-propylamine, and *n*-butylamine applied, but did not metabolize the other amines significantly. Hyphae accumulated ³H-labeled (-) *sec*-butylamine to a level 5 to 10 times greater than that in the medium. Metabolic products were not detected. Uptake of (-) *sec*-butylamine by the hyphae was inhibited by 2,4-dinitrophenol, azide, cyanide, or incubation under N₂, indicating that the amine was actively transported into the hyphae. Accumulated (-) *sec*-butylamine effluxed rapidly from the hyphae.

The genetics of seedling emergence, postemergence damping-off, and final stand in cotton. K. M. EL-ZIK & L. S. BIRD (Texas A&M Univ., College Station). Data were obtained from an eight-parent diallel cross of *Gossypium hirsutum* grown in a controlled-temp tank and at three locations in Texas. The temp tank results confirm the differential ability of some strains to germinate at lower temp. Seedling emergence, velocity of emergence, and final stand were greater at 20 C than at 16.7 C. Postemergence damping-off was more severe at 20 C. Variation detected in velocity of emergence was caused mainly by environment. The additive genetic components were not significant for any of the traits. Dominance was in the direction of higher emergence, damping-off, and final stand. Emergence was controlled by at least four genes and final stand by three genes exhibiting overdominance. One effective factor controlled postemergence damping-off. Field test data, for 2 years, indicated that genetic variation in final seedling stand was due to dominance. The field data suggested a min of two effective factors controlling plant stands. Overdominance was exhibited in all tests, and heritability estimates were relatively low. Seedling-stand ability in cotton is inherited polygenically, and environment plays a major role in its expression.

A quick-dip method for inoculation of tomato seedlings with tobacco mosaic virus. D. A. EMMATTY & C. A. JOHN (H. J. Heinz Co., Bowling Green, Ohio.). Inoculation of tomato seedlings at the cotyledonary stage by a dipping method was compared to a rubbing method. When the cotyledons and the upper part of the stem of susceptible Heinz 1370 tomato seedlings were dipped in the crude sap extracts containing the aucuba strain of tobacco mosaic virus, 99% were infected. Rubbing the cotyledons with cotton swabs dipped in the inoculum also resulted in 99% infected plants. The resistant line, Alexander 683201, remained free of symptoms when inoculated by either method. At temp of 29-35 C, the aucuba strain induced necrosis in addition to the typical yellow mosaic symptoms in susceptible seedlings; however, more necrosis occurred on plants inoculated by rubbing than by the dip method. At 15.5 to 21 C, mosaic without necrosis was observed in plants inoculated by either method. The dipping technique was also highly effective in infecting plants with the Ohio TMV strain V. This quick and effective dip method can be substituted for the rubbing method as a reliable way to determine the resistance or susceptibility of large numbers of tomato seedlings to strains of TMV.

*The pathogenicity of *Helminthosporium* sp. on chrysanthemum, rose, snapdragon, and carnation.* A. W. ENGELHARD (Univ. Fla., Gulf Coast Exp. Sta., Bradenton). In January 1969, *Helminthosporium* sp. was isolated in Florida from the petals of a spider-type chrysanthemum seedling from a commercial grower. Pathogenicity tests in the laboratory indicated that the blooms of Yellow Knight, Tinsel, Southern Comfort, and Yellow Shasta cultivars of chrysanthemum were susceptible, whereas Iceberg, Yellow Iceberg, Blue Chip, Bronze Chip, Dolly, and Jackstraw were not. Additional tests indicated that blooms of rose (Tropicana, Improved Red American Beauty [IRAB]), snapdragon

(Hawaii), and carnation (Goldilocks) were also susceptible, whereas salmon- and lavender-colored blooms of King aster were not. Inoculated rose (IRAB) blooms became infected when incubated in polyethylene bags for 48 hr at 7, 13, 18, 24, 29, and 35 C, with max disease development in the 24- to 29-C range. Initial symptoms on chrysanthemum petals included irregular, reddish-brown spots up to 0.2 mm in length; on rose and snapdragon, tan, necrotic areas up to 2 mm in diam; on carnation, marginal tissue necrosis. Initial symptoms on all suspects were followed by general necrosis if spotting was severe. *Helminthosporium* sp. was reisolated from inoculated plants. This constitutes, to the author's knowledge, the first report of *Helminthosporium* sp. inciting disease on these hosts.

Cell wall degradation by a bean pathogen initiated by a "wall-modifying enzyme". P. D. ENGLISH, K. G. KEEGSTRA, J. A. MAGLOTHIN, & P. ALBERSHEIM (Univ. Colo., Boulder). When the fungus, *Colletotrichum lindemuthianum*, is grown on cell walls isolated from bean hypocotyls, several polysaccharide-degrading enzymes are secreted sequentially into the culture medium. Pectinase and α -arabinosidase are secreted first, followed by cellulase and β -xylosidase, then β -glucosidase, and, finally, α -galactosidase. The culture medium enzyme solution is able to degrade bean hypocotyl cell walls. A highly purified preparation of the pectinase is able to cause some cell wall degradation, but highly purified preparations of α -arabinosidase and α -galactosidase and partially purified cellulase cannot degrade cell walls by themselves. The action of the pectinase on walls is required before the α -arabinosidase or cellulase can cause degradation. The order of enzyme secretion may reflect the sequence in which the enzymes must work to degrade cell walls. The pectinase appears to have the properties of a "wall-modifying enzyme". Wall-modifying enzymes are likely to play an essential role in diseases in which cell wall degradation is important.

Enhancing efficacy of benomyl to control Venturia inaequalis. A. H. EPSTEIN (Iowa State University, Ames). Four rows of 3-year-old nursery trees of the crabapple variety Hopa (highly susceptible to *Venturia inaequalis*) were sprayed 4 times during the growing season with benomyl [methyl-1-(butylcarbamoyl)-2-benzimidazolecarbamate] 4 oz active ingredient/100 gal. Two of the four rows were sprayed with a liquid polyethylene sticker (Plyac), 6 oz/100 gal with the benomyl. Sprays were applied 9 June, 1 July, 21 July, and 18 August. Terminal shoots were collected on 23 September. Lesions were counted on 50 leaves selected randomly from five shoots from each tree. Counts were made on five trees randomly selected in each row. Addition of Plyac to sprays containing benomyl resulted in significant reduction (1% level of confidence) in the average number of scab lesions per leaf, as compared to treatments in which no Plyac was used. Trees sprayed with benomyl alone averaged 10.0 lesions/leaf, while those sprayed with benomyl plus Plyac averaged 1.1 lesions/leaf. Unsprayed control trees in two additional rows were severely defoliated because of severe *Venturia inaequalis* activity; hence, no data are presented from them.

Effect of nitrogen on development of seedling disease caused by Fusarium moniliforme in sorghum. I. D. ERINLE & L. K. EDMUNDS (Kansas State Univ., ARS, USDA, Manhattan). Four nitrogen sources, $(\text{NH}_4)_2\text{SO}_4$, NH_4NO_3 , NaNO_3 , and urea, were tested at the equivalent of 0.8 g actual N/kg of infested soil amended with mature plant residues. Two, 4, 6, and 8 weeks after soil infestation, inoculum densities were estimated by soil-dilution plates on Nash-Snyder medium. After 8 weeks, the soil was planted with Spur Feterita. Three weeks later, root damage and height and wt of top growth were recorded. Pathogen population declined more rapidly in nitrogen-supplemented treatments, but no direct correlation was observed between inoculum density and subsequent disease severity. Plants grown in nitrogen-supplemented (especially NaNO_3) soil

were significantly more diseased than plants in residue-only infested soil. Nitrogen levels of 0, 50, 100, 200, and 400 ppm from NH_4NO_3 were tested on 3 sorghums, RS 610, Redlan, and Spur Feterita grown in sand infested 3 days before being planted. Disease development was greatest at 200 ppm N; 400 ppm N were phytotoxic. Effects on pathogen virulence or host susceptibility and antagonisms between *F. moniliforme* and other microflora, alone or in combination, may have accounted for the various responses to sources and levels of nitrogen supplied.

Translocation of 2-(4-thiazolyl)benzimidazole in cotton. D. C. ERWIN, M.-C. WANG, & J. J. SIMS (Univ. Calif., Riverside). When 2-(4-thiazolyl)benzimidazole (TBZ) was applied to sand or soil in which cotton plants were growing, a fungitoxic substance presumed to be TBZ was detected by an agar-diffusion bioassay method in xylem and bark tissue of stems but less frequently in leaves. When 1.2 μC of ^{14}C -TBZ was applied in the root zone of 3-week-old plants, radioactivity was detected 3 days later in the stems and to a lesser extent in leaves. Scintillation counts per min per 10 mg dry tissue at 6 days were: roots (1.2 cm below surface) 599; stems (2.5 cm above) 557; (5.0 cm) 417; (10.0 cm) 201; (15.0 cm) 137; and (20 cm) 9. Radioautograms of plants indicated a similar pattern of distribution. Binding of ^{14}C -TBZ to a plant constituent is postulated, since there was no redistribution of ^{14}C -TBZ in plants that were heated to 60 C. Mobility by thin-layer chromatography (TLC) of the ^{14}C -constituent(s) from crude extracts (acidic methanol) was different from authentic ^{14}C -TBZ. After hydrolysis of the ^{14}C -constituent(s) with hot 6 N HCl, a radioactive compound with a similar R_F value (TLC) to that of authentic ^{14}C -TBZ was obtained. ^{14}C -TBZ (0.3 μC in acetone), applied to a defined spot near the base of a leaf, moved toward the tip of the leaf but not to the petiole. When applied to the base of the stem, movement was upward only.

Interaction of Pratylenchus penetrans and Meloidogyne incognita acrita as cohabitants on tomatoes. R. A. ESTORES & TSEH AN CHEN (Rutgers Univ., New Brunswick, N. J.). Interaction of *Pratylenchus penetrans* and *Meloidogyne incognita acrita* infecting tomatoes alone and as cohabitants was studied. Although initial penetration of *Pratylenchus* was not affected by the presence of *Meloidogyne*, the subsequent development and reproduction of *Pratylenchus* were inhibited. Sixty days after inoculation, the number of *Pratylenchus* was 57% higher when alone than when combined with *Meloidogyne*. Even when *Meloidogyne* was introduced later than *Pratylenchus*, the result was the same as when they were introduced at the same time. Split-root experiments indicated that the inhibitory effect of *Meloidogyne* on the development of *Pratylenchus* involves more than a competition for feeding sites. Thirty-six days after inoculation, the number of *Pratylenchus* increased 82% when one-half the root system was left noninoculated, as compared to a 26% increase when one-half the root system was infested with *Meloidogyne*. In the presence of *Pratylenchus*, the galls formed by *Meloidogyne* were smaller and fewer than those formed by *Meloidogyne* alone. Although plant growth was reduced by the infection of either nematode species or combination of both, *Meloidogyne* alone caused a more severe stunting than when combined with *Pratylenchus*.

Reaction of selected sorghum and millet hybrids, cultivars, and accessions to strains of sugarcane mosaic virus and maize dwarf mosaic virus. S. FAZZI, R. W. TOLER, & A. J. BOCKHOLT (Texas A&M Univ., College Station). Thirteen sorghum and millet genotypes were evaluated both in the greenhouse and field after virus inoculation with an artist's airbrush. Effects of sugarcane mosaic virus (SCMV) strains A, B, D, H, and I, in addition to maize dwarf mosaic virus strain A (MDMV-1), were investigated. Disease reaction ratings included symptomology, per cent plants infected, and degree of disease severity. Differential

reactions were observed among the cultivars ranging from symptomless, mottling, chlorosis, reddening, and necrosis to hypersensitive lethal. Amount of infection varied from 0 to 97%. Only two of the genotypes exhibited resistance to the different strains and viruses. The cultivars Wiley and Martin offer some degree of resistance to all viruses and strains tested. These two sorghum cultivars offer potential multiple resistant genotypes, and may be used as sources of resistance to SCMV and MDMV.

A fungal inducer of soybean phytoalexin from Phytophthora megasperma var. sojae. J. A. FRANK & J. D. PAXTON (Univ. Ill., Urbana). Bioassays utilizing soybean cotyledons indicated that fungal extracts of *P. megasperma var. sojae* were capable of inducing the yellow-green fluorescent soybean phytoalexin. This inducer was present in small amounts in 7-day-old cultures grown on V-8 juice broth. The amount of inducer could be increased when the fungus was removed from the V-8 broth after 7 days and placed in filter-sterilized juice from a resistant variety (Harosoy 63). Juice from a susceptible variety (Harosoy) did not appreciably increase the amount of inducer. Autoclaved soybean juice was no longer capable of stimulating fungal inducer, but the inducer itself appeared to be relatively heat stable.

Multiple disease resistance in exotic sorghum lines. R. A. FREDERIKSEN & D. T. ROSENOW (Texas A&M Univ., College Station). Several genotypes of sorghum obtained from tropical introduction³ possess high levels of multiple disease resistance. The lines were developed in a program in which tall, late-maturing tropical sorghums were converted to dwarf, early-maturing lines. By screening and selecting partially converted sorghums under conditions of natural infection for head smut (*Sphacelotheca reiliana*), downy mildew (*Sclerospora sorghi*), anthracnose (*Colletotrichum graminicola*), maize dwarf mosaic, and several common foliar pathogens, seven experimental lines with resistance to these diseases were developed. Although the level of resistance to any one disease may not exceed that of common open-pedigree breeding lines and cultivars, none was previously known to possess such high levels of resistance to all of the diseases. Resistance, particularly to foliar diseases, appears to be horizontal. The multiple disease-resistant sorghums were derived from the following exotic sorghums: IS2579, IS2816, IS12610, IS12661, and IS12664.

Comparative biochemistry of nine soybean cultivars and its relationship to susceptibility to Macrophomina phaseoli. S. GANGOPADHYAY & T. D. WYLLIE (Univ. Mo., Columbia). *Macrophomina phaseoli*, the cause of charcoal rot of soybean and many other crops, has become increasingly important on soybeans throughout Missouri. Protein nitrogen, total nitrogen, total and free amino acids, total sugars, acid hydrolyzable polysaccharides, and total and water soluble phenols were determined on nine diverse soybean genotypes in an attempt to explain observed differences among the cultivars in the responses of root, internodal, and nodal tissue to infection and sclerotial formation by *M. phaseoli*. Greater quantities of total nitrogen, soluble protein, total sugars, total and free amino acids were found in the more susceptible cultivars and also in the more susceptible nodal tissues of each variety. Inverse correlations of acid hydrolyzable polysaccharides and total and water soluble phenols were observed. A nutritional basis for degrees of susceptibility of specific tissues of specific cultivars to sclerotial formation by *M. phaseoli* is postulated.

Protection against Helminthosporium victoriae toxin and evidence for proteins as toxin receptors. J. M. GARDNER & R. P. SCHEFFER (Mich. State Univ., E. Lansing). *Helminthosporium victoriae* (HV) toxin causes rapid changes in the plasma membranes of susceptible oat cells. The degree of toxin-induced damage was measured as increased electrolyte loss from cells. Pretreatment of tissues with cycloheximide (CH) (2-5 µg/ml) for 12 hr reduced sensi-

tivity to toxin by 80-90%, but when tissues were removed from CH, toxin sensitivity reappeared in 48 hr. Tissues infiltrated with N-ethylmaleimide (NEM) and dinitrofluorobenzene (DNFB) solutions (5 µM) 40 sec prior to toxin treatment were 20-40% less sensitive to toxin; tissues pretreated for 30 min were 80-90% less sensitive than were control tissues. DNFB but not NEM protection was reversed with 80 µM mercaptoethanol. NEM protection was reduced 20-80% when toxin breakdown products (0.2-0.8 mg/ml), including the peptide portion of the toxin molecule, were added with the NEM. A particulate fraction from susceptible oats, prepared by differential centrifugation in 0.5 M sucrose between 12,000-50,000 g, bound significant amounts of ¹⁴C-NEM. Toxin breakdown products reduced the ability of this preparation to bind ¹⁴C-NEM. These results suggest that toxin receptors in the cell are proteins.

Transmission of exocortis virus to various citrus plants by knife-cut inoculation. S. M. GARNSEY & R. WHIDDEN (ARS, USDA, Orlando, Fla.). Seedlings and cuttings of various citrus species, hybrids, and a citrus relative were tested for susceptibility to knife-blade contamination with exocortis virus. Stems of 10 or more healthy plants were cut 6 to 10 times with a knife blade freshly contaminated by cutting stems of Etrog citron (*Citrus medica*) systemically infected with exocortis virus. Six to 12 months later, the plants inoculated by knife-cut were indexed on citron indicators to determine whether infection had occurred. Nearly all Etrog citron, Rangpur lime (*C. limonia*), Eureka lemon (*C. limon*), trifoliolate orange (*Poncirus trifoliata*), and Carrizo and Morton citrange (*C. sinensis* × *P. trifoliata*) plants were infected. No plants of Orlando tangelo (*C. reticulata* × *C. paradisi*) or Rusk citrange, and less than 5% of Duncan grapefruit (*C. paradisi*) plants were infected. Plants of sweet orange (*C. sinensis*), sour orange (*C. aurantium*), Mexican lime (*C. aurantifolia*), and rough lemon (*C. jambhiri*) were of intermediate susceptibility with infection rates of 20 to 70%. Noninoculated plants of all test varieties indexed negatively.

Antagonisms between indigenous Pythium myriotylum and introduced Rhizoctonia solani and peanut pod breakdown. K. H. GARREN (ARS, USDA, Va. Polytechnic Inst., Holland). Many peanut (*Arachis hypogaea*) fruit were rotted by *Pythium myriotylum* when grown in a potting mixture of soil, vermiculite, and peat moss (3:1:1). The soil, from a field in which much pod breakdown was found in 1968, was pasteurized with moist heat before the mixing. When the potting mixture was infested with massive doses of *Rhizoctonia solani* originating from decayed peanuts, pod breakdown occurred. These results met the requirements of Koch's postulates and established *R. solani* as a pathogen of peanut pod breakdown. Antagonism between the indigenous *P. myriotylum* and the introduced *R. solani* was also noted. At one of four temp regimes, indigenous *P. myriotylum* clearly dominated over introduced *R. solani* in causing pod breakdown. Important field losses from pod breakdown by *R. solani* should occur when conditions favor this pathogen and keep the endemic *P. myriotylum* quiescent.

Further investigations upon some biologically active bark components in apple. J. E. GATES, J. CIMANOWSKI, & D. F. MILLIKAN (Univ. Mo., Columbia). Previous investigations indicated the presence of two groups of endogenous constituents in the apple bark tissue possessing biological activity against *Phytophthora cactorum*. These components were isolated by gel-filtration of an alkaline extract of inner bark on Sephadex G-26 by column elution with 0.02 M phosphate buffer (pH 6.7). The substances under one peak were found to be stimulatory and the other inhibitory to the in vitro growth of the fungus. When combinations were made on the basis of ultraviolet absorption spectra of the 20-ml eluates under these peaks, it was found that only a portion of the stimulatory peak was active. The addition of sodium sulfite (2%) to the ex-

tracting solution improved the separation of the active substances on the Sephadex column and increased its yield. A second inhibitory substance was found in the original stimulatory peak. This additional inhibitor gave a positive Lowry and Kjeldahl test, and paper chromatography indicated the presence of at least five free amino acids. Both inhibitory substances appear to inhibit other species of *Phytophthora*, and another bark pathogen, *Endothia parasitica*.

Amino acid analysis of five strains of sugarcane mosaic virus. A. G. GILLASPIE, JR., & V. L. FRAMPTON (ARS, USDA, Houma, New Orleans, La.). Several samples of each of five strains of sugarcane mosaic virus (SCMV) were purified for amino acid analysis. Four strains (A, B, D, and H) infecting sorghum were extracted in 0.3% ascorbic acid and 2-mercaptoethanol with 0.01 M sodium diethylthiocarbamate, and one strain (I) on corn was extracted in 0.5 M sodium citrate, pH 7.5, with 0.3% 2-mercaptoethanol. One-third volume of chloroform was used for clarification. Purification was accomplished by one high-speed ultracentrifugation followed by sucrose density gradient centrifugation (2 hr rate and then 17 hr "equilibrium"). The virus was centrifuged out of the sucrose, resuspended, and stored frozen. Samples for amino acid analysis were thawed, mixed with 6 N HCl, sealed in ampoules in a N₂ atmosphere, and hydrolyzed at 110 C for 18 hr. Three replications of each sample were analyzed. Methionine, phenylalanine, proline, tyrosine, and valine were either present in small amounts or not present. These amino acids are more abundant in other viruses. It may be possible to differentiate strains of SCMV using amino analysis.

Aphid transmission of pea seed-borne mosaic virus. L. C. GONZALEZ & D. J. HAGEDORN (Univ. Wis., Madison). Pea seed-borne mosaic virus (PSBMV), sometimes called pea fizzle-top virus, was transmitted by *Myzus persicae*, *Acyrthosiphon pisum*, and *Macrosiphum euphorbiae* when apterae or alatae of these aphid species were allowed single probes, 10-90 sec long, on infected source plants. Transmission seldom occurred when any of the three aphid species were given 3-hr acquisition feedings. The retention period of PSBMV after uptake by *A. pisum* was less than 5 min in feeding and slightly longer in fasted vectors. The results clearly indicate that PSBMV is transmitted in a stylet-borne manner. Pea (*Pisum sativum*) and broad bean (*Vicia faba*) appeared to be equally adequate as sources of the virus for uptake by aphids. Preliminary evidence suggested that, of the aphids tested to date, *M. persicae* may be the most efficient vector.

Serological identification of potato virus Y and tobacco etch virus using immunodiffusion plates containing sodium dodecyl sulfate. G. V. GOODING, JR., & W. W. BING (N.C. State Univ., Raleigh). The recent use of techniques for degrading flexuous, rod-shaped viruses into serologically active, diffusible units has facilitated their identification by immunodiffusion. In this investigation, sodium dodecyl sulfate (SDS) and sodium dibutylphthalenesulfonate (SBNS) at 0.1, 0.5 and 1.0% concn and 0.5 M ethanolamine, pH 10.5, were compared for the serological detection of potato virus Y and tobacco etch virus from *Nicotiana tabacum*, *Lycopersicon esculentum*, and *Capsicum frutescens*. SDS or SBNS were incorporated into a medium containing 0.8% Purified Difco agar and 1% sodium azide; crude juice was used as the antigen source. Ethanolamine treatment consisted of grinding 1 g of tissue in 2 ml of ethanolamine; antigen prepared in this way was used in an agar-gel containing 0.8% Purified Difco agar and 1% sodium azide. Both viruses were consistently detected in all hosts using 0.5% SDS, and nonspecific precipitation occurred infrequently. Some of the other treatments resulted in good precipitin line formation with some virus-host combinations. No other treatment, however, was as good over-all as 0.5% SDS, and more nonspecific reactions occurred with them.

Similarity of ultrastructural modifications in tobacco leaf tissue caused either by pathogenic bacteria or exogenous ammonia. R. N. GOODMAN, S. B. PLURAD, & L. LOVREKOVICH (Univ. Mo., Columbia). Identical gross symptoms were caused when tobacco leaves were infiltrated with 10⁸ cells/ml of *Erwinia amylovora*, *Pseudomonas tabaci*, *P. pisi*, or exposed briefly to ammonia gas. Tissue showing the earliest evidence of symptom development (water-soaking and flaccidity) was prepared for electron microscopic examination. Studies of thin sections revealed virtually identical ultrastructural modifications caused by either the bacteria or ammonia. Subcellular organelles most drastically altered are the chloroplasts, mitochondria, and microbodies. Chloroplasts displayed a characteristic swelling at granal margins which subsequently vesiculated. The primary effect seems to be on the bounding membranes of these organelles. A hypothesis is proposed that implicates ammonia produced by the bacteria as a necrotoxin which alters tertiary structure of membrane protein.

The relationship of a maize virus isolate from Ohio to sugarcane mosaic virus strains and the B strain of maize dwarf mosaic virus. D. T. GORDON & L. E. WILLIAMS (Ohio Agr. Res. Development Center, Wooster). Maize virus isolate 13B from Ohio, previously shown to be related but not identical to maize dwarf mosaic virus strain A (MDMV-A), has been found to be more closely related to strain B of MDMV (non-Johnson grass strain) and sugarcane mosaic virus (SCMV) strains A, B, D, E, and H. Antiserum of 13B cross-absorbed with MDMV-A reacted in microprecipitin tests with each of the SCMV strains, MDMV-B and 13B. MDMV-A antiserum cross-absorbed with 13B reacted with MDMV-A but not with the SCMV strains, MDMV-B or 13B. Inoculation of *Sorghum bicolor* cultivar Rio with SCMV strains, MDMV-B or 13B produced a systemic mottle, while MDMV-A produced severe stunting, mottle, and necrosis. Only MDMV-A produced systemic symptoms in Johnson grass, *Sorghum halepense*. On Atlas sorghum, SCMV strains E and H, MDMV-B and 13B produced local lesions on inoculated leaves and limited systemic red striping; MDMV-A and SCMV-A and B produced systemic mosaic, necrosis, and stunting, but not necrotic lesions on inoculated leaves. It is concluded that the 13B isolate is closely related to MDMV-B and strains E and H of SCMV. The results indicate that MDMV designations are most accurate for virus isolates which are serologically related to MDMV-A and which readily produce systemic mottle in Johnson grass and maize.

Conidial germination and population of Aspergillus flavus in the geocarposphere of peanut. G. J. GRIFFIN (Va. Polytech. Inst., Blacksburg). Under greenhouse conditions, pegs of Virginia Bunch 46-2 peanut plants were introduced into small pots containing nonsterile loamy fine sand. This soil was infested with conidia of *A. flavus* isolated from peanut fruit. Dilution plate analysis 2 weeks after infection indicated that the 0.5-mm layer of soil adhering to pegs (P) had a slightly higher *A. flavus* population than soil (S) from pots with no pegs (P/S = 1.3); the P/S value for total fungi was 4.7. After 10 weeks, the population of *A. flavus* isolated from the 0.5-mm layer of geocarposphere soil of pods (G) was slightly higher than in S (G/S = 1.1); the G/S value for total fungi was 5.9. The populations of bacteria and actinomycetes were much greater in P and G than in S. For plants maintained in a growth chamber at 30 C, microscopic observation indicated that conidia of *A. flavus* did not germinate in P and trace germination occurred in G (<0.1%) when samples were examined at intervals over periods of 24 and 81 days, respectively. When soil was artificially infested with *Fusarium oxysporum*, chlamydospore germination was observed in P. Germination of conidia of *A. flavus* was high (>50%) in G after 16 hr when a 4- to 6-mm² area of pod surface was superficially injured and inoculated with infested soil.

Citrus leaf pieces as traps for soil-borne Phytophthora spp. G. R. GRIMM & ANN F. ALEXANDER (ARS, USDA, Orlando, Fla.). Citrus leaf pieces proved to be a substitute for lemon fruit in detecting and isolating citrus *Phytophthora* spp. from soil. Soil samples, known to contain *Phytophthora* spp., were placed in small containers, and sufficient water was added to provide 1 to 2 cm of water above the soil level. Citrus-leaf pieces, 3 to 5 mm², were floated on the surface of the water. Sporangial development occurred in 3 to 4 days along the cut edge of floating leaf pieces, but was absent from whole leaves or cut pieces that sank. Leaf pieces from sweet orange (*Citrus sinensis*), Sour orange (*C. aurantium*), Yuma citrange (*C. sinensis* × *Poncirus trifoliata*), *P. trifoliata*, and several other citrus species and hybrids were equally effective as traps. Interference from other fungi with identification and isolation of *Phytophthora* spp. was negligible. Leaf pieces are especially adapted for use in small containers and are continuously available, whereas unblemished, unsprayed lemon fruit are available in Florida for only short periods each year.

Effect of initial inoculum level on epidemics of common blight of Phaseolus vulgaris. JERRY H. HAAS (Can. Dep. Agr., Harrow, Ontario). A suspension of *Xanthomonas phaseoli* var. *fuscans* was sprayed on primary leaves of Sanilac bean plants shortly after emergence. Initial inoculum levels (IIL) were established with 0, 1, 5, 90, 500, and 1, 3, 10 inoculated plants/1,000-plant field plot in 1968 and 1969, respectively. Disease was assessed weekly by measuring lesion area (LSA) and leaf area/plant on a random sample of plants from each of at least five replicate plots/treatment. At the 1/1,000 plant IIL, LSA increased from 0.001 to 0.37 cm²/plant during the first 35 days after inoculation in 1968. In 1969, the LSA reached 3.8 cm² in 37 days from the same IIL. Each year, the highest IIL had the lowest rate of LSA increase. The proportion of leaf area with lesions (*X*) also increased most slowly with the highest IIL. With 1 inoculated plant/1,000, *X* increased in 1968 from 10⁻⁷ to 10⁻³ between 9 and 35 days after inoculation. At the same time but with 50 inoculated plants, the increase was from 10⁻³ to 10⁻². In 1969, *X* progressed from 10⁻⁵ to 10⁻² between 7 and 43 days after inoculation of 1 plant/1,000. The epidemics were limited by normal senescence, not by lack of green leaf area. The relative infection rate was also inversely correlated with the IIL. Therefore, the IIL had less effect than expected on the final amount of disease.

The role of dyes and other DNA intercalating compounds in activating genes for phytoalexin production. L. A. HADWIGER & M. E. SCHWOCHAU (Wash. State Univ., Pullman). The increased synthesis of the phytoalexin and pisatin, and the activity of phenylalanine ammonia lyase (PAL), an enzyme in its metabolic pathway, are stimulated by an array of microbial metabolites and chemical compounds. We have proposed that compounds with the potential to change the conformation of double-stranded DNA can derepress the genes controlling these responses. The following compounds are listed in decreasing order of PAL induction potential: chromomycin A₃, quinine, nile blue, quinacrine, actinomycin D, taurine, nogalamycin, 2,7 diamino fluorene, thionine, 9-amino acridine, acridine orange, methylene blue, pyrogallol red, chloroquine, azure A, methylene green, pyronine Y, neutral red, methyl green, auramine O, brilliant cresyl blue, and ethidium bromide. Optimal concn of each of these chemicals induce PAL to activity levels 3 to 15 times those found in controls. Induction potential appears related to substitutions on planar 3 ring systems, and to the ability of the compound to intercalate into double-stranded DNA. Several of these components can induce resistance in host tissue to infection by various plant pathogens.

Replication of tobacco mosaic virus in barley. R. I. HAMILTON & J. A. DODDS (McGill Univ., Macdonald Col-

lege, Que., Canada). Tobacco mosaic virus (TMV) infected barley (*Hordeum vulgare* 'Black Hulless'), when seedlings, inoculated at the one- to two-leaf stage, were incubated at 30 C. TMV could be detected in inoculated leaves by analytical density-gradient centrifugation at 6 days; the amount of infectious virus increased during the interval of 6 to 10 days. Systemic spread of TMV could be detected in barley by infectivity assay or by analytical density-gradient centrifugation only when extracts from leaves were concd 10-fold. In mixed inoculation with barley stripe mosaic virus (BSMV), 60 to 70% of the inoculated plants were infected with TMV as determined by microprecipitin and infectivity assays of crude extracts from the third leaves. The proportion of TMV to total viral nucleoprotein in systemically infected third and fourth leaves was estimated to be 0.4 and 0.6, respectively. This is the first report of TMV infecting barley. The mixed infection of BSMV and TMV in barley is similar to other mixed virus systems, with the exception that (i) temp may be an important factor in establishing TMV infection; and (ii) the estimated concn of TMV in systemically infected barley leaves after inoculation with TMV alone is inordinately low for this virus.

The influence of age on neutral carbohydrate components in root exudates from alfalfa plants grown in a gnotobiotic environment. R. A. HAMLIN, F. L. LUKEZIC, & J. R. BLOOM (Pa. State Univ.). Because of their importance in *Meloidogyne* egg hatch and fusaria chlamydospore germination, investigations were completed on the neutral carbohydrate fraction of root exudates of DuPuits alfalfa. Plants were grown gnotobiotically in a nutrient solution in washed riverbank sand, and after 6 weeks' growth were clipped weekly to a constant height. Distilled water leachings from roots in situ and root dry-wt determinations were obtained from 4-, 6-, 8-, 10-, 12-, 14-, and 16-week-old plants. Trimethylsilyl derivatives of the carbohydrates in the leachates were analyzed with gas chromatography and confirmed by mass spectrometry. Based on mg/leachate sample arabinose, ribose, xylose, fructose, and mannose increased initially, then disappeared. Glucose and inositol also increased initially, but later decreased to a constant level. Unknowns U₁ and U₃ increased for 16 weeks, while U₂ rapidly disappeared. Sucrose and maltose were detected for 12 weeks, then disappeared. However, based on dry root wt (mg/g), fructose, mannose, U₂, sucrose, maltose, and pentoses, but not ribose, decreased with age. Glucose, inositol, U₁, and U₃ decreased initially, but became constant with time. Glucose initially was the major component, but was replaced by U₁ with increase in age.

Quinone accumulation in tobacco infected with tobacco streak or tobacco ringspot viruses. R. E. HAMPTON & L. L. WANG (Univ. Ky., Lexington). Acetone powders of tobacco streak virus-(TSV) or tobacco ringspot virus-(TRSV) infected plants contained a protein-quinone complex which was absent in healthy plants. Acid or enzymatic hydrolyses of the complex contained caffeic acid, a hydrolysis product of chlorogenic acid. The relative concn of the complex was highest in inoculated and systemically infected leaves of TSV-infected plants. Similar leaves from TRSV-infected plants contained less of the complex. Leaves in which the symptoms of either virus were masked contained only traces of the complex. Although polyphenol oxidase and peroxidase increased following infection, there was no apparent correlation between the magnitude of the increase in enzyme activities at the time of harvest and the concn of quinone-protein complex.

Localization of crystals in diseased oats treated with uranyl salts. PENELOPE HANCHEY & H. WHEELER (Colo. State Univ., Fort Collins; Univ. Ky., Lexington). Uranyl salts (UO₂⁺⁺) were shown to suppress electrolyte leakage in oats (*Avena sativa*) treated with victorin, the patho-

toxic product of *Helminthosporium victoriae*. Ultrastructure investigations have been conducted to study the nature of the suppressive effect. Nine-day-old oat leaves were allowed to take up an aqueous solution containing 0.001 M uranyl acetate and either 10 units/ml of victorin or an equivalent amount of deactivated victorin at the rate of 0.1 ml of solution/500 mg of tissue. In leaves with uptakes of UO_2^{++} and deactivated victorin, electron-dense crystals were abundant on cell walls and near plasma membranes of cells in vascular bundles. Crystals occurred predominantly on cell walls in leaves treated with UO_2^{++} and victorin, and less deposition was found near plasma membranes. Fewer crystals were found intracellularly in leaves given either treatment, and were completely absent in leaves which had not been treated with UO_2^{++} . These results support a previous suggestion that the suppressive effect of uranyl salts on victorin-induced electrolyte leakage may be based on a reaction at the cell surface.

Alterations of chemical constituents of flue-cured tobacco as a result of infection with tobacco mosaic virus. G. E. HARMAN, G. V. GOODING, JR., & T. T. HEBERT (N.C. State Univ., Raleigh). Five cultivars of flue-cured type tobacco were inoculated at transplanting with tobacco mosaic virus (TMV) and grown under normal field conditions. After conventional flue-curing, the laminae tissue from TMV-infected and virus-free tobacco was analyzed for a number of constituents. Mean values for total N for the 5 cultivars increased from 2.4% of the dry wt in the uninfected tissue to 3.0% in the infected tissue. Starch decreased from 1.8 to 0.7%, and reducing sugar levels decreased from 9-13% to 7-9% in the infected tissue, as compared to the healthy tissue. Small but consistent increases were found in the Mg and Ca concn of diseased tissue. No consistent changes were found in the P, K, alkaloid, pectic, or phenolic contents. From this study, it appears that the only significant change resulting from TMV infection from the standpoint of the quality of the tobacco and the health of the consumer is in the increase in nitrogenous compounds.

Phytophthora and Pythium species from old growth forest sites in Kentucky, North Carolina, and Tennessee. FLOYD F. HENDRIX, JR., & W. A. CAMPBELL (Univ. Ga.; USDA Forest Service, Athens, Ga.). Although species of *Phytophthora* and *Pythium* are common to agricultural soils in the South and East, few attempts have been made to determine those native to the area. Old growth forest stands "representative of the original vegetational land cover" were located in Kentucky, North Carolina, and Tennessee, mainly in mountain coves. Soil samples from these areas were assayed for pythiaceae fungi by the apple technique and by two selective media. The fungi isolated are considered indigenous to these areas, and include: *Phytophthora heveae* from two stands, one in Tennessee and one in North Carolina; *P. cinnamomi* from one site in Tennessee; *Pythium sylvaticum*, *P. intermedium*, unidentified heterothallic isolates, and *P. irregulare* which were common to all areas; and *P. echinocarpon*, *P. heterothallicum*, *P. parocandrum*, and *P. vexans* from soils at two of four sites. The *Pythium* species at only one site included *P. helicandrum*, *P. middletoni*, *P. torulosum*, and *P. ultimum* var. *sporangiferum*. The common soil *Pythiums* such as *P. irregulare*, *P. vexans*, and the heterothallic species which are widespread in agricultural situations apparently are native to the areas sampled.

Disc-plate method for selective isolation of Rhizoctonia from soil. L. J. HERR (Ohio Agr. Res. Development Center, Wooster). A selective disc-plate method was devised for isolation of *Rhizoctonia* from soil in the field and in the presence of undisturbed living plants. Paper discs (6.35 mm diam) were soaked in a nutrient solution containing 5 ppm Cu from cupric sulfate, 5 ppm active benomyl, 100 ppm rose bengal, and 100 ppm streptomycin sulfate, then dried at 37 C. Discs were placed over holes (ca. 4 mm diam) drilled in aluminum plates (3 x 4 inches) in three rows of

six holes/row and covered with strips of vinyl tape. Sterilized disc-plates were inserted vertically into soil, incubated, and recovered. Discs were plated on water agar containing selective inhibitors for 2 days, then examined for *Rhizoctonia*. Pathogen-free soil was artificially infested with graded concn of *Rhizoctonia* inoculum and assayed by the disc-plate and pre-emergence seedling kill methods. Standard log dosage-probit response curves were fitted to the results. The disc-plate method exhibited greater sensitivity than the diseased seedling method. Best results were obtained with soil disc-plate incubations of 5 to 7 days.

Dodder transmission of a mycoplasma from ash trees with yellows-type symptoms. C. R. HIBBEN & B. WOLANSKI (Brooklyn Bot. Garden, Ossining, N.Y., Boyce Thompson Inst., Yonkers, N.Y.). By means of dodder (*Cuscuta subinclusa*), an agent was transmitted from witches'-broom diseased American ash (*Fraxinus americana*) to *Vinca rosea*, in which it caused symptoms typical of a yellows-type infection. Transmissibility was confirmed by graft transmission tests between yellowed and healthy *V. rosea*, and by dodder (*C. campestris*) transmission of the agent from *V. rosea* to *Daucus carota*. Sap transmission attempts failed. Thin sections of *F. americana* and *V. rosea* vein tissue, and of dodder strands connecting these hosts, were examined by electron microscopy. Virus particles were not seen in any sections. Typical mycoplasma-like bodies were present in large numbers in the phloem cells of all three species examined. Mycoplasma-like bodies were also found in thin sections of roots of *F. americana* and *V. rosea*. These bodies were morphologically indistinguishable from mycoplasmas known to cause yellows-type diseases in other plants. On the basis of these results, a mycoplasma etiology is suggested for the ash witches'-broom disease.

Some properties of papaya mosaic virus and its isolated constituents. E. HIEBERT (Univ. Fla., Gainesville). Papaya mosaic virus (PMV), a flexuous rod-shaped virus with a normal length of 533 m μ , was purified from papaya leaves by chloroform and *n*-butanol clarification followed by polyethylene glycol precipitation and differential centrifugation in neutral 0.1 M potassium phosphate buffer. Papaya mosaic virus has a sedimentation coefficient of 118.7 Svedberg units (S), an isoelectric point around pH 5.3, an absorbancy index at 260 m μ of 2.85 cm²/mg, and an ultraviolet absorption spectrum typical of nucleoprotein. Phenol extracts of virus yielded one sedimenting species of RNA which was infectious and had a sedimentation coefficient of 31.8 S. The nucleic acid has a base composition in mole per cent of 34.2% Ap, 20.3% Gp, 22.2% Up, and 23.3% Cp. Protein was isolated by either treatment with 67% glacial acetic acid or with 2.5 M guanidine HCl.

Purification and identification of alfalfa phytoalexin. V. J. HIGGINS, R. L. MILLAR, D. G. SMITH, & A. G. MCINNES (Univ. Toronto, Toronto, Ont., Cornell Univ., Ithaca, N. Y., Nat. Res. Council Can., Halifax, N. S.). Alfalfa phytoalexin was isolated from alfalfa (*Medicago sativa*) leaves by the drop-diffusate technique using spore suspensions of *Helminthosporium turcicum* as inoculum, and was purified by partitioning diffusates with CCl₄, followed by preparative Silica Gel-TLC using pentane-ethyl ether-acetic acid (75:25:1, v/v) as the developing phase. The phytoalexin was crystallized with chloroform and heptane as the solvent pair, giving a yield of approx 10 mg from 700 ml of diffusate. The phytoalexin was identified as (-)-3-hydroxy-9-methoxypterocarpan (demethylhomopterocarpan), a compound previously isolated from several tropical leguminous trees. The identification was made on the basis of the mass spectrum, proton magnetic resonance spectra of both the phytoalexin and its monoacetate, infrared and ultraviolet spectra, and the physical constants. We propose that this phytoalexin be assigned the trivial name "medicarpin."

Absorption and movement of benomyl into cotton bolls. R. B. HINE, L. J. ASHWORTH, JR., & J. L. MCMANS

(Univ. Ariz., Tucson; Univ. Calif., Berkeley; ARS, USDA, US Cotton Res. Sta., Shafter, Calif.). Peduncles of detached, immature cotton bolls in laboratory studies did not absorb and translocate sufficient concn of benomyl [methyl-1-(butylcarbamoyl)-2-benzimidazolecarbamate] from aqueous suspensions to protect locules from infection by *Aspergillus flavus*. The chemical was not detectable in carpal tissue, lint, seed coat, or embryo tissue by use of a bioassay method utilizing *Penicillium expansum* or in chloroform extracts. Benomyl was not detectable in interior boll tissues 1 and 3 weeks after foliage; nearly mature but green bolls were sprayed (2,000 ppm-active) to runoff in the field. Benomyl was readily detected in leaves, stems, and exterior boll walls. Benomyl formulated in glycerine, Volck oil, and Puregrow Supreme 90 oil (5%, v/v), and sprayed in the greenhouse on plants with squares but no flowers or bolls, was not detectable in boll tissue formed after application. Benomyl has not been detected in bolls from plants growing in soil containing as high as 500 ppm (active) on an air-dry, soil wt basis.

The effects of water potential on zoospore production in Aphanomyces euteiches. H. C. HOCH & J. E. MITCHELL (Univ. Wis., Madison). Osmotic and matric water potentials ($\psi\pi$, ψ_m) influenced the extent and quantity of zoospore differentiation in vitro in *Aphanomyces euteiches*. Typical zoospores were produced when 24-hr-old mycelia grown in peptone (2%)-glucose (0.5%) broth were washed for a period of 4 hr at 25 C with glucose, sucrose, mannose, sorbitol, fructose, and polyethylene glycol (PEG) 4,000 osmotic at $\psi\pi$ greater than -2 bars. The extrusion of primary spores was stopped at -3 bars. Differentiation within the hyphae proceeded until an $\psi\pi$ equal to -5 bars was imposed upon the hyphae. Potentials below -5 bars did not allow any detectable differentiation. When the osmoticum was replaced within 2 hr by one of greater potential, sporulation proceeded normally. Osmotica of mannitol, *i*-inositol, *i*-erythritol, and glycerol did not completely inhibit zoospore production until the $\psi\pi$ equalled -8 bars. In contrast to the $\psi\pi$ results, matric potentials higher than -0.01 bars were required for zoospore production with very little differentiation below -0.05 bars ψ_m . Mycelia immersed in PEG-20,000 solutions or separated from it by a dialysis membrane produced zoospores only at potentials higher than -0.05 bars, and prevented spore differentiation at potentials below -0.25 bars. This indicated PEG-20,000 exerted a ψ_m rather than an $\psi\pi$.

Phytophthora species involved in rhododendron wilt and sources of resistance. H. A. J. HOITINK & A. F. SCHMITT-HENNER (Ohio Agr. Res. Dev. Center, Wooster). *Phytophthora cactorum*, *P. cinnamomi*, *P. citricola*, and an unidentified *Phytophthora* sp. were isolated from wilted rhododendrons in Ohio. *Phytophthora cinnamomi*, *P. citricola*, and the *Phytophthora* sp. invade roots and move through the crown into the lower branches with subsequent wilting. All three are equally virulent. *Phytophthora cactorum* normally infects the upper portion of the plant, but will infect crowns under flooding conditions causing wilt. The *Phytophthora* sp. grows at temp from 6 to 37 C, and has nonpapillate sporangia and hyphal swellings, but no chlamydospores. It is heterothallic, and forms abundant oospores when paired with compatibility type A1 of *P. cryptogea* and *P. drechsleri*; thus, it has characteristics of both species. *Rhododendron maximum*, *R. carolinianum*, and 40 hybrids of *R. catawbiense* origin were susceptible to wilt when inoculated with *P. cinnamomi* in the greenhouse and watered with an automatic drip watering system. Hybrids, van der Hoop, D. A. Koster, and one strain of English Roseum were resistant. Another strain of English Roseum was susceptible. Roots of wilt-resistant varieties were rotted by *P. cinnamomi*, but crowns were not invaded. Plants did not wilt, as new roots developed at the soil surface from healthy crowns.

Electron microscopy of wheat affected by wheat spindle streak mosaic. G. R. HOOPER & M. V. WIESE (Mich. State Univ., E. Lansing). Leaf tissues of wheat (*Triticum*

aestivum 'Genesee') affected by wheat spindle streak mosaic (WSSM) were examined in the electron microscope. Sectioned preparations of affected plants contained viruslike rods, pinwheel and plate inclusions, and X-bodies in mesophyll, epidermal, and vascular parenchyma cells. The rod-shaped particles were 22-24 m μ in diam and of undetermined lengths in sectioned preparations. They occurred as cytoplasmic aggregates or as single particles spaced along pinwheel arms. Aggregates of plates occurred in diseased plants, and appeared to fragment to give rise to pinwheels. The plates were attached at one end to endoplasmic reticulum (ER) which when curved back on itself became the pinwheel core and the attached plates became the arms. X-bodies in affected plants were highly organized membranous structures composed of tubes or sacs with projections into the cytoplasm. The projections were usually coated with ribosomes similar to rough ER, and appeared to be associated with pinwheels. The membranous material in the X-bodies occasionally was sheetlike, with "loops" of membranes on both surfaces. X-bodies, pinwheels, and viruslike particles were especially abundant in cells from chlorotic or necrotic tissues. The organelles in such cells were severely disrupted.

Microflora responsible for volatile inhibitory factors in soil fungistasis. T. S. HORA & RALPH BAKER (Colo. State Univ., Fort Collins). Previous studies have demonstrated the presence of a volatile inhibitory factor associated with the phenomenon of soil fungistasis. To determine the soil microorganisms responsible for the production of such a factor, conidia of 10 fungi commonly found in soil were placed on water agar discs and suspended over sterilized soils infested with mixed soil bacteria, fungi, or actinomycetes isolated from natural soils. A reduction in spore germination of 30 to 100% was observed in 8 soils (pH 5.5-8.4) 15 days after infestation with actinomycetes when compared with the noninfested sterilized soils. Slight inhibition developed in soils infested with bacteria or fungi. Ten odoriferous *Streptomyces* spp. were cultured individually on glucose (2%)-peptone (0.5%) agar (2%) medium. Volatiles from these cultures decreased conidial germination. When some natural soils were incorporated with chitin (2,500 ppm), cellulose (1,000 ppm-C), or glucose (1,000 ppm-C), and infested with mixed *Streptomyces* spp., the germination of spores was reduced. No reduction in spore germination was evident when lignin (2,500 ppm) was added to soils. These observations suggest that soil actinomycetes produce a significant portion of volatiles responsible for the inhibition of conidial germination in soil.

Aerobiology of Fusarium spp. associated with stem rot of Dianthus caryophyllus. R. K. HORST, P. E. NELSON, & T. A. TOUSSOUN (Cornell Univ., Ithaca, N.Y., Pa. State Univ., Univ. Park). Fifteen-min exposures of petri dishes containing Nash's medium were made during a period of 6 months to air inside and outside commercial carnation greenhouses at Salinas, Calif. A variety of *Fusarium* spp. including *F. roseum*, *F. solani*, and *F. oxysporum* were collected. Similar clones of these fungi were also isolated from soil adjacent to the greenhouses, from dust particles lodged in polyethylene tubes of the air distribution system, and from dust in filters on fans located in the greenhouse ridges. Clones of *F. roseum*, *F. solani*, and *F. oxysporum* were regularly isolated when the medium was exposed 0, 21, or 49 inches above the soil surface inside carnation greenhouses in southeastern Pa. These fungi were also isolated from air outside these Pa. greenhouses and from wood fibers of filter pads used in the greenhouse cooling system. These results show that *Fusarium* propagules are widely dispersed through the air. We suggest that air dispersal of *Fusarium* spp. may be associated with dust particles. The importance of this phenomenon, which is not restricted to semiarid regions such as Calif., must be evaluated in the epidemiology of the disease.

Strawberry fruit rot caused by Colletotrichum fragariae. C. M. HOWARD (Univ. Fla., Strawberry & Vegetable Field

Lab., Dover). During recent years, a fruit rot of strawberry which has previously been attributed to *Gloeosporium* sp. has caused extensive losses in plantings of the cultivar Tioga in Florida. The disease is characterized by dark brown, circular, firm-rot lesions up to 2 cm in diam occurring anywhere on the fruit. The lesions sometimes bear buff-colored spore masses. Isolation from the lesions usually yields only a fungus which closely resembles *Colletotrichum fragariae*. *Colletotrichum fragariae* apparently has not previously been tested for pathogenicity to strawberry fruits. Spore suspensions from an isolate obtained from a fruit rot lesion and from a *C. fragariae* isolate were used to inoculate Tioga plants and ripe, pink, and green fruits. Plants inoculated with either isolate developed anthracnose symptoms. Two small drops of spore suspension were placed at different points on each surface-sterilized fruit. Within 5 days, typical rot lesions had developed at many of the points of inoculation with either isolate regardless of fruit maturity. The fungi were reisolated from all infected pink and green fruits. No lesions developed on control fruits. It thus appears that this fruit rot is caused by *C. fragariae*.

Alfalfa yields as influenced by air quality. R. K. HOWELL & D. F. KREMER (ARS, USDA, Beltsville, Md.). Eight alfalfa cultivars were seeded 1 May 1969 in 4-inch pots and cultured in carbon-filtered (CF) and in nonfiltered (NF) greenhouse air and in nonfiltered air outside of the greenhouse (O) during the summer of 1969 at the Plant Air Pollution Laboratory, Beltsville. Yields, as dry wt for each harvest date 8 August, 4 September, and 16 October, were obtained. The cultivars Lahontan, Peruvian, and Vernal produced equally well under CF and O air, but yields were reduced in NF as compared to those from CF air by 62%, 59%, and 33% for Lahontan, Peruvian, and Vernal, respectively. The varieties African, DuPuits, Grimm, Iroquois, and Williamsburg yielded 69%, 84%, 69%, 58%, and 81%, respectively, as much forage from plants in O as from plants in CF air; however, the yields of the same cultivars in NF air were only 38%, 45%, 48%, 36%, and 52% of the yields of those plants in CF air. Dry matter production of all cultivars was reduced by NF air as compared to O air. The data demonstrate the limitations of usefulness of plant growth under CF and NF air conditions as being an indication of air quality.

An evaluation of host reactions of Valencia peanut accessions grown at two locations to the blackhull disease fungus in vitro. D. C. HSI (N. Mex. State Univ., Clovis). A standard check and 137 accessions of Valencia type peanut, *Arachis hypogaea*, were evaluated for reactions to the fungus, *Thielaviopsis basicola*, which is the main organism causing blackhull disease in peanuts. The accessions were world collections mainly from South and Central America. They were grown in Puerto Rico in 1968 and in New Mexico in 1969. The evaluations were made on steamed unblemished pods in petri plates. The pods of individual accessions (three/plate) were moistened with sterile water and incubated at room temp for max development of transferred plugs from 7- to 10-day-old cultures. Rate of growth of the plug located in the center of each pod was measured at 1- to 2-day intervals for 10 days after inoculation. Average discoloration spread ranged from 35 to 100% of the pod surface in 1969, using the Puerto Rico-grown materials, and from 37 to 100% in 1970, using the New Mexico-grown materials. The color of the fungal growth varied from very light gray to intense black. Twenty-five introductions in 1969 and 27 in 1970 had less than 75% of the pod surface discolored by the fungal growth. Only 7 introductions showed less than 75% blackhull in both years.

Effect of pH on viral inoculations of monolayers of insect vector cells. H. T. HSU & L. M. BLACK (Univ. Ill., Urbana). The field variety of potato yellow dwarf virus transmitted by *Aceratagallia sanguinolenta* but not by *Agallia constricta* can infect monolayers of cells from both insects, although cells of the nonvector are less susceptible.

The opt pH of inoculum for infection of cells of both species is near 6.0. Another field variety of the virus with vector relationships the reverse of those above can also infect cells from both insect species; in this case, the opt pH of inoculum for cells of both species is near 5.3. The marked influence of pH on inoculation appears to be mediated through the virus and not through the host.

Sensitivity of fungal hyphae to soil fungistasis. S. C. HSU & J. L. LOCKWOOD (Mich. State Univ., E. Lansing). Hyphae of 20 different fungi were incubated on cellophane placed on natural Conover loam soil amended with different amounts of alfalfa extract. Differences in hyphal growth were determined after 4 hr. Analyses of dosage-response curves permitted determinations of hyphal sensitivity to soil fungistasis. Among the fungi, hyphal sensitivity to fungistasis was inversely correlated with growth rate on potato-dextrose agar (PDA) ($r=0.91$) and with hyphal diam ($r=0.82$). Hyphal diam was directly correlated with growth rate ($r=0.88$), and with spore volume ($r=0.84$). Fungistasis sensitivities of spores and hyphae of the same species were also correlated ($r=0.74$). Survival of hyphae of 10 fungi incubated on membrane filters on natural soil was determined by periodic transfer to acidified PDA. After 8 days, fewer than 10% of the hyphae of any of the fungi remained viable. The conidial portion of germinated spores of *Helminthosporium sativum*, *H. victoriae*, *Curvularia lunata*, *Fusarium solani* f. sp. *passi*, and *F. roseum* f. sp. *cerealis* 'Culmorum' survived longer than the hyphae themselves. When germinated conidia of *H. sativum* and *H. victoriae* were incubated alternately on soil and acidified PDA, the original conidia were able to regerminate at least 5 times on acidified PDA following complete lysis of hyphae on soil.

Fomes annosus conidia on Ponderosa pine. R. S. HUNT & F. W. COBB, JR. (Univ. Calif., Berkeley). Selectively cut old growth Ponderosa pine (*Pinus ponderosa*) stumps were inoculated with *Fomes annosus*. Bark shrinkage of these stumps was such that many tubular chambers were formed behind the bark due to parenchyma disintegration. Within such chambers and adjoining bark beetle (*Dendroctonus valens*) galleries, the *Oedocephalum* stage of the fungus was found in the field on about 50% of the infected stumps. Dye solutions poured on the tops of such stumps indicated the possibility that rain could wash conidia down to the roots before the fungus mycelium reached the roots. *Fomes annosus* mycelium and *D. valens* were found together infrequently. When this did occur, the beetles seemed to be dead or dying. Attempted isolations from living *D. valens* taken from the stumps indicated their potential as vectors for *F. annosus* to be low.

Comparative concentration of maize dwarf mosaic virus in corn. R. K. JONES & S. A. TOLIN (Va. Polytechnic Inst. State Univ., Blacksburg). The ability of corn to support the synthesis and accumulation of maize dwarf mosaic virus (MDMV) under various conditions was compared. As little as 2 g corn leaf tissue from which the midrib had been removed was homogenized in 0.1 M sodium citrate and 0.5% 2-mercaptoethanol. After chloroform clarification, the sap was analyzed by sucrose density-gradient centrifugation and photometric scanning. The area under the virus peak was converted to mg MDMV/g fresh wt. The validity of this method was confirmed in initial tests by systemic infectivity assays. Age of the plants at inoculation and sampling, leaf position, cultivar, and host nutrition all affected MDMV content. In greenhouse tests, susceptible Hy × C103 and DeKalb 805A, and field-resistant T8 × Oh7B, contained 0.01-0.03 mg/g 10 to 14 days after inoculation, and less thereafter. The similarity in virus content of the three hybrids indicates that the mechanism of resistance of T8 × Oh7B to MDMV does not involve virus synthesis. Field-grown Hy × C103 sampled over a 2 to 3 month period gave about 5 times more virus than the highest amount it gave in the greenhouse. The higher MDMV content in field-grown corn was prob-

ably the result of more favorable nutrient levels and other environmental conditions.

Induction of Verticillium wilt symptoms in cotton cuttings by lipopolysaccharide but not by endopolygalacturonase from Verticillium albo-atrum. N. T. KEEN, M-C. WANG, & M. LONG (Univ. Calif., Riverside). Cell-free, dialyzed fluids from log phase conidial cultures of *V. albo-atrum* produced interveinal flaccidity on excised cotton leaves which was similar to that caused by fungus infection of intact plants. In attempts to identify the active factor(s), chromatographically, ultracentrifugally, and electrophoretically homogeneous preparations of endopolygalacturonase (endo-PG) were isolated from culture fluids. The enzyme had a molecular wt of 30,000, was rich in basic amino acids, and liberated galacturonic acid equivalents from sodium polypectate at 2,150 μ moles/min per mg protein. The purified endo-PG had no visible effects, however, on cotton cuttings or excised leaves at concn up to 100 μ g/ml. Typical *Verticillium* wilt symptoms were produced when cuttings or excised leaves were placed in 50 μ g/ml or higher concn of a lipopolysaccharide (LPS) isolated from log phase cultures. The LPS had a mol wt between 1.5 and 15 million, and contained equal amounts of lipid, protein, and polysaccharide. It accumulated to about 0.75 mg/ml in log phase cultures. The LPS produced severe wilt symptoms on the wilt-susceptible cotton cultivar Delta Pine Smooth Leaf, intermediate symptoms on the moderately susceptible cultivar SJ-1, and mild symptoms on the resistant cultivar Seabrook Sea Island.

Role of oil sprays in production of aerial strands by Erwinia amylovora. H. L. KEIL & T. VAN DER ZWET (ARS, USDA, Beltsville, Md.). *Erwinia amylovora* produced abundant aerial strands on Bartlett pear trees inoculated in the greenhouse and sprayed with 1 to 2% commercial Superior '70' oil 1 to 4 days prior to or after inoculation. Few short (2-12 mm) strands were observed on succulent shoots of unsprayed, inoculated trees, whereas sprayed trees appeared as if wrapped with long cottony fibers. Strand production was most abundant on leaf petioles and lower surfaces of mid ribs. Strands on sprayed trees ranged from 30-75 mm long and 12-36 μ (avg 19 μ) in diam. They were easily detached from the plants by air currents, and disintegrated in high relative humidity or free water. Strands maintained their shape, however, when mounted in undiluted '70' oil and shattered like glass when pressed between two hard surfaces. Several short pieces (2-6 mm) of strands, placed in a water drop in the leaf axils of greenhouse-grown Bartlett and Jonathan trees, caused more infection when the stem tissue was injured by needle puncture through the drop than noninjured tissue. Bacterial cells in the strands remained viable and virulent after 3 months' storage in the laboratory. The use of oil as a pesticide may aid in production of bacterial strands, especially in the drier regions, which could play a significant role in the dissemination of fire blight.

Relationship between motility and loss of virulence in Pseudomonas solanacearum. A. KELMAN & J. HRUSCHKA (Univ. Wis., Madison). Virulence in cultures of *P. solanacearum* was lost in nonaerated but not in aerated broth media containing 1% glucose. Avirulent, butyrous colony-type variants appeared and increased rapidly in nonaerated media after 96 hr. Cells of fluidal, virulent isolates were usually nonmotile and nonflagellate when grown in tryptone-yeast extract-glucose broth (TYG) or on peptone-casamino acids-glucose agar (PCG). Conversely, cells of avirulent, butyrous colony variants were highly motile in TYG and on PCG plates. In a semisolid motility medium (0.01% tryptone, 0.01% glucose, and 0.35% agar), migration distances of avirulent cells away from a central point at which ca. 1×10^8 cells were placed were significantly greater than those of virulent cells. Aerotaxis of avirulent but not of virulent cells was demonstrated in the motility agar in 5-cm glass tubes. Nonmotile virulent cells grown on PCG agar remained nonmotile when transferred to a

liquid medium with 1% glucose; in contrast, they became actively motile within 2 hr if the medium contained 0.01% glucose. Loss in virulence in culture appears to be related to the high motility and capacity for aerotaxis of avirulent cells and the absence of motility of virulent cells at glucose levels of 1% or above.

Photosynthetic efficiency of soybean leaves affected by toxins produced in vivo by Pseudomonas glycinea. B. W. KENNEDY & V. CARDWELL (Univ. Minn., St. Paul). Systemic toxemia is the apparent cause of a chlorosis on bacterium-free trifoliolate leaves of soybean resulting from an earlier infection on the leaf immediately below. Photosynthesis in young unfolding leaflets was estimated by analyzing for net CO_2 uptake after they were enclosed in microchambers that were attached to an infrared gas analyzer. Affected leaflets were reduced in both area and photosynthetic capability per unit area by 45 to 55%. After 2 days, photosynthesis was reduced by about 10% and leaf area 25 to 30%. Leaves returned to normal in most cases after 5 to 7 days, recovery of photosynthetic ability being faster than recovery to normal size.

Variation among Rhizoctonia bataticola isolates from Urid bean plant parts and soil. M. N. KHARE, N. K. JAIN, & H. C. SHARMA (J. Nehru Krishi Vishwa Vidyalaya, Jabalpur, India). *Rhizoctonia bataticola* was found associated with root, stem, leaf, pod, and seeds of Urid bean (*Phaseolus mungo*). Six isolates from various plant parts and soil exhibited differences in virulence. The soil isolate proved most pathogenic, followed by the pod and root isolates. In vitro morphological studies at pH 6.5 and 25 C revealed differences in growth patterns and sclerotia size. When grown on differential media (potato-dextrose agar without or with extracts from soil, seed, root, stem, leaf tissue, and oatmeal agar), all six isolates differed from each other in their growth patterns and rate of growth. The soil isolate had the least amount of growth in almost all media, and the pod isolate, the most. The leaf isolate developed the largest sclerotia (96 \times 86 μ) and the soil isolate the smallest (79 \times 63 μ) after 72 hr. The isolates varied in their growth rates at different temp and pH levels. The best pH for growth of all isolates was 5.5. The variation among the isolates may be due to various biochemical processes in the plant parts.

The occurrence of virus particles in tubules in leaf cells of systemic and local lesion hosts infected with bean pod mottle virus. K. S. KIM & J. P. FULTON (Univ. Ark., Fayetteville). Viruslike particles were readily observed within tubules of leaf cells infected with bean pod mottle virus (BPMV). In a local lesion host, *Phaseolus vulgaris* 'Pinto', particles were in a single straight line within tubules which occurred commonly between the cell wall and the plasmalemma. In a systemic host, *P. vulgaris* 'Cherokee Wax', similar tubules with particles were present but appeared to be within the cell wall, often associated with abnormal cell wall protrusions. In both hosts, the tubules with particles were noted in plasmodesmata. The tubule consists of an outer unit membrane, an inner layer of material, and the line of particles. Tubules were 60 to 70 μ in diam. The tubules did not extend into the cytoplasm in either of these hosts.

Production of extracellular H₂O₂ and peroxidase by wood-rotting basidiomycetes. J. W. KOENIGS (Forest. Sci. Lab., Research Triangle Park, N.C.). Formation of extracellular H_2O_2 by wood-rotting fungi has been postulated by others but not demonstrated. Since H_2O_2 , alone or with catalytic amounts of certain metals, can attack cellulose and hydroxylate phenols, evidence of its formation was of interest. Furthermore, H_2O_2 and extracellular peroxidase (P), if the latter were also produced, comprise a system which could oxidize the phenolic groups in lignin. Extracellular H_2O_2 was detected by growing fungi on a heated blood-malt extract medium and by measuring catalase

inactivation by aminotriazole. Peroxidase was assayed spectrophotometrically in culture filtrates. After 1 week, all but 5 of 31 species of brown (B) and white (W) rot fungi produced H_2O_2 ; by 2 weeks, 4 of the 5 produced some. None of 9 B, but 10 of 22 W, produced P. Eight W produced H_2O_2 and P, but only *Fomes pini* produced sufficient H_2O_2 and P to confound interpretation of the Bavendamm reaction. That nearly all B produced H_2O_2 suggests a possible role in cellulose decomposition. Since no B, but some W, produced P and H_2O_2 , this system may attack lignin; polyphenol oxidase from W could furnish more H_2O_2 , thus coupling the two enzymes.

The effects of sugarcane mosaic virus (strain H) and Pythium graminicola, singly and in combination, on growth of sugarcane. HIDEO KOIKE & SHAW-MING YANG (ARS, USDA, Houma, La.). In replicated greenhouse tests, the interaction between sugarcane mosaic virus strain H, the most prevalent strain found in Louisiana, and the root rot fungus *Pythium graminicola* isolated from roots of sugarcane grown in field soil, was studied on four commercial sugarcane cultivars. The virus and fungus each caused significant reductions in fresh wt of top growth (in three and four varieties, respectively), dry wt of top growth (in all varieties), and total height of shoot per pot (in three and four varieties, respectively), when compared to noninoculated controls. The fungus reduced growth significantly more than the virus in some cultivars. When both pathogens were present, an additive effect was noted; reductions in fresh wt of top growth (in two varieties), dry weight of top growth (in three varieties), and total height of shoots per pot (in three varieties) exceeded those caused by either pathogen alone. The results suggest that under field conditions, greater reductions in growth may result from the presence of both pathogens.

Cowpea chlorotic mottle virus hypersensitive reaction in soybean inhibited by 2-thiouracil. C. W. KUHN (Univ. Ga., Athens). Treatment with 2-thiouracil (TU) caused cowpea chlorotic mottle virus (CCMV) lesion area on hypersensitive soybean to be enlarged 8 to 75 times. Comparisons were made with paired half leaves floated on TU ($0.5 \times 10^{-3} M$) or water. Lesion enlargement was detected 48 hr after inoculation, and increased rapidly for 6 to 8 days. Although soybean becomes more resistant (fewer and smaller lesions) with increasing age, TU caused lesions to reach a similar size regardless of host age. Compared to controls, lesion area increased 2, 13, and 4 times at 21, 27, and 32 C, respectively. Increased lesion area was noted when TU treatment began at 0, 12, and 24 hr after inoculation, but not at 48 hr. The full effect, however, occurred when treatment began between 0 and 12 hr. When lesion area was increased 10 to 13 times, the relative infectivity was increased 33 to 38 times. The increase in lesion size could be prevented when uracil ($10^{-3} M$) was added to TU ($10^{-4} M$). 2-Thiouracil treatment caused CCMV to move from one half-leaf to another, but no systemic movement from one leaf to another was detected. Cowpea chlorotic mottle virus multiplication was enhanced 3 times, whereas tobacco mosaic virus was inhibited 7 times, by TU in *Chenopodium amaranticolor*, a local lesion host for both viruses.

Preliminary characterization of virus inhibitors in aphids. P. E. KYRIAKOPOULOU & C. E. YARWOOD (Univ. Calif., Berkeley). The failure to recover a virus from viruliferous aphids by mechanical inoculation is usually attributed to virus inhibitors in the aphids. When *Myzus persicae*, which had fed on purified tobacco mosaic virus (TMV) preparations, were ground in 1% K_2HPO_4 , 2% magnesium trisilicate, and 0.2% activated charcoal, the suspension was infective for *Phaseolus vulgaris*. The high speed (73,449 g, 60 min) supernatant of *M. persicae* (12 to 15 days old, reared at 15 C on healthy *Raphanus sativus*) homogenate was more inhibitory than the pellet. Upon successive re-

suspensions and high speed centrifugations of the pellet, the inhibitory principle(s) was progressively but incompletely removed from the pellet into the supernatant. The inhibitory activity of the supernatant was abolished by heating for 30 min at 90 C. Chromatography of the supernatant on Sephadex G-100 column revealed two inhibitory fractions, the first narrow and the second extending over several aliquots. Their absorption peak at 280 m μ and their reaction with biuret indicate that the two inhibitory fractions are proteins. The first inhibitory fraction appears to have a mol wt between 100,000 (exclusion limit of Sephadex G-100) and above 67,000 (mol wt of bovine albumin), and the second between 67,000 and 13,000 (mol wt of cytochrome c).

Visualization of maize dwarf mosaic virus in corn leaf tissue. W. G. LANGENBERG & H. F. SCHROEDER (ARS, USDA, Univ. Neb., Lincoln). Maize dwarf mosaic virus could be readily and consistently visualized in sweetcorn leaf-tissue thin-sections if the fixative contained one of several enzyme inhibitors or thioglycolate. Fixation in phosphate buffered 6% glutaraldehyde followed by 1% osmic acid, or in a modified Randolph's fixative, were equally effective when an enzyme inhibitor or thioglycolate was present during all preparatory steps until dehydration of the tissue. When the modified Randolph's fixative at pH 7.0 was used alone, the virus often had a "washed out" appearance. Sodium thioglycolate (0.2%) and the inhibitors 1% potassium polyvinylsulfate, 0.01 M diethyl pyrocarbonate, and 0.01 M iodoacetate helped preserve virus when used with either fixative. Maize dwarf mosaic virus was present in sections as band-shaped inclusions, sheetlike mats of virus, and individual particles in the cytoplasm of all cell types of the maize leaf. Chloroplasts, nuclei, and mitochondria were free of virus inclusions. Tubular and pinwheel type inclusions were commonly present in tissue fixed in the presence or absence of the enzyme inhibitors, but bands and sheets of virus were absent if the tissue was fixed in fixative only.

Evaluation of postharvest-prestorage fungicidal treatments for the control of Fusarium tuber rot of potatoes. S. S. LEACH (ARS, USDA, Presque Isle, Maine). Three experiments, each with four replicates, were conducted during the past 2 years to evaluate various fungicidal dusts and dips for the control of *Fusarium* tuber rot. Whole tubers were wounded with a vegetable grater and immediately inoculated with a spore suspension prepared from 4-week-old cultures of *Fusarium roseum* 'Sambucinum'. Twenty-four hr later, lots of 50 inoculated tubers were placed in wire mesh bags and treated with either a fungicidal dust or a 2-min fungicidal dip. Dusts were applied at 1 lb./cwt and dips at 2 lb./100 gal H_2O formulated materials. After treating, the inoculated tubers were placed throughout a bin of bulk stored potatoes, to simulate natural storage conditions, and stored for 5 months at 6 to 8 C. In these studies, the use of Daconil 2787 (tetrachloroisophthalonitrile) resulted in 93% control in 1968 and 50% in 1969; Polyram [complex of zinc ethylenebis (dithiocarbamate) and polyethylene thiuramdisulfide], 71% and 100%; and Dithane M-45 [zinc ion-manganous ethylenebis (dithiocarbamate) coordination product], 88% and 100%, respectively. Thiabendazole [2-(4'-Thiazolyl)-benzimidazole] and benomyl [methyl-1-(butylcarbamoyl)-2-benzimidazole carbamate] resulted in 100% control in the two tests in which they were included.

The effect of races of Heterodera glycines on nodulation and nitrogen fixation in soybean. P. S. LEHMAN, K. R. BARKER, & D. HUISINGH (N.C. State Univ., Raleigh). Soybeans infected with *Heterodera glycines* from Wilmington, North Carolina (race 1), show severe nitrogen deficiency symptoms, whereas plants infected with many other races do not show these symptoms. Using initial inoculum densities of 100, 200, and 400 yellow cysts/10-cm pot, *H. glycines* from North Carolina (race 1), Virginia (race 2), and Arkansas (race 4) were compared in silica sand for

their effect upon N_2 fixation in Lee soybeans at 55 days after inoculation. The effect of race 1 on N_2 fixation was quantitatively determined at 25, 40, 60, and 80 days. Nitrogen fixation was measured with an acetylene-ethylene gas chromatographic assay. Compared to noninoculated plants, race 1 reduced nodulation and N_2 fixation at all inoculum densities; whereas race 2 and race 4 caused no significant reduction in nodulation except at the highest inoculum density. When assayed at 25, 40, 60, and 80 days, high population densities of race 1 caused a severe reduction in nodulation and total N_2 fixed/plant. However, nematode infection resulted in increased nodular efficiency of the few nodules present; i.e., an increase in N_2 fixed/mg fresh wt of nodule.

Effect of mineral amendments on Aphanomyces root rot of peas. J. A. LEWIS (ARS, USDA, Beltsville, Md.). In an attempt to reduce root rot of peas caused by *Aphanomyces euteiches*, various minerals were added to soil infested with the pathogen in the greenhouse. Water-soluble salts of Al, Ca, Cu, and Zn at an element concn of 100 ppm reduced root rot more than 80% with no inhibitory effect on pea emergence. Salts of Mo, B, Co, Ba, Mg, or Mn were not as effective in controlling the disease, and reduced pea emergence. Addition to soil of water-insoluble salts of Al, Ca, Cu, or Zn did not reduce root rot. Disease severity was not appreciably reduced when chelates of Ca, Cu, or Zn were added to soil at an element concn of 100 ppm. A complete NPK fertilizer (20-20-20) had no effect on the control obtained with Al, Ca, Cu, or Zn. Three of the cations effective in reducing disease also had a detrimental effect on various phases in the life cycle of the pathogen. Growth of *A. euteiches* in a liquid medium was prevented by 5 ppm Al and Cu and 100 ppm Zn, whereas Ca even at 300 ppm did not inhibit growth. Zoospore formation was prevented in the presence of 1 ppm Cu and 10 ppm Al or Zn. Ca at 100 ppm inhibited zoospore formation by 50%. Al and Cu at 20 ppm completely inhibited zoospore germination, but 100 ppm of Ca and Zn only slightly inhibited germination.

Biosynthesis of F-2 (zearalenone) by Fusarium roseum 'Graminearum'. J. R. LIEBERMAN & C. J. MIROCHA (Univ. Minn., St. Paul). Studies of F-2 (zearalenone) biosynthesis have been complicated by the unavailability of a defined medium on which the estrogen F-2 can be produced in amounts comparable to those obtained on moist, autoclaved grain. Moreover, high moisture contents of media interfere with accumulation of F-2. Ordinarily, F-2 is produced by incubating *Fusarium*-infested grain at 15 C or less for 2 to 6 months; yields can be increased by raising the temp to 27 C for the last few days of incubation. Six cultures of *F. roseum* 'Graminearum' grown on moist, autoclaved rice for 2 weeks at room temp and incubated for 7 weeks at 10-13 C were combined, and aliquots were exposed to 1,000 ppm of aq acetate, malonate, shikimate, combinations thereof, beta-methyl crotonate, glucose, or mevalolactone. After 2 days of incubation at 27 C, the biomass which had been treated with mevalolactone yielded 699 ppm F-2, whereas the yields from the other treatments ranged from 367 to 524 ppm. Mevalolactone thus appears to be a precursor in the biosynthesis of F-2.

Loss of rice yield caused by stem rot. G. D. LINDBERG (La. State Univ., Baton Rouge). An epidemic of stem rot caused by *Leptosphaeria salvinii* developed in the rice crop in Louisiana in 1969. Infected lower nodes rotted, causing extensive leaf kill and premature panicle ripening. Panicles were light in wt, and grain quality was poor. Sclerotia embedded in rotted nodes permitted positive identification of the fungus. Eight cultivars in replicated plots were scored according to the amount of leaf kill and severity of nodal infections and lodging. Bluebonnet, Nova 66, and La Crosse were most resistant, Saturn, Belle Patna, and Bluebelle most susceptible, and Nato and Dawn showed

intermediate reactions. Large, dark lesions developed on inoculated rice. Many inoculated stalks were killed and the disease spread to noninoculated tillers. Leaf kill was extensive and panicles were smaller and lighter in wt than were the healthy controls.

Transpiration of corn seedlings infected with maize dwarf mosaic virus. D. W. LINDSEY, R. T. GUDAUSKAS, & BETTY L. KLEPPER (Auburn Univ., Auburn, Ala.). Effect of maize dwarf mosaic virus (MDMV) infection on transpiration of *Zea mays* 'Hy X C103' was measured indirectly by wt loss of soil-grown plants in pots enclosed in waterproof plastic bags. Seedlings (two-leaf stage) were inoculated by rubbing with MDMV-containing corn sap; one set of control plants was rubbed with healthy sap, an additional set was untreated. All rubbed plants showed increased water loss 1 day after inoculation, but little difference in water loss between rubbed and untreated plants occurred 2 to 4 days after inoculation. On the 5th day, MDMV symptoms appeared and transpiration of infected plants was 18 and 35% less than healthy-rubbed and untreated plants, respectively. Transpiration of infected plants was reduced 22 to 36% over days 6 to 8. Nine days after inoculation, all noninfected plants were wilted; MDMV-infected plants were turgid. Average wt loss over 9 days was 75.2 g for infected plants, 90.4 g for healthy-rubbed, and 96.6 g for untreated. Reduced water loss was associated with reduction of stomatal apertures in infected leaves.

Ultrastructure of Melampsora lini infection in flax. L. J. LITTLEFIELD & C. E. BRACKER (Purdue Univ., Lafayette, Ind.). Electron microscopy of compatible flax rust infection sites showed invagination and continuity of host plasma membrane around haustoria of the pathogen. This invaginated membrane showed the same ultrastructural characteristics as plasma membrane against the host wall when treated with ordinary electron stains, but the invaginated portion showed different staining properties in response to a periodate-phosphotungstate-chromic acid staining procedure. The fungal cell wall was continuous between the haustorial mother cell and the haustorium, although the wall of the haustorial neck often had a densely staining ring midway between the host wall and the haustorium. No pores were observed in the haustorial wall. The host wall stained differently in advance of the fungal penetration peg, and a collar of host wall material was deposited at many penetration sites around the base of the haustorial neck. Rarely, the entire haustorium was engulfed by elaborations of collar material. Normally, the haustorium was enclosed by an amorphous sheath between the haustorial wall and the invaginated host plasma membrane. The sheath material was not continuous with the collar. Host endoplasmic reticulum occurred in the vicinity of the haustorial sheath, whereas host dictyosomes were often associated with the collar.

Antigenic and immunodiffusional analysis of Erwinia carotovora and Erwinia atroseptica. S. C. Y. LIU (E. Mich. Univ., Ypsilanti). Antisera against *Erwinia carotovora* and *E. atroseptica* were produced in rabbits by immunization with sonicated cellular extracts. Antisera thus obtained contained precipitating and agglutinating antibodies to both sonicated and nonsonicated cells. With sonicated cells, the antisera gave titers of 1/640 and 1/420 to *E. carotovora* and *E. atroseptica*, respectively. Cross reactivity was observed in heterologous antigenic systems. Heating cellular extracts at 56 C for 30 min considerably reduced their serological reactivity when used as antigen. Immunodiffusional studies were done in Ouchterlony plates, with sonicated antigens. In homologous systems, four specific precipitin bands were observed with *E. carotovora*, whereas three bands were formed with *E. atroseptica*. In heterologous systems, one of these bands showed identity and the other bands indicated a range of nonidentity to partial identity. Heating sonicated cells at 56 C for 30 min removed the innermost band of *E. carotovora*, and also caused the others to be coalesced. Reciprocal cross absorption was

done in the cold; then absorbed sera were tested for cross reactivity. In homologous systems, antigenic components were removed, whereas in heterologous systems the absorption reduced the number of bands formed. Antigenic and immunodiffusional studies indicated that the two bacteria had antigenic components in common.

Suppression of rot on cranberry fruit by nitrogen storage. C. L. LOCKHART (Can. Dep. Agr., Kentville, Nova Scotia). At 3 C, there was less fungal rot in cranberries stored in N₂ than in air. At 3 weeks, cranberries in air and in N₂ had 16.3 and 3.8% rot, respectively. At 6 weeks in air and in N₂, there was 43.2 and 11.4% rot, respectively. A comparison of 3 weeks in air followed by 3 weeks in N₂ and 3 weeks in N₂ followed by 3 weeks in air gave 33.1 and 17.6% rot, respectively. There was little or no increase in rot in cranberries from these treatments stored in N₂ and in air for an additional 3 to 6 weeks. Sterile breakdown was more prevalent in cranberries stored in N₂ than those stored in air.

A selective medium for the assay of Botrytis allii in organic and mineral soils. J. W. LORBEER & G. M. TICHELAR (Cornell Univ., Ithaca, N.Y., Inst. Phytopathol. Res., Wageningen, The Netherlands). Populations of *Botrytis allii* in New York organic soils cropped to onion (*Allium cepa*) or artificially infested with the fungus were assayed by the soil dilution and plate count procedure, using a selective medium consisting of Martin's rose bengal agar amended with 50 or 100 ppm PCNB (pentachloronitrobenzene), 1 ppm Dithane M-45 (zinc ion and manganese ethylene bisdithiocarbamate coordination product), 1 ppm Polyram (active ingredients are a mixture of 83.9% ammoniates of ethylenebis[dithiocarbamate] zinc and 16.1% ethylenebis [dithiocarbamic acid] bi- and trimolecular cyclic anhydro- and disulfides), and 2 ppm aureomycin (chlorotetracycline HCl). The complete medium was necessary for assay of *B. allii* in New York organic soils, but in mineral soils of The Netherlands the fungus was satisfactorily assayed when Polyram and Dithane M-45 were omitted and PCNB was used at 100 ppm. In the complete medium, growth of the fungus was somewhat suppressed but sporulation was not. Uniformly distributing the soil suspension over the agar surface resulted in a more sensitive assay than did mixing it with the melted medium. It was necessary to increase the concn of the two antibiotics in the medium for satisfactory assay of the fungus in certain organic soils with high populations of bacteria.

Evidence for strains of maize dwarf mosaic virus in southern Ohio. R. LOUIE & J. K. KNOKE (ARS, USDA, OARDC, Wooster, Ohio). Four dissimilar virus isolates from southern Ohio were recovered by bioassays of corn leaves with uncommon maize dwarf mosaic (MDM) symptoms. In greenhouse tests, these isolates continued to induce atypical symptoms on Oh28 and N20 corn lines. Common mosaic, chevron, ring, mild mosaic, or fleck symptoms which differentiated MDMV strain A (MDMV-A), and each of the four isolates, respectively, were most obvious on N20. Single local lesions induced by MDMV-A and each isolate on inoculated leaves of Pa32 and IIIA were subcultured 8 times to obtain pure cultures. Similarities in host range, serological reactions, and morphological and transmission characteristics indicated the isolates to be related to MDMV-A. Each isolate caused systemic infection in *Sorghum halepense* and reacted with MDMV-A antiserum cross absorbed with 13B (MDMV-B) but not with 13B antiserum cross absorbed with MDMV-A. MDMV-A and each of the four isolates, however, were distinguished by their reactions on corn lines CG1, C.I.44, E38-11-11-5, Ky61:2335, Ky66:2500, Ky226, M14, Mo18W, N6J, N7B, Oh41, Oh514, Pa405, and Va35. Differences in symptoms, physical properties, and reactions of corn lines indicated that the four isolates are sufficiently distinct from

each other and MDMV-A to be designated MDMV-C, MDMV-D, MDMV-E, and MDMV-F.

Certain biochemical comparisons in wheat stem rust disease reactions controlled either at the Sr6 or Sr11 loci. P. LUDDEN & J. M. DALY (Univ. Neb., Lincoln). Previous studies of the temp-sensitive resistance imparted by the Sr6 allele for resistance of wheat to race 56 of *Puccinia graminis tritici* have indicated that there is no net synthesis of phenolic compounds. Although peroxidase activity does increase, it does not appear to be causally related to resistance. It is possible that the observed effects are unique to the Sr6 allele. Using the same techniques, isogenic lines of wheat carrying the Sr11 alleles for resistance and susceptibility were analyzed for total phenolic compounds, total peroxidase activity, and isozyme patterns. As with the Sr6 alleles, phenolic compounds did not increase significantly in either resistant or susceptible reactions. Total peroxidase increased in resistant reactions. The same isozyme found in the case of resistance due to the Sr6 locus appears to be associated with resistance controlled at the Sr11 locus. In agreement with the genetic evidence for different mechanisms for expression of resistance, the Sr11 allele is not influenced by temp or ethylene as is the Sr6 allele. Thus, peroxidase activity appears to be a consequence of nonspecific incompatibility caused by a prior biochemical event in resistant reactions.

Polygalacturonase production by Sclerotinia sclerotiorum in young cultures and in bean tissue during the early stages of pathogenesis. R. D. LUMSDEN & ROBERTA L. DOW (ARS, USDA, Beltsville, Md.). Endopolygalacturonase (endoPG) was detected in 100- μ sections of *Sclerotinia sclerotiorum*-infected bean hypocotyls. Sections were placed on 1.0% sodium pectate (NaP) medium that contained 0.5% ammonium oxalate and 0.01% thiomersalate. After 24 hr at 30 C, opaque halos developed at sites of enzyme activity when the plates were flooded with 5 N HCl. EndoPG was most active in sections taken from 1- to 3-cm lesions on hypocotyls harvested 24-48 hr after inoculation. After 48 hr, or when the lesions had advanced further than 3 cm, enzyme activity was variable and appeared to be present only at the tips of the lesion margins. EndoPG was not detected in older portions of these lesions, nor in tissue completely invaded by the fungus. Exopolygalacturonase was present as early as 24 hr after inoculation, and was most abundant after 72 hr, when mycelial growth of the fungus was evident on the hypocotyls. Two polygalacturonases were present in cultures of *S. sclerotiorum* grown in a basal medium with 1.0% NaP as the substrate. EndoPG activity reached a max after 2 to 3 days, and had an apparent pH optimum of 5.0. A greater second peak of enzyme activity had an apparent pH optimum at pH 4.5, and was most active at the time of greatest mycelial growth.

Control of starch accumulation by ADP-glucose pyrophosphorylase in wheat leaves infected with Puccinia striiformis. P. W. MACDONALD & G. A. STROBEL (Mont. State Univ., Bozeman). Starch content of wheat leaves inoculated with stripe rust decreased during flecking, increased during sporulation to twice that of healthy leaves, and decreased thereafter. Starch granules in chloroplasts adjacent to fungal hyphae 12 days after inoculation were observed by electron microscopy. Sugar phosphates, ATP, and inorganic phosphate (Pi) were measured during the infection process. In vitro activity of partially purified ADP-glucose pyrophosphorylase (ADP-PP) from healthy leaves could be correlated with the pattern of starch accumulation in diseased leaves during the infection process. The accumulation of starch could be explained on the basis of regulation of ADP-PP by changes in metabolite levels at critical times during the infection process. The reduced level of Pi 10 to 11 days after inoculation could depress ADP-PP, allowing starch synthesis to occur. The decrease in starch content of diseased leaves at 12 to 14 days after

inoculation may result from increased levels of Pi which inhibit starch biosynthesis, and to activity of starch-degradative enzymes.

Uptake and distribution of Fusarium spores in Fusarium wilt-susceptible or resistant tomatoes. M. E. MACE & J. A. VECH (ARS, USDA, Beltsville, Md.). Studies on the uptake of *Fusarium oxysporum* f. sp. *lycopersici*, race 1, spores and their subsequent distribution and development into mycelium within tomato plants indicate that resistance to *Fusarium* wilt may be attributed to the inhibition of spore germination or growth in resistant tomato stems. Microconidia were introduced directly into the vascular system through a transverse cut in the lower hypocotyl of both susceptible and resistant 30-day-old tomato plants. The rate of spore uptake and distribution in susceptible and resistant plants was the same. After a 15-min inoculation period, the spores were transported several cm into the stem. Longer inoculation periods did not proportionally increase the transport distance. Similarly inoculated plants were allowed to incubate for various times, then examined for the presence of mycelium. Extensive fungus colonization was found in the susceptible stems; only slight colonization occurred in resistant stems. It appears that the induction of host morphological responses are of minor importance in restricting the systemic distribution of the pathogen in either host. Factors that may account for the inhibition of spore germination or growth are currently under investigation.

Weight gain of caryopses from wheat plants with and without stem rust. M. E. MADSON, L. CALPOUZOS, & R. W. ROMIG (Univ. Minn., St. Paul). Gain in dry wt of caryopses was measured in three cultivars, Purdue 5481-C, Mindum, and Klein Lucero, from three replicates/treatment in the field. In one treatment, plants were inoculated artificially with *Puccinia graminis tritici*, and severe rust developed. In the other treatment, plants were kept nearly rust-free with fungicides. Caryopses were sampled at 3- to 4-day intervals. Dry wt of caryopses became significantly different at early dough, 18-21 days after the start of the log phase of the disease progress curve. In the rust-free plots, caryopses of Purdue and Klein Lucero reached a dry wt max 10 to 13 days before harvest, followed by a 16% drop. In the rust plots, caryopses of Mindum and Klein Lucero reached a dry wt max 13 to 16 days before harvest, followed by a drop of 23% and 11%, respectively. Between treatments, differences in disease severity and caryopses wt at harvest were ca. 2:1 (96% and 50% in Purdue; 88% and 44% in Mindum; and 85% and 54% in Klein Lucero).

Ozone and infection of geranium flowers by Botrytis cinerea. W. J. MANNING, W. A. FEDER, & I. PERKINS (Univ. Mass., Waltham). Flowering plants of geranium cultivars Enchantress, Red Perfection, and White Mountain were exposed to 0.2, 0.35, and 0.55 ppm ozone for 4-hr periods at 20 C in a greenhouse fumigation chamber in March 1970. Three fully-opened flower heads were sprayed with a spore suspension of *Botrytis cinerea* at 2,000, 1,000, or 500 spores/ml immediately before exposure to ozone began. Sterile distilled water was sprayed on noninoculated flower heads. All flowers were examined for evidence of infection 24 hr after the end of the ozone-exposure periods. All flower heads were then removed and placed in wet, loosely tied plastic bags and incubated at 20 C for 72 hr, with examinations at 24-hr intervals for evidence of infection. Ozone at 0.2 ppm did not injure the plants or prevent or inhibit flower infection by *B. cinerea* at all inoculum levels. Natural infection also occurred on some noninoculated flowers. Ozone at 0.35 ppm did not injure the plants or prevent infection, but did inhibit pathogenesis at the 500-spore/ml inoculum level and on noninoculated flowers. Ozone at 0.55 ppm caused moderate injury on all plants. Ozone at this level did not prevent infection, but did restrict pathogenesis on all inoculated and noninoculated flowers.

Indoleacetic acid transport in Pseudomonas savastanoi. J. L. MARLOW & T. KOSUGE (Univ. Calif., Davis). A system for indoleacetic acid (IAA) transport has been detected in *P. savastanoi*. Results of isotopic competition experiments indicated that IAA transport is inhibited by several indole derivatives such as indolepyruvic acid, but not by 18 common amino acids including L-tryptophan, from which IAA is formed by this bacterium. Such results distinguish this system from the tryptophan transport system previously described. Initial rates of IAA transport were dependent upon temp, pH, and substrate concn. When intact cells were subjected to osmotic shock, IAA- and tryptophan-binding activities were detectable in the osmotic shock fluid and could be separated by various protein fractionation procedures. On the basis of these results, it is concluded that *P. savastanoi* possesses a specific transport system for IAA that is separate from the one for tryptophan.

The reproduction of russet crack in Jersey Orange sweetpotatoes by grafting on plants affected with either sweetpotato virus leaf spot or internal cork. W. J. MARTIN (La. State Univ., Baton Rouge). Russet crack-free Jersey Orange scions were cleft-grafted into plants of cultivar Porto Rico sweetpotatoes affected with virus leaf spot but free of internal cork. In 1968, russet crack developed in over 50% of the sweetpotatoes produced by Jersey Orange plants grown from cuttings made from the developing scions 3 months after grafting. The check plants, established from cuttings of the Jersey Orange vine used to provide the scions for grafting, produced sweetpotatoes free of russet crack. Plants were maintained under cages covered with 32 x 32 mesh screen to exclude known vectors of internal cork virus. In 1969, Jersey Orange scions from plants free of russet crack were cleft-grafted into additional virus leaf spot-affected Porto Rico plants, and internal cork-affected Goldrush plants. Russet crack developed in Jersey Orange potatoes produced by plants established from developing scions from both leaf spot-affected Porto Rico and internal cork-affected Goldrush stocks. Jersey Orange sweetpotatoes from check plants were free of russet crack. Symptoms of russet crack were not evident in any of the Porto Rico or Goldrush potatoes used. Apparently, an infectious agent present in leaf spot-affected Porto Rico and internal cork-affected Goldrush plants incited russet crack in Jersey Orange sweetpotatoes.

Relationship of chemical structure to toxicity and action of oxathiin systemic fungicides. D. E. MATHRE (Mont. State Univ., Bozeman). Various analogs and chemicals related in structure to carboxin (5,6-dihydro-2-methyl-1,4-oxathiin-3-carboxanilide) were tested for their toxicity to growth of *Rhizoctonia solani* and germination of teliospores of *Ustilago nuda*. Their effect on respiration and metabolism of ¹⁴C-acetate was also determined. Oxidation of the sulfur atom greatly decreased the toxicity of the parent compound. These oxidized compounds also had a decreased ability to inhibit respiration and metabolism of acetate by these basidiomycetes. At 10⁻⁵ M, carboxin inhibited metabolism of acetate 65-80%, while 10⁻⁵ M of the monoxide and dioxide forms inhibited acetate metabolism 0-5%. When the benzene ring was eliminated from the carboxin molecule, the resulting compound (5,6-dihydro-2-methyl-1,4-oxathiin-3-amide) was only slightly inhibitory to growth or spore germination, and 10⁻⁴ M had no effect on respiration or acetate metabolism of either fungi. When a thiazole structure was substituted for the oxathiin moiety, the resulting compound (2,4-dimethylthiazole-5-carboxanilide) was inhibitory to growth, spore germination, respiration, and acetate metabolism. Thus, the oxathiin moiety per se is not necessary for inhibition of growth and metabolism of sensitive organisms.

Further observations of tobacco ringspot virus within Xiphinema americanum. J. M. MCGUIRE, K. S. KIM, & L. B. DOUTHITT (Univ. Ark., Fayetteville). Tobacco ringspot viruslike particles were present in the esophageal

lumen of *Xiphinema americanum* that had transmitted the virus. There was a concn of particles near the cuticular wall lining the circular lumen of the anterior esophagus immediately posterior to the connection with the stylet extension. Only scattered particles were present in the lumen at other levels of the anterior esophagus. Large numbers of particles were present throughout the triradiate lumen of the esophageal bulb. When the lumen was slightly open, several rows of particles were visible. The greatest supply of virus seemed to be located in this region. The esophageal lumen of nonviruliferous *X. americanum* contained a mucuslike material, but no viruslike particles. Virus particles may be held in the mucuslike material in viruliferous nematodes.

Distribution of lipid bodies in Erysiphe graminis. W. E. MCKEEN (Univ. W. Ontario, London, Canada). Many lipid bodies which stain with Sudan Black B and Sudan IV appear in parts of the colonial growth of *Erysiphe graminis* f. sp. *hordei* race CR3 growing on the susceptible commercial Keystone cultivar of barley. They are readily observed by means of the light and electron microscope after osmium tetroxide staining, and are abundant in conidiophores, conidia, and mycelium except in the haustorial mother cells, where they are usually absent. Lipid bodies are lacking at the very growing hyphal tip, but gradually increase in number and size farther back. Electron micrographs show that they are intracytoplasmic, intravacuolar, and up to 1 μ in diam. When the colony is washed with acetone or alcohol rather than with aqueous buffer after glutaraldehyde fixation and prior to osmium fixation, the osmiophilic bodies are removed, indicating that they are lipids.

Histological studies of the Pythium-root knot nematode complex in tobacco. P. L. MELÉNDEZ & N. T. POWELL (N. C. State Univ., Raleigh). Invasion and colonization by *Pythium ultimum* were studied in both *Meloidogyne incognita*-infected and noninfected tobacco roots. No fungus invasion was observed in root knot-susceptible plants unless the roots were inoculated with *M. incognita* 3 or 4 weeks prior to inoculation with the fungus. *Pythium ultimum* had penetrated galled tissues of such plants 24 hr after inoculation and progressed into three to four layers of cortical cells in 48 hr. Three days after inoculation, six cortical cell layers were invaded. The fungus had colonized the stele of galled roots after 6 days, and invaded giant cells were beginning to lose their cytoplasmic contents. Nongalled tissues from nematode-infected roots were also invaded by the fungus, but colonization was less than in galled regions. *Pythium ultimum* did not colonize tissues of root knot-resistant plants in any treatment.

The Pythium-root knot nematode complex in flue-cured tobacco. P. L. MELÉNDEZ, & N. T. POWELL (N. C. State Univ., Raleigh). The role of *Pythium ultimum* in premaria breakdown of tobacco roots infected with *Meloidogyne incognita* was investigated. Root knot-susceptible and -resistant plants were given various inoculation treatments with either one or both organisms in pot tests in the greenhouse and in gnotobiotic cultures in growth chambers. No significant necrosis occurred in resistant plants with any treatment. Root decay of root knot-susceptible plants increased with increasing time between nematode and fungus inoculation, and max necrosis occurred when nematodes were added 4 weeks prior to fungus inoculation. Roots of plants maintained aseptically reacted similarly, except that the extent of necrosis was greater in all treatments. Only two *M. incognita* egg masses were necessary to predispose nematode-susceptible roots to decay by *P. ultimum*, although necrosis increased with increasing nematode inoculum. *M. incognita* populations were depressed in nematode-susceptible roots to which the fungus was added 2 weeks before the nematode, but increased with time as *M. incognita* preceded the fungus.

Effect of benomyl and ethirimol on primary infection of wheat by Erysiphe graminis f. sp. tritici. H. C. MELLINGER,

R. S. SLESINSKI, & A. H. ELLINGBOE (Mich. State Univ., E. Lansing). Wheat seeds (cultivar Little Club) were immersed in acetone suspensions of benomyl [methyl-1-(butylcarbomoyl)-2-benzimidazolecarbamate] and ethirimol (5-butyl-2-ethylamino-4-hydroxy-6-methylpyrimidine) and shaken until the acetone evaporated. Treated and control seeds were planted, and 6-day-old seedlings inoculated with conidia of *Erysiphe graminis* f. sp. *tritici*. Inoculated plants were held under conditions favoring synchronized development of the fungus, namely, 17 C and 100% relative humidity (RH) for the 1st hr without light, followed by 35 hr at 22 C and 65% RH, with low light (240 ft-c) supplied the 2nd through 6th and after the 20th hr. Development of primary infection was followed. Spore germination was inhibited on the inoculated leaf surface by ethirimol at concn above 6,000 ppm, but was not affected by benomyl at concn up to 10,000 ppm. Ethirimol at 4,000 ppm caused swelling and distortion of germ tubes and immature appressoria, while benomyl at 4,000 ppm caused stunting but no distortion. Both compounds prevented appressorial development at 4,000 ppm. No elongating secondary hyphae were formed on seedlings treated with benomyl at 2,000 ppm or ethirimol at 4,000 ppm. No sporulation was observed 21 days after inoculation of plants treated with these fungicides.

Toxicity to rats of Alternaria sp. isolated from seeds, flour, and feeds. R. A. MERONUCK & C. M. CHRISTENSEN (Univ. Minn., St. Paul, Minn.). Eighty-five isolates of *Alternaria* from seeds of corn, peanuts, rye, sorghum, sunflower, and wheat, and from wheat flour were grown on sterile moist corn-rice, and incubated for 14-20 days at room temp. The resulting *Alternaria*-infested grain was ground and fed to rats for 2 weeks, either as their sole ration, or as 50% of a balanced diet. Sixty-eight per cent of the *Alternaria* isolates tested in this manner were lethal to rats within 3 to 10 days. Symptoms included anorexia, wt loss, hemoglobinuria, hemorrhaging in the intestinal lumen, and death. Rats that succumbed to *Alternaria*-infested diets ate an average of 15 g of feed and lost an average of 13 g of body wt. Rats that survived on *Alternaria*-infested diets ate an average of 99 g of feed and gained an average of 19 g of body wt.

Microorganisms in the bases of dead branches of Pinus strobus. W. MERRILL (Pa. State Univ., Univ. Park). Resin-impregnated wood from bases of dead branches of *Pinus strobus* completely inhibited spore germination of several Hymenomycetes, whereas blue-stained wood immediately distal stimulated germination over that obtained on fresh sapwood, heartwood, or artificial media. As an initial step in determining the basis for this phenomenon, microorganisms present in these two areas and the interface between them were determined. Isolations from several branches from trees from five sites were made onto glucose-yeast extract agar, acidified weak malt agar, or pine sawdust agar; replicate plates were incubated at 15, 21, or 28 C. Results indicate the presence of a rather consistent microflora in the bases of dead branches. Initial invaders appear to be *Cephalosporium* sp. and a nonsporulating Hyphomycete which are dominant in the resin plug. These appear to be followed and replaced by *Haplosporella* sp., *Trichoderma viride*, and three other nonsporulating Hyphomycetes which are dominant in the blue-stained wood. Low populations of several other species of fungi and yeasts occur at the interface; lower populations of some of these species also occur in the blue-stained wood. An unidentified Hymenomycete frequently was isolated from wood distal to the most intensely blue-stained zone.

Effect of host cultivar on the resident phase of Pseudomonas glycinea. T. W. MEW & B. W. KENNEDY (Univ. Minn., St. Paul). Three pathogenic races were sprayed on leaves of three soybean cultivars in the greenhouse. Care was taken to avoid introduction of bacteria into plants as a result of injury or water congestion. Assays of bacterial populations on symptomless leaves after 4, 24, 120, and 240

hr indicated that bacteria increased >1,000-fold 1 to 2 weeks after suspensions had been fogged on susceptible leaves. Populations were unchanged or declined on leaf surfaces of resistant cultivars. Combinations in the intermediate range resulted in an increase similar to susceptible series during the 1st week, but populations declined during the 2nd week. Although it is difficult to conclude with certainty that bacteria never entered or multiplied within the host, indications are that a distinct magnitude of specificity is conferred to races on leaf surfaces.

Phytoalexin production in soybean roots. Wm. A. MEYER, P. N. THAPLIYAL, J. A. FRANK, & J. B. SINCLAIR (Univ. Ill., Urbana). Two, 6, and 12 days following germination, disease-free Amsoy, Harosoy, and Harosoy-63 soybean seedlings were transplanted into a specially designed system. Seedling roots were allowed to grow for 10 days in sterile, distilled water with or without a zoospore suspension of *Phytophthora megasperma* var. *sojae*. When the susceptible Amsoy and Harosoy cultivars were transplanted into the system, infection of all seedlings occurred within 8 to 10 days. When the resistant Harosoy-63 cultivar was treated similarly, all seedlings remained uninfected up to 10 days. Cheesecloth wicks, with one end in the water suspension, showed a reddish-brown coloration at 3 days and a dark brown color at 6 to 7 days at the opposite end of the wicks for the three cultivars. No color was noted on wicks in noninoculated controls. Chromatographic and spectrophotometric analyses of the wick material confirmed the presence of the yellow-green fluorescing soybean phytoalexin. This is the first report of phytoalexin production in soybean roots.

Bacterial leaf spot of *Ixora* caused by *Xanthomonas* sp. J. W. MILLER (Fla. Dep. Agr. Conserv. Service, Gainesville). *Ixora coccinea*, a popular shrub in the warmer regions of Florida, is grown both as a hedge and for its showy clusters of red flowers. During 1969, leaf specimens were received exhibiting irregular, vein-delimited, necrotic spots having a water-soaked margin. Isolations from a number of leaf specimens yielded a yellow bacterium which proved to be a species of *Xanthomonas*. Successful inoculations were made either by puncturing the leaves with a ring of needles through a drop of bacterial suspension or spraying the inoculum onto unwounded leaves. Approximately 5 days following inoculation, circular, light-colored spots were seen on the young leaves, which soon developed water-soaked centers. The water-soaking spread, and the center of the spots became brown and necrotic. Spots coalesced to form large necrotic areas which caused leaf distortion. Of three bactericides applied as protective sprays, copper-maneb (a mixture of basic copper sulfate + a coordination product of zinc ion and manganous ethylenebis-[dithiocarbamate]) and Agri-strep (streptomycin sulfate) provided good control, but Kocide 101 (cupric hydroxide) was not effective in controlling the bacterial leaf spot.

Mechanical transmission of the coconut palm lethal-yellowing pathogen from frozen or fresh inocula prepared in two buffers. M. E. MILLER & D. A. ROBERTS (Univ. Fla., Gainesville). Necrotic tissues from coconut palms infected with the lethal-yellowing pathogen were ground, with mortars and pestles, in sodium diethyldithiocarbamate (Na-DIECA) plus 2-mercaptoethanol, pH 8.0, or in Tris[tris-(hydroxymethyl)-amino methane]-HCl, pH 7.2. The resulting slurries, to which 500-mesh Carborundum was added, were applied to the basal portions spear leaves of 1.5- to 3-year-old palms by the gauze-pad method of inoculation. Fresh inocula prepared in Na-DIECA infected 4 of 22 palms, whereas those prepared in Tris infected 11 of 45. Inocula prepared in Tris from necrotic tissue that had been frozen for as long as 18 weeks infected 12 of 42 palms. These results confirm that lethal yellowing of coconut palms is caused by a mechanically transmissible agent, and support the theory that lethal yellowing is a viral disease.

Selective medium for the isolation of *Erwinia amylovora* and other *Erwinia* spp. T. D. MILLER & M. N. SCHROTH (Univ. Calif., Berkeley). *Erwinia amylovora* was obtained from apparently healthy flowers, young fruits, and leaves of pear with a selective medium containing cycloheximide, sodium taurocholate, thallium nitrate, bromothymol blue, and tergitol 7 (sodium heptadecyl sulfate). The plating of a dilution series on this medium and other standard media such as potato-dextrose peptone agar showed that the efficiency of recovery was equal to the standard media. Most bacteria found on pear trees other than *E. amylovora* were inhibited by the selective medium. Pure cultures of *E. amylovora* often were obtained from washings of apparently healthy flowers and other pear parts. The colonies were readily distinguished from other bacterial colonies after 2- to 5-days' incubation by a characteristic colony morphology and a red color. Although the medium was highly selective for *E. amylovora* when assaying the microflora on pear trees, laboratory experiments indicated that it was in effect selective for most *Erwinia* spp. Other *Erwinia* spp. that grew on the medium were *E. quercina*, *E. dieffenbachiae*, *E. carotovora*, *E. aroideae*, *E. nigrifluens*, *E. magniferae*, and an unidentified *Erwinia* from insects. *Erwinia chrysanthemi* did not grow on the medium. Each *Erwinia* spp. has a characteristic colony morphology and color.

Modifications in buffer extraction of leaf proteins for the improvement of gel disc electrophoretic identification of proteins. D. F. MILLIKAN, V. BOJNANSKY, & J. A. ROSS (Univ. Mo., Columbia). In our studies of virus-induced or genetic-controlled graft failures there were very weak or few bands of proteins when leaf tissues were extracted with 0.1 M borate-HCl (pH 8.0) or 0.1 M [2-amino-2-(hydroxymethyl)-1-3-propanediol] (pH 8.0) supplemented with 12.5% sucrose and 0.1% ascorbic acid. When the buffer was changed to combinations of phenol (P), acetic acid (A), water (W), and 0.2 M mercaptoethanol (M), good bands were obtained. PAMU (2:1:1 made in 5 M urea) was superior to other combinations. Peroxidase bands were obtained from proteins extracted with either borate-HCl and Tris[tris(hydroxymethyl) amino methane]-HCl, and increased amounts of peroxidase isozymes were associated with both types of graft failure. No differences in proteins due to the incompatible combinations were found by disc electrophoresis, in spite of the marked differences when the protein content was estimated by Kjeldahl or the Lowry method. When the PAMU-extracted proteins were compared with those estimated by the Lowry method, we found that the quantitative differences associated with graft failure were not extracted by the PAMU buffer.

Effects of CO₂ and O₂ on growth and sporulation of several species of *Phytophthora*. D. J. MITCHELL & G. A. ZENTMYER (Univ. Calif., Riverside). The effects of aeration on growth and sporulation of 8 species of *Phytophthora* were determined by incubating cultures in continuous flow (100 ml/min) atm containing different concn of CO₂ plus 1, 5, or 20% O₂. Species used for studies on growth (G), oospore production (OP), or sporangium production (SP) were *Phytophthora cactorum* (OP, SP), *P. capsici* (G, OP, SP), *P. cinnamomi* (OP, SP), *P. citrophthora* (G, OP, SP), *P. drechsleri* (OP), *P. megasperma* var. *sojae* (OP), *P. palmivora* (G, OP, SP), and *P. parasitica* (G, OP, SP). Linear growth of most isolates on sucrose-asparagine agar was max at 5% O₂ and 0% CO₂; lower levels of O₂ reduced growth in comparison to the ambient air control. Decreasing the O₂ level below 20% reduced growth of most isolates in sucrose-asparagine liquid medium. Growth on agar was stimulated by 5% CO₂ when the O₂ level was 1%. Growth of most isolates in liquid or agar was reduced by CO₂ concn greater than 5% when compared to growth at the same O₂ level without added CO₂. SP on mycelial mats grown in dextrose-nitrate liquid medium and rinsed with deionized H₂O was reduced by decreasing O₂ concn or

increasing CO₂ levels above that in air. OP in dextrose-nitrate agar was greater at 1 or 5% O₂ than in air.

Carbon dioxide stimulates germination of basidiospores of Polyporus dryophilus and Fomes rimosus. T. P. MCG & H. L. MORTON (Univ. Mich., Ann Arbor). Basidiospores of four isolates of *Polyporus dryophilus* and two of *Fomes rimosus* were cast onto 2% water agar (pH 5.8-6.8) and incubated at 24 C in closed bottles containing industrial-grade CO₂ at concn between 0 and 100%. After 14 days, agar plugs containing the spores were stained with lactophenol cotton blue to stop germination. Per cent spore germination at each CO₂ level was determined by counting at least 300 spores. Germination occurred with four of the six isolates tested. Germination was highest at CO₂ levels above 25%. Maximum germination of 92% and 41% occurred at 65 and 100% CO₂ for *P. dryophilus* and *F. rimosus*, respectively. Spores of these isolates were also cast onto 2% water agar (pH 5.7-6.8) and incubated at 24 C in bottles continuously flushed with a gaseous mixture containing 15, 25, or 65% CO₂. Germination occurred with four of the five isolates tested. After 14 days, max germination of 15% and 31% occurred at 15% CO₂ for *P. dryophilus* and *F. rimosus*, respectively. Three tests were run, using the spores of both fungi in which sterile filter paper, paper bearing live spores, and paper bearing heat-killed spores were exposed to 300 µc of ¹⁴CO₂ in an atm containing 15% CO₂ for 14 days. In all three tests, the live spores appeared to fix carbon dioxide.

Effect of nematode-trapping fungi, media, and temperature on the morphometrics of Aphelenchus avenae. H. L. MONOSON (Bradley Univ., Peoria, Ill.). The parthenogenic nematode, *Aphelenchus avenae*, was maintained with four species of nematode-trapping fungi on three different media at 4 temp for 7 days. De Man's procedures were utilized for the measurement of nematodes, and the data was analyzed statistically. The two statistical methods used in this study were analysis of variance and coefficient of determination. The data suggest that the culture variables of the study accounted for more of the variability in "V" than did "L"; "a", "b", or "c" measures when "V" is the position of the vulva expressed as a percentage of body length measured from the anterior end; "L" is body length expressed in µ; "a" is body length divided by the greatest body width; "b" is body length divided by the distance from the anterior end of the nematode to the base of the esophagus; "c" is body length divided by the length of the tail from the anus or cloaca to the terminus. The "V" values were considered the best taxonomic parameter for the identification of *A. avenae*.

Unsaturated fatty acids as natural stimulants of rhizomorph production by Armillaria mellea. A. R. MOODY & A. R. WEINHOLD (Univ. Calif., Berkeley). Production of rhizomorphs, the infection structures of *Armillaria mellea*, is stimulated by several unsaturated fatty acids. To determine the possible role of these compounds in stimulating rhizomorph production in nature, roots of ponderosa pine, peach, Douglas fir, white fir, and incense cedar were analyzed for fatty acids. Extraction of ground roots with cold ethyl ether was followed by three extractions with ethyl ether under reflux. The combined extracts were then bioassayed and analyzed with gas chromatography. The bioassay consisted of growing the fungus on defined media with the lipid extract. No rhizomorphs were produced in the absence of extract, and the dry wt of rhizomorphs varied with the amount of extract added. When the lipid extracts were saponified and the fatty acids removed, the resulting extracts were inactive. The gas chromatographic data were then compared with the data from the bioassay. Roots of ponderosa pine and peach contained relatively high amounts of unsaturated fatty acids, and the bioassay yielded relatively large quantities of rhizomorphs. White

fir, incense cedar, and Douglas fir contained less unsaturated fatty acid, and fewer rhizomorphs were produced. It appears that unsaturated fatty acids may stimulate rhizomorph production in nature.

Effects of aqueous soil extracts on zoospore production by Aphanomyces euteiches. R. H. MORRISON & T. H. KING (Univ. Minn., St. Paul). Aqueous, unsterilized extracts from two soils were compared with water and a salt solution (SS) containing Ca, Mg, and K for their effects on the asexual production of zoospores by *Aphanomyces euteiches*. Greater numbers of zoospores were formed in SS than in water by five isolates of *A. euteiches*. Soil extract (SE) from both soils induced motile and/or nonmotile zoospores in all isolates. SE, undiluted or diluted 1:10 with water, induced motile zoospores in three isolates capable of producing abundant motile zoospores in SS. SE induced as many or more motile zoospores in an isolate which produced few motile zoospores in SS, but did not induce them in an isolate lacking the ability to produce motile zoospores in SS. SE (1:10) yielded more motile and/or nonmotile zoospores than did undiluted SE. Two prolific motile zoospore-producing isolates were tested in SE dilutions of 1:2, 1:5, 1:10, 1:100 and 1:1,000 in water. Most zoospores were produced in dilutions of 1:2, 1:5, and 1:10. In dilutions of 1:100 and 1:1,000 and in undiluted SE, the same numbers or a few more zoospores were produced than in water.

Endopolygalacturonate trans-eliminase production by a potato dry-rot pathogen. J. M. MULLEN & D. F. BATEMAN (Cornell Univ., Ithaca, N. Y.). *Fusarium roseum* 'Avenaceum', which causes a dry rot of *Solanum tuberosum*, produced an extracellular endopolygalacturonate trans-eliminase when cultured on a potato broth-pectic acid medium at 22 C. This enzyme is calcium-dependent and exhibits max activity at pH 9.0. It was purified 112-fold by ammonium sulfate fractionation (60-80% fraction) and electrofocusing (pH range 7-10). The enzyme appears to consist of one component with an isoelectric point of pH 8.2, and has a mol wt of ca. 32,000 as determined by gel-filtration in Sephadex G-75. Both crude and purified preparations of the enzyme caused maceration and cell death of potato tuber tissue.

Effect of Chrysomyxa pirolata cone rust on dispersal and viability of Picea pungens seeds. D. L. NELSON & R. G. KREBILL (USDA Forest Service, Intermountain Forest Range Exp. Sta., Logan, Utah). Our recent discovery of *Chrysomyxa pirolata* cone rust on blue spruce (*Picea pungens*) in Utah focused attention on the desirability of understanding the impact of this disease on spruce regeneration. Reports from North America and northern Europe indicate that partial to complete destruction of seed occurs in rust-infected cones. On six sample plots in central Utah, nonrusted cones averaged 204 seeds/cone, whereas, rusted cones averaged 188. Seeds that were easily dislodged from mature cones by tapping averaged 113 seeds for nonrusted and only 13 for rusted. The remaining seeds were extracted by breaking the cones apart. All seeds were stratified at 1 C for 3 months, then tested for germination by incubating at temp of 25-C day (8 hr) and 15-C night. Of seed extracted by tapping cones, 71.0% germinated from nonrusted cones as compared to 48.9% from rusted cones. Of seeds extracted by breaking cones apart, 53.3% from nonrusted cones germinated, as compared to 34.8% from rusted cones. These results suggest that there is little reduction in the number of seed produced in rusted cones, but that malformation of these cones may significantly interfere with seed dispersal. Also, seed from rusted cones may be less viable but not totally destroyed by the disease.

Characteristics of false broomrape disease and properties of the etiological agent. M. A. NEWMAN & R. W. TOLER (Texas A&M Univ., College Station). Tumors from false broomrape infected tobacco were collected at three locations. The three isolates of false broomrape were obtained from Kentucky, North Carolina, and Georgia. Isolate num-

bers are Ky 101, NC 201, and G 301, respectively. False broomrape inoculum was prepared by homogenizing false broomrape tumors from *Nicotiana tabacum* cultivar Hicks in 0.1 M pH 7 phosphate buffer at a 2:1 buffer to tissue ratio. Inoculations were made onto Hicks tobacco by rubbing the inoculum onto Carborundum-dusted roots with cotton swabs. The tobacco was grown at a greenhouse temp of 30 ± 3 C. Incubation time was measured from inoculation to symptom development; i.e., teratomas recognizable by the unaided eye. Incubation time studies revealed no differences among the isolates of false broomrape. The mean incubation period was 21 ± 3 days. All three isolates failed to infect eggplant, bell pepper, collard, coleus, petunia, squash, bean, tomato, pea, and cucumber. Inoculations were the same as for tobacco controls. The thermal inactivation end point of the false broomrape isolates was 90 C for 10 min. The aging of the frozen false broomrape tumor tissue (-20 C) end point was 15 days.

Growth reduction in scarlet oak due to oak decline in Virginia. C. R. NICHOLSON & J. M. SKELLY (Va. Polytechnic Inst., Blacksburg). In order to determine the amount of annual increment lost due to the slow killing disease called oak decline, 5 dead scarlet oak trees in each of 4-, 6-, 8-, 10-, and 12-inch diam classes were felled, and a 1-inch disc was removed at 4.5 ft above the ground. Crown class, total height, age diam breast height year of death, and decay fungi sporophores present were recorded. Annual increments were measured with the use of a dendrochromograph equipped with a $\times 30$ stereoscopic microscope. The average number of years trees were in decline and their average annual increment during these years was 11 and 0.57 mm (4-inch class), 9 and 0.72 mm (6-inch class), 5 and 0.79 mm (8-inch class), 4 and 0.84 mm (10-inch class), and 5 and 1.27 mm (12-inch class). The average annual increment for the same number of years before decline as after decline was 1.12 mm, 1.29 mm, 1.74 mm, 1.58 mm, and 1.70 mm for trees in each respective diam class. In some instances decline was initiated, then stopped with increased radial growth; then further decline ended with death. The smaller-diam trees that were suppressed had a more uniform average annual increment than dominant trees. *Polyporus gilvus* was the most abundant decay fungus present.

Phenylalanine ammonia lyase activity in tobacco tissue inoculated with Pseudomonas pisi and Pseudomonas tabaci. A. NOVACKY & G. ACEDO (Univ. of Mo., Columbia). To induce the hypersensitive reaction (HR), one-half the tobacco leaves were inoculated with a suspension of *Pseudomonas pisi* containing 10^8 cells/ml. The remaining half was injected with either water or heat-killed bacteria and used as the control. Phenylalanine ammonia lyase (PAL) activity was assayed at 1-hr intervals after inoculation. Approximately 3 hr after inoculation, the enzyme activity was 3 to 4 times more than the controls. A similar experiment was completed using *P. tabaci*, which causes the wildfire disease. The PAL activity increased to an identical level, but max activity was reached at 15 to 18 hr instead of 3 hr, as observed with *P. pisi*. A drop in PAL activity in leaves inoculated with both organisms was detected with the appearance of symptoms. Based on the comparisons of PAL activity, the only difference between HR and the wildfire disease is the time needed for a max increase in activity.

Evaluation of preharvest benomyl applications on postharvest Monilinia rot of peaches and nectarines. J. M. OGAWA, B. T. MANJI, & D. J. RAVETTO (Univ. Calif., Davis). Captan-[N-(trichloromethylthio)-4-cyclohexene-1,2-dicarboximide] and benomyl-[methyl-1-(butylcarbamoyl)-2-benzimidazolecarbamate] sprayed fruits were each given the following three postharvest treatments: (i) dry nylon brush defuzzing; (ii) water wash, wet brush defuzzing, and spray of Botran (2,6-dichloro-4-nitroaniline) (1 lb. active/100 gal) and wax; or (iii) 0.8% Na o-phenylphenate foam, water rinse, and waxing. After treatment,

seven replications of 50 fruits each were commercially packed in plastic cups and stored for 1 week at 1 C and ripened at 20 C and 85% relative humidity for 6 days. Control peaches developed 21% decay. Peaches receiving benomyl sprays 21 days and 1 day before harvest followed by treatment (i) developed 3.5% decay; treatments (ii) or (iii) less than 2%. Two preharvest captan sprays significantly reduced decay in treatments (i) or (ii) but not in (iii). A benomyl spray 21 days before harvest followed by treatment (i) was as effective as two preharvest benomyl sprays. A single captan spray was not effective. On nectarines, after 7 days of ripening, benomyl sprays 14 and 1 day before harvest reduced decay from 50 to less than 2% regardless of the postharvest treatments. Two preharvest captan sprays reduced decay significantly with each postharvest treatment, but not as effectively as benomyl.

Low temperature maintains virulence of dry Colletotrichum trifolii inoculum in storage. S. A. OSTAZESKI, T. A. CAMPBELL, T. E. DEVINE, & C. H. HANSON (USDA, Beltsville, Md.). Production and use of a dry *Colletotrichum trifolii* inoculum (DI) have been described elsewhere. Adoption and use of DI by plant breeders in screening alfalfa for resistance to anthracnose depend on maintaining virulence under long-term storage. Three sublots of one batch of DI were subjected to storage temp of -20 , 7.5, or 21 C. After 1, 2, 4, and 10 months, samples were taken from each sublot and tested on flats of 2-week-old alfalfa seedlings. Seedlings in each flat consisted of row plots of an anthracnose-resistant selection, ARS-1; two moderately resistant cultivars, Team and Sonora; and a highly susceptible cultivar, Saranac. Check flats were inoculated with freshly comminuted 2-week-old agar cultures of the pathogen. Plants were scored 2 weeks later (1 = no infection; 5 = plant dead) to obtain infection indices for check inoculum and for DI in each storage treatment. Infection indices of the four inocula differed significantly (.01 level) after 2, 4, and 10 months of storage. Virulence declined with time, and inoculum stored 4 months at 21 C was nearly avirulent (index 1.30). After 10 months, DI stored at -20 and 7.5 C was as virulent as freshly prepared agar inoculum (indices 3.64, 3.61, and 3.70, respectively).

Effect of stunt and root knot nematodes on Verticillium wilt of tomato. A. J. OVERMAN & J. P. JONES (Univ. Fla., Gulf Coast Exp. Sta., Bradenton). Seedlings of four tomato stocks (Floradel, Tropic, 393-9, and VFN 6428) were grown at 23 C in pots of methyl bromide-treated, composted Leon fine sand infested with factorial combinations of two nematodes (*Tylenchorhynchus capitatus* and *Meloidogyne incognita*) and two isolates of *Verticillium albo-atrum*. Highest incidence and severity of *Verticillium* wilt symptoms occurred in Floradel plants infested with *T. capitatus* at 23 C, with *M. incognita* at 29 C. A significant difference occurred in the increase of *T. capitatus* during the test period in pots inoculated with the two isolates of *V. albo-atrum*: the nematode population density attained in combination with one isolate was similar to the 13-fold increase of the inoculum level attained in pots without the fungus; with the other isolate, the nematode population increased 9-fold. Foliar symptoms of the disease did not appear on Tropic or VFN 6428 exposed to the fungus and either nematode.

Suppression of Aphanomyces root rot of peas by soil amendments, fungicides, and fumigants. G. C. PAPAIVAS & J. A. LEWIS (ARS, USDA, Beltsville, Md.). Cruciferous amendments such as leaves and stems of cabbage, kale, and mustard at 0.5% of the oven-dry wt of soil controlled root rot of peas caused by *Aphanomyces euteiches* in the greenhouse. Only kale was effective in the field. Attempts to control the disease with the fungicides 2-(4'-thiazolyl)benzimidazole (TBZ), 2,6-dichloro-4-nitroaniline (Botran); [methyl-1-(butyl-carbamoyl)-2-benzimidazole carbamate] (benomyl); and zinc ethylenebis (dithiocarbamate) (zineb)

failed. Only the fungicide *p*-dimethylamino benzenediazo sodium sulfonate (Dexon) was effective in both greenhouse and field. We obtained very good control in the greenhouse with methylisothiocyanate (MIT) and other fumigants that decompose to MIT in soil. Sodium *N*-methylthiocarbamate (Vapam) and three formulations of tetrahydro-3,5-dimethyl-2H-1,3,5-thiadiazine-2-thione (DMTT-I, DMTT-II, and DMTT-III) controlled root rot effectively at 50-200 ppm. Root rot was suppressed with a lower concn of fumigants when the soil containers were closed in polyethylene bags for 2 weeks after fumigant application than when left uncovered. The fumigants were effective in suppressing root rot of several pea cultivars and at a soil temp range of 17-32 C. All fumigants tested were effective in the field.

Isolation of a phytotoxin from Pseudomonas phaseolicola. S. S. PATIL (Univ. Hawaii, Honolulu). Cultures of an isolate of race 2 of *Pseudomonas phaseolicola* grown in synthetic and nonsynthetic media were harvested during the late log phase of growth. Filtrates were concd in vacuo and desalted with methanol. The methanol extracts were evaporated to dryness and the residue was dissolved in water and passed through a column of Sephadex G-10 (82 × 5 cm) at rate of 1 ml/min. Aliquots from alternate fractions were tested for halo-inducing activity on bean leaves. The active fractions were combined and passed through a column of DEAE cellulose (30 × 1.2 cm). The column was first washed with several bed volumes of water and then, in a batchwise manner, with 0.01, 0.05, and 0.1 M NaCl. The eluates were desalted on Sephadex G-10 (90 × 1.2 cm) at a flow rate of 0.15 ml/min. Halo-inducing activity was eluted by 0.01 M NaCl. With Sephadex column chromatography and paper and thin-layer chromatography it was established that in synthetic and nonsynthetic media the same compound was produced. Finally, by subjection of extracts of bean leaves inoculated with *P. phaseolicola* to the same isolation procedure, a halo-inducing compound was obtained.

Reactions of eleven Chenopodium species to seven sap-transmissible grass viruses. A. Q. PAULSEN (Kansas State Univ., Manhattan). Soil-borne wheat mosaic virus induced chlorotic and necrotic local lesions on six *Chenopodium* species with best symptoms on *C. amaranticolor*, *C. ambrosioides*, and *C. quinoa*. Barley stripe mosaic virus caused chlorotic and necrotic local lesions on eight species. Local lesions on four species, and local and systemic symptoms in six more, were induced by brome mosaic virus. Systemic invasion was indicated in apical leaves by chlorotic spotting and leaf distortion. Foxtail mosaic virus infected *C. ambrosioides* locally, while nine other species showed local and systemic symptoms. The systemic symptoms (chlorotic rings and line patterns) appeared on mature leaves above inoculated ones. No species was infected by either Panicum mosaic or wheat streak mosaic virus, or by strains A or B of maize dwarf mosaic virus. *Chenopodium bonus-henricus* was not infected by any one of the seven viruses. Combinations of *C. amaranticolor*, *C. ambrosioides*, *C. bonus-henricus*, *C. murale*, and *C. quinoa* may be useful to assay and diagnose the viruses tested. Some infection occurred on *C. album*, *C. capitatum*, *C. foetidum*, *C. rubrum*, and *C. urticum*, but symptoms were often faint and the plants exhibited undesirable growth habits.

Pratylenchus thornei, a nematode pest of wheat in Sonora, Mexico. J. B. PEREZ B., S. D. VAN GUNDEY, L. H. STOLZY, I. J. THOMASON, & R. J. LAIRD (Centro de Investigaciones Agrícolas del Noreste, Sonora, Mexico, Univ. Calif., Riverside, Centro Internacional de Mejoramiento de Maíz y Trigo, Mexico City, Mexico). *Pratylenchus thornei* has been found widely distributed and causing serious yield reductions in the wheat-growing areas of western Sonora, Mexico, during the last 3 years. Damage was greatest in fields planted early in the season, usually prior to the recommended planting dates for wheat, 15 November to 15 December. An experiment was established

in the Yaqui Valley on a *P. thornei*-infested soil to determine the effects of soil fumigation and time of planting on growth and yield of wheat. Six cultivars of wheat replicated 3 times were planted on fumigated (462 liter/ha of D-D [1,3-dichloropropene; 1,2-dichloropropane]) and non-fumigated soil. There were five planting dates at 2-week intervals starting 27 October. A significant increase in tillering, plant growth, and yield in response to soil fumigation occurred in the first two plantings when the soil temp at 15 cm were above 23 C. When soil temp were below 13 C at the last planting, no significant response to soil fumigation was observed. Cultivar responses to soil fumigation were observed in the earlier plantings. These observations indicate that correlation of planting date with soil temp may provide an economical control of this nematode in Mexico.

Fungal invasion of peanut kernels as influenced by harvesting and handling procedures. R. E. PETTIT & R. A. TABER (Texas A&M Univ., College Sta.). Peanuts were dug from field plots at Yoakum and Stephenville, Texas, during the years 1967 through 1969, and were subjected to the following treatments: field-dried in inverted (pods in the upright position) and random windrows for different time periods; cured on the vine with forced air; combined at kernel moisture levels above 25% and either flash-dried or stored at 10 C and/or dried at different air flow rates with and without supplemental heat. Following each treatment replicated, 100 kernel samples were surface-sterilized and plated on rose bengal-streptomycin agar. The degree of fungal invasion increased when the average drying rate was less than 0.12%/hr. This rate was influenced by the air velocity, heavy dew and shower frequency, air temp, and relative humidity. The best drying treatment was on the vine in bins with forced air, and the second best was where 25 CFM of air were passed through several different volumes of bulk peanuts. Peanuts from the inverted windrow were less severely damaged by fungi. Peanut pods in contact with the soil surface beneath the random windrow were more heavily invaded. High-moisture peanuts held in bulk containers over 18-24 hr without adequate aeration, and those stored at 10 C for over 48 hr were heavily invaded.

Damage of lettuce seedlings related to crop residue decomposition. D. J. PHILLIPS (Univ. Calif., Berkeley). Lettuce seedlings, grown in the field and greenhouse in soil containing green crop residues, show damage to their root systems. Damage occurred on both primary and secondary root tips and on mature primary root tissue. The primary incitant of the root lesions was difficult to determine. The response of seedlings growing in soil containing an organic amendment was a retardation or prevention of root cell elongation and collapse and darkening of intact tissues. *Pythium* infection, when associated with this damage, caused disintegration of tissue and cells. Root damage was reduced by incorporating a mixture of Benlate [methyl-1-(butylcarbamoyl)-2-benzimidazolecarbamate] and Dexon (*p*-dimethylaminobenzenediazo sodium sulfonate) into the amended and nonamended field soil. Isolation of fungi from the roots suggested that in field soils a correlation exists between residue-related damage and the presence of *Pythium* spp. Soil treated with a steam-air mixture at 60 C for 50 min and amended with green crop residues was damaging to lettuce roots.

Effect of temp on brown stem rot of soybean. D. V. PHILLIPS (Univ. Ga., Ga. Exp. Sta., Experiment). Brown stem rot (BSR) symptom development was studied in field plots and in growth chambers at 15, 21, 27, and 32 C. Several soybean cultivars (maturity groups IV through VIII) were inoculated with different isolates of *Cephalosporium gregatum* (from North Carolina, Illinois, Iowa, and Mexico) by the basal stem puncture method. BSR symptom ratings were made 21 to 30 days after inoculation in growth chambers and just before maturity in field plots. In five experiments, the mean symptom ratings

(max rating = 4.0) for all inoculated plants were 3.0, 2.8, 3.1, and 0.5 at 15, 21, 27, and 32 C, respectively. Symptom ratings were lower at 32 C than at lower temp for all combinations of soybean cultivars and *C. gregatum* isolates tested. In field plots, internal browning averaged 50% of the stem length for all inoculated plants. Air temp recorded near the field plots from 1 June to 1 November indicated that temp were above 32 C for 4%, 27 to 32 C for 22%, 15 to 27 C for 63%, and below 15 C for 11% of the growing season. Since BSR symptoms developed extensively between 15 and 27 C and were not completely inhibited at 32 C, temp occurring during the growing season probably do not limit disease development where soybeans in maturity groups IV through VIII are grown.

Control of decay in Anjou pears with hot water and with heated and unheated suspensions of benomyl or thiabendazole. C. F. PIERSON & H. M. COUEY (ARS, MQRD, USDA, Wenatchee, Wash.). Anjou pears were dipped for 3 min in water at 21, 52, or 54 C, in 250- and 500-ppm suspensions of benomyl [methyl-1-(butylcarbamoyl)-2-benzimidazolecarbamate], or in 500- and 1000-ppm suspensions of thiabendazole [2-(4-thiazolyl)benzimidazole], at 21 C. Treatments were also made with these chemicals at 52 C at one-half the concn used at 21 C. Examinations were made for flesh rot and stem (pedicel) decay after storage for 6 months at 0 C. Flesh rot was significantly reduced by all treatments except water at 52 C. Stem decay was substantially reduced by hot water and by the heated fungicide solutions. Although the benomyl treatments were superior to thiabendazole at 21 C, neither material produced commercially significant disease control at that temp. Colonies of *Penicillium expansum* and *Alternaria* sp. were isolated in equal numbers from stems of check fruits. *Alternaria* sp. was the principal organism isolated from stems treated with benomyl or thiabendazole at 21 C, while *P. expansum* was the major organism from stems treated with hot water. The control of stem decay appears to depend on a combination of chemicals and heat, while control of flesh rot can be achieved with chemicals alone.

Microbial antagonism encouraged by the monosodium salt of hexachlorophene. J. A. PINCKARD (La. Agr. Exp. Sta., Baton Rouge). Field applications of a formulation of the monosodium salt of hexachlorophene [2,2'-methylenebis(3,4,6-trichlorophenol)] (Isobac 20) at 4 oz/acre to cotton plants and soil have resulted in significant increases in disease control. In vitro tests of the formulation against five species of fungi listed below showed fungicidal activity ranging from 2 to more than 500 ppm. But when a 10% soil-water mixture from a field showing disease control was shaken with Isobac 20 at concn of 1, 2, 4, 8, or 16 ppm for 24 hr, then plated on potato-dextrose agar or water agar, a different bacterial flora developed from that on untreated checks. It resulted in the appearance on the plates of a fast-growing chalk-white bacterium antagonistic to species of *Rhizoctonia*, *Pythium*, *Phytophthora*, *Fusarium*, *Trichoderma*, and to other unidentified fungi of the soil. Since hexachlorophene is known to be selectively toxic to certain bacteria at low dosages, these results suggest that the observed field control of several diseases of cotton by Isobac 20 is indirect, and related to its ability to alter the ecological balance of certain soils.

Inhibition of corn root and crown rot pathogens in vitro with fungitoxicants systemic in corn. T. E. POCKLINGTON & M. B. LINN (Univ. Ill., Urbana). Five chemicals reported to be systemic in plants other than corn (*Zea mays*) were tested for toxicity to three corn root and crown rot pathogens, *Fusarium moniliforme*, *Gibberella zeae*, and *Diplodia zeae*. The toxicants were OM-2424 [5-ethoxy-3-(trichloromethyl)-1,2,4-thiadiazole]; carboxin (5,6-dihydro-2-methyl-1,4-oxathiiin-3-carboxanilide); chloroneb (1,4-dichloro-2,5-dimethoxy benzene); TBZ [2-(4-thiazolyl) benzimidazole]; and benomyl [methyl-1-(butylcarbamoyl)-2-benzimidazolecarbamate]. All cultures were grown on

potato-dextrose agar (PDA) and transferred to PDA impregnated with fungitoxicant using the agar plug method. Benomyl and TBZ completely inhibited growth of *F. moniliforme* and *G. zeae* at 25 ppm, while OM-2424 and chloroneb caused little or no inhibition at 700 ppm. Carboxin stopped growth of *F. moniliforme* and *G. zeae* at 500 ppm. Benomyl, TBZ, and chloroneb at 50 ppm and OM-2424 and carboxin at 300 ppm prevented growth of *D. zeae*. TBZ at 150 ppm and carboxin at 50 or 150 ppm showed systemic activity in aboveground parts of corn.

The endocarpic mycoflora of two types of windrow-dried peanut fruit. D. M. PORTER (ARS, USDA, Va. Polytechnic Inst., Holland). The endocarpic mycoflora of peanut (*Arachis hypogaea*) fruit dried in either a random windrow (plants left as they fell from the digger) or an inverted windrow (plants inverted to expose fruit to sunlight) is distinctly different, both quantitatively and qualitatively, from that of freshly dug fruit. The proportion of shells and seed infested with microorganisms was reduced 25% and 48%, respectively, after field-drying for 5 to 7 days in random and inverted windrows. *Chaetomium* (21%), *Penicillium* (18%), *Trichoderma* (14%), *Rhizoctonia* (6%), and *Fusarium* (6%) were the dominant fungi (isolation frequency of 6% or more) found associated with shells (pericarp) of freshly dug fruit. The dominant mycoflora of shells of windrowed fruit included *Chaetomium* (21%), *Fusarium* (14%), *Rhizoctonia* (11%), *Alternaria* (10%), and *Sclerotium* (8%). Seeds of freshly dug fruit were dominated by *Penicillium* (28%) and *Aspergillus* (9%). *Penicillium* was the only dominant fungus found associated with seed of windrowed fruit. Fewer isolates of *A. flavus* were obtained from fruit dried in the inverted windrow than from fruit dried in the random windrow. The isolation frequency of fungi from shells and seed increased greatly provided plants were subjected to rainfall while in the windrow.

Early excess of barley stripe mosaic virus ribonucleic acid in infected tissue. D. R. PRING (Univ. Nebr., Lincoln). Phenol extracts were made from healthy and barley stripe mosaic virus-infected leaves one (the first green leaf) and two at 24-hr intervals 1 to 8 days after inoculation of the mature leaf one. The preparations were analyzed on log-linear sucrose gradients. Viral RNA (21 Svedberg units) was detected at 2 days in leaf one, and at 3 days in leaf two. At 3 days, there was a 150-fold excess of free viral RNA over that encapsulated in leaf one, while a 60-fold excess was apparent in leaf two. The specific infectivity of the 21 Svedberg units RNA from infected leaves was the same as that of RNA from purified virus. The total viral RNA in leaf one or two reached max levels at 4 to 5 days, then declined steadily. Maximum virus nucleoprotein concn occurred in 7 to 8 days. Even though the max level of viral RNA in leaves one and two was comparable, the yield of intact virus in the systemically invaded leaf two was 3 to 4 times that recovered from leaf one. Less than 20% of the max level of viral RNA was recovered as intact virus. The specific activity of ³²P-labeled viral RNA was 10-fold that of host RNA species during the period of rapid viral RNA production. A severe reduction in the rate of ³²P incorporation into host RNA was apparent at 2 days, and the amount of total host RNA in infected tissues was reduced by about 60% at 8 days.

*Studies of infection of male sterile barley by *Claviceps purpurea*.* S. B. PURANIK & D. E. MATHRE (Mont. State Univ., Bozeman). The seriousness of ergot in male sterile barley used for production of hybrid barley seed prompted further study of the biology of this pathogen. Clipping of the upper portion of the glume followed by the addition of conidia from a fine capillary dropper was the best method of inoculation. The optimum time for inoculation was 4 days after the spikes emerged from the boot and as the spikelets were opening. Suspension of conidia in a 2% sucrose solution did not increase the level of infection as compared to suspension of the conidia in water. Infection

occurred with as few as 10 conidia/ml, but max infection occurred with 10^5 through 10^7 conidia/ml of water, using 20-30 μ liters/floret. Occasionally, infection occurred with no honey dew secretions or sclerotial development, but conidia were produced on the ovary surface. Glucose and fructose occurred in both honey dew and an ethanol extract of ovaries. Sucrose and an unidentified disaccharide also were present in the ovary extract. The pH of squashed ovaries was 5.6 to 6.0. Germination of conidia in water was nil, but good germination occurred in 0.2 M glucose, fructose, or sucrose. Conidia germinated equally well in glucose at pH 5.6 and 6.5.

The effect of presporodochial benomyl and oil spray applications on the development of almond brown rot caused by Monilinia laxa. D. C. RAMSDELL, B. T. MANJI, & J. M. OGAWA (Univ. Calif., Davis). Benlate 50W (benomyl) [methyl-1-(butylcarbamoyl)-2-benzimidazolecarbamate] at 1 lb. and 0.5 lb. plus superior oil at 1.5 gal/100 gal water and benomyl at 1 lb./100 gal water without oil was applied prior to sporodochial development on Jordanolo almond trees infected with *M. laxa* twig and blossom blight. A complete randomized block design plot consisting of 24 trees/replication was sprayed with an air-blast sprayer at 6 gal/tree on 18 December 1969. Sporodochial development on previously blighted flowers and peduncles was evaluated at bloom on 6 February 1970. The 1-lb. plus oil treatment resulted in a 95 and 83.9% reduction in numbers of sporodochia on blighted blossoms and peduncles, respectively. The 0.5-lb. plus oil treatment gave a 64 and 64.3% reduction, respectively, while the treatment without oil afforded reductions of 50.4 and 64.3%. Evaluation of disease control was made on 26 March 1970. The 1-lb. plus oil, 0.5-lb. plus oil and 1-lb. without oil applications reduced the number of blighted shoots 68.8, 63.7, and 44.4%, respectively, as compared to the control. The addition of oil to benomyl seems to enhance the degree of control of almond brown rot.

Cotton disease control with benomyl and thiabendazole. C. D. RANNEY (ARS, USDA, Delta Branch Exp. Sta., Stoneville, Miss.). Field studies indicated that a degree of seedling disease control can be obtained with relatively low rates of either methyl-1-(butylcarbamoyl)-2-benzimidazolecarbamate (benomyl) or 2-(4-thiazolyl) benzimidazole (thiabendazole). Thiabendazole appeared to be more phytotoxic than benomyl, particularly when on, or immediately adjacent to, the cottonseed. Multiple applications, seed, soil, and early season foliage sprays of both systemic fungicides, significantly reduced incidence of *Verticillium* wilt and in some instances significantly increased yield of seed cotton. Both fungicides, applied as foliage sprays late in the season at low rates, reduced losses due to boll rot. A higher level of disease control and higher yields were consistently obtained when cotton was treated with benomyl rather than with an equal rate of thiabendazole. Results indicate that control of several cotton diseases can be obtained from timely and effectively placed applications of these systemic fungicides at relatively low total rates.

Isozyme changes in pea wilt. M. N. REDDY & M. A. STAHMANN (Univ. Wisc., Madison). During pathogenesis of *Fusarium oxysporum* f. sp. *pisii* race 1 on peas, isozyme patterns of various enzymes were examined by polyacrylamide and starch-gel electrophoresis. The isozyme bands from diseased plants were generally more intense than those from healthy plants. The number of isozymes of catalase, acid and alkaline phosphatases, glutamic dehydrogenase, and 6-phosphogluconate dehydrogenase were not changed. Positively charged peroxidase isozymes were increased. The number of esterase isozymes increased, and their mobility and intensity were altered. New isozymes of NAD- and NADP-dependent malate dehydrogenases and glucose-6-phosphate dehydrogenase were observed.

The development of cabbage yellows in organic and mineral soils of Ontario. A. A. REYES (Can. Dep. Agr.,

Vineland, Ont.). Field surveys in Ontario showed that cabbage yellows (*Fusarium oxysporum* f. sp. *conglutinans*) was less severe on plants grown in organic soils than on those grown in mineral soils. To test the validity of this observation, disease development was compared in an organic (muck) and 3 mineral soils (clay, loam, clay-loam) at two levels of fertility. Seedlings of moderately-resistant Marion Market cabbage were inoculated with a mixture of 7 Ontario isolates of the fungus. Plants were grown at 60% relative humidity for 4 weeks at soil and air temp of 26 C and 22 C, respectively, and illuminated for 14 hr with artificial light at 1,000 ft-c. Disease intensity was determined by severity of symptoms on the leaves, degree of vascular discoloration in the stem, decrease in wt of plants, and ease of isolation of the fungus from inoculated plants. Results showed that unfertilized plants grown in the organic soil had milder disease symptoms than plants with the same treatment grown in any of the three mineral soils. This effect was not observed when the plants were given a weekly application of a 20-20-20 fertilizer solution. There were no consistent differences in symptoms on plants grown in any of the mineral soils irrespective of the fertilizer treatment.

A simple method for maintaining Venturia inaequalis inoculum. A. E. RICH (Univ. N.H., Durham). Conidia of *Venturia inaequalis* were washed from infected apple leaves with tap water, frozen, and held at -10 C until needed for inoculation. The conidial suspension was thawed, diluted to approximately 100 conidia/low-power ($\times 10$) microscope field, and atomized onto healthy young leaves of McIntosh apple seedlings which were then incubated at 18 C and 100% relative humidity for 24 hr. The plants were then removed to a greenhouse bench where apple scab symptoms developed in 10-14 days, thus providing more inoculum which could be used immediately or frozen for later use. This method of storing frozen inoculum is simpler than is maintaining viable inoculum on diseased plants or artificial media. Frozen inoculum remained infective for 6 months to 1 year. Freezing conidia in situ on infected leaves proved unsatisfactory.

The susceptibility of potato varieties to ozone in the field. S. RICH & A. HAWKINS (Conn. Agr. Exp. Sta., New Haven, Univ. Conn., Storrs). Potato foliage turned yellow in many fields in Connecticut, following periods of air pollution in June 1968. The older leaves were most severely injured. The yellowed leaves also developed necrotic spots. These symptoms were duplicated in the greenhouse by exposing young potato plants to ozone at 20 ppm for 2 to 3 hr. In the field, the most severely injured cultivars were Alamo, Alaska, Bake King, Katahdin, Kennebec, Lenape, and Wauseon. Some injured leaves of Wauseon were glazed on the undersurface. The cultivar Superior and the late-maturing cultivars Houma, Ona, and Peconic were less injured than the other cultivars. By the first week of July, new, green foliage developed on all cultivars, hiding the older, yellow leaves.

Properties of a high molecular weight toxin produced by Corynebacterium insidiosum. S. M. RIES & G. A. STROBEL (Mont. State Univ., Bozeman). A toxin causing stem and leaf wilt in alfalfa and tomato plants was isolated in a homogeneous form from shake cultures of *Corynebacterium insidiosum*. Toxin production was opt after 4 days' growth, which corresponded to late logarithmic or early stationary growth phase of the organism. One liter of culture yielded 1.4 g of purified toxin. Biological activity was determined on 10- to 12-day-old tomato seedlings with the wilt-o-meter which quantitatively measures stem wilt. The toxin retained biological activity after heating at 121 C for 2 hr, and could not be inactivated by dialysis against pH 2.0 and 12.0 buffers. Purification was accomplished by passage of the toxin through Dowex 1 and 50, and by acetone and ammonium sulfate precipitations. Criteria of homogeneity was indicated by gel filtration (Sephacrose 2B) and with high voltage paper and zone electrophoresis. Sepharose 2B,

light scattering, and ultracentrifugation indicated a mol wt of approx 5×10^6 . The toxin had a blue chromophore in the visible spectrum due to copper chelation at a concn of 1 $\mu\text{gCu}/\text{mg}$ toxin. Removal of Cu by EDTA treatment altered the absorbance by the toxin at 280 and 630 μm without affecting biological activity. Acid hydrolysis indicated that the toxin is a galactose-fucose polymer.

Inhibition of *Erwinia amylovora* by *Erwinia herbicola*. J. H. RIGGLE & E. J. KLOS (Mich. State Univ., E. Lansing). A yellow bacterium obtained from fire blight cankers on pear was shown to be *Erwinia herbicola* as defined by Dye. Preliminary comparison of this bacterium with Dye's *E. herbicola* and Goodman's 35-A indicated that the three were closely related. In a local orchard, the yellow bacterium and *E. amylovora* occurred together within fire blight cankers. Both organisms had a peak in isolation frequency at about 3 inches within the dead tissues during February. Isolations of *E. herbicola* and *E. amylovora* were obtained only slightly less often in 1-year-old wood than in older wood. A reduction in pear blossom infections was obtained when a suspension of *E. herbicola* was spray-inoculated into the blossoms 24 hr prior to inoculation with *E. amylovora*. An attempt was made to duplicate this effect in vitro. *Erwinia herbicola* was grown for 24 hr on a simulated nectar medium. After filter sterilization, the remaining solution would not support growth of *E. amylovora*. Growth of *E. amylovora* resumed when amino acids were supplied and the pH was adjusted to neutrality.

Viral infection of apparently uninjured leaves as influenced by particle morphology and host species. D. A. ROBERTS (Univ. Fla., Gainesville). The polyhedral viruses of southern bean mosaic and tobacco necrosis moved through the xylem and systemically infected, apparently uninjured trifoliate leaves of local-lesion bean hosts following hypodermic-needle injection of virus into stems below portions whose phloem had been killed by scalding. No such infections occurred in 95, 75, and 46 bean plants similarly inoculated with, respectively, the rod-shaped viruses of alfalfa mosaic, tobacco mosaic, and bean yellow mosaic. But infection of uninjured leaves is not entirely a function of particle morphology. Tobacco necrosis and cucumber mosaic viruses did not systemically infect a total of 330 cowpea plants inoculated by the stem-injection method. Tobacco ringspot virus did not infect leaves of 92 injected cowpea plants or 82 bean plants, but caused systemic necroses in leaves of approx one-fourth of the injected plants of a local-lesion host, lima bean (*Phaseolus lunatus* L. 'Jackson's Wonder'). Viral infection of uninjured leaves thus depends on species of host plant as well as particle morphology.

General resistance (slow mildewing) to *Erysiphe graminis* f. sp. *tritici* in Knox wheat. J. J. ROBERTS & R. M. CALDWELL (Purdue Univ., ARS, USDA, Lafayette, Ind.). Seedlings of Knox wheat have been highly susceptible to powdery mildew since its origin over 25 years ago. At maturity in field test nurseries over this period, it has suffered only 30% or less infection under heavy inoculum loads when adjacent cultivars suffered severe infection. The Knox resistance has been subjected to a critical test for general resistance through the production of the Knox cultivar or Knox 62 on over 1.7 million acres annually from 1954 to 1969. During this period, the original level of the Knox general resistance has remained undiminished, while several specific (monogenic) resistances have been broken by new virulent races in the greenhouse and field at Lafayette. Replicated natural infection tests of mature plants in the greenhouse have confirmed the field results. The general resistance of Knox has been transmitted to many of its progeny in the Purdue-USDA breeding program. It has been easily detected in conventional progeny-row tests under heavy inoculum loads in breeding nurseries. Both infection numbers and pustule size on Knox appear to be of lesser magnitude than in highly susceptible varieties, although the two may not be associated in hybrid progeny.

Enzymatic activities in soil from single and mixed culture of *Rhizoctonia solani* and *Fusarium oxysporum* f. sp. *vasinfectum*. R. RODRIGUEZ-KABANA & E. A. CURL (Auburn Univ., Auburn, Ala.). Enzymatic activities were determined directly in soil of either single or mixed cultures of *Rhizoctonia solani* (RS) and *Fusarium oxysporum* f. sp. *vasinfectum* (FOV). Both fungi showed activities of amylase, α -glucosidase, aryl phosphatase (substrate = disodium phenyl phosphate), acetyl esterase (substrate = phenyl acetate), and catalaselike activity. Optimal pH range for acetyl esterase activity was 6.5-7.0; other activities were max at pH values between 5.5-6.5. Enzymatic activities per g of dry soil were higher in FOV than in RS soils. When mycelial homogenates of each fungus were added to autoclaved soil, a linear relationship was found between enzymatic activity and amount of mycelial dry matter. Changes in enzymatic activity with time in monocultures of the fungi in soil revealed characteristic lag, log, and stationary phases of growth. In a mixed culture of the fungi in soil maintained for 8 days, growth of each fungus was followed by assaying for specific enzymatic activities. Maximal growth rate for both fungi was attained during the first 4 days after inoculation. Growth of RS was much faster than that of FOV, which did not develop to a great extent in the mixed culture. In the mixed culture, most nutrients were removed during the first 4 days of the experiment.

Gradients in horizontal dispersal of cereal rust uredospores. A. P. ROELFS (Cooperative Rust Lab., Univ. Minn., St. Paul). Gradients were determined with uredospores of *Puccinia graminis* and *P. recondita* dispersed in and around a 72-m diam source plot of *Triticum aestivum* 'Lee' infected with stem and leaf rust. Rod impaction traps, 15 cm above the crop canopy, were stationed on annuli inside and outside of the plot to sample the dispersed spores. Inside the source, numbers of spores increased within the first 9 m from the "upwind" edge of about 85% of the number trapped over the "downwind" edge of the source. At 9 m or more inside the source, however, the numbers of spores per station on all annuli were statistically similar. The omnidirectional horizontal gradient observed outside the source showed that 14% of stem rust and 24% of leaf rust spores leaving the source were still in this horizontal plane at 100 m. A larger proportion of spores from leaf rust than from stem rust reached each annulus outside the source, indicating some difference in the spore dispersal of the two rusts. The gradient for the number of uredospores per cm^2 on the downwind axis from the source showed that 10% of the spores of each rust species reached 100 m. This downwind gradient, described by the regression equation $\log \hat{Y} = \log a + bX$, approximated Gregory's prediction.

Comparison of inoculum levels of *Ceratocystis paradoxa* on disease development in pineapple fruit and planting material. K. G. ROHRBACH & W. J. APT (Dole Co., Honolulu & PRI, Wahiawa). Chlamydospore-conidia inoculum was applied at various densities to fresh fruit and planting material to induce fruit rot and butt rot, respectively. Spores were also applied to steam-treated and untreated field soil in which pineapple was planted. Disease development on fresh fruit was measured by counting the number of infected fruit, the number of infections per fruit, and the per cent of each fruit rotted after a 7- to 9-day incubation period. Disease development in planting material was measured in field studies by assessing mortality and the wt of survivors. The rotted area of longitudinal sections was measured in laboratory tests of disease development. The number of infected fruit increased significantly with 1×10^4 spores/fruit. In contrast, 1×10^6 spores were required to significantly increase the severity and number of infections per fruit. In a field test, 2,000 spores/quarter crown section were required to obtain a significant increase in disease when compared to a noninoculated check. In laboratory tests with whole crowns, 20,000 spores/g of field soil and 200 spores/g of steamed soil were required to obtain a significant increase in disease incidence.

Fungal colonization of cottonseed prior to harvest. R. W. RONCADORI & S. M. MCCARTER (Univ. Ga., Athens). From September through November 1969, fluffed seed cotton samples were collected from 13 locations throughout Georgia to determine the fungi present, their survival in storage, and their relationship to germination and free fatty acid (FFA) content of seed. Excessive moisture at harvest resulted in poor seed germination ranging from 11 to 81%, with 3 lots germinating at 70% or more. FFA content varied from 1.2 to 15.3%, with only three lots below 2.0%. Poor germination and high FFA were usually associated with extensive fungal colonization. Frequency of fungal isolation from whole seed after harvest ranged from 27 to 88%, with eight lots having more than 50% of the seed invaded. The incidence of embryo infection was lower, varying from 8 to 67%, with seven lots having more than 30% invasion. Fifteen genera of fungi were identified, the most prevalent being *Alternaria* sp., *Diplodia gossypina*, *Fusarium moniliforme*, *F. roseum*, *Glomerella gossypii*, and *Phomopsis* sp. Storage of the seed at 15 C and 7.0 to 8.5% moisture content for 4 to 5 months reduced fungal survival by 20 to 30% in some lots. Abundant inoculum was still present the next growing season, and could be involved in seedling disease problems.

Corn stunt agent isolated from sorghum. E. ROSENKRANZ (ARS, USDA, Miss. State Univ., State College). Since 1948, the known host range of the corn stunt agent (CSA) has been limited to maize and teosinte. In 1969, several severely stunted plants of sorghum cultivar DeKalb FS 24 were noticed in the field at State College, Miss. On one of these plants, two leaves exhibited marginal chlorosis and laminar constrictions, symptoms typical of the Mississippi corn stunt in greenhouse-grown corn. *Graminella nigrifrons*, but not *Dalbulus maidis*, was able to acquire the etiological agent from this sorghum plant and to transmit it to two diverse maize genotypes: sweetcorn cultivar Seneca Chief and dent corn inbred Mp 486. The CSA from sorghum (CSA-S) was recovered repeatedly with noninfective *G. nigrifrons* from corn test plants originally inoculated by leafhoppers which had fed on the field-diseased sorghum. Symptoms of disease in test plants were (i) proportional stunting resulting in miniature plants one-fourth to one-third normal size; (ii) rudimentary tassel; (iii) bare ear; and (iv) a high incidence of "tassel seed". The shortest observed incubation period of CSA-S in corn was 10 days. CSA-S appears to be identical with the Ohio corn stunt agent discovered by the author in 1968. Thus, sorghum is the second economically important crop to prove susceptible to corn stunt and the third species in the host range of CSA.

Effect of a mycorrhizal Endogone on soybean yield. J. P. ROSS & J. A. HARPER (ARS, USDA, & N. C. State Univ., Raleigh). An isolate of *Endogone*, a vesicular-arbuscular mycorrhizal fungus producing chlamydospores, was obtained from a Mississippi field with a history of high soybean yields. The fungus was cultured monoxenically on soybean roots and increased on soybean roots in the greenhouse. Soil and roots from these cultures were used to infest 1 × 1.5 × 0.9-m deep plots containing sandy loam, previously fumigated with methyl bromide, chloropicrin, and methylisothiocyanate. Two hundred or 1,900 g of soil and roots containing *Endogone* constituted treatments; roots and soil free of *Endogone* were added to control plots. Yields from three-replicate plots infested with low and high inoculum levels were 37 and 43% higher, respectively, than yields from controls. Mycorrhizal plants accumulated twice as much phosphorus and significantly greater amounts of nitrogen, calcium, copper and manganese in their foliage as did nonmycorrhizal plants. In fumigated field trials, the addition of *Endogone* increased yields 29% over those from fumigated, noninfested controls. Addition of *Endogone* to nonfumigated soil did not increase yields over the controls. *Endogone* mycorrhizae can have a significant influence upon soybean yields, and

its effect may be markedly affected by competing soil microorganisms.

Combining seedling and adult resistance to Puccinia graminis f. sp. avenae. P. G. ROTHMAN (ARS, USDA, Univ. Minn., St. Paul). Race 6AFH of *P. graminis* f. sp. *avenae*, which has virulence for all known hexaploid genes, has been reported in the USA, Canada, and Kenya. A search for a gene in the host conferring adequate protection to this race has been unsuccessful. Recessive genes described recently offer effective resistance to this race if combined in a common background. Gene pg 12 confers seedling resistance to all prevalent races of oat stem rust, but does not condition mature plant resistance. Gene pg 11 confers resistance in the adult plant but not in the seedling. F₁ plants were obtained from the cross C.I. 3034 (pg 11)/C.I. 8250 (pg 12). F₂ plants were tested in seedling and adult stages to 6AFH. Four classes of F₂ progenies indicated that both genes were independent. F₃ progeny lines were field-planted and tested to race 6AF and 6AH. Resistant reactions were recorded on 81 adult plants. Seedling tests on F₄ progenies of these lines to 6AFH screened out the susceptible lines and confirmed the adult resistance of the remaining lines.

In vitro effects of potassium azide on soil-borne fungi. M. C. RUSH, C. W. AVERRE, S. F. JENKINS, & A. KHAN (N. C. State Univ., Raleigh). Potassium azide (KN₃) is a broad spectrum biocide having herbicidal, nematocidal, and fungicidal activity. In agar incorporation tests using fungal plant pathogens, it was found that the compound possessed both fungistatic and fungicidal properties at concn from 5 to 500 ppm. KN₃ was incorporated into an agar medium (1% glucose-2% yeast extract-1.5% agar) at various concn, and the plates were inoculated with fungi and incubated at 27 C for 6 days. The final pH of the test media varied from 6.8 to 7.5. Complete inhibition of hyphal growth was observed at concn of 10-50 ppm. The compound was fungicidal at 100-500 ppm with the following fungi: *Fusarium moniliforme*, *F. roseum*, *F. oxysporum*, *Verticillium albo-atrum*, *Rhizoctonia solani*, *Sclerotium rolfsii*, *Thielaviopsis basicola*, *Pythium aphanidermatum*, *P. debaryanum*, and *P. ultimum*. KN₃ was more effective and broader in spectrum than captan [*N*-(trichloromethylthio)-4-cyclohexene-1,2-dicarboximide], PCNB (pentachloronitrobenzene), and Dexon (*p*-dimethylaminobenzenediazo sodium sulfonate). These results indicate that KN₃ has considerable promise as a soil fungicide.

Mycorrhizal enhancement of water transport in soybean. G. R. SAFIR, J. S. BOYER, & J. W. GERDEMANN (Univ. Ill., Urbana). Whole plant resistances to water transport, determined by recovery and steady transpiration methods, were lower in mycorrhizal than in nonmycorrhizal soybeans. Stem plus leaf resistances were similar in mycorrhizal and nonmycorrhizal plants. Therefore, the differences observed were due to differences in root resistance to water transport. The reduced resistance of mycorrhizal soybeans appeared after stimulation of shoot growth by the mycorrhizae, but could not be attributed to stimulation of root growth.

Osmotic potential, a controlling factor in the development of bacterial spot diseases. J. M. SASSER, R. W. MILLER, & D. J. FIELDHOUSE. (Univ. Del., Newark). *Xanthomonas vesicatoria* will not divide normally at osmotic potentials occurring in pepper leaf intercellular fluid. Water congestion (water-soaking) of leaves during periods of high relative humidity and rainfall serves to reduce this osmotic potential by dilution, allowing bacterial division and expression of pathogenicity. Thus, the osmotic potential of pepper leaf intercellular fluid under nonwater-congested conditions normally inhibits population increase of *X. vesicatoria*. This hypothesis proposes that the primary effect of water congestion is not predisposition of pepper leaves to disease

development, but that water congestion diminishes the repressive effect of a high osmotic potential in intercellular fluid on bacterial multiplication.

Sugar accumulation associated with Hypoxylon mammatum canker development in aspen. A. L. SCHIPPER, JR. (USDA Forest Service, St. Paul, Minn.). Quaking aspen (*Populus tremuloides*) saplings with cankers caused by *Hypoxylon mammatum* were sampled for free sugar content during 1968 and 1969. An accumulation of free sugars was found in the xylem 3 to 6 cm above the apex and at the base of each canker. Gas chromatography of these sugars indicated that they were from the phloem translocation stream rather than being products of cellulose and hemicellulose hydrolysis. This accumulation is probably the result of phloem necrosis rather than of active accumulation by the fungus. But it seems likely that *H. mammatum* uses these sugars to support its growth in the xylem, as this region of sugar accumulation corresponds with the location of the advancing edge of the fungus mycelium.

Viral antigen in clover yellow mosaic virus infections. D. E. SCHLEGEL & D. DELISLE (Univ. Calif., Berkeley). Clover yellow mosaic virus-infected *Vicia faba* leaf cells contain structures filled with amorphous material in both the cytoplasm and the vacuole. Electron microscope immunoradiography of such leaf tissue treated with virus-specific ^{125}I -labeled antibodies showed that these structures contained substantial amounts of virus protein. The structures first appeared ca. 4 to 5 days after inoculation of leaves 2 whorls below those sampled, and were not found in noninoculated tissue. The leaves sampled were approximately 1 cm long at the time the plant was inoculated. The appearance of such structures coincided with the first recovery of infectivity from these leaves. As time after inoculation increased, they tended to disappear and were not seen in 12-day infections. No virus particles could be distinguished in the amorphous material.

Effect of sequence and time of inoculation with soybean mosaic and tobacco ringspot viruses on yield of soybean cultivars. A. F. SCHMITTHENNER & D. T. GORDON (Ohio Agr. Res. Development Center, Wooster). In small field plots, 11 soybean cultivars were inoculated with soybean mosaic virus (SMV) and/or tobacco ringspot virus (TRSV) during five successive periods, beginning on the half-expanded, second trifoliolate leaf and continuing on each successive trifoliolate when half expanded up through the sixth. SMV reduced yields (11%) over all cultivars for periods 1-4 only. TRSV reduced yields 60% for periods 1-3, 49% for period 4, and 31% for period 5. Simultaneous inoculation with SMV and TRSV reduced yields 84% for periods 1 and 2, 75% for period 3, 63% for period 4, and 43% for period 5. Amsoy, Beeson, Calland, Corsoy, Harosoy 63, and Wayne had losses over all periods of 50-69% for TRSV and 76-86% for SMV plus TRSV: A-100, Chipewa 64, Ford, Clark 63, and Cutler were less susceptible with losses of 37-40% for TRSV and 47-69% for SMV plus TRSV. Four cultivars inoculated with SMV at period 1 followed by TRSV at period 2 or 3 yielded similarly to those inoculated with both viruses at period 2 or 3. Results were the same with the sequence of SMV and TRSV inoculated in reverse order. However, sequential inoculations beginning with period 3 resulted in greater yield losses than simultaneous inoculations at periods 4 and 5, except for one cultivar.

Cross-protection in Gossypium hirsutum infected with vascular wilt fungi related to host-pathogen specificities. W. C. SCHNATHORST (USDA, Univ. Calif., Davis). The induction of gossypol and related compounds was reported to be a nonspecific response of cotton (*Gossypium hirsutum*) tissue. Induction of these chemicals in verticillium wilt-tolerant and -susceptible cottons should lead to protection from inoculations of highly virulent vascular pathogens. In the greenhouse, susceptible Delta Pine 15 cotton was root-inoculated with weakly virulent isolates of *Verti-*

cillium albo-atrum and *V. nubilum* and challenged several weeks later on roots or stems with the virulent T-1 strain of *V. albo-atrum*. Fusarium-resistant Auburn 56 and verticillium-tolerant Acala 4-42 were used in reciprocal cross-protection tests with *Fusarium oxysporum* f. sp. *vasinfectum* and SS-4 and T-1 strains of *V. albo-atrum*. Acala 4-42 inoculated with SS-4 or milder isolates and challenged with T-1 served as the control protection responses. All experiments except the controls failed to demonstrate a long-term protective response. Cross-protection appears to be a specific response associated only with certain strains of *V. albo-atrum* and tolerant cottons, and resembles the specificity that I earlier reported for the formation of inhibitor(s) in xylem fluid.

Interactions between several viruses and the satellitelike virus of tobacco ringspot virus. I. R. SCHNEIDER (Plant Virol. Lab., USDA, Beltsville, Md.). Interactions between viruses, including strains of tobacco ringspot virus (TRSV), and a new satellitelike virus (Satellite) resembling TRSV, were explored. Satellite was not activated by tobacco necrosis virus or by tomato ringspot virus. Satellite was activated by all strains of TRSV tested except the Eucharis mottle strain, which is serologically related to, but not serologically identical to, the others. When the ST-strain of TRSV activated a source of Satellite, many lesions appeared from which no infectious virus could be extracted. These lesions, which contained mostly Satellite, were phenotypically distinct from those containing only ST-TRSV. In contrast, when the WS-strain of TRSV activated the same source of Satellite, the lesions from which no infectious virus could be extracted were phenotypically indistinguishable from those containing only WS-TRSV. The density-gradient profile of purified Satellite varied significantly when the strain of TRSV used in the inoculum to increase Satellite was changed from the ST- to the WS-strain. The phenotype of lesions containing almost 100% Satellite was more affected by the strain of TRSV than by the previous history of the Satellite used in the mixed inoculum.

Ribonucleic acid synthesis during the development of zoosporengia of Aphanomyces euteiches. C. L. SCHOUTIES & C. Y. YANG (Univ. Ky., Lexington). The role of ribonucleic acid (RNA) synthesis in the development of zoosporengia of *Aphanomyces euteiches* was studied. The fungus was induced to sporulate by replacing the medium with a solution having the following ionic composition: $1.5 \times 10^{-3} \text{ M CaCl}_2 \cdot \text{H}_2\text{O}$, $9.2 \times 10^{-4} \text{ M MgSO}_4 \cdot 7\text{H}_2\text{O}$, $1.5 \times 10^{-3} \text{ M KCl}$. Under these conditions, mycelial mass decreased, whereas in noninduced controls, mycelial mass increased. Levels of RNA remained constant during the period of zoosporengial development, while RNA levels in noninduced controls decreased. An inhibitor of pyrimidine biosynthesis, 6-azauracil, at concn of 1-100 $\mu\text{g/ml}$ did not inhibit sporulation, suggesting that synthesis of additional pyrimidine nucleotides was not required for differentiation. Complete inhibition of sporulation was obtained with actinomycin D at 25 $\mu\text{g/ml}$, indicating that new species of RNA were required for differentiation.

Resistance to watermelon mosaic virus 2 in Pisum sativum conditioned by the gene for resistance to bean yellow mosaic virus. W. T. SCHROEDER & R. PROVIDENTI (N. Y. State Agr. Exp. Sta., Cornell Univ., Geneva). Plants of *Pisum sativum* resistant to bean yellow mosaic virus (BYMV) are also resistant to watermelon mosaic virus 2 (WMV-2). Thirty pea cultivars (*mo mo*) resistant to BYMV were resistant to WMV-2. A comparable number of BYMV-susceptible cultivars were also susceptible to WMV-2. Population halves from F_1 and F_2 families of crosses between resistant and susceptible pea cultivars were inoculated with BYMV or WMV-2. Respective segregations indicated that *mo* gene conditioned the plants for resistance to each virus. Analogous reactions of clonal cuttings from F_2 plants to the two viruses authenticated a common gene for resistance. WMV-2 also exhibited two other characters

common to BYMV on pea; namely, influence of temp on symptoms in heterozygotes and appearance of thermal mutants that overcome *mo mo* resistance.

Pathogenic variation and overlapping host ranges in Pseudomonas phaseolicola, Pseudomonas glycinea, and Pseudomonas mori. M. N. SCHROTH, D. C. HILDEBRAND, & VILMA VITANZA (Univ. Calif., Berkeley). Inoculation of different germ plasm of *Phaseolus vulgaris*, *Phaseolus lunatus* f. *macrocarpus*, *Phaseolus limensis*, and *Glycine max* with varying strains of the nomenspecies *Pseudomonas phaseolicola*, *P. glycinea*, and *P. mori* revealed no clear distinction in the host ranges. None of the nomenspecies were host-specific; however, there was a pronounced tendency of the strains to segregate along their commonly accepted host lines. *Pseudomonas phaseolicola* strains usually attacked fewer soybean varieties and were less virulent than *P. glycinea* strains. The reciprocal also was true. Both species were highly virulent on lima bean. *Pseudomonas mori* isolates produced water-soaked lesions and symptoms identical to *P. glycinea* and *P. phaseolicola* on both lima bean and French bean. None of the *P. phaseolicola* and *P. glycinea* strains infected mulberry. There was considerable variation in virulence among strains of the three nomenspecies. Virulence differed in two ways. Certain strains were less virulent than others on all hosts tested, whereas virulence of some strains depended solely upon the host. Nutritional studies of the nomenspecies indicated that strains with dissimilar host ranges differed in nutritional requirements.

Component ratio differences in strains of alfalfa mosaic virus. F. W. SCHWENK, S. H. SMITH, & H. E. WILLIAMS (Univ. Calif., Berkeley, Calif. Dep. Agri., Sacramento). Viruses isolated from four woody ornamentals, *Hebe* sp. 'Co-ed', *Ilex cornuta* 'Rotunda', *Viburnum opulus*, and *V. tinus*, were shown to be related to alfalfa mosaic virus (AMV) by serology and particle morphology. Based on differential host reactions, the isolates from the two *Viburnum* spp. are considered distinct strains. Due to lack of observable differences, the isolates from *Hebe* sp. 'Co-ed' and *I. cornuta* are considered to be a single strain. This latter strain was distinguishable from each of the other two. The isolates were partially purified from tobacco by polyethylene glycol precipitation and density-gradient centrifugation. When gradients were removed through a spectrophotometer measuring at 260 m μ and attached to a recorder, three strongly absorbing component bands appeared in comparable places in the gradient for each isolate, though the relative component concn varied. The upper two components appeared within 4 days after inoculation; the lowest component appeared by the 6th day. The isolates from *Hebe* sp. 'Co-ed' and *I. cornuta* produced similar patterns. Each of the strains tested, including one other known strain of AMV, was identifiable by its absorption pattern. This is suggested as a method of AMV identification and of strain differentiation.

Peroxidase isozymes in resistant and susceptible reactions of wheat stem rust controlled at the Sr6 locus. P. SEEVERS & J. M. DALY (Univ. Neb., Lincoln). Previous studies have indicated increases in total peroxidase activity, at 20 C, of isogenic lines of wheat carrying the temp sensitive *Sr6* allele for resistance to race 56 of *Puccinia graminis tritici*, but not in susceptible lines. Temperature or ethylene treatments cause reversal to susceptibility in resistant lines without decreases in peroxidase levels. The existence of multiple forms of peroxidase allows for the possibility that a specific form associated with resistance may be affected by temp or ethylene. Quantitative spectrophotometric determinations of peroxidases separated by gel electrophoresis showed a min of 14 peroxidases in healthy wheat tissue. Resistant and susceptible reactions caused similar increases in several isozymes, but only one detectable band was consistently associated with resistance. But temp and ethylene treatments did not appear to influence its activity during reversion from resistance to susceptibility.

Biological and physico-chemical properties of an aberrant mutant of tobacco mosaic virus. O. P. SEHGAL (Univ. Mo., Columbia). A tobacco mosaic virus (TMV) mutant was isolated from a population of TMV-RNA molecules inactivated to 0.1% survival by nitrous acid treatment. This mutant is very poorly transported in Samsun tobacco and induces chlorotic spots, oak-leaf patterns, ringspots, and necrosis. Its specific infectivity is only one-twentieth that of the parent strain. Rate zonal sucrose density-gradient and equilibrium centrifugations in sucrose and CsCl indicate that the purified mutant preparations contain a heterogeneous population of particles. These vary in length, but ca. 15 to 20% are infectious rods of standard (300 nanometers) length. Purified RNA from the mutant contain an overabundance of 7 to 8 Svedberg units (S) and a few 30 S molecules; only the latter are infectious. Compared to the parent strain, the mutant coat protein contains two additional residues of arginine and one of glycine, but lacks one residue each of glutamic acid, serine, and threonine. These observations, and other data based on temp sensitivity, exposure to urea or formamide, and serology, suggest that the mutant particles are inherently unstable and undergo rapid fragmentation and depolymerization into the viral protein and nucleic acid moieties.

Correlation between buoyant density and ribonucleic acid content in viruses. O. P. SEHGAL, JONG-HO JEAN, R. B. BHALLA, MEEI MEEI SOONG, & G. F. KRAUSE (Univ. Mo., Columbia). Isopycnic CsCl centrifugation method was used to determine the possible correlation between ρ and RNA content of eight viruses with nucleic acid content from 4 to 42%. These viruses, barley stripe mosaic, tobacco mosaic, cucumber mosaic, bromegrass mosaic, southern bean mosaic, MS2, pea enation mosaic, turnip yellow mosaic, and tobacco ringspot, showed a highly significant correlation ($R^2 = 0.99$) between ρ and RNA content. The functional relationship indicated that the ρ of viruses increased at a greater than linear rate as the RNA content increased, but ca. 89% of this variation was directly attributed to the absolute amount of RNA in the virions. Based on these observations, a mathematical formula was derived to estimate the RNA content of a virus from its ρ value. Reliable estimate of the RNA content of a virus can be made by this method if such experiments are supplemented with appropriate infectivity tests, even of viruses not amenable to rigorous purification procedures.

Multiple infectious forms of a free-RNA plant virus. J. S. SEMANCIK & L. G. WEATHERS (Univ. Neb., Lincoln, Univ. Calif., Riverside). Phenolized preparations from exocortis virus-infected (CEV) *Gynura aurantiaca* displayed multiple-sedimenting forms of infectious RNA after rate sedimentation in log-linear sucrose density-gradients. The major area of infectivity at 9 Svedberg units (S) was more homogeneous than the secondary regions at 19 and 36 S. Fractionation of the phenol extracts by differential salt precipitation and "winding" of DNA did not enrich the preparations for any of the specific sedimenting forms. The 2 M LiCl supernatant preparation, containing principally soluble-RNA, displayed the greatest infectivity with a dilution threshold of 0.04 μ g/ml when assayed on *G. aurantiaca*. Formaldehyde treatment destroyed the infectivity in all preparations. LiCl supernatant preparations heated to 60 and 100 C, fast-cooled, and centrifuged in log-linear gradients, displayed increasing levels of infectivity in the 19- to 36-S region in addition to the typical 9-S moiety. The infectivity distribution of CEV, after equilibrium sedimentation in Cs₂SO₄, was more heterogeneous and at a lower density (1.575) than was single-stranded cowpea mosaic virus RNA (1.615). These properties suggest that the infectious CEV-RNA represents an atypical viral nucleic acid characterized by either a low mol wt or circular structure or by unusual aggregation properties.

Mycotoxins in Aspergillus. G. SEMENIUK, G. S. HARSFIELD, C. W. CARLSON, C. W. HESSELTINE, & W. F. KWOLEK

(S. Dak. State Univ., Brookings, ARS, USDA, N. Regional Res. Lab., Peoria, Ill.). The toxigenicity of 392 strains of *Aspergillus*, representing 134 species and 17 varieties, was assessed on six chicks and six mice fed iso-caloric 50% basal feed mixes of wheat and soybeans molded by these strains. On the basis of death numbers, low wt gain of survivors, and low feed consumption over a 4-week period, 164 of the strains, representing 73 species and 9 varieties, were toxigenic. Ten species (*A. alliaceus*, *A. avenaceus*, *A. clavato-flavus*, *A. janus*, *A. melleus*, *A. ochraceus*, *A. parasiticus*, *A. quercinus*, *A. sclerotiorum*, and *A. sulphureus*) had strains that were markedly toxic (three to six deaths) to one or both animal types on both feeds; the other 62 species and 9 varieties had strains that were moderately to mildly toxic (zero to two deaths) to one or both animal types on one or both feeds. In retests of some strains, toxigenicity varied between successive preparation of molded feed.

Pepper leaf intercellular fluid composition after inoculation with Xanthomonas vesicatoria. M. G. SINCLAIR, J. M. SASSER, & T. J. GULYA (Univ. Del., Newark). The intercellular fluid of plants consists of nutrients which are readily available for utilization by pathogens. Experiments were conducted to determine which constituents might increase during pathogenesis. Leaves of pepper plants susceptible to *Xanthomonas vesicatoria* were inoculated by injection of 10^8 cells/ml of the bacterium at 12-hr intervals up to 96 hr. The intercellular fluid was collected by centrifugation after the leaves were vacuum-infiltrated with water. Following filtration to remove the bacteria, the extract was analyzed for carbohydrates, amino acids, protein, cond, pH, and various cations. Carbohydrates, amino acids, protein, and cond all increased 10-fold over their initial levels. The pH rose slightly over the 96-hr period. Magnesium and K increased to a level 10 times higher than originally present, while Na increased 3-fold. Little change was recorded for Mn, Cu, Ca, and Fe. Steadily increasing levels of some of the constituents of the intercellular fluid suggest that the infected leaf accumulates materials normally available to the remainder of the plant.

Occurrence, symptoms, and diagnostic hosts of strains of potato spindle tuber virus. R. P. SINGH (CDA, Fredericton, N.B., Canada). Two hundred and thirty-seven potato samples suspected of being infected with spindle tuber virus were tested, using the double-inoculation technique. Two hundred and two samples were found to be infected; 174 of these were infected with the mild strain and 28 with the severe strain. Symptoms produced in virus-free stocks of three potato varieties in the greenhouse were stunting of plants, rugosity of leaves, and necrosis of stems. These symptoms were severe when stocks were inoculated with the severe strain, and slight to moderate when the mild strain was used. About 100 species belonging to the family Solanaceae were screened for diagnostic hosts for both strains. The plants of the tomato cultivar Allerfrühestefreiland, *Scopolia cariviolicca*, *S. lurida*, *Solanum aviculare*, *S. aviculare* var. *albiflorum*, and *S. depilatum* reacted with diagnostic symptoms to the severe strain, and the first three species also reacted to the mild strain. The symptoms caused by either strain were rugosity of leaves, stunting of plant, and extensive veinal and stem necrosis. Other species were symptomless carriers, and a few were immune to both strains.

A geographically isolated source of oxides of nitrogen and its effect on surrounding conifer species. J. M. SKELLY, L. D. MOORE, & L. STONE (Va. Polytechnic Inst., Blacksburg). Four species of conifers found in proximity to a source of moderately high concn of nitrogen (NO_x) were examined for symptoms. Ambient amounts of NO_x were recorded at seven monitoring stations, and the highest 1-hr concn was > 0.58 ppm at a station located 0.5 miles from the source. Levels of SO_2 at the same station were 0.67 ppm for a 2-hr collection period. A synergistic effect

of these two oxidants may have been in evidence. At a distance of 200 yards from a source of NO_x , *Pinus strobus* seedlings planted 3 years prior to this investigation were severely stunted and showed no appreciable growth since planting. A witches'-broom effect at the tips of branches and tipburn had occurred. At a distance of 1 mile or more from the source, native *P. strobus* had developed significant chlorotic mottle on older needles and some tipburn, and highly sensitive trees were in evidence as indicated by severe chlorosis and significant height growth reduction. Damage to *P. taeda*, *P. echinata*, and *P. virginiana* consisted of chlorotic mottle and, in some instances, severe chlorosis and tipburn. Significant growth losses of individual trees were not evident. Of the four species examined, *P. strobus* was apparently most susceptible.

Susceptible and hypersensitive reactions in tobacco and their prevention by cell-free extracts of Pseudomonas tabaci and Pseudomonas glycinea. H. C. SLEESMAN, J. E. PERLEY, & H. A. J. HOITINK (Ohio Agr. Res. Development Center, Wooster). The susceptible reaction of tobacco leaves (White Burley) to *Pseudomonas tabaci* was delayed 60 hr when leaves were infiltrated with heat-killed cells of *Pseudomonas tabaci* or *P. glycinea* 18 hr before challenging with live cells (5×10^7 cells/ml). Heat-killed cells prevented the hypersensitive reaction induced by live cells of *P. glycinea* (5×10^8 cells/ml). Protection was not systemic. Tests were made on plants grown at 16 hr of light, 2,000 ft-c, and 24 to 27 C. Heat-stable protective fractions were obtained from cells of *P. tabaci* strains Pt 1, Pt 3, and Pt 5, and also from *P. glycinea* strain Ohio 40 by (i) extraction in phenol using the Westphal phenol-water procedure (68 C) and (ii) sonication and fractional precipitation of supernatants using ethanol or $(\text{NH}_4)_2\text{SO}_4$. Insoluble cell wall constituents also induced protection. A heat-labile fraction inducing a hypersensitive reaction was extracted from sonicated cells of *P. glycinea* strain Ohio 40, and small quantities were extracted from culture filtrates. A similar fraction which caused water-soaking followed eventually by necrosis was obtained from *P. tabaci* isolates. The activity was in the 25 to 55% $(\text{NH}_4)_2\text{SO}_4$, and the 30 to 50% ethanol fractions. Pronase destroyed activity of both fractions.

Prevention of powdery mildew on container-grown squash utilized for virus research. D. H. SMITH (Univ. Ga., Ga. Sta., Experiment). Powdery mildew is a serious problem on container-grown squash plants (*Cucurbita pepo* 'Golden Crookneck', 'Early Yellow Summer Crookneck', and 'Caserta') which are utilized in cucurbit virus research. The objective of this study was to develop an effective method for routine control of mildew on container-grown squash. Golden Crookneck squash were treated with benomyl [methyl-1-(butylcarbamoyl)-2-benzimidazole-carbamate]. A benomyl seed treatment (seed immersed in 50 ml H_2O + 1 g benomyl + 1 ml surfactant F for 5 min) gave no mildew control. Excellent mildew control was obtained with a single soil drench application (200 mg benomyl/8-inch pot in 200 ml H_2O) at the first-true-leaf stage of development. Although containers were top-watered twice daily, a single soil drench prevented mildew for 6 to 7 weeks. Weekly soil drenches were equally effective. Weekly and biweekly foliar sprays (5 g benomyl + 1 ml surfactant F + 1 gal H_2O) gave mildew control equal to soil drenches. Marginal chlorosis of the oldest two leaves occurred when benomyl was applied as a soil drench, but not when applied as a seed treatment or foliar spray. Benomyl had no deleterious effects on infection and symptom expression by cucumber mosaic virus, squash mosaic virus, or watermelon mosaic virus.

Control of brown rot and Rhizopus rot of inoculated peaches with hot water or hot chemical suspensions. W. L. SMITH, JR. (USDA, Beltsville, Md.). Peaches treated with hot water or hot suspensions of 2,6-dichloro-4-nitroaniline

(botran) and methyl-1-(butylcarbamoyl)-2-benzimidazole-carbamate (benomyl) (both at 100 ppm) developed significantly less brown and rhizopus rots than those treated with unheated water (21 C) or unheated chemical suspensions. The addition of either chemical to 52 C water did not improve decay control obtained by 52 C water alone. But chemical suspensions in 46 C water did improve decay control over that obtained by 46 C water alone. Peaches treated at 52 C developed less decay than those treated at 46 C. Unheated suspensions of benomyl effectively controlled brown rot development, but did not control rhizopus rot. Unheated suspensions of botran did not control either decay. Unheated or heated suspensions of botran or benomyl did not injure the fruit. In other studies, fruit were often severely injured when treated in 50 C or 52 C water containing *N*-(trichloromethylthio)-4-cyclohexene-1,2-dicarboximide (captan). Fresh captan residues also caused injury on fruit treated in 50 C or 52 C water.

Inactivation in anoxia of germinating spores of post-harvest fungi. N. F. SOMMER & R. J. FORTLAGE (Univ. Calif., Davis). The postharvest fruit fungi, *Rhizopus stolonifer*, *Botrytis cinerea*, *Penicillium expansum*, *Monilinia fructicola*, *Cladosporium herbarum*, and *Gloeosporium perennans* have low oxygen requirements for germination and growth. Responses to anoxia have varied in different reports, possibly because of difficulty in eliminating oxygen contamination. To obtain nearly complete anoxia, anaerobic jars were evacuated and filled with N₂ gas 4 times followed by replacement of the N₂ by H₂ and the catalytic reduction of remaining traces of oxygen. Freshly harvested spores on potato-dextrose agar failed to germinate in anoxia but remained viable. If incubated at 25 C in air before exposure to anoxia, germinants and young colonies became increasingly sensitive as growth progressed. Depending upon the species, max sensitivity to anoxia was reached after about 15 to 30 hr of aerobic incubation. With longer periods of growth, tolerance to anoxia returned. With *M. fructicola* and *B. cinerea*, inactivation was more rapid at 35 C than at lower temp. A prior heat treatment sensitized spores to anoxia.

Relative resistance of tomatoes at various stages of ripeness to bacterial soft rot. D. H. SPALDING & C. S. PARSONS (USDA, Beltsville, Md.). Early work indicated that green tomatoes are more susceptible to bacterial soft rot than red tomatoes. In the present tests, 40% of red tomatoes inoculated with *Erwinia carotovora* (ATCC No. 25271) remained free of bacterial soft rot symptoms during 6 days at 13 C, while 5% of pink tomatoes and none of the turning and green tomatoes remained free of symptoms. Inoculated but decay-free tomatoes held an additional week at 13 C usually did not develop soft rot. At 18 C, 8% of red tomatoes and no green tomatoes remained healthy following inoculation. At 7 C, 100% of red tomatoes and 72% of green tomatoes remained healthy after inoculation. Green-to-red tomatoes did not show statistically significant differences in pH or total phenols, but total acids were less in red tomatoes than in green, turning, or pink tomatoes. Antibiotic sensitivity tests of the four phenolic acids reported present in tomato fruit showed that 0.5 mg of ferulic acid, *p*-coumaric acid, or chlorogenic acid in a filter paper disc inhibited growth of *E. carotovora*, but 1.0 mg of caffeic acid/disc did not inhibit growth.

Association of ammonia with electrolyte leakage from leaves of Capsicum annuum. R. E. STALL, A. A. COOK, & C. B. HALL (Univ. Fla., Gainesville). Volatiles from *Xanthomonas vesicatoria* (XV) cause necrosis of pepper leaves, and NH₃ is among the volatiles produced. The ammonia content and electrolyte leakage from leaves hypersensitive and susceptible to XV were determined following exposure to NH₃ vapors or injection with the bacterium. Both treatments resulted in electrolyte leakage from both types

of leaves. At similar levels of electrolyte leakage, however, the amount of NH₃ obtained from hypersensitive leaves exposed to NH₃ vapors was notably greater than from such leaves inoculated with bacteria. Conversely, at similar levels of electrolyte leakage the NH₃ content of susceptible leaves exposed to NH₃ vapors was similar to the NH₃ content of such leaves inoculated with bacteria. It was concluded that NH₃ is probably not a causal factor in hypersensitivity, but may be involved in susceptibility of pepper leaves to XV.

The resistant reaction in cabbage to Xanthomonas campestris. T. STAUB & P. H. WILLIAMS (Univ. Wis., Madison). Plants of a resistant cabbage inbred line and of the susceptible cultivar Sanibel were inoculated in the growth room with washed suspensions of *Xanthomonas campestris* (OD = 0.5) by either cutting out with a razor blade 2 mm "V"-shaped portions of the marginal hydathode regions under the bacterial suspension (notch inoculation) or by placing 1 µl of suspension on the guttating hydathodes. After notch inoculation of resistant plants, the bacteria moved along the veins and caused vein blackening similar to that in susceptible plants; however, the bacterial movement was slower than in susceptible plants and was temp-dependent. After hydathode inoculations, a restricted marginal necrotic lesion with a dark rim and no vein blackening were characteristic of resistant plants. At 16, 20, 24, and 28 C, the advance of the lesions in resistant leaves (R) between 9-18 days relative to the advance in the susceptible leaves (S) was (R/S) 0.10, 0.36, 0.45, and 0.88, respectively, for the notch inoculations, and 0.37, 0.07, 0.07, and 0.25 for the hydathode inoculations. The bacterial concn required for max infection in both resistant and susceptible plants were 30 times higher for the notch than for the hydathode inoculations.

Sources of resistance to Alternaria alternata in the genus Nicotiana. J. R. STAVELY & G. W. PITTARELLI (ARS, USDA, Beltsville, Md.). We tested 65 of the 67 species in the genus *Nicotiana* for reaction to *Alternaria alternata*, incitant of tobacco brown spot. Greenhouse-grown plants averaging 13 weeks of age were inoculated with a virulent conidial suspension (120,000/ml) containing 0.01% Triton B-1956 (77% modified phthalic glyceryl alkylid resin). They were then incubated for 1 week in plant growth rooms under opt humidity and temp conditions for infection. Symptom severity on stems and leaves was rated 16 to 18 days after inoculation. No species was entirely free of brown spot symptoms in all replications but *N. bonariensis*, *N. nesophila*, *N. stocktonii*, one accession of *N. suaveolens*, and *N. wigandioides* were highly resistant. *Nicotiana acaulis*, *N. debneyi*, *N. goodspeedii*, two accessions of *N. longiflora*, *N. noctiflora*, and *N. repanda* were less resistant. All species in subgenus *Tabacum* and all but *N. thyriflora* in subgenus *Rustica* were susceptible. All resistant, some highly susceptible, and many intermediate reacting species were in subgenus *Petunioides*. Among the most susceptible species were *N. acuminata*, *N. corymbosa*, *N. fragrans*, *N. glutinosa*, *N. otophora*, *N. raimondii*, and *N. trigonophylla*. *N. tabacum* cultivars 'Coker 187 Hicks', 'NC95', and 'PD121' were rated susceptible by the criteria used in these tests.

Resistance to the pea seed-borne mosaic virus. W. R. STEVENSON, D. J. HAGEDORN, & E. T. GRITTON (Univ. Wis., Madison). Field and greenhouse studies of 143 processing pea cultivars from nine seed companies and 551 plant introductions (P.I.'s) were made in search of resistance to the pea seed-borne mosaic virus (PSbMV), sometimes called pea fizzle-top virus. Seedlings 3 to 5 inches tall were inoculated with an atomizer at 16 psi in the field or by rubbing in the greenhouse. Symptoms included characteristic vein clearing, leaf cupping, and stunting reactions. All commercial cultivars and all but two P.I.'s, 193586 and 193835, were susceptible. These two introductions were

tested repeatedly in the greenhouse, using inoculation and back inoculation procedures. P.I. 193835 was a mixture of pink-axiled susceptible plants and green-axiled resistant plants. All seedlings from P.I. 193586 were resistant. Repeated attempts to recover the virus from symptomless plants of either P.I. number, and electron microscope dip preparations from similar plants, failed to indicate the presence of the virus. F_1 progenies of reciprocal crosses between susceptible pea cultivars (A45, Elf, Sprite, Alaska, Alsweet, New Era, and New Season) and the resistant P.I. accessions were all susceptible, showing that resistance is recessive. F_1 hybrids between the two P.I.'s were resistant, indicating that the same gene(s) for resistance are carried by both accessions.

Influence of temperature on symptom expression of pea seed-borne mosaic virus in pea. W. R. STEVENSON & R. E. RAND (Univ. Wis., Madison). The influence of temp on the symptom development of the pea seed-borne mosaic virus (pea fizzle-top virus) in peas was investigated. Alaska and Dark Skin Perfection cultivars were germinated at 24 C and inoculated mechanically by rubbing 14 days after planting. Immediately thereafter they were moved into temp-controlled greenhouses set at 16, 20, 24, and 28 C, where symptom progression was carefully noted. Similar reactions were found for both susceptible pea cultivars. Symptom progression, in order of appearance, included tendril curl, vein clearing, veinbanding, necrosis of inoculated leaves, mosaic, cupping of leaves, plant stunt, and apical malformation. Vein clearing, the most striking early symptom, reached max severity 3 days after inoculation at 28 C, 4 days at 24 C, and 5 days at 20 and 16 C. Apical malformation, generally the final symptom, was evident at all temp within 4 weeks after inoculation, but was apparent first at higher temp. Results indicated that (i) symptom progression was accelerated at higher temp; and (ii) over-all and final severity of symptoms seemed not to be significantly affected by temp.

Isolation and purification of phytoactins from fermentive concentrates. M. F. STONER & S. O. GRAHAM (Wash. State Univ., Pullman). Simple, efficient, and inexpensive means for isolating and purifying phytoactins from crude fermentive conc produced by *Streptomyces hygroscopicus* (NRRL-2752) are not available for physiologic research, selective media, and disease control. The method reported here may be modified to isolate small or large quantities of suitable purity. First, remove solvents from conc in vacuo. Decant any oily supernate and dissolve the remaining semisolids (phytoactins, impurities) in ethanol. Add chilled petroleum ether (30 parts to 1 part extract) and let stand for 10 hr at 2 C. Discard petroleum ether phase. Extract the residue with methyl ethyl ketone (25 ml/g residue) and filter through solvent resistant membrane (0.45 μ pores). Discard the residues and add filtrate to chilled petroleum ether (1:20) to precipitate phytoactins. Collect them on a solvent resistant membrane. Repeating the methyl ethyl ketone-petroleum ether step increases purity. Up to 95% of contained phytoactins have been isolated from conc by this method. These phytoactins can then be separated by reported methods to obtain purified phytoactins A, B, and C.

A new systemic fungicide for the control of rusts. G. E. STORY, J. C. SHARP, & M. W. MOON (The Upjohn Co., Kalamazoo, Mich.). 4-Amino-6-chloro-2-(methylthio)pyrimidine has demonstrated effective control of *Uromyces phaseoli* and *Puccinia graminis* var. *tritici* under greenhouse conditions. A single soil application as either a drench or dust of 5 mg/plant has resulted in rust control for a period of 3 to 4 weeks. At present, our tests have not shown foliar or curative properties.

*Effect of ultraviolet radiation and cholesterol on growth and sporulation of *Cytospora leucostoma* and *C. cincta*.* R. E. STUCKEY & A. L. JONES (Mich. State Univ., E.

Lansing). Cultures of *Cytospora leucostoma* and *C. cincta* were grown on potato-maltose agar and potato-maltose broth adjusted to pH 6. Experiments were conducted in growth chambers maintained at 24 C and 50% relative humidity. Dry wt of cultures irradiated continuously with ultraviolet light (UV) at 320-460 nm (max 365 nm) were less than those irradiated with cool-white fluorescent lamps (400-500 ft-c). Isolates of *C. cincta* grew more slowly than *C. leucostoma* regardless of treatment, and were inhibited more by the UV. Pycnidial formation in UV-irradiated cultures appeared earlier and increased 3 to 100-fold depending on the isolate. Moreover, the percentage of sporulating pycnidia was increased nearly 2-fold in cultures irradiated with UV. Cultures grown in continuous darkness failed to form pycnidia. Addition of 100 ppm cholesterol to the medium had little or no effect with the exception of one *C. leucostoma* isolate (Ma4). Cultures of Ma4 grown under UV or fluorescent lamps produced conidia-like structures in the absence of pycnidia in 4 days, and in 3 weeks produced sporulating pycnidia. The conidia-like structures were more variable in size and shape than spores produced in pycnidia. Ultraviolet light appears to favor the development of pycnidia and shorten the time for sporulation.

Cabbage head rot in the Georgia Coastal Plain. D. R. SUMNER (Univ. Ga., Coastal Plain Exp. Sta., Tifton). Cabbage head rot frequently occurs in both winter and spring crops in southern Georgia. Some heads show no external discoloration when picked, but deteriorate during shipment. Bacteria similar to *Pseudomonas cichorii*, *P. maculicola*, *Xanthomonas campestris*, *X. vitians*, and *Erwinia* spp. were found in diseased heads from one or more fields. All except *X. vitians* caused head rot. Symptoms similar to those most commonly seen in naturally infected heads were produced in inoculated heads only by *P. cichorii* and *P. maculicola*. Only weakly virulent cultures of head-rot bacteria were recovered from 1,160 seeds of 10 cultivars. Gray to purplish-black lesions 1 to 2 mm in diam were observed on 46% of one or both cotyledons of 1,788 seedlings. No virulent cultures were isolated from 269 cotyledons with lesions and 199 without lesions. But virulent cultures of *P. cichorii* and *P. maculicola* were recovered from cotyledon lesions on seedlings grown from artificially infested seed. More seedlings grown from seed infested with *P. maculicola* were deformed and showed yellow-to-white cotyledon lesions than from noninfested seed. Virulent cultures of *P. cichorii* and *P. maculicola* were isolated from surface-disinfested seeds 203 days after artificial infestation.

*Production and germination of sporangia of *Sclerophthora macrospora* from corn.* M. H. SUN & A. J. ULLSTRUP (Purdue Univ., Lafayette, Ind.). *Sclerophthora macrospora* rarely produces sporangia on infected corn plants in the field. They can be obtained by incubating excised young diseased leaves showing "green island" symptoms in water. Seedlings grown from infected kernels are ideal for sporulation studies, as the fungus is readily transmitted through corn seeds maintained at 10 to 12% moisture and stored for up to 6 months at 1 C. Sporulation occurs abundantly on young succulent leaves, but declines markedly as the plant becomes older. No sporangia are produced on intact leaves, suggesting that the depletion of host nutrients may be a prerequisite for sporangial formation. Sporulation takes place in complete darkness within a temp range of 8 to 28 C with an optimum of 24 to 28 C. Sporangia may be continuously produced from the same host substrate for 6-15 days, depending on temp. Germination of sporangia is most commonly achieved by zoospore production, although direct germination by germ tube is occasionally observed. The type of germination is apparently not influenced by temp. The number of zoospores released from a single sporangium ranges from several to over 100, with 40 to 60 the most frequent number observed. The optimum temp for sporangial germination is 12 to 16 C. The zoospore germinates by a germ tube.

A chlorosis-inducing toxin from a pseudomonad pathogenic to timothy. P. A. TAYLOR, R. D. DURBIN, & D. P. MAXWELL (Univ. Wis., Madison). A fluorescent *Pseudomonas* sp. causing chlorotic lesions on timothy leaves, *Phleum pratense*, produces a toxin when grown in Woolley's medium. Toxin concn in culture reaches 100 μ M (glycine equivalents) at the end of log phase, and declines sharply thereafter. The toxin was purified from lyophilized culture filtrates by methanol extraction, column chromatography on cellulose and polyamide, and preparative layers of Silica Gel H. It is chromatographically identical to the toxin from *P. tabaci* and *P. coronafaciens*, and likewise yields tabtoxine, threonine, and serine (M ratios of 1.0:1.0:0.12, respectively [glycine equivalents]) after hydrolysis in 6N HCl for 30 min at 121 C. Mild hydrolysis (pH 3.1 for 30 min at 100 C) gives 2 ninhydrin-positive products which are also present in culture filtrates. A gas-chromatographic method for separating derivatives [*N*,*O*-bis-(TMS)-trifluoroacetamide-pyridine, 1:1 for 20 min at 60 C] of toxin and its degradation products was developed using a column of 2% DC-11 liquid phase on Gas-Chrom Q solid support coupled to a Finnigan 1015 mass spectrometer. This confirmed the presence of two tabtoxine-containing compounds, one with threonine and one with serine. It has not been determined whether both compounds can induce chlorosis.

Seed transmission of barley stripe mosaic virus in relation to the length of time parental plants are infected. R. G. TIMIAN (ARS, USDA, N. Dak. State Univ., Fargo). Seed transmission of several strains of barley stripe mosaic virus (BSMV) in 4 barley cultivars was determined. BSMV transmission through pollen and ovules of plants infected for various periods of time prior to anthesis was also resolved. Some strains of the virus were readily transmitted through the seed of some varieties but not in others. Some strains were not transmitted through the seed of any of the cultivars tested. BSMV was not transmitted through the ovules of plants unless the parental plants were infected with the virus at least 12 days prior to fertilization. A longer time between infection and anthesis was necessary for virus transmission through the pollen. Genetic characters in some cultivars of barley that prevent seed transmission of BSMV are adequate for control of the virus, even though the variety is susceptible to infection through mechanical inoculation.

Performance of sweetcorn under natural infestation of maize dwarf mosaic virus. R. W. TOLER, A. J. BOCKHOLT, & D. J. BLASINGAME (Texas A&M Univ., College Station). Data were obtained in 1968 and 1969 on the reaction of 122 commercial sweetcorn hybrids, cultivars, and accessions to natural infection of maize dwarf mosaic virus, strain A (MDMV-A). The trials were located in Wharton County, Texas. The principal vector was the cornleaf aphid, and the virus reservoir was over-seasoning Johnson grass. In 1968, 30 cultivars and hybrids were evaluated with a number of accessions showing resistance to infection. The percentage of infection ranged from 83% at 50 days after seeding to 100% at harvest. The first accessions that showed some tolerance were E6550, Merit Ex 75703, Aristogold Bantam Evergreen, and Sweet No. 2. In 1969, of the 92 cultivars and hybrids screened, only Georgia Special was resistant to virus infection. Those entries that showed acceptable tolerance or resistance to disease development were Keystone 4L 001, SRS Valley Gold, Niagara Golden Charm, Rogers Golden Queen, and Ferry Morse E31076. This indicates new and additional sources of resistance and tolerance in the sweetcorn genome to MDMV-A.

Three fungi associated with cantaloupe roots in Arizona. J. L. TROUTMAN & J. C. MATEJKA (Univ. Ariz., Yuma). Two cantaloupe crops are produced annually in Arizona and under these intensive farming practices there is buildup of root pathogens. Surveys made during the past 3 years indicate that root rots of cantaloupe are commonly caused

by *Rhizoctonia solani*, *Verticillium albo-atrum*, and one unidentified fungus. These root pathogens are associated with previous cropping histories; *R. solani* follows alfalfa, *V. albo-atrum* follows cotton, and the unidentified fungus follows small grains. The aboveground symptom expression is similar for each disease and usually coincides with fruit development. A short period of wilting is followed closely by chlorosis of the leaves with an eventual collapse of the plants. In contrast, root symptoms are very distinctive. *Rhizoctonia* induces a watery soft rot of the lower portions of the tap roots. This rot progresses upward. *Verticillium* induces mild vascular discoloration during early phases of the disease, but causes no appreciable root decay. The unidentified fungus causes decay of secondary roots typified by numerous scattered, small, round, black bodies. All three fungi attack cantaloupe under greenhouse conditions.

An ultrastructural study of potato virus M-induced local lesions in Red Kidney bean. J. C. TU & C. HIRUKI (Univ. Alberta, Edmonton, Canada). The primary leaves of Red Kidney bean (*Phaseolus vulgaris*) were mechanically inoculated with an Alberta isolate (AP-1) of potato virus M (PVM). Local lesions were sampled for electron microscopy 8 days after inoculation. Thin sections of local lesions revealed an abnormal thickening in the inner portion of the secondary cell wall. This thickening appeared to be one of the major ultrastructural alterations associated with the formation of PVM local lesion. The thickening was initiated in those cells at the periphery of the lesion. These cells showed individual virus particles, virus masses, accumulation of ribosomes, and an extensive endoplasmic reticulum. In the later stages of infection, chloroplasts were abnormally large due to the increased size and number of starch grains. Degenerate cells were electron-opaque and virus particles were not observed in these cells.

Life cycle and spread of ash rust in Texas. E. P. VAN ARSDEL & A. CHITZANIDIS (Texas A&M Univ., College Station). *Puccinia peridermiospora* grows on naturally separated alternate hosts in littoral Texas. In 1969, aecial hosts were *Fraxinus pennsylvanica* and *F. velutina*. The coastal salt marsh grass telial hosts are *Spartina patens* and *S. bakeri*; *S. cynosuroides*, *S. alterniflora*, and *S. spartinae* were not infected. Constant term germination tests at 5-C intervals from 0 to 35 C showed germination of aeciospores 10 to 30 C, urediospores 15 to 30 C, teliospores 15 to 25 C, sporidia 15 to 25 C. All opt temp were 25 C. Sporidia release occurred at night, 16-28 April. A large release 16-17 April (peak 1-4:00 AM) was correlated with meteorological events to reasonably explain the 1969 ash infection pattern. Rust intensity on ash declined with distance from salt marshes in a stepped decline. On the average, the number of infections decreased as the sq root over each distance of 7 miles. The first step in the curve at 12 miles could be due to frequent low nocturnal land breeze counter currents. The 45-mile step could be explained by the southeast wind's blowing of the spores northwest for 3 hr at 15 mph. An advancing cold front's updrafts could loft the spores to 5,000 ft, where south winds rapidly moved them up to 130 miles northwest, where they were brought down by rain and downdrafts.

Ethanol stimulation of growth of Armillaria mellea correlated with ethanol suppression of phenol accumulation. C. P. VANCE & M. O. GARRAWAY (The Ohio State Univ., Columbus). *Armillaria mellea* was grown for 7 days on a glucose-L-asparagine medium supplemented with ethanol (500 ppm); then groups of 15 thalli (average dry wt per thallus, 6-8 mg) were transferred either to a medium without ethanol or to similar media supplemented with different concn of ethanol. The dry wt of transferred thalli were determined at various incubation times from zero to 72 hr. Concurrently, similarly treated thalli were extracted with 80% ethanol, and the total phenol content was determined for each incubation time with Folin-Denis reagent. The average dry wt per thallus after 72 hr of incubation

on a glucose medium or on one supplemented with 50, 100, 250, 500, or 1,000 ppm ethanol was, respectively, 9.2, 10.4, 11.1, 14.1, 17.0, and 17.2 mg. The corresponding phenol concn were 6.7, 5.6, 5.5, 4.0, 3.0, and 3.2 $\mu\text{g}/\text{mg}$ dry wt, respectively. The ethanol-induced increases in growth and the accompanying decreases in phenol concn were linear with time. Chromatographic and spectrophotometric analyses revealed that ethanol also caused qualitative changes in *A. mellea* phenols. These data indicate that concn of ethanol supplement which stimulate growth suppress phenol accumulation and alter the kinds of phenols synthesized.

Possible modification of susceptibility of tomato to Fusarium wilt by a Chaetomium sp. P. S. VERMA & C. C. ALLISON (Ohio State Univ., Columbus). *Chaetomium* spp. have been found in roots of plants and in soil. Previous observers suspected that some *Chaetomium* isolates present in soils and as internal residents in tomato seedlings were highly effective in delaying and retarding *Fusarium* wilt symptoms of tomato. Several new and old isolates were selected for further investigation in 1969. Work continued in 1970 has demonstrated that certain isolates are effective in retarding and inhibiting symptom development of *Fusarium* wilt of Bonny Best tomato seedlings. Using the host plant in a selective capacity, isolates of *Chaetomium* were selected that varied in their capacity to retard and inhibit symptom development of *Fusarium* wilt. Isolates of *Chaetomium* can be readily isolated as internal residents from stems and petioles of tomato seedlings inoculated with *Fusarium oxysporum* f. sp. *lycopersici* but without external and internal symptoms of wilt. It is proposed that the tomato seedlings become resistant due to the effects, either physiological or morphological, of the *Chaetomium* fungus growing internally in the plants inoculated 48 hr previous to inoculating with *Fusarium*.

Stimulation of Rhizobium and inhibition of Fusarium by certain components of alfalfa root exudate. H. E. VOEGEL, JR., & F. L. HOWARD (Univ. R.I., Kingston). Alfalfa seedling root exudates contained 4 α -amino nitrogen (α AN)-containing compounds (A, B, C, and D). Compounds A and B constituted over 83% of the α AN of each exudate. "Synthetic" exudates containing C (lysine) and D (histidine) were prepared and adjusted to pH 7.0. Six amino acids suspected of being compounds A and B were added in pairs in all combinations and bioassayed with *Rhizobium meliloti*. Amino acid concn were comparable to components in "natural" exudate. After inoculation with *R. meliloti*, the solutions were incubated at 35 C for 24 hr, and growth was determined by spectrophotometer absorbance at 470 nm. Similar amino acid formulations were incorporated into min nutrient agar, inoculated with *Fusarium oxysporum* and incubated at 25 C, and colony diam was measured daily. Cysteine alone stimulated *Rhizobium* growth. Bioassay of 1 to 280 ppm cysteine by *R. meliloti* indicated a growth stimulation of 9 times the controls at 1 to 16 ppm, and inhibition above 40 ppm. Cysteine concn from 200 to 280 ppm inhibited fusarial growth, but had no effect below 200 ppm. Chemically-related methionine increased bacterial growth 26 and 38 times the controls at 120 and 280 ppm, respectively, but had no effect on the growth of *F. oxysporum*.

Sensitivity of tobacco varieties to tobacco mosaic virus. H. L. WALKER & T. P. PIRONE (Univ. Ky., Lexington). The tobacco cultivars X73, Samsun NN, and Havana 425, which produce local lesions in response to TMV infection, were compared with the systemically infectible cultivars T.I. 787, Samsun nn, Gold Dollar, and Kentucky 26 for sensitivity to infection by the common strain of TMV. Test plants in the five- to six-leaf stage were dusted with 600 mesh Carborundum, and the youngest expanded leaf was inoculated with 2.5 μl iters of inoculum diluted in 0.01 M phosphate buffer, pH 7. About 50 plants of each cultivar were inoculated with purified virus at three different concn. The number of TMV particles in the inoculum was determined by particle counts of electron micrographs. The

cultivars X73, Havana 425, T.I. 787, and Gold Dollar were more sensitive than the others tested. Percentages of infected plants in three experiments with these cultivars ranged from 40 to 60% with inoculum which contained 4.4×10^4 particles; from 3 to 11% with 4.4×10^3 particles; and from 0 to 5% with 4.4×10^2 particles. Over-all, the systemic varieties were no more sensitive than the local lesion varieties.

The association of cellulolytic and pectolytic enzymes with Diplodia boll rot of cotton. SY-YING C. WANG & J. A. PINCKARD (La. Agr. Exp. Sta., Baton Rouge). Greenhouse-grown cotton bolls were surface-sterilized and incubated in sterilized culture dishes with moist filter papers. The bolls were slightly wounded and inoculated with *D. gossypina*. After periods of incubation at 30 C, the growing mycelium on the surface of bolls, the rotten carpel walls, and the rotted locks were extracted separately and assayed for enzyme activities. Cellulase (C_x) and pectic enzymes were not detected in noninoculated bolls. Very high C_x occurred in rotted carpel walls and locks with the same pH optimum at 4.5 as C_x from mycelium. High activities of pectin methyl-esterase (PME), poly-galacturonate trans-eliminase (PGTE) and pectin trans-eliminase (PTE) were found in rotted carpel walls and locks with pH optima at 6.5, 9.0, and 9.0, respectively. The same pH optima were obtained with mycelial extracts. The enzymes were stimulated by Ca^{++} , but inhibited by EDTA. The activities of C_x , PME, PGTE and PTE increased as the degree of boll rot increased. Exopolysaccharonase (exoPG) could only be detected at very early stages of infection. The enzyme had an optimum pH at 4.5, and was inhibited by Ca^{++} and EDTA. The activity of exoPG decreased rapidly as boll rot increased. Fresh or heated extracts of healthy carpel walls slightly inhibited exoPG and PME but not PGTE, PTE, and C_x .

Relationship of Fusarium, fertilizer, and crop residue to seedling blight of wheat. H. L. WARREN & THOR KOMMEDAHL (Univ. Minn., St. Paul). Wheat (*Triticum aestivum* 'Chris') grew in continuous wheat plots in the field in which crop residues were either retained or removed, with and without fertilizer application (5:20:20 NPK, 243 kg/ha). Seedlings were evaluated for blight 3 times during the season by examining shoots and roots, three replicates each time, 50 plants/replicate. Blighted seedlings averaged 1 to 2% when fertilizer and residue were present and 11 to 14% when both were absent. Based on a disease index (scale 0-100) of roots, presence of fertilizer and residue gave indices of 30-31, but absence of both gave indices of 52-66. Fertilizer alone gave a lower blight incidence, lower root disease indices, and fewer *Fusarium*-infected roots than did residues alone. *Fusarium* spp. were isolated from 12 to 14% of roots where both fertilizer and residues were present and from 19 to 24% of roots where both were absent. *Fusarium roseum* comprised 90% of the *Fusarium* isolates from roots, followed by *F. oxysporum* and *F. solani*. Graminearum comprised 70% of *F. roseum* cultivars; Avenaceum and Culmorum, 15% each. Graminearum was most abundant where fertilizer and residue were absent. Thus, fertilizer and residues together gave the lowest blight incidence and least root infection; fertilizer reduced blight more than did residue.

The effect of soil temperature on Pythium ultimum damage to lettuce seedlings. A. G. WATSON (Univ. Calif., Berkeley). Sixteen thermocouples recording temp on a continuous multipoint recorder were buried in soil at depths of 0.25, 0.50, 1 and 2 inches in four locations in two fields. Each week during a 17-week period, 1,400 lettuce (*Lactuca sativa*) seed were planted in seven replicates of 200 seed each, at 0.25 inch depth in the locations of the thermocouples. The seedlings were washed free of soil after 7 days, and the percentages showing *Pythium* injury to the main root-tip were recorded. *Pythium ultimum* populations were determined by Stanghellini's method at planting

and when the seedlings were removed. The *Pythium*-lettuce interaction was extremely temp-sensitive. An increase in the mean soil temp at 0.25 inch from 18 to 24 C resulted in an increase in the percentage of *Pythium* damaged roots from 10 to 65% when the *Pythium* population was 200 propagules/g soil.

Influence of previous nematode environment on sex differentiation of Meloidogyne graminis. A. J. WEBBER & J. A. FOX (Va. Polytechnic Inst., Blacksburg). Populations of *Meloidogyne graminis* reared on Tifgreen Bermudagrass under different temp and nutrient regimes were found to differ in their innate sex differentiation. Tifgreen Bermudagrass sprigs from plants grown at 20 to 26 C and a high nutrient level were rooted in distilled water. Rooted sprigs were inoculated with 150 larvae from nematode populations maintained for 4 months on host plants at low or high nutrient levels. Inoculated sprigs were incubated at 26 C. Populations preconditioned at a low host nutrient level averaged 32% males while those preconditioned at a high host nutrient level averaged 1% males. The effect of temp was studied with sprigs inoculated with 150 larvae from nematode populations maintained for 4 months on host plants at 20 or 26 C. Inoculated sprigs were incubated at 26 C. Populations preconditioned on the host at 20 C averaged 6% males, while those preconditioned on the host at 26 C averaged 20% males. Variation in sex differentiation may be the result of genetic selection or physiological adaptation during the previous temp and nutrient regimes.

The Hemileia vastatrix coffee rust disease established in the American tropics. F. L. WELLMAN, R. DESROSIERS, & E. SCHIEBER (N.C. State Univ., Raleigh, US-AID, Brazil; Ministerio Agr., Guatemala). The causal fungus of the devastating coffee rust disease, *H. vastatrix*, was found in January 1970 on Arabica coffee, *Coffea arabica*, by A. G. Medeiros, near Itabuna in the Bahia state of Brazil. Specialists found it to be established in that state as well as in Espirito Santo and Minas Gerais. Brazilian officials arranged consultation and field trips with the authors for the purpose of studying possibilities of eradication; rust was collected on 3 cultivars of *C. arabica*, and on *C. canephora* cultivars Quillou and Robusta (freely occurring on fruits of the latter). Its severity indicates that it has been present for a few years at least. Eradication appears almost impossible: (i) infection centers are well scattered in numerous fazendas of irregular terrain; (ii) the large region of occurrence has a climate well suited to the rust; and (iii) it is evidently now spreading. The rust, firmly established, is a serious potential threat to coffee in the rest of Brazil and in all tropical American countries.

Growth of Aspergillus repens in stored flue-cured tobacco. R. E. WELTY (ARS, USDA, N.C. State Univ., Raleigh). *Aspergillus repens* is one of the predominant species of fungi isolated from marketed, damaged, and nondamaged flue-cured tobacco *Nicotiana tabacum*. Some time between on-farm curing and marketing, the fungus becomes associated with the leaf. Factors that determine whether or not *A. repens* grows in cured tobacco are likely temp and moisture content (MC). To determine the critical MC for fungal growth and the effect of temp and MC on fungal growth rates, tobacco inoculated with *A. repens* was stored at 20 and 30 C and 75, 80, 85, 87, and 95% relative humidity (RH). These 5 RH were previously found to reproduce MC in the range commonly encountered by flue-cured tobacco during marketing. Samples from these storage treatments were taken weekly for 4 weeks and evaluated for mold growth and MC. *Aspergillus repens* did not grow in tobacco at or below 25% MC at either 20 or 30 C. Tobacco between 25 and 30% MC became mold-damaged at 20 C in 3 to 4 weeks and at 30 C in 2 to 3 weeks. Above 30% MC at either 20 or 30 C, fungal damage occurred after 1 week. The critical MC for growth of *A. repens* in stored tobacco appears to be near 25%.

Distortion of Polyporus tomentosus germ tubes by autoclaved spruce bark. R. D. WHITNEY (Can. Dep. Fisheries

Forest., Winnipeg, Manitoba). Dry-autoclaved (10 min at 5 lb.) slices of inner bark of white spruce (*Picea glauca*) stimulated spore germination of basidiospores of *Polyporus tomentosus*, but germ tubes were very short and distorted. Hot-water extracts from inner bark (Soxhlet extraction of 10 g bark in 150 ml distilled water) also distorted germ tubes. Soon after emergence, germ tubes developed swellings of various shapes and became angular, contorted, and excessively septate. Swollen spores were about 4 times the size normally attained during germination. Wet-autoclaved inner bark slices did not distort adjacent germ tubes, but germ tubes on media containing the filtrate from this material were distorted. Apparently heating the bark produces the inhibiting substance which can be leached from the bark in boiling water. Slight early distortion occurred in media containing cold water extracts, indicating weak formation of the inhibiting substance in cold water. Unsterilized bark and wood slices stimulated germination; germ tubes were normal, indicating that the distorting substances are absent in untreated bark. Distorted germ tubes grew normally when transferred to malt extract agar.

Response of certain tobacco cultivars to root, stem, and leaf inoculations with two races of Phytophthora parasitica var. nicotianae. W. H. WILLS & L. D. MOORE (Va. Polytechnic Inst., Blacksburg). Several cultivars of tobacco were inoculated with two races of the black shank pathogen by standard root inoculation, by a leaf-strip inoculation technique, and by four methods of stem inoculation. Disease development among the cultivars was related to the inoculation technique and the resistance of the host. Stem resistance in all cultivars to both races was correlated with resistance observed in plants inoculated by standard methods through the roots. The cultivar L8 showed high root and stem resistance and no leaf resistance to Race 0, even though the progenitor of L8, *Nicotiana longiflora*, exhibited high root, stem, and leaf resistance to Race 0. Both *N. longiflora* and L8 are susceptible to Race 1. Uninjured stems of resistant cultivars were not invaded. Penetration of uninjured stems of a susceptible cultivar was successful in about 50% of the inoculations. When stems were injured by peeling back the epidermis, colonization of the cortex was as extensive as when deep cortical wounds were made. Inoculation in the pith produced more extensive necrosis than inoculation in the cortex. The epidermis seems to provide a primary barrier to stem penetration which is more than mechanical. Lack of resistance in leaves of L8 suggests that more than one mechanism may be involved.

Mycoplasma-like artifacts in healthy and yellows-diseased plants. B. S. WOLANSKI & K. MARAMOROSCH (Boyce Thompson Inst. Plant Res., Yonkers, N.Y.). Negatively stained preparations of extracts from healthy plants of rye, corn, sugarcane, aster, and tobacco revealed the presence of artifacts that were indistinguishable from structures found in yellows-diseased plants and presented by several authors as mycoplasmas. These artifacts were similar to negatively stained animal mycoplasmas. No such artifacts could be found in either healthy or diseased plant extracts which were prefixed with osmium tetroxide, then stained with phosphotungstic acid. The findings stress the unreliability of the negative-staining technique as a rapid diagnostic tool for the detection of presumptive mycoplasma infections in plants. The slower method of examining thin sections of plant tissues remains the only reliable test available.

Isolation and characterization of a virus from Penicillium brevi-compactum. H. A. WOOD, R. F. BOZARTH, & P. B. MISLIVEC (Boyce Thompson Inst., Yonkers, N.Y., Food Drug Admin., Washington, D.C.). Spherical viruslike particles measuring approximately 44 m μ in diam were found associated with a single isolate of *Penicillium brevi-compactum*. Isolation and purification of the particles were made by chloroform extraction of the mycelium, followed by differential and sucrose density-gradient centrifugation.

Two sedimenting sizes of particles were found which had sedimentation coefficients of 144 and 123 Svedberg units. Free-boundary and gel electrophoresis indicated the presence of three electrophoretic components. Purified preparations had a typical nucleoprotein ultraviolet absorption spectrum and contained double-stranded ribonucleic acid components with mol wt of 2.18 and 1.99×10^6 daltons. It is surmised from the data that the *P. brevi-compactum* investigated contains a previously unreported virus.

Effect of three fungicides on vegetative growth of Verticillium malthousei and Agaricus bisporus isolates. P. J. WUEST & H. COLE, JR. (Pa. State Univ., Univ. Park). *Verticillium malthousei* is a serious pathogen of the cultivated mushroom *Agaricus bisporus*. Zineb [zinc ethylenebis (dithiocarbamate)] is the standard fungicide used commercially for control of *Verticillium*. Recently, difficulties in control were experienced by certain growers. The reasons for the failures were not known. *Verticillium* isolates from a problem site and various other sources were evaluated for zineb resistance in agar culture. Two possible replacement fungicides, maneb [manganous ethylenebis (dithiocarbamate)] and benomyl [methyl-1-(butylcarbamoyl)-2-benzimidazolecarbamate], were evaluated along with the zineb standard against *Verticillium*. The fungi were grown on potato-dextrose agar amended with test fungicides at levels from 1 to 500 µg/g. There was no evidence of strong resistance to zineb by any of the isolates tested. Slight differences in isolate sensitivity were noted. With maneb, slight shifts in isolate sensitivity also occurred. But with benomyl at 100 ppm, up to 6-fold growth differences between isolates were noted. *Agaricus bisporus* was tolerant to zineb, maneb, and benomyl at the dosages tested.

Enzyme-induced germination of Aphanomyces oospores. C. Y. YANG (Univ. Ky., Lexington.). The proteolytic enzymes pronase and chymotrypsin are able to induce germination of oospores of the pea root-rot pathogen, *Aphanomyces euteiches*. The germination varied with the isolates of the fungus and with the length of dormancy of the oospores. Enzyme-induced germination was successful with physically separated oospores, as well as with oospores in intact fungal cultures. Up to 50% germination was achieved with chymotrypsin at the concn of 250 units/ml. Two modes of germination were observed: indirect germination through zoosporangial formation and direct germination by germ tubes. Preliminary evidence indicates that the mode of germination is related to the age of the oospore. Direct germination seemed to be the dominant form among oospores older than 4 weeks. Growth of the germ tubes into hyphae proceeded at a rate almost triple that of the initial rate of growth recorded during the first 3 hr. Several microchemical methods as well as birefringent analysis indicated that the oospore wall is not made of homogeneous cellulose components.

The formation of elongating secondary hyphae of Erysiphe graminis f. sp. hordei as a criterion for the identification of segregating genes for resistance in barley. S. L. YANG & A. H. ELLINGBOE (Mich. State Univ., E. Lansing). Barley plants grown from seed derived from heterozygous parent plants with genotypes *Mlmla*, *Mlgmlg*, *Mlpmlp*, or *Mlkmk* were inoculated with Race CR 3 of *Erysiphe graminis* f. sp. *hordei*. Barley plants from homozygous recessive and homozygous dominant parents were used as controls. Inoculated plants were held under environmental conditions known to favor parasite development. The percentage of conidia that produced elongating secondary hyphae (ESH) on individual plants was recorded 28 hr after inoculation. The genotype of each progeny plant was determined by the segregation of disease reaction among its progenies. The range of the per cent ESH was greater among plants derived from plants heterozygous for an *Ml* gene than among plants derived from a plant homozygous for the *Ml* gene. The per cent ESH on progeny plants from homozygous recessive parent plants was always higher than 70. The

per cent ESH on homozygous recessive plants from heterozygous parent plants ranged from 45 to 85. The per cent ESH on plants homozygous dominant for an *Ml* gene was also dependent on the genotype of the parent plant. The per cent ESH formed on *MlaMla* plants, for example, ranged from 0 to 15 if the parent plant was *MlaMla* and 0 to 40 if the parent plant was *Mlmla*. The results suggested a carryover effect from heterozygous parent to homozygous progenies.

A virus resembling tobacco mosaic virus in oak. C. E. YARWOOD & E. HECHT-POINAR. (Univ. Calif., Berkeley). Tobacco mosaic virus (TMV) in oak species is indicated by symptoms, transmission, electron microscopy, serology, and the K_2SO_3 color test, none of which is entirely satisfactory. Symptoms were yellowing, rosetting, early breaking of dormancy, and necrotic spotting which have been observed in some *Lithocarpus densiflora* trees. Inoculations from *Lithocarpus* to *Chenopodium quinoa* have yielded 783 TMV-like lesions in 69 tests (725 plants with about 8,700 inoculated leaves) over 6 years. Most inoculated plants yielded no lesions. The most effective method of inoculation was by brush extractions, and the most effective supplement seemed to be 0.1% K_2SO_3 + 0.2% caffeine. Similar infection resulted from transmissions twice from *Quercus douglasii* and once from *Q. robur*. TMV-like rods have been observed in *Lithocarpus* and in *Q. alba*, *ceris*, *douglasii*, *ilex*, *lobata*, *obtusata*, *phellos*, and *robur*, but most abundantly in *Q. douglasii*. Suspensions of *Lithocarpus* leaves reacted with TMV antiserum, but no healthy controls were known to be available. Suspensions of leaves of *Q. alba*, *ceris*, *douglasii*, *ilex*, *lobata*, *macrocarpa*, *mongolica*, *obtusata*, and *robur* yielded a bright purple color with K_2SO_3 , but no healthy controls were known to be available.

Dynamics of fungal spore germination in soil. D. L. YODER & J. L. LOCKWOOD (Mich. State Univ., E. Lansing). Conidia of *Helminthosporium sativum*, *Penicillium frequentans*, and *Aspergillus ustus* showed 50% germination on sterilized soil in 1.5, 10, and 16 hr, respectively. Conidia did not germinate in 24 hr when placed on membrane filters on natural soil, or on glass beads saturated with water or subsoil extract at 24 C. Conidia exposed to these conditions for 24 hr, then placed on sterilized soil, germinated in ca. 30% less time than untreated controls. Spores were exposed to sterilized soil for 75 to 85% of the time required for germination, transferred to natural soil for 6 days, and subsequently returned to sterilized soil. Germination time of these spores, based on total incubation time on sterilized soil, was 50 to 70% longer than that of spores not exposed to natural soil. *Penicillium frequentans* conidia, which required amendments of glucose and ascorbic acid for germination on natural soil, were incubated on natural soil containing 0.3% glucose for 12 hr, then placed on non-amended soil for 0, 2, 4, 6, and 8 days. When these conidia were transferred to soil containing 0.01% ascorbic acid for an additional 12 hr, germination was 95, 77, 62, 8, and 15%, respectively. The germination process appears to have a water-uptake phase, followed by a reversible phase requiring an exogenous energy source.

Effect of Helminthosporium carbonum toxin on nitrate reductase activity in susceptible corn embryonic axes. O. C. YODER (Mich. State Univ., E. Lansing). Nitrate reductase (NR) is substrate-induced. Resting axes were placed in water or toxin solution (2 µg/ml) for 12 or 24 hr, then in NO_3^- solution for 8 hr. NR activity was not affected by toxin in the 12-hr set, but was reduced 66% in the 24-hr set. Axes which had imbibed water for 36 hr were placed in NO_3^- solutions with and without toxin. Toxin (0.2-200 µg/ml) caused 30-70% increase in NR activity after 2-4 hr. After 4-hr toxin pretreatment, the initial rate of NR induction increased. Toxin did not affect NR activity in the absence of NR synthesis (without NO_3^-), nor did toxin

change the rate of NR degradation. Thus increased activity appears to be caused by increased synthesis. The possibility was tested that toxin, like helminthosporol (another bioactive product of *Helminthosporium* spp.), has a stimulatory effect similar to that of gibberellic acid (GA). Toxin and GA were compared in three standard tests for GA. GA promoted dark germination of tobacco seeds, elongation of cucumber hypocotyls, and synthesis of α -amylase by barley endosperm; toxin at 0.01-100 $\mu\text{g}/\text{ml}$ was not active in these tests. The stimulatory effect of toxin is unlike that of helminthosporol and GA, but its effect on protein synthesis may be similar to that reported for several plant infections.

Production of indoleacetic acid in culture by Pythium debaryanum. KAZUHIRO YOSHII & J. C. WALKER (Univ. Wis., Madison). *Pythium debaryanum* grown in potato-dextrose broth produced a heat-stable compound inhibitory to root growth of rape seedlings. The compound was extracted with ether from the culture filtrate at pH 3.0, eluted with ethyl acetate 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. The compound, recrystallized from water, was identified as indole-3-acetic acid (IAA) by its melting point, chromatographic behavior, ultraviolet and infrared spectra, fluorimetry, and biological activity. *P. debaryanum* produced 1.4 $\mu\text{g}/\text{ml}$ IAA after 7 days of incubation in potato-dextrose broth. At this concn, IAA caused about 80% inhibition of root growth of rape seedlings. IAA may possibly be involved in pathogenesis of root rot caused by this fungus. *Pythium irregulare*, *P. oedocephalum*, *P. paroecandrum*, *P. sylvaticum*, *P. ultimum*, and *P. vexans* also produced IAA in potato-dextrose broth, but *P. aphanidermatum*, *P. myriotylum*, *P. rostratum*, and *P. torulosum* did not.

Electron microscope studies on the effect of 2-aminobutane on germinating conidia of Penicillium digitatum. A. I. ZAKI, J. W. ECKERT, & R. M. ENDO (Univ. Calif., Riverside). Germlings of *Penicillium digitatum* were fixed in glutaraldehyde and OsO_4 , dehydrated, and embedded in Epon 812. Ultrathin sections were stained with uranyl acetate and lead citrate. Micrographs of untreated germlings revealed the presence of electron-dense bodies (DB) associated with myelinlike structures within unit membrane-bounded vacuoles. Appearance and staining characteristics suggest that both structures are composed of phospholipoproteins and may be related. It is proposed that they may function in the assembly of membranes for organelles formed during germination. Electron-dense bodies appeared to disintegrate and eventually disappear from 2-amino-

butane treated germlings. Vacuoles simultaneously enlarged, and the contrast of various organelles decreased. It is hypothesized that 2-aminobutane deprives the conidium, through a direct or indirect action, of materials which normally are precursors of membranes for the developing organelles during early stages of germination.

Titers of two virus diseases of celery affecting field spread. T. A. ZITTER (Univ. Fla., Belle Glade). Cucumber mosaic virus (CMV) and western celery mosaic virus (WCMV) are two stylet-borne, aphid-transmitted viruses affecting celery in south Florida. CMV concn in celery plants singly and doubly infected with WCMV were determined at weekly intervals for 7 weeks by local lesion assay on *Chenopodium amaranticolor* and by aphid transmission with *Myzus persicae*. Starved aphids were given 1-min acquisition periods prior to transfer to healthy pepper plants (*Capsicum annuum* 'Early Calwonder'). WCMV titers in singly and doubly infected plants were assayed weekly by aphid transmission to celery (*Apium graveolens* var. *dulce* 'Utah 52-70-2-14'). CMV titers in singly infected plants reached a peak 2 weeks after inoculation and rapidly decreased. Parallel results were obtained with both methods. CMV titers in plants infected with both viruses followed a similar pattern, except that the peak occurred 3 weeks after inoculation. Maximum WCMV activity was reached at 3 weeks and remained nearly constant throughout the experiment. In older, double-infected plants, only WCMV was recovered by aphids. The behavior of CMV in celery conforms to the pattern of this virus in other hosts, and can account for limited field spread. The more constant and reasonably high titers of WCMV explains its more rapid spread.

Separation on sweet sorghum cultivars of six mosaic-inducing virus isolates found infecting corn, Johnson grass, and sorghum in the United States. N. ZUMMO (ARS, USDA, Meridian, Miss.). Differentiation of virus isolates from California, Virginia, Georgia, Mississippi, and Frankfort and Quicksand, Kentucky, was possible on the basis of their effect on growth and leaf chlorosis pattern of sweet sorghum (*Sorghum bicolor*) plants, cultivar Rio. Virginia and Quicksand, Kentucky, isolates severely stunted the plants and killed the growing points. The Mississippi isolate severely stunted the plants but did not kill the growing points. California and Frankfort, Kentucky, isolates caused only moderate stunting of plants. The Georgia isolate had no apparent effect on growth of Rio. Leaf symptoms varied from a mild linear chlorosis by the Georgia isolate to severe chlorosis and necrosis by the Quicksand and California isolates. The percentage infection was of no value in separating the virus isolates.