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Resistance to Phytophthora cactorum in apple seedlings from controlled crosses. H. S. ALDWINCKLE, J. N. CUMMINS, & H. L. GUSTAFSON (New York State Agr. Exp. Sta., Geneva). Some promising parents for the breeding of new apple rootstocks were tested for resistance to *Phytophthora cactorum*, the incitant of collar rot and crown rot, by the inoculating of dormant sticks, stripped to the cambium, with *P. cactorum*-bearing agar. M.IX, M.VII, MM.106, Antonovka Poltorafontowaja (AP), and Antonovka Kamienaja (AK) were resistant; M.26, Alnarp 2, and P.I. 274842 were susceptible. Progeny from controlled crosses among these parents were screened for resistance as seedlings. At the two-leaf stage, seedlings were inoculated by a flooding to the midhypocotyl level with a mixed suspension of zoospores of *P. cactorum* isolates. Most susceptible seedlings died within 2 weeks, from necrosis of the hypocotyl and/or roots. In some crosses, resistance appeared to be inherited as a single dominant gene, but in others, where a good fit to simple ratios was not obtained, modifying genes were probably important. M.IX, M.27, and AP transmitted resistance more consistently to their progeny than did M.VII, MM.106, and AK. Tests of the reaction of the *P. cactorum*-resistant seedlings to artificial inoculation with *Venturia inaequalis* showed that some were also scab-resistant.

Comparison of incidence of Fomes annosus infection of loblolly pine with field capacity, soil texture, and pore space of two soil hazard types. S. A. ALEXANDER & J. M. SKELLY (Va. Polytech. Inst. & State Univ., Blacksburg). Incidence of *Fomes annosus* in loblolly pine plantations on two soil hazard types correlates with field capacity, soil texture, and pore space. To determine the incidence of *F. annosus*, two roots were removed from each of 138 and 219 trees located on 10 high and 10 low hazard sites, respectively. Isolations were made on ortho-phenyl-phenol media and incubated at 21 C for 14 days. Soil texture (sand-silt-clay) at 8-12 inches deep was determined by the pipette method of analysis. The average per cent sand-silt-clay for these high and low hazard sites were 76-18-6% and 38-33-29%, respectively. The per cent capillary and noncapillary pore space and field capacity were determined for samples removed at 8- to 12-inch soil depth at 0.10 or 0.33 atm. Average per cent capillary and noncapillary pore space for high and low hazard sites were 17 and 21, and 6 and 40%, respectively. Incidence of *F. annosus* increased with a decrease in field capacity and capillary pore space and with the associated increase in noncapillary pore space. Per cent infection of trees located on soils with field capacities of 6 to 13% averaged 32.4%, whereas higher field capacities of 19 to 35% yielded 5.9% infection.

The in vitro culture of loblolly pine callus tissue. S. A. ALEXANDER & J. C. WHITE (Va. Polytech. Inst. & State Univ., Blacksburg, La. Tech. Univ., Ruston). In order to facilitate a study of *Cronartium fusiforme* under controlled laboratory conditions, it is essential that initially the host tissue be cultured. Stem explants of *Pinus taeda* were cultured in vitro on a modified Harvey's chemically defined medium containing (g/liter): $\text{Ca}(\text{NO}_3)_2$, 0.5; MgSO_4 , 0.14; KH_2PO_4 , 0.3; $(\text{NH}_4)_2\text{SO}_4$, 0.025; FeSO_4 , 0.014; MnSO_4 , 0.0035; glucose, 20.0; 2,4-D, 0.002; and 0.6% Bacto agar in double distilled water, pH 5.6. Stem sections from

1-month-old seedlings were surface-sterilized 2 min in 1/1,000 HgCl_2 solution, twice washed in sterile distilled water, cut into 5-mm sections, and implanted in the medium. Cultures were incubated under constant light, 250 and 350 ft-c, at 18 to 22 C. Primary callus cultures were light green in color with a rough texture, and produced enough callus to be subcultured in 3 to 4 weeks. Subcultures were grown under similar conditions with Harvey's vitamin complex added to the medium. Subcultures appeared to double in size in ca. 4 weeks.

Development of Fusarium wilt disease in a susceptible introduction of Lycopersicon pimpinellifolium. E. H. ALLEN & W. P. WERGIN (USDA, ARS, Beltsville, Md.). Seeds of *Lycopersicon pimpinellifolium* (P.I. 126927) were sown in loam, perlite, and peat (4:2:1, v/v) in 10-cm pots and placed in a greenhouse at ca. 27 C. Three weeks after germination, the potted soil containing 10- to 15-cm plants was drenched with bud cells, chlamydospores, and mycelium of *Fusarium oxysporum* f. sp. *lycopersici* race 1 without injury to the roots prior to or after inoculation. Eight to 10 days after inoculation, the cotyledons and two or three of the lowest leaves became chlorotic. Four to 6 days later, all leaves became permanently wilted. However, shoots began developing in the axils of the wilted leaves and grew normally for 2 to 3 weeks before developing typical wilt symptoms. The distribution of the pathogen in the plants was determined with a bioassay and an electron microscope. The rate of disease development observed in P.I. 126927 was compared to the rate in susceptible cultivars of *L. esculentum* (Bonny Best, Pearson Select, and Marglobe). P.I. 126927 was classified as extremely susceptible; Bonny Best, susceptible; Pearson Select and Marglobe, tolerant. Severe symptoms in Bonny Best developed ca. 1 week later than in P.I. 126927. However, only a trace of yellowing occurred in Pearson Select and Marglobe 3 weeks after inoculation.

Increased glucose exudate and damping off in sugar beets in soils treated with herbicides. J. ALTMAN (Colorado State Univ.). Pyramin [5-amino-4-chloro-2-phenyl-3-(2H)pyridazinone] and Ro-Neet (S-ethyl-N-ethylthiocyclohexanecarbamate) used as preplant herbicides at 4 lb./acre preplant (2ppm) resulted in increased glucose exudates at the hypocotyl-soil interface. This increase in glucose was detectable at 2, 4, and 10 weeks after treatment. Hypocotyl extracts were analyzed for glucose spectrophotometrically using the Kornberg-Horecker method for true glucose. Sugar beets grown in herbicide-treated soil infested with *Rhizoctonia solani* showed 50% increased damping off when compared with infested but nontreated soil. In laboratory tests, *R. solani* grew in graded concentrations of herbicides up to 10,000 ppm with little or no inhibition. This is additional evidence that herbicides may predispose plants to increased disease.

Penetration and host-parasite relationships of Macrophomina phaseolina on Glycine max. V. AMMON & T. D. WYLLIE (Univ. Mo., Columbia). Primary roots of Adelpia soybean seedlings were surface inoculated with 17-day-old sclerotia of *Macrophomina phaseolina*. Samples were collected after 1, 2, and 3 days' incubation and prepared for scanning electron microscopic examination. Scanning micrographs revealed that the hyphal strands

radiated out in all directions from the germinated sclerotia without any recognizable pattern of growth over the root surface. Contact with the root by the tips of primary and secondary hyphal strands resulted in the development of flask-shaped appressoria. Light and electron micrographs of sections from other infected samples indicated that penetration of root surfaces occurred most frequently between epidermal cells, aided by chemical softening and mechanical pressure. Growth of the pathogen through cortical tissue and into the stele occurred within 3 days. Initial growth of *M. phaseolina* through root tissue was primarily confined to intercellular spaces and the middle lamella, with intracellular colonization occurring later. Deterioration of the middle lamella, presumably by the action of fungal-secreted pectolytic enzymes, accompanied intercellular growth of the pathogen. Loss of cytoplasmic organization and recognizable organelles in root cells containing the fungus were commonly observed.

Aspects of the specificity of the plant proteins which inhibit pathogen secreted endopolygalacturonases. ANNE ANDERSON & P. ALBERSHEIM (Univ. Colo., Boulder). The α , γ , and δ races of *Colletotrichum lindemuthianum* secrete endopolygalacturonases (endo-PG) which behave in an identical manner during purification on several column materials. The endo-PG secreted by each of these races is completely inhibited by proteins associated with the cell walls of Red Kidney, Small White, and Pinto beans. These bean cultivars differ in their response to the α , γ , and δ races of the pathogen. Crude extracts from the hypocotyl cell walls of each of the bean cultivars possess approximately equal abilities to inhibit each of the endo-PG's. The proteins from Red Kidney and Pinto beans, which inhibit the endo-PG, purify identically, and the purified protein preparations possess equal specific activities. These proteins, purified for their ability to inhibit the endo-PG secreted by the α race, are equally able to inhibit the endo-PG's from the γ and δ races. It is concluded that the endo-PG's secreted by the three races of *C. lindemuthianum* are identical, and that their inhibitor proteins present in the three bean cultivars are also identical.

Development of cylindrical inclusions in tobacco etch virus-infected plants. J. H. ANDREWS & T. A. SHALLA (Univ. Calif., Davis). Roots of tobacco plants (*Nicotiana tabacum* 'Havana 425') systemically infected with the severe etch strain of tobacco etch virus were subdivided, fixed in glutaraldehyde-osmium, dehydrated in acetone, and embedded in Spurr's plastic. Electron-microscopic examination of thin-sections from apical meristems, regions of elongation, and root hair zones revealed a sequence of infection stages as determined by the size and number of inclusions. These cytopathic alterations, extensive in the root hair zone, diminished in less differentiated cells nearer the root tip. In the elongation and root hair regions, cylindrical inclusions were usually appressed vertically to the cell wall and were frequently in intimate association with plasmodesmata. It appears either that early in development they become juxtaposed to plasmodesmata due to cytoplasmic movement or that the inclusions originate there in situ. The latter hypothesis is favored, based on cytological evidence obtained thus far.

Influence of desiccation on viability of microsclerotia of Verticillium albo-atrum. L. J. ASHWORTH, JR., & O. C. HUISMAN (Univ. Calif., Berkeley). Viability of *Verticillium albo-atrum* microsclerotia was reduced 76% (range, 41 to 94%) in nine naturally infested soils by air drying and storage for 2 to 3 months at 25 to 30 C. Microsclerotia from infected cotton plants had an initial germination of ca. 51% when

seeded on potato-dextrose agar. After 2 weeks at 20 C, subsamples of these microsclerotia held in atmospheres of 0, 42, 58, 80, and 90% relative humidity had germination percentages, respectively, of 4.5, 4.6, 7.6, 13.9, and 51.4%. Although germinability of microsclerotia is adversely and quantitatively affected by desiccation, the fungus is known to persist in culture and in soil for many years in the absence of known susceptible hosts. These data suggest that loosening and drying soil, to the extent that it equilibrates with the relatively dry air of the San Joaquin Valley, Calif., may in a short time result in a drastic reduction in the inoculum density of the fungus.

Benomyl controls Fusarium wilt and is translocated in mimosa seedlings. A. ATTABHANYO & G. E. HOLCOMB (La. State Univ., Baton Rouge). Benomyl was tested for its efficiency in controlling wilt of mimosa (*Albizia julibrissin*) seedlings inoculated with *Fusarium oxysporum* f. *perniciosum*. Tests were conducted under greenhouse conditions in soil and sand cultures. Benomyl gave complete disease control at concentrations of 50, 100, and 200 ppm (based on dry soil or sand). A partial therapeutic effect of benomyl was observed, in that some plants survived when inoculated 3 and 7 days before being transplanted to soil containing benomyl. Translocation of benomyl or its fungitoxic derivative was detected in seedlings by use of a bioassay utilizing a species of *Penicillium*. Benomyl was detected in roots, stems, and leaves with the highest concentration in leaves. Levels of fungitoxicant detected were higher in plants grown in sand than in plants grown in soil. Benomyl could still be detected in leaf tissue 34 days after seedlings had been transplanted from benomyl-treated sand to untreated sand.

A phytotoxic substance produced by Alternaria tenuis which affects passion fruit vines in Hawaii. M. Y. AZALDIN & S. S. PATIL (Univ. Hawaii, Honolulu). An isolate of *Alternaria tenuis* pathogenic to passion fruit vines (*Passiflora edulis* 'Flavicarpa') produces an extracellular toxin in modified Richard's solution. It was purified according to the method previously reported for tentoxin produced by the bean isolate of *A. tenuis*. The toxin induces chlorosis in cotyledons of cucumber seedlings. A dosage response curve between toxin concentration ranging from 0.0006 to 160 $\mu\text{g/ml}$ and the chlorophyll content of hypocotyls and cotyledons of germinated cucumber seedlings was established. Visible chlorosis was evident at low toxin concentration of 1.6 $\mu\text{g/ml}$, and complete chlorosis of cotyledons at 16 $\mu\text{g/ml}$. Unlike tentoxin, the toxin from the passion fruit isolate is extracellular, is produced in the presence of glucose instead of sucrose, and has no ultraviolet absorption.

Development of a selective medium for isolation of Sclerotium rolfsii. P. A. BACKMAN & R. RODRIGUEZ-KABANA (Auburn Univ., Auburn, Ala.). The ability of *Sclerotium rolfsii* (SR) to produce and tolerate high levels of oxalate was investigated as the basis for isolation of SR. SR cultured on a basal salts agar medium containing increasing amounts of oxalate (0-0.54 M, pH 4.2) grew rapidly but sparsely, and lysed after 5 days. A similar response was found with basal salts alone. Addition of glucose (0-30 g/liter) to the oxalate medium led to good SR growth, but in direct proportion to the amount of glucose added. When glucose was held constant (30 g/liter) and oxalate was varied (0-0.054 M, pH 4.2), no change in mycelial production was detected; however, production of sclerotia was reduced. When aliquots of natural soil suspensions were plated on these media, glucose-oxalate

medium supported a variety of fungal contaminants, whereas the oxalate medium showed few. Gallic acid (130 mg/liter) added to selectivity observed with oxalate alone when a soil suspension was plated. Gallic acid stimulated the growth of SR above that shown on oxalate alone, and no lysis was observed. A few restricted colonies of *Aspergillus niger* and *Penicillium* sp. constituted the only contaminants in the gallate-oxalate medium.

Effect of soil fumigation on root density of peach trees on a short life site. P. A. BACKMAN, E. J. WEHUNT, B. D. HORTON, & W. M. DOWLER (USDA, Clemson, S.C., Byron, Ga.). Soil fumigation previously has been shown to extend peach tree life in short life (decline) orchards. To determine the relationship between soil fumigation and root density, soil cores of uniform depth and volume were removed at specific sites from beneath 4-year-old Coronet peach trees grown in soil fumigated with 1,2-dibromo-3-chloropropane and in nonfumigated soil. The dry weight of roots per unit soil volume, number of *Pythium* propagules/g of soil, and per cent feeder rootlets invaded with *Pythium* spp. were determined. Trees grown in nonfumigated soil had significantly less roots ($P=.01$) than trees grown in fumigated soil when sampled in June and July. Differences were not significant when sampled in September. Trees grown in nonfumigated soil versus fumigated soil had fewer feeder roots, and these roots were more frequently invaded with *Pythium* spp. ($P=.05$). Numbers of *Pythium* propagules/g of soil did not vary significantly between the soil treatments. Whether *Pythium* spp. were involved as primary or as secondary invaders in root density reduction could not be determined. These data indicate a direct relationship between fumigation and root density which may be a factor in tree longevity.

Biological indications of large-scale mutations in tobacco mosaic virus. J. G. BALD (Univ. Calif., Riverside). Repeated attempts were made under greenhouse conditions to obtain the *Nicotiana glauca* form of tobacco mosaic virus (TMV) (type strain, U5) by maintaining U1 in *N. glauca*. All attempts made at that time failed. Bawden, on the other hand, induced reversible mutations between a legume and tobacco form of TMV. The contrast led to a study of mutation-selection processes in TMV. Criteria of change were host susceptibility and symptoms, cytological reactions, and occasional serological tests. Single lesion isolates of U1 and U5 were inoculated to tobacco, tomato, *N. glauca*, and local lesion hosts, and separate lots of inoculated plants were submitted to different temperature regimes. Yellow spots on U1-infected plants were frequent sources of mutants. Spots failed to appear on any plants at temperatures below 27 C. Individual spots contained clusters of mutants, some of which were unstable. Continuing instability, due presumably to very high mutation rates, was a feature of all major changes in the TMV populations tested. The direction and end point of these changes were host-determined. The transition from one natural form (serotype) of TMV to another under controlled conditions now seems feasible.

Evidence that the oat blue dwarf virus propagates in the aster leafhopper. E. E. BANTTARI & R. J. ZEYEN (Univ. Minn., St. Paul). A long incubation period after virus acquisition and persistence of virus in the vector suggest that the oat blue dwarf virus (OBDV) propagates in the aster leafhopper, *Macrostelus fascifrons*. This hypothesis was supported by serial dilution-inoculation experiments and electron microscopy of thin sections of viruliferous adults. In serial dilution-inoculations, aster leafhoppers were injected with buffer-diluted extracts of ground viruliferous

leafhoppers that had been similarly injected 2 weeks earlier and incubated on an immune host. Eight of 25 leafhoppers transmitted OBDV after the fifth serial inoculation in which the equivalent dilution of the original virus would have been 1 in 10^{10} . Original extracts of viruliferous leafhoppers were not infectious when diluted more than 1 in 10^5 . Electron microscopy of thin-sections of viruliferous adults revealed aggregates and paracrystalline formations of particles that correspond to the size and shape of OBDV in brain tissue and fat bodies.

A histoid enation disease of Trifolium in the USA. O. W. BARNETT & CHI-CHANG CHEN (Clemson Univ., Clemson, S.C.). Enations were observed on the upper surface of leaves of greenhouse-maintained *Trifolium* plants. In most enation diseases of viral (including a clover enation disease in Italy) or probable viral origin, the proliferations occur on the lower surface of leaves. On *T. repens*, enations were usually scattered over the leaf lamina, but sometimes concentrated over the midrib or lateral veins. Many enations were elongated, but small papillae 1-2 mm in length also occurred. Occasionally a few small enations occurred on the lower surface. Interspecific hybrids with *T. occidentale* parentage had the same pattern of symptoms, with longer enations. The disorder was graft-transmitted to *T. incarnatum*, *T. occidentale*, *T. pratense*, *T. repens*, and *Vicia faba*. Systemic chlorotic vein-banding and veinal necrosis, and death eventually occurred on *T. incarnatum*. Symptoms on the other plants occurred only near the graft site. Shoots proliferated on *T. occidentale*. Protuberances occurred on the corners of *V. faba* stems. Tissues in the enations had an undifferentiated internal structure, and the cells varied in size and shape. There was no vascular tissue under interveinal enations. Long enations on the midrib had areas of disorganized vascular tissue with no connection to the vascular bundle in the vein.

Occurrence of aflatoxins and aflatoxin-producing strains of Aspergillus flavus in soybeans. G. A. BEAN, J. A. SCHILLINGER, & W. L. KLARMAN (Univ. Md., College Park). Above-average rainfall in Maryland during August, September, and October, 1971, resulted in heavy mold growth in soybeans. Twenty-eight samples, including three cultivars and 16 experimental lines collected from field plots, and four samples used in poultry feed, were assayed for aflatoxins and aflatoxin-producing strains of *A. flavus*. Aflatoxins were identified by thin-layer chromatography, absorption spectra, and chick embryo bioassay. Samples with the highest percentage of moldy kernels were from an area having the greatest rainfall; however, there was no correlation between percentage moldy grain and the occurrence of aflatoxins. Aflatoxins were found in 14 samples, two of which were used in poultry feed. To isolate *Aspergillus* spp., soybeans were surface-sterilized and placed on potato-dextrose agar containing 6.0% NaCl and incubated at 40 C. *Aspergillus* spp. were isolated from 11 samples; five of these isolates produced aflatoxins in liquid medium. No attempt was made to identify the species of *Aspergillus* present. Of the cultivars, Wayne contained more aflatoxin than either Callard or Cutler. Roasting soybeans did not affect aflatoxin levels.

Localization of infections in tomatoes having single-dominant-gene resistance to Fusarial wilt. C. H. BECKMAN, D. M. ELGERSMA, & W. E. MAC HARDY (Univ. R.I., Kingston, Phytopathol. Lab., Baarn, The Netherlands, Univ. Guelph, Guelph, Ont., Can.). The buildup and distribution of *Fusarium oxysporum* f. sp. *lycopersici* in near-isogenic lines of susceptible Pearson and resistant Pearson

VF-11 tomato were investigated. When intact root systems were drenched with inoculum, no symptoms appeared in either isolate, indicating equal resistance of these isolines to invasion. When *Fusarium* microspores and sporesize red tracer particles were taken directly into the vascular systems, the initial distribution was similar in the two isolines. Buildup of the pathogen, when calculated for initially infected vessels only, was comparable in both isolines. Thus, these isolines appeared to function comparably through the period of pathogen buildup in the infection court. Secondary distribution, however, remained limited in VF-11, whereas in Pearson most vessels in most vascular bundles eventually became infected. A microscopic study showed tylose occlusion of infected vessels 2 days after inoculation in VF-11, whereas occlusion was often delayed until day 7 or later in Pearson. It was concluded that the single-dominant-gene for resistance in VF-11 somehow modifies the process of tylose formation to provide a rapid sealing off of infected vessels, and thus prevents secondary distribution of the pathogen.

Snow mold abatement of golf courses with nonmercurial fungicides. K. M. BECKMAN & G. E. STORY (The Upjohn Co., Kalamazoo, Michigan). The loss of mercurial compounds through public clamor and restrictive legislation has necessitated a rapid discovery of suitable nonmercurial replacements for the abatement of the cold weather diseases caused by pink and gray snow mold, *Fusarium nivale* and *Typhula itoana*, respectively. Acti-dione TGF (cycloheximide), 3-[2-(3,5-Dimethyl-2-oxocyclohexyl)-2-hydroxyethyl]-glutarimide; Benlate 50W (benomyl); Demosan 65W (chloroneb); Tersan 75 (thiram); and Topsin M (thiophanate-methyl) 1,2-Bis(3-methoxycarbonyl-2-thioureido) benzene were evaluated individually and in dual or triple combinations. Organism specificity was a limiting factor with the individual compounds. Efficacy was improved between 35 and 80% with several of the combinations. Specific formulations, application timing and interval, and turfgrass cultural practices significantly influenced disease control. The cycloheximide-thiram-benomyl combination resulted in near-perfect control and best all-around performance.

Control of postharvest decay of mechanically harvested apples. S. V. BEER (Cornell Univ., Ithaca, N.Y.). Mechanically harvested McIntosh apples, which had been treated with fungicides before or after harvest, and untreated fruit were placed in cold storage (CS) at 0-1 C for 4 months or outdoor storage (OS) for 6 weeks. The fruit was graded for extent of decay, and isolations were made from each decayed fruit. Postharvest dips in either benomyl (with or without Cl₂) or Thiabendazole [2-(4-Thiazolyl)-benzimidazole] (TBZ) + Cl₂ were more effective in reducing decay than preharvest sprays with benomyl or captan under both storage situations. All treatments resulted in some reduction of decay as compared to that of untreated fruit. The most effective treatments for CS apples were dips in 225 ppm benomyl + 500 ppm Cl₂ or 1,000 ppm TBZ + 500 ppm Cl₂. Benomyl at 450 ppm + 500 ppm Cl₂ was most effective for OS apples. Pathogens most frequently isolated from untreated decayed fruit of both storage treatments were *Penicillium* sp. and *Botrytis* sp. High proportions of these fungi also generally were isolated from decayed fruit which had been preharvest-treated with benomyl or captan. *Alternaria* sp. was most frequently isolated from CS-decayed fruit which had been dip-treated with benomyl + Cl₂ or TBZ + Cl₂. Bacteria and yeastlike organisms were most prevalent in similarly treated OS fruit.

Effect of light intensity and quality on peroxidase activity

associated with resistance of tomato cultivars to Septoria lycopersici. W. G. BENEDICT (Univ. Windsor, Ontario, Can.). Peroxidase activity in leaves of tomato, *Lycopersicon esculentum* 'Bounty', 'Campbell 1327', 'Heinz 1350', and 'Rutgers', infected by *Septoria lycopersici* and exposed to various intensities and wavelengths of light, was assayed by means of acrylamide flat gel electrophoresis, followed by staining with 3-amino 9-ethyl carbazole. Inoculated plants of Bounty, Campbell 1327, Heinz 1350, and Rutgers, in that order, showed increasing leaf peroxidase activity and increasing resistance to the early blight disease as evidenced by the decreasing numbers of lesions produced by the leaf spot pathogen. When grown in decreasing light intensities, all four tomato cultivars showed increasing leaf peroxidase activity and increasing resistance to the disease, with a notable exception occurring at a light intensity of 14,200 ergs/cm² per sec. When grown in a light intensity of about 12,000 ergs/cm² per sec utilizing various color filter glasses to give specific wavelengths of light, all four tomato cultivars showed increasing peroxidase activity and disease resistance according to the color and wavelength of the light, being lowest in green (515 nm), followed by orange (640 nm), red (720 nm), yellow (456-550 nm), and blue (430-436 nm).

Enzyme profiles and virulence in phosphatidase mutants of Erwinia carotovora. L. BERAHA, B. A. BILLETTER, & E. D. GARBER (ARS, USDA, Dep. Biol., Univ. Chicago, Chicago, Ill.). An autoclavable medium of nutrient agar and 0.1% refined homogenized soybean lecithin was suitable for the detection of excreted phosphatidase in situ around colonies of *Erwinia carotovora*. Hg Cl₂ (1-1,000) enhanced the opaqueness of the circular zone of hydrolysis. In both the wild type, virulent phosphatidase (+) strain and an avirulent phosphatidase (-) strain, N-Methyl-N-nitro-N-nitrosoguanidine-induced mutants of the following additional types were selected: virulent, phosphatidase (-) and avirulent, phosphatidase (+). Phosphatidase (+) mutants of the avirulent strain were virulent and had levels of polygalacturonase (PG), pectate lyase (PL), and cellulase (Cx) as high or higher than the wild type. Mutants having partial restoration of phosphatidase activity and low levels of PG, PL, and Cx were reduced in virulence or avirulent. In mutants made from the wild type strain, avirulence was always associated with no phosphatidase activity and low levels of PG, PL, and Cx. Mutants that were phosphatidase (-) could be either virulent or avirulent, depending only on the relative activities of PG, PL, or Cx. Therefore, if phosphatidase is involved in the virulence of *E. carotovora*, it is not by itself the sole enzyme determining virulence.

Helminthosporium turcicum incidence and infection rates in sweet corn. R. D. BERGER (Univ. Fla., Belle Glade). Disease incidence (x) and infection rates (r) of *Helminthosporium turcicum* in 18 replicated sweet corn lines were followed in a large, sprayed cornfield. Lesion numbers times average lesion size divided by leaf area provided accurate estimates of x when x was less than 1%. The degree of disease tolerance of most lines was exhibited after the first flush of observed lesions, although some escapes were present. As the disease progressed, the most tolerant, intermediate, and susceptible lines were satisfactorily detected at a very early stage in the epidemic when x was 0.25-0.6, 0.6-0.9, and 0.9-2.6%, respectively. Most lines retained their relative position in disease severity through the season, and at harvest, x was 5.2-6.2, 6.4-8, and 9.4-15% for tolerant, intermediate, and susceptible lines, respectively. The highest r values (0.18-0.27) occurred at layby, when daily leaf area increase was maximal because infections occurred on the rapidly emerging foliage in blight-favorable weather.

Low r values (0.01-0.11) were found after tasseling because few new lesions developed on the sprayed corn, and most disease increase was primarily from enlarging lesions. Both x and r values were useful in determining varietal susceptibility and spray program effectiveness.

Relationship between per cent dry matter content of potato tubers and susceptibility to bacterial soft rot. W. L. BIEHN, D. C. SANDS, & L. HANKIN (Conn. Agr. Exp. Sta., New Haven). Severe loss of Connecticut potatoes in 1971 due to bacterial soft rot was associated with wet soil conditions just before harvest. Excessive soil moisture near the end of the growing season produces tubers that are easily injured and of low specific gravity. Therefore, tubers with a low per cent dry matter content may be more susceptible to bacterial soft rot. Potato discs cut from the inner parenchyma of Katahdin tubers obtained from five sources were inoculated separately with two isolates of *Erwinia carotovora*. Tubers with 13.5-17.0 and 18.5-21.5% dry matter averaged 51.3 ± 4.0 and $30.4 \pm 4.7\%$ maceration, respectively, after 26- to 30-hr incubation. This difference was statistically significant. These studies suggest that Katahdin tubers with a relatively high dry matter content are generally less susceptible to soft rot caused by *E. carotovora* than tubers with a lower dry matter content.

Repression of pectic enzymes and pathogenesis in Erwinia carotovora. W. L. BIEHN, D. C. SANDS, & L. HANKIN (Conn. Agr. Exp. Sta., New Haven). Since pectic enzymes produced by *Erwinia carotovora* are considered important in pathogenesis, compounds that repress pectic enzyme synthesis should protect potato tubers against bacterial soft rot. Previous work has demonstrated that glucose represses in vitro pectate lyase production by this bacterium. In our experiments, glucose and sucrose greatly repressed in vitro pectate lyase and endopolygalacturonase synthesis in *E. carotovora* (ATCC 8061) and in isolates obtained from rotted potatoes. Potato discs, vacuum infiltrated with 5% solutions of glucose, sucrose, and H_2O , and inoculated with *E. carotovora*, showed an average maceration of 5, 5, and 43%, respectively, after 30-hr incubation. Even glucose at the 2% level was effective in reducing maceration of potato discs. These studies suggest that compounds which greatly repress pectic enzyme production by *E. carotovora* may also inhibit soft rot in potato tubers.

Differences in host-cell responses to the reniform nematode. W. BIRCHFIELD (ARS, USDA, La. State Univ., Baton Rouge). The reniform nematode, *Rotylenchulus reniformis*, parasitizes many dicots in tropical and semitropical areas, but few monocotyledonous plants. Centennial sweet potato, Pinto bean, Black Eye pea, Long Green cucumber, Creole tomato, Golden Cross Bantam corn, and C. P. 44-101 sugarcane were studied to determine differences in host parasite relations. Plants were grown 3 months in infested soil in the greenhouse; the roots were removed, washed, killed, fixed, embedded in paraffin, stained, and examined for infection, location of parasite, and symptoms. The parasite preferred the pericycle of all plants studied except corn and sugarcane. Enlarged cell nuclei, nucleoli, and granulated cytoplasm which stained differently were observed near the feeding site of all host plants. Cells of the parasitized pericycle of sweet potato were enlarged $3 \times$ normal size in small feeder roots, but storage roots were not infected. The parasite fed throughout the cortical parenchyma of corn, but no enlargement of these cells occurred. Sugarcane was not parasitized. A few females were embedded in the root tissue of all host plants studied.

Aphids repelled and virus diseases reduced in peppers planted on aluminum foil mulch. L. L. BLACK & L. H. ROLSTON (La. State Univ., Baton Rouge). Aluminum foil mulch was compared to black polyethylene mulch and no mulch for its effectiveness in repelling aphids and reducing virus infection of *Capsicum annuum*, bell peppers. Aphids were trapped on yellow sticky boards 18 inches above the rows. Tobacco etch virus (TEV), potato virus Y, and cucumber mosaic virus were identified in the test plots. The number of aphids trapped over the aluminum foil mulch plots was less than 10% of those trapped over rows with black polyethylene or no mulch during the first 3 weeks after planting. During the next 5 weeks, the percentage gradually increased to about 50%. At the first harvest, 10% of the plants grown in aluminum foil plots were showing mosaic symptoms as compared to 85% of the plants in black plastic and 96% on no mulch plots. Plants in aluminum foil plots yielded 58% more than those on black polyethylene, and 85% more than those on no mulch. In a separate experiment, *C. frutescens* 'Tabasco' was planted on the same treatments, and numbers of plants killed by TEV were recorded over an 8-week period. Two months after planting, 42% of the plants grown on aluminum foil plots had died as a result of TEV infection as compared to 96% on black plastic and 98% without mulch.

Effect of fertilization, site, vertical position, and age of loblolly pine seedlings on susceptibility to fusiform rust. R. L. BLAIR & E. B. COWLING (Intern. Paper Co., Bainbridge, Ga., N.C. State Univ., Raleigh). During the basidiospore-flight-period of 1969 (1 April-30 June), 9- to 22-week-old loblolly pine seedlings were exposed in duplicate greenhouse flats to the natural inoculum of *Cronartium fusiforme* at 0.5 and 0.3 m above ground on three sites that differed markedly in hazard of fusiform rust in 1964-65. Half the seedlings in each flat were fertilized by a watering twice (1 March and 1 May) with a 12-6-6, N-P-K nutrient solution. A comparison was made of the average percentage of shoots with fusiform rust galls (APG) on seedlings suspended above the sites and adjacent 6-year-old seedlings of the same loblolly pine family planted on the sites. The results indicate that: (i) Fertilization strongly predisposed the seedlings to fusiform rust (APG = 43% for fertilized and 12% for nonfertilized seedlings); (ii) the extent of predisposition was greatly influenced by the site above which the seedlings were exposed (effect of fertilization varied from 2- to 6-fold on the three sites); (iii) the hazard of rust infection was not greatly different at 0.5 than at 3 m aboveground (APG = 11% at 0.5 m and 13% at 3 m); and (iv) shoots of nonfertilized 9- to 22-week-old and adjacent 6-year-old seedlings were not greatly different in susceptibility to fusiform rust (APG = 12% compared to 7%, respectively).

Ultrastructural comparisons of lesions produced by Race T of Helminthosporium maydis on susceptible and resistant cultivars of corn. R. O. BLANCHARD (Univ. Ga., Athens). Ultrastructural observations of lesions (up to 48 hr after inoculation) produced by Race T of *Helminthosporium maydis* on susceptible and resistant cultivars of corn inbreds (B37T, B37N) show several pronounced differences. Epidermal cells in lesions of the susceptible cultivar do not change shape appreciably, but similar cells of the resistant cultivar are generally collapsed. Accompanying the collapsed epidermis in the resistant cultivar is an unidentified matrix material which appears to fill the intercellular spaces. This material precedes fungal proliferation. No evidence of a similar material was found in the susceptible cultivar. Fungal hyphae in both cultivars appear to proliferate intercellularly. Large amorphous accumulations are characteristically found in

hypae within resistant tissue, but not in hyphae within susceptible tissue. Chloroplasts of lesions in resistant cultivars do not appear to be immediately affected, and are seen intact well into the lesions. At the periphery of lesions in susceptible cultivars, large vacuoles develop in the chloroplasts. Chloroplasts with internal disorganization usually show disruption of the outer membranes in the resistant cultivar, but these membranes tend to remain intact in the susceptible one.

Synthesis of heterocaryons between field isolates of Rhizoctonia solani. H. BOLKAN & E. E. BUTLER (Univ. Calif., Davis). New techniques have shown that heterocaryotic field isolates of *R. solani* [= *Thanatephorus cucumeris*] will form new heterocaryons when paired with each other. The isolates studied here were obtained from nature and shown to be heterocaryons through analysis of colonies obtained from single basidiospores. When heterocaryotic isolates were paired in various combinations on potato-dextrose agar containing 1% charcoal, compatible pairings gave rise to mycelial tufts where the two isolates met. Colonies originating from hyphal tips from the tufts were morphologically distinct from either parent isolate. When lithium-sensitive and lithium-tolerant field isolates were paired, all hyphal tip cultures from the tuft mycelium showed an intermediate response to lithium, indicating that the tuft mycelium was heterocaryotic. These lithium-intermediate isolates did not form tufts in pairings with the parent isolates, suggesting that the lithium-intermediate heterocaryons contained nuclei from both parents. Continuous transfer and hyphal tip analysis showed that the synthesized heterocaryons remained true to type.

Viruslike particles in virulent strains of Helminthosporium maydis. R. F. BOZARTH, H. A. WOOD, & R. R. NELSON (Boyce Thompson Inst., Yonkers, N.Y., Pa. State Univ., University Park). Nine *Helminthosporium maydis* isolates were screened for the presence of viruslike particles (VLPs) by extraction of 10 g dry weight mycelium with 0.1 M, pH 7.0, phosphate buffer followed by differential and sucrose density-gradient centrifugation. Ultraviolet-absorbing fractions from the gradients were negatively stained and examined by electron microscopy. Isolates of Race T collected from corn in North Carolina, Guiana, and Nigeria contained VLPs. Isolates from fescue, rice, and corn collected in The Netherlands, Thailand, and El Salvador which caused severe blight in corn also contained VLPs. VLPs were not observed in weakly pathogenic isolates from crabgrass, *Poa*, and corn from Mexico, Italy, and Georgia, respectively. All VLPs were isometric and 35-40 nm diam. Sucrose density-gradient analysis indicated that these VLPs contained several centrifugal components and that there were at least three different types of VLPs involved.

Isometric viruslike particles in maize with stunt symptoms. O. E. BRADFUTE, R. LOUIE, & J. K. KNOKE (Ohio Agr. Res. Development Center, ARS, USDA, Wooster). Three maize plants with shortened upper internodes and blotchy, yellow-green upper leaves were collected from the field in southern Ohio. One maize seedling with a disease agent transmitted by *Graminella nigrifrons* from Johnson grass collected in northern Kentucky was received from T. P. Pirone (Univ. Ky., Lexington). Isometric viruslike particles (IVLP) were found in all four plants by electron microscopy of leaf tissue thin-sections. The IVLP (26-31 nm) were larger than maize ribosomes and distinctly angular. They appeared individually, and aggregated in the cytoplasm and central vacuole of phloem cells. IVLP were observed in cells outside the bundle sheath in one section. Dense-staining cellular

inclusions, similar to the X-material in tissue infected with tobacco mosaic virus, and other cytopathic changes were found in cells with IVLP. Neither cylindrical inclusions and flexuous rods associated with maize dwarf mosaic nor the mycoplasma-like bodies associated with Rio Grande and Louisiana corn stunt were observed. Host and vector relationships of the IVLP and their role in the maize disease complex are to be determined.

Correlation of grapevine shoot growth with maturity of Phomopsis viticola pycnidia. A. J. BRAUN (N. Y. State Agr. Exp. Sta., Geneva). Average shoot length was a reliable indicator of the maturity of pycnidia of *Phomopsis viticola*, the cause of dead arm disease of grapevines. In 1968, a single application of Orthocide 50W (captan) at 4 lb./50 gal per acre applied when the mean shoot length was 4.9 inches gave 99.9% control of the cane spot phase. Single application at the 1.0-, 2.2-, and 3.0-inch stages were less effective. In 1970, Orthocide 50W at 2 lb./25 gal/acre at the 6.0-inch stage gave equally good control. Sprays applied at the 3.6-, 8.4-, and 14.2-inch stages were less effective. A study of shoot orientation and elongation indicated that infection beyond the seventh internode seldom occurs because the susceptible portion of the shoot is out of reach of splashing raindrops carrying spores from the infected canes. In view of the excellent control with a single application at the time when the pycnidia first mature, the previously recommended application at the 1- to 2-inch stage can be omitted without an impairing of control.

Inheritance of resistance to Puccinia arachidis in peanut. K. R. BROMFIELD & W. K. BAILEY (ARS, USDA, Frederick, Beltsville, Md.). In the 1970-71 USDA peanut nursery, Isabela, Puerto Rico, a single peanut plant in a planting of rust-resistant P.I. 298115 was observed to differ markedly from the others in rust reaction. Because this plant also produced red testa in contrast to the white testa of the others, it was assumed to be an F_1 hybrid that arose from a natural cross between a maternal rust resistant plant of P.I. 298115 and a paternal rust susceptible plant of unknown identity. All seeds produced by the atypical plant were planted in the greenhouse at Beltsville, Md. Emerged F_2 plants were subsequently transplanted to the field for seed increase. Detached leaflets of each of the 108 resulting F_2 plants were individually tested for reaction to two cultures of peanut rust at Frederick, Md. Leaflets from an individual plant reacted similarly to both rust cultures. Seven of the plants were resistant, seven were highly susceptible, and the others were intermediate in reaction. The data support the assumption of hybrid origin of the atypical plant, and indicate bigenic control of rust reaction in this cross with resistance recessive. Members of the F_3 generation will be observed for rust reaction in the field in Puerto Rico.

Persistence of benomyl in oranges from grove application and its effect on green mold. G. E. BROWN & L. G. ALBRIGO (Fla. Dep. Citrus, Univ. Fla., Lake Alfred). Methyl 2-benzimidazole-carbamate (MBC), the breakdown product of benomyl, was recovered from orange fruit after the trees were sprayed with benomyl. MBC was detected in the peel, and slight amounts were also present in the juice 1 day after applying benomyl to the trees. Movement of MBC from the fruit surface into the peel was favored more by rainfall than by dew. Translocation of MBC from the foliage into the fruit was not detected. MBC was still detected in the peel of Hamlin oranges at 70 days, and in Valencia oranges at 86 days, after the trees were sprayed with 300 and 500 $\mu\text{g}/\text{ml}$ of benomyl, respectively. Some control of green mold, resulting from artificial inoculations, was obtained in Hamlin oranges

picked 70 days after spraying, and in Valencia oranges picked at 84 days. Benomyl applied with emulsified oil, Nu-Film 17, or Biofilm to the Hamlin trees provided better control of mold than an application of benomyl alone. Control of green mold in the Valencia oranges was also enhanced by mixing 1 or 3% Vapor Gard with the benomyl when it was sprayed on the trees.

Improved procedures for the preparation of pathological specimens for scanning electron microscopy. M. F. BROWN (Univ. Mo., Columbia). Procedures commonly employed for drying pathological specimens for scanning microscopy involve removal of incorporated water by air drying or in vacuo as the sample surface is coated with a conductive metal. Due to surface tensions which develop at the liquid-gas interface, these procedures result in specimen shrinkage, cell collapse, or other distortions of normal morphology. Experience with a variety of fungal pathogens grown in vitro or in vivo has led to preparative procedures in which these artifacts are minimized. Specimens are fixed at 4 C in 6% glutaraldehyde in a 0.1-M phosphate buffer at pH 7.2 for 6-24 hr, and postfixed at 4 C in 2% OsO₄ in the same buffer for 12-48 hr. After several rinses in distilled water, the samples are processed by freeze drying (FD) or Anderson's critical point drying (CPD) method. Specimens for FD are quenched for 5 min in isopentane, cooled to -150 C in liquid nitrogen, and dried under a vacuum of 0.1 Torr at -55 C. FD is satisfactory for bacteria and fungal samples, with thin, unpigmented cell walls and low vacuolar volumes. CPD yields excellent preservation of highly vacuolated plant tissues and fungi with thick, pigmented, or hydrophobic walls, whereas these types of specimens often have drying artifacts when preserved by the FD procedure.

Aflatoxin inhibition and detoxification by a culture filtrate of Aspergillus niger. CHARLOTTE BURNETT & G. W. RAMBO (Univ. Md., College Park). *Aspergillus flavus* is a common storage mold of peanuts, rice, and other food products. The fungus produces aflatoxins, the most potent naturally occurring carcinogens known. A culture filtrate of *A. niger* inhibited synthesis of aflatoxin by *A. flavus* on peanut seed and in liquid medium (200 g sucrose, 7 g yeast extract, 3 g KNO₃, and 0.5 g MgSO₄/liter distilled water). *A. niger* detoxified aflatoxin in liquid culture. After 8 days' growth, only traces of aflatoxin were found in the medium and in the mycelial mat. The synthesis-inhibiting and detoxifying substance was heat-stable, but could not be extracted with chloroform, ether, hexane, or chloroform-methanol (2:1) at pH 5.6. Addition of methionine did not reverse the action of the inhibitor. Citric, fumaric, gallic, gluconic, and oxalic acids, organic acids produced by *A. niger*, were added to liquid medium prior to its inoculation with *A. flavus* to test these acids as possible aflatoxin inhibitors. The molar concentrations added were .005, 0.01, 0.05, and 1 M. Only fumaric acid inhibited aflatoxin synthesis.

Factors affecting overwintering and epidemiology of Helminthosporium maydis in Illinois corn. E. E. BURNS & M. C. SHURTLEFF (Univ. Ill., Urbana). Viable spores of *Helminthosporium maydis*, race T, were recovered from infected corn leaves stored 30 cm above the soil surface in nylon mesh bags from 7 December 1970 until 1 July 1971. Race T survived burial in the soil at 10 or 45 cm up to March 1971. Spores of race O were not isolated from infected leaves, stored either above or below the soil surface, after 1-month exposure to winter weather conditions in east-central Illinois. Crop tillage practices, such as chiseling or zero-tillage, favored the survival of race T. Viable conidia of

race T were obtained from infected corn debris left at the soil surface from harvest in 1970 until May 1971. Race T appeared first on susceptible corn planted in the zero-till plots and last in the plowed plots. Race T survived in stored crib corn. Seedling blight by race T occurred in less than 1% of heavily infected seed in greenhouse tests. Chlamydospores formed within conidia under dry conditions, and may contribute to the overwintering of race T. Spread of the fungus was correlated with spore trap and weather data. Infected corn leaf sections, stored dry at room temperature for 1 year, produced a new "crop" of conidia when moisture was added. Spore production under controlled conditions was optimal at 25 C and 100% relative humidity.

Relative half-life of certain races of Puccinia coronata avenae. E. BUSTAMANTE-R. & J. A. BROWNING (Iowa State Univ., Ames). Although race 264A has the widest virulence of any race of *Puccinia coronata avenae*, races 216, 326, and 264B have predominated successively in the USA. We investigated competitive ability of these four races using mixtures of monouredial isolates on the common host *Avena byzantina* 'Bond'. Although called races herein, our isolates do not necessarily represent these races in nature. We studied Bond-race mixture interactions for four generations under three day:night temperature regimes, 21/12, 26/17, and 32/19 C. We indexed the ratio of resistant-to-susceptible infection types in the mixtures on Iowa oat isolines X-421 and C-649, and used van der Plank's method for calculating relative half-life. Race 326 was most aggressive; other races mixed with it decreased. Race 216 increased in mixture with 264A and 264B, and 264B decreased relative to others. Race 264A, competing with 326, had a relative half-life of 0.9-2.5 generations that varied directly with temperature. Race 216, competing with race 326, had a relative half-life of 2.5-7.5 generations, without a definite temperature relation. Under the conditions of this study, the race with fewer virulence genes was not always most aggressive; therefore, factors for aggressiveness seem to be inherited independently of virulence.

Diagnosis of stubborn disease of citrus by chemical methods. R. L. CALDWELL & R. M. ALLEN (Univ. Ariz., Tucson). Identification of stubborn disease of citrus by some chemical methods was attempted. Methanol or water extracts of leaves plus acid hydrolysate of these samples were separated by thin-layer chromatography. Detection by ultraviolet (UV), ninhydrin, and diazotized sulfanilic acid followed. Only UV detection of cultivar Sexton tangelo (greenhouse samples), of several cultivars tested, gave consistent differences for healthy and diseased plants. Additional testing followed modified procedures of other workers. Water extracts of citrus bark were subjected to two successive separations by paper chromatography. UV absorption ratios for healthy and diseased tissues at 280 and 320 nm were variable. Bark samples from Frost Valencia and Frost Washington navel (orchard trees) gave results consistently different for healthy and diseased trees. Differences between healthy and diseased tissues appeared most consistent by both methods (leaf and bark) when samples were taken in summer. Samples from injured plants yielded results like those from stubborn plants. These chemical methods probably are of limited usefulness in diagnosing the presence of stubborn disease, under Arizona conditions, except as a supplement to data obtained by other diagnostic processes.

Ultrastructural characterization of races O and T of Helminthosporium maydis. O. H. CALVERT & J. A. WHITE

(Univ. Mo., Columbia). Three different isolates of race O and of race T of *Helminthosporium maydis* were studied in an attempt to differentiate between the two races on the basis of conidial ultrastructure. No differences in either dormant or germinating conidia could be resolved by light microscopy. Conidia of the two races were collected from cultures and prepared for electron microscopy. Ultrastructural examination revealed that race O and race T were, in terms of over-all subcellular organization, quite similar. However, racial differences were found in the wall structure of the conidia and in the occurrence of unidentified, osmiophilic bodies in the cytoplasm. Scanning electron microscopy revealed an extracellular sheath surrounding the conidiophores of both races, and indicated that a similar material may be associated with immature conidia. Examination of ultrathin sections confirmed the presence of this sheath.

Inhibition of movement of Ceratocystis ulmi in elm stems by wounding. R. J. CAMPANA & A. L. PRATT (Univ. Maine, Orono). Host recovery from Dutch elm disease is enhanced by delayed downward movement of conidia in vessels. Interference of vertical movement by wounding was tested in 400 inoculated small stems, and 64 inoculated or infected trees. Inoculations were made above or below wound cuts after wounding of xylem tissue; recovery of the fungus was attempted above or below cuts after 2, 4, or 6 weeks. Small trees were inoculated above saw cuts 2 weeks after wounding. Large infected elms were cross-wounded in the trunk on infected sides at 90-240 cm above ground level by chain saw cuts 2.5 cm deep. The fungus could not be detected beyond wound cuts in small twigs after 2 weeks, but was detected in about 50% after 4 weeks, and 100% after 6 weeks. In small trees, fungal movement was delayed long enough to allow recovery in two of four trees. Wounding did not prevent vertical movement beyond severed tissue, but delayed the fungal distribution to the base of trunks, and some trees recovered.

Transmission of sweet potato russet crack virus. R. N. CAMPBELL, NANCY M. MELINIS, & D. H. HALL (Univ. Calif., Davis). Russet crack is characterized by distinct lesions on the roots of sweet potatoes, especially Jersey Orange. The agent is maintained when sweet potato is vegetatively propagated, and can be graft transmitted to and recovered from sweet potato, *Ipomoea setosa*, *I. nil*, and *I. tricolor*. The foliar symptoms on these hosts do not differ significantly from those produced by sweet potato feathery mottle virus that is aphid-borne and causes no russet crack. Russet crack is stylet-borne by *Myzus persicae*. Russet crack is also mechanically transmissible with difficulty from sweet potato to *I. nil* or from *I. nil* to sweet potato, but easily from *I. nil* to *I. nil* with the same symptoms as when graft-inoculated. No symptoms have developed on hosts in other families after mechanical inoculation from *I. nil*. Flexuous rods, about 800 nm long, are associated with russet crack and with feathery mottle viruses. The preferred hypothesis is that russet crack is caused by a strain of the sweet potato feathery mottle virus.

Serology of Xanthomonas vesicatoria. R. CHARUDATTAN & R. E. STALL (Univ. Fla., Gainesville). Antigenic analyses were made on 72 isolates of *Xanthomonas vesicatoria* from pepper and tomato. Antisera were prepared in rabbits against sonicated bacterial suspensions of seven isolates, four belonging to the tomato group and three to the pepper group. Heated (2 hr at 100 C in a water bath) and unheated whole cell suspensions of bacteria (14×10^8 cells/ml) as well as sonicated suspensions of bacteria were tested against antisera in agar double-diffusion tests. Two

serotypes were distinguished among the isolates tested, based on the presence or absence of specific precipitin bands. Serotype I isolates formed 1-3 specific arclike bands close to antigen wells with unheated antigens and a thick band near the antiserum well with heated antigens. Antigens and antisera of serotype II isolates did not form these arclike bands. There were other nonspecific antigens common to the two serotypes. There was no correlation between serotypes and the host of origin of the isolates; tomato and pepper isolates included both serotypes. Also, pathogenicity, ability to hydrolyse starch, streptomycin resistance, and age of isolates in culture had no correlation to the serotypes.

The effect of leaf and plant leachates from five tomato cultivars varying in resistance to Botrytis cinerea on germination of B. cinerea spores. L. G. CHOU (Ohio Agr. Res. Development Center, Ohio State Univ., Wooster). The carbohydrate content of leachates obtained from aboveground portions of tomato plants or individual leaves in 1,000 ml or 150 ml of water, respectively, ranged from 1.0 to 6.6 µg/ml and varied with different cultivars. Germination of *Botrytis cinerea* spores ranged from 0 to 71% in tenfold plant or fifteenfold leaf concentration of the leachates. Leaf leachates from various cultivars contain both stimulatory carbohydrate and inhibitory phenol substances. Petroleum ether, but not ethyl ether fractions of acidified leachates, inhibited spore germination in solutions of 100 µg/ml glucose. Quantitative determination of phenols in the petroleum ether fraction of leachates from the various cultivars showed a lack of correlation between phenol content of leachates and susceptibility to *B. cinerea*. Susceptibility of different cultivars to *B. cinerea* could not be explained on the basis of differences in carbohydrate or phenol content of their leachates.

Comparative properties of tomato ringspot virus isolates associated with Prunus stem-pitting disease. E. L. CIVEROLO & S. M. MIRCETICH (USDA, Beltsville, Md.). Stone fruit isolates of tomato ringspot (TomRSV-SF) associated with *Prunus* stem pitting disease induced local necrotic lesions in *Phaseolus vulgaris*, whereas TomRSV (ATCC-78) and peach yellow bud mosaic virus (PYBMV) induced local chlorotic rings, ringspots, and line patterns. In *Vigna sinensis*, the TomRSV-SF isolates induced local chlorotic and necrotic lesions, necrotic rings, and local vein necrosis followed rapidly by systemic terminal necrosis and collapse of the entire seedling; whereas TomRSV (ATCC-78) and PYBMV induced local chlorotic lesions, necrotic rings, or chlorotic line patterns followed by systemic chlorotic spots, mottling, and stunting, but not systemic terminal necrosis. TomRSV-SF isolates and TomRSV (ATCC-78) produced single congruent precipitin lines in agar double-diffusion tests when reacted against TomRSV (elderberry isolate) antiserum. TomRSV-SF isolates were serologically related to, but distinct from, PYBMV. Partially purified preparations of TomRSV (PV-78), PYBMV, and TomRSV-SF (apricot isolate) contained two virus-specific components corresponding to the 52-54S and 124-128S components reported for a raspberry isolate of TomRSV. Peach and apricot seedlings were infected with TomRSV-SF (apricot isolate) by mechanical inoculation or implantation of infected cucumber tissue beneath the bark.

Longevity of ergot sclerotia in cold storage. R. L. CLARK (USDA, ARS, Iowa State Univ., Ames). After 10 years in cold storage, germination of ergot (*Claviceps* sp.) sclerotia was 60% in contaminated seed of *Agropyron caninum* (P.I. 208062); 54%, after 11 years in *A. repens* (222520); 20%, after 13 years in *A. elongatum* (222958); 40%, after 13 years in *A. intermedium* (222961); and 16%, after 11 years in

Bromus carinatus (232205). Sclerotia were selected from seed lots held in storage for at least 10 years. Storage had been in glass jars tightly capped (not sealed) at 6 C and 70-75% relative humidity. Sclerotia were surface-sterilized in 0.5% sodium hypochlorite for 15 min, placed on water agar slants, and incubated at 15 C. Germination readings were taken after 3 months and again after 6 months. The North Central Regional Plant Introduction Station, Ames, Iowa, maintains more than 1,300 lines of grass. Many of our grass seed harvests contain large numbers of ergot sclerotia which must be removed by hand. Since longevity of ergot sclerotia under field conditions is limited, we hoped to eliminate the field problem of viable sclerotia by long term seed storage. These results show that this is not feasible. As far as is known, this is the longest reported survival of ergot sclerotia.

Internal rib necrosis and rusty brown discoloration produced in lettuce, variety Climax, by lettuce mosaic virus infection. STELLA M. COAKLEY & R. N. CAMPBELL (Univ. Calif., Davis). Internal rib necrosis (IRN) occurs only in the lettuce cultivar Climax, and is characterized by necrosis of parenchyma cells in the midrib. With the light microscope, it is easily distinguished from ammonia damage that causes necrosis in the vascular elements. Climax and Vanguard or Calmar lettuce plants were inoculated with lettuce mosaic virus (LMV) in growth chambers operating at 65-45 F during photo- and dark periods, and indexed for infection on *Chenopodium quinoa*. IRN was induced in 75% of 51 infected Climax plants, but not in other cultivars. IRN first appeared in a minimum of 21 days and in the youngest leaves present at the time of inoculation. Rusty brown discoloration (RBD) always developed when infected Climax heads were stored at 32-34 F for 7 days, whereas virus-free heads remained normal. RBD seems to be more severe the longer plants have been infected. Similar results were obtained when plants were inoculated in the field.

Indoleacetic acid oxidase activity in sunflowers stunted by Plasmopara halstedii. YIGAL COHEN & W. E. SACKSTON (Macdonald Coll. McGill Univ., Ste. Anne de Bellevue, Que., Can.). Sunflower plants stunted by downy mildew (*Plasmopara halstedii*) did not show phototropic or negative geotropic responses. Application of exogenous indoleacetic acid (IAA) or kinetin to inoculated plants before or after appearance of mildew symptoms did not counteract stunting; gibberellic acid (GA_3) counteracted stunting to a slight extent. Activity of IAA-oxidase in stem sections of *Plasmopara*-infected sunflowers was positively correlated with intensity of stunting. IAA-oxidase activity of leaves was low and essentially equal in healthy and infected plants.

Zoosporangia of Plasmopara halstedii in epidemiology of downy mildew of sunflowers. YIGAL COHEN & W. E. SACKSTON (Macdonald College, McGill Univ., Ste. Anne de Bellevue, Que., Can.). Zoosporangia of *Plasmopara halstedii* survived several days in soil and induced systemic infection through belowground tissues of sunflower seedlings. Zoosporangia also induced systemic infections through stem apices of sunflowers, and induced local infections on leaves. Heavy infection occurred when germinating seeds were immersed in zoosporangial suspensions for 3 to 6 hr. Infection was induced by zoospore concentrations as low as 200/ml at the optimum temperature of 15 C. Higher concentrations were required to induce infection at 10 and 20 C.

Drought stress as a factor stimulating the saprophytic activity of Helminthosporium sativum on bluegrass debris. P.

F. COLBAUGH & R. M. ENDO (Univ. Calif., Riverside). The highest incidence of *Helminthosporium* leaf spot and foot rot occurred within dry areas of bluegrass lawns; sporulation was often abundant on the drought-stressed plants and debris. In the well-watered areas, *Helminthosporium* activity was usually detected as an occasional leaf spot. Studies of the factors underlying the saprophytic activity of *H. sativum* (isolate T54) on dry debris were undertaken. *H. sativum* grew only at relative humidities above 90%. Conidia did not germinate on moist debris, but when removed from the debris, the spores germinated rapidly in deionized water. If the debris was allowed to dry, and subsequently was remoistened, per cent conidial germination was greatest immediately after re-wetting. As the time between remoistening the debris and application of conidia was increased, percentage of germination decreased. Leached material collected from the dried debris after remoistening was richer in soluble nutrients than that from debris kept continuously moist. The release of soluble nutrients after remoistening dried turf debris appeared to be an important factor in stimulating the saprophytic activity of the fungus.

Ultrastructural changes in Pythium aphanidermatum zoospores and cysts during encystment, germination, and penetration of primary lettuce roots. W. M. COLT & R. M. ENDO (Univ. Calif., Riverside). Roots of 4-day-old lettuce seedlings (*Lactuca sativa* 'Calmar') grown aseptically in aerated nutrient solution in the dark were immersed in a suspension of *Pythium aphanidermatum* zoospores. Roots with encysted zoospores were examined by transmission and scanning electron microscopy (SEM) at various times after immersion. The liquid CO₂ critical point method was used in sample preparation for SEM. When zoospores encysted, mucilage was deposited on the surface of the cyst. Mucilage may function in adhesion of the cyst to the root, thus facilitating germ tube penetration. Mucilage appeared to originate from vesicles which contained granular material; these vesicles occurred in great abundance near the zoospore plasmalemma. Striated "fingerprint" inclusions appeared to partially disintegrate during encystment and germination, and were subsequently discharged into the developing vacuole within the cyst. Lipid-containing vesicles and other cytoplasmic organelles were also released into the cyst vacuole. In germ tubes and in penetrating hyphae, only lipid vesicles were discharged into small hyphal vacuoles.

Electrolyte leakage from Texas male sterile and normal cytoplasm corn leaves infected with Helminthosporium maydis races O and T. J. C. COMSTOCK & C. A. MARTINSON (Iowa State Univ., Ames). Texas male sterile (Tcms) and normal (N) cytoplasm corn leaves were inoculated with 10-15 *Helminthosporium maydis* race T or O conidia/mm² leaf surface. Rate of leakage after 20 hr was 4.5 and 1.5 μ mos/hr, respectively, for Tcms and N leaves inoculated with race T, and 1.1 and 0.9 μ mos/hr, respectively, for Tcms and N leaves with race O. Leakage rate of noninoculated checks of both lines was 0.05 μ mos/hr. With race T, the first measurable increase in leakage occurred 13-14 hr after inoculation for Tcms leaves versus 18-20 hr for N leaves. Tcms-specific toxin (T-toxin) produced by race T was detected in the infection droplet by 18 hr. Leakage rate from Tcms leaves inoculated with race O increased 2.5 \times when inoculum was suspended in partially purified T-toxin; there was no increase from N leaves with T-toxin. Leakage rate from Tcms leaves treated only with T-toxin was slightly higher than from water controls; T-toxin did not affect leakage from N leaves. When Tcms corn leaves resistant to *H. carbonum* were inoculated with *H. carbonum* plus T-toxin, the rate of electrolyte leakage was 5.0 μ mos/hr after 30 hr

compared to 1.8 μ mhos/hr without T-toxin. T-toxin is selective in its effect on Tcms leaf tissue, and probably is responsible for the increased virulence of race T on Tcms corn.

Occurrence of raspberry bushy dwarf virus in Rubus species in the United States. R. H. CONVERSE (ARS, USDA, and Ore. State Univ., Corvallis). Raspberry bushy dwarf virus (RBDV), an isometric virus originally described in Scotland and previously unknown in the United States, occurs in commercial fields of red and black raspberry in Oregon and Washington and in commercial red raspberry in eastern USA. Several cultivars are infected, all without recognizable symptoms. Canby red raspberry and Munger black raspberry cultivars are particularly heavily infected. RBDV also occurs in boysenberry in California. RBDV was identified in these cultivars by sap transmission from suspect leaves to *Chenopodium quinoa* seedlings. Such infected *C. quinoa* plants developed chronic oak-leaf patterns and leaf distortion without necrosis. Sap from RBDV-infected *C. quinoa* plants reacted strongly with antisera against RBDV obtained from Scotland in agar gel tests under conditions where nonspecific reactions did not occur. In Oregon, RBDV rapidly infects healthy Munger plants planted in infected Munger fields (25% infection/year). In Canby the virus infects 22% of open-pollinated seedlings.

Hypersensitivity in Capsicum annum cultivars caused by the tomato strain of Xanthomonas vesicatoria. A. A. COOK (Univ. Fla., Gainesville). Isolates of the tomato strain of *Xanthomonas vesicatoria* were inoculated into leaves of two pepper cultivars that were (i) hypersensitive; and (ii) susceptible to race 2, pepper strain, of the same bacterium. Multiplication in vitro, electrolyte loss patterns from inoculated leaf tissues, and visible symptoms were considered evidence of a hypersensitivity-inducing mechanism distinct from that caused by race 2, pepper strain. Temperature, but not light, was found to influence the time required for symptom expression and pattern of electrolyte loss. Results obtained with different isolates were indicative of distinguishable subtypes of the tomato bacterium and consistently different hypersensitive responses of the two pepper cultivars.

Ozone-induced isoenzyme changes in bean leaves detected by isoelectric focusing. C. R. CURTIS (Univ. Md., College Park). *Phaseolus vulgaris* 'Pinto III' plants were grown in filtered air for 14 days, then exposed to 18 ppm of ozone for 3 hr. Leaves were harvested 24 hr after exposure. Controls were not exposed to ozone. Deveined leaves or leaf discs were homogenized in 0.1 M Tris [tris(hydroxymethyl)amino methane] buffer, pH 8.0, and centrifuged at 30,000 g for 30 min at 2-4 C. The extract was layered on 5% polyacrylamide gels for isoelectric focusing in a pH 3-10 gradient. Gels were prepared with enzyme grade acrylamide and photopolymerized with riboflavin. Samples of extract (50 μ liter) were focused isoelectrically at a constant 110 v for 5 to 6 hr. The gels were then tested for peroxidase and esterase activities. The separated peroxidase isoenzyme bands from ozone-treated leaves were clearly more intense when viewed visually than the same bands from control leaves. Visual differences in esterase isoenzyme activity were more difficult to detect, but gel scans revealed subtle changes in activity of certain esterase bands. The significance of the detected changes in isoenzyme activities is unknown, but they probably reflect deleterious biochemical alterations in leaf metabolism caused by ozone.

Growth regulating and phytotoxic effects of a metabolite

produced by Fusarium moniliforme. H. G. CUTLER, R. J. COLE, B. R. BLANKENSHIP, J. W. KIRKSEY, & B. DOUPNIK, JR. (Univ. Ga. Coastal Plain Exp. Sta., ARS, USDA, Tifton, Ga.). A crystalline, water-soluble compound was isolated from *Fusarium moniliforme* found on Southern blight-damaged corn seed and cultured on cracked corn. It was bioassayed on corn, tobacco, cucumber roots, and wheat coleoptiles in dilution series. Aqueous solutions were either introduced into "funnels" of corn seedlings or sprayed on tobacco seedlings in the four-leaf growth stage, and ranged from 2,000 to 20 ppm (actual amounts per plant: corn, 200 μ g-2 μ g; tobacco, 2 mg-20 μ g). At 2,000 and 200 ppm necrosis, interveinal chlorosis, stunting, and morphological irregularities occurred. Cucumber root and wheat coleoptile extension bioassays were made in vitro from 200-2 ppm. At 200 and 20 ppm, cucumber roots were inhibited 68 and 19%. Wheat coleoptiles were inhibited 57 and 24% at 200 and 20 ppm, respectively. Chemical data suggest a structurally new growth inhibitor.

An assay for Helminthosporium victoriae toxin based on the induction of electrolyte leakage. K. E. DAMANN, JR., & R. P. SCHEFFER (Mich. State Univ., East Lansing). Toxin from *Helminthosporium victoriae* (HV) induces rapid loss of electrolytes from susceptible but not from resistant plants. Cheesecloth bags containing oat leaves were incubated in several toxin concentrations for 1 hr, rinsed 10 min, and leached in distilled water. Toxin-induced electrolyte loss was determined by electrical conductance of leaching solutions read at 0.5-hr intervals for up to 6 hr. The following characteristics of toxin-induced electrolyte leakage were noted: Dosage-response curves showed saturation kinetics; leakage rates were linear for at least 3 hr; and leakage rates increased with increases in toxin concentration over four orders of magnitude. Thus, the rate of induced leakage can be used to measure HV-toxin concentration in solutions. Other factors affecting toxin-induced electrolyte loss were toxin exposure time, weight of tissue, infiltration method, and age of tissue. Approximately equal concentrations of toxin were detected by root growth bioassay and electrolyte leakage assay; a toxin preparation diluted 10⁷ times gave significant responses in both cases. The electrolyte loss assay has several distinct advantages, including the short time required for completion.

A thin-layer chromatographic method for purification and detection of Helminthosporium victoriae toxin. K. E. DAMANN, JR., & R. P. SCHEFFER (Mich. State Univ., East Lansing). A rapid, reliable method was developed for routine preparation of *Helminthosporium victoriae* (HV) toxin in a state of high purity. Toxin-containing eluates from alumina columns were spotted on plates of Silica Gel GF containing an indicator which fluoresces at 254 nm. Chromatograms were developed in three solvents: *n*-butanol, acetic acid, water (3:1:1); *n*-propanol, acetic acid, water (200:3:100); or 2-butanone, propionic acid, water (15:5:6). Exposure to ultraviolet light of 254 nm revealed several fluorescing and quenching spots. Sequential sections of chromatograms were eluted with water and bioassayed for ability to induce electrolyte leakage from susceptible, but not resistant, oat tissue. Purified, highly active, host-selective HV toxin was found in areas with R_F values of 0.45, 0.75, and 0.90 for chromatograms developed with the three solvents, respectively. In each case, toxicity was correlated with a quenching spot visible during irradiation at 254 nm. Thus, small amounts of the labile HV toxin can be processed for individual experiments.

Motility of helical filaments produced by a

mycoplasma-like organism associated with corn stunt disease. R. E. DAVIS & J. F. WORLEY (ARS, USDA, Beltsville, Md.). The coiled filaments diagnostic of corn stunt (CS) infection in corn plants exhibit movements previously undescribed for filaments produced by mycoplasmas. Juice expressed from plants with CS symptoms was examined by phase contrast microscopy at about 21 C and found to contain high numbers of helical filaments. These filaments often whirled or spun rapidly about the long axis of the helix and, although we have observed no gliding or swimming, the filaments also exhibited flexing, bending, and curling motions reminiscent of those described for some spirochaetes. The helical filaments associated with CS, however, possess no cell wall, envelope, axial filament, or flagellae. Yet our observations, documented by cinematography, suggest that the movements are not the results of external forces and indicate the possibility of internal ultrastructure not yet revealed by electron microscopy. The findings constitute another characteristic, in addition to helical morphology, by which the mycoplasma-like organism associated with CS may be recognized and by which, in addition to natural habitat, it differs from descriptions of organisms now in the Mollicutes.

Apparent synchronous growth of plant viruses achieved by temperature manipulation. W. O. DAWSON & D. E. SCHLEGEL (Univ. Calif., Berkeley). Differential temperatures were used to separate the processes of systemic infection from virus synthesis, allowing massive synthesis in cells in approximately the same stage of infection. Primary leaves of cowpea, *Vigna sinensis*, mechanically inoculated with cowpea chlorotic mottle virus were maintained at an optimum temperature for virus synthesis (30 C), while the noninoculated trifoliolate leaves were maintained at a temperature (3-5 C) which allowed movement of the infectious agent into them but was too low for detectable virus synthesis. After 3 days when substantial amounts of virus had accumulated in the mechanically inoculated leaves, the entire plants were exposed to a temperature (32 C) which allowed virus synthesis. The lag period before the exponential phase of the infectivity curve was reduced to about 12 hr, and the maximum infectivity was observed at 24 hr. This is compared to the normal lag period of 24 hr and the peak of infectivity, 96-120 hr, after mechanical inoculation. Trifoliolate leaves, detached at the time of exposure to 32 C, contained no detectable infectivity; however, virus increased in these leaves at about the same rate as in attached leaves. This system also worked for tobacco mosaic virus in *Nicotiana tabacum*.

Positive correlation between extranuclear inheritance and mycovirus transmission in Ustilago maydis. P. R. DAY, S. L. ANAGNOSTAKIS, H. A. WOOD, & R. F. BOZARTH (Conn. Agr. Exp. Sta., New Haven, Boyce Thompson Inst., Yonkers, N.Y.). Two strains of *Ustilago maydis* (P1 and P3) contain spherical, viruslike particles (VLPs) with a diameter of 41 nm. Partial purification was achieved by extraction in 0.1 M, pH 8.0, phosphate buffer followed by differential and sucrose density-gradient centrifugation. Five components were observed with approximate sedimentation rates of 110 to 160 S. The VLPs were not serologically related to either of the 2 VLPs from *Penicillium stoloniferum* or the VLP from *P. chrysogenum*. The VLPs from the P1 and P3 strains appeared identical. P1 strains produce a proteinaceous substance that kills sensitive cells but is insensitive to this substance. P3 strains do not produce the killer substance and are insensitive to it. No VLPs were found in five cultures of a third strain (P2) which does not produce, and is sensitive to the killer substance. A P2 strain, converted to P1 by heterokaryon transfer, contained VLPs. The results confirm earlier data that the killer and insensitivity factors are

extranuclear, and show a positive correlation between the presence of VLPs and the toxin insensitivity factor in *U. maydis*.

Uso de plantas indicadoras en la determinación de aeropolutos en la Ciudad de México. L. I. DE BAUER (Colegio de Postgraduados, Chapingo, México). Durante los meses de abril a septiembre de 1971, se expusieron plantas indicadoras de lechuga, espinaca, frijol, alfalfa, tabaco y chile, en seis zonas urbanas y una industrial de la Ciudad de México así como en los invernaderos de Chapingo. De acuerdo con los síntomas observados, después de un periodo de 3 semanas de exposición, se determinó que: en cinco de las seis zonas urbanas y en la zona industrial existen concentraciones tóxicas de ozono. En una de las zonas urbanas se detectaron síntomas de toxicidad debidos al nitrato de peroxiacetilo; y en la zona industrial los síntomas en las plantas indicadoras correspondieron a niveles tóxicos de etileno, bióxido de azufre y ozono. En todos los casos, las plantas mostraron una reducción marcada en desarrollo, lo cual podría atribuirse al efecto del bióxido de nitrógeno. En los grupos expuestos dentro y fuera de los invernaderos, no se observó ninguno de los síntomas correspondientes a los compuestos antes mencionados.

Changes in sapwood of longleaf pine logs stored under water sprays for 4 months. R. C. DE GROOT (USDA Forest Serv., Gulfport, Miss.). Sapwood of longleaf pine logs stored under a continuous water spray during the summer in southern Mississippi was rapidly invaded by gram-negative, fermentative bacteria during the 1st month of storage. Absorptivity and radial air permeability of the sapwood increased only slightly during the 1st month of storage, markedly during the 2nd month of storage, and very little thereafter. Sharp increases in absorptivity and permeability were associated with destruction of cell walls of ray parenchyma cells. No significant changes in maximum crushing strength or in stress at proportional limit, as tested by compression parallel to the grain, occurred in the sapwood during 4 months of storage. A slight, but significant, linear decrease in pH of the sapwood did occur. Neither quantity of hot and cold water extractables nor yields or relative composition of the terpene component of the logs significantly changed during storage.

Effects of dimethylsulfoxide on the production and identification of Aspergillus flavus aflatoxin grown on soybean seed. D. P. DEMYERS (Univ. Md., College Park). *Aspergillus flavus* (ATCC 2221) produced aflatoxins on soybean seed in the presence of dimethylsulfoxide (DMSO) at concentrations of 0, 5, 6, 7, 8, 9, 10, and 25 mg/ml. Fifty g of soybean seed were soaked for 30 min in each DMSO concentration. After soaking, the seeds were autoclaved and inoculated with a spore suspension of *A. flavus*. After incubation for 7 days at 24 C, aflatoxins were extracted using 70% acetone and chloroform separation. Spectrophotometric and TLC analysis indicated that four different aflatoxins were produced at lower DMSO concentrations. Aflatoxin production was stimulated by DMSO concentrations of 5-10 mg/ml. Inhibition of total aflatoxin production occurred at 25 mg/ml DMSO and above. Aflatoxin B₂ and G₂ were inhibited more readily at 10-25 mg/ml DMSO, suggesting the need for additional research on how DMSO affects aflatoxin production. DMSO concentrations of 5 mg/ml and above also caused loss of pigmentation of conidia.

Immuno-specific grids for electron microscopy of plant viruses. K. S. DERRICK (La. State Univ., Baton Rouge). Plant viruses are specifically attached to specimen grids

coated with a film of dried antiserum. Antiserum to potato virus Y (PVY) and normal serum were dialyzed against and diluted 1:50 with distilled water. Small drops of diluted sera were placed on specimen grids coated with carbon-backed Parlodion films and allowed to air-dry for 1 hr. The grids were then floated on dilutions of an extract of PVY-infected tobacco leaves in neutral phosphate containing 0.15 M NaCl. Following a 10-min reaction time, the grids were washed by floating for 5 min on 0.15 M NaCl and 3 times on distilled water for 1 min. The grids were prepared for examination by electron microscopy by shadow casting. A 1:4 dilution of PVY-infected sap gave a mat of virus particles that completely covered grids prepared with immune serum. The number of virus particles per unit area appeared to decrease linearly with dilution, and were readily found at a dilution of 1:1024. Examination of similar dilutions of PVY-infected sap on normal-serum grids revealed a few virus particles at a dilution of 1:4, and no particles were found at dilutions greater than 1:128. Immuno-specific grids have also been prepared for tobacco mosaic virus, cucumber mosaic virus, and tobacco etch virus.

Nature and concentration of propagules of Verticillium dahliae in field soils relative to the prevalence of Verticillium wilt in cotton plants. J. E. DE VAY & LINDA L. FORRESTER (Univ. Calif., Davis). Propagules of *Verticillium dahliae*, which were persistent in soils air-dried at 30-50% relative humidity for at least 6 weeks, were directly observed and their frequency determined. Seventy-five soil samples were collected at a depth of 10 to 20 cm from specific areas in 13 cotton fields that varied from 0- to 100%-wilted plants just before harvest. Three 10-mg samples of dry soil from each collection were evenly distributed onto cellophane discs on Menzies soil-extract agar. After 16 hr at 23 C, sterile camels' hair was used as a marker for locating germinating propagules; positive identity of *V. dahliae* was made within 10 days based on the formation of microsclerotia. Propagules ranged from 11 to 225 μ in greatest dimension, and consisted of irregular masses of mainly hyaline cells (7-8 μ in diam). The majority of cells of the propagules were viable; up to 36 germ tubes were produced/propagule. Soils varied from 0 to 2,730 propagules/g soil. Pathogenicity of 40 randomly selected isolates of *V. dahliae* was tested by stem inoculations of at least three cotton plants, var. Acala SJ-1, with conidial suspensions. Thirty-six of the isolates were pathogenic, including one defoliating strain; four were nonpathogenic. Propagule frequency in soil was not a reliable index for determining prevalence of *Verticillium* wilt in cotton.

Observations on ectodesmata and the virus infection process. G. A. DE ZOETEN, G. GAARD, & W. S. W. MERKENS (Univ. Wis., Madison). Ectodesmata were visualized by means of electron microscopy in outer epidermal cell walls of *Nicotiana tabacum* 'T.I. 787' by means of a silver nitrate stain (3% AgNO₃ for 5 min preceding glutaraldehyde fixation). Reduced silver was found in epidermal outer walls associated with structures (ectodesmata) in which the cellulose micelles were pulled apart. This caused a localized decrease in wall density and a thickening of the cell wall resulting in a local elevation on the leaf surface. Phase contrast and differential interference contrast microscopy corroborated the data obtained by electron microscopy. After potato virus X inoculation, virus particles could not be observed in ectodesmata by direct observations in the electron microscope. However, virus particles were seen attached to, but not penetrating, the cuticle of inoculated tobacco leaves.

Red clover clones with hypersensitive resistance to a common variant of bean yellow mosaic virus. S. DIACHUN & L. HENSON (Univ. Ky., Lexington). Plants of red clover clone KyC40-1 inoculated with a common variant of bean yellow mosaic virus (204-1) form localized necrotic spots; systemic infection occurs rarely. The hypersensitive resistance is controlled by a single dominant factor. Inbreds of clone KyC40-1 have been developed and cloned. Several of the inbreds are homozygous for the resistance factor. They offer promise as breeding stocks for development of virus-resistant populations of red clover.

Estudio y caracterización de un mosaico del frijol de costa (Vigna sinensis) en El Salvador. ANTONIO J. DIAZ (Centro Nacional de Tecnología Agropecuaria, Sta. Tecla, El Salvador). El cultivo del frijol de costa es relativamente reciente en El Salvador. En 1971, se observó en el país un mosaico virótico atacando cultivos comerciales de frijol de costa. En los estudios realizados se logró transmisión mecánica y por insectos crisomelidos. *Diabrotica balteata*, *Ceratoma ruficornis* y *Systema* sp., transmitieron el virus. No se logró transmisión por semilla. Aparentemente este virus esta relacionado serológicamente al virus del mosaico rugoso ("rugose mosaic") y a razas del virus del "cowpea mosaic" de Arkansas y Nigeria. En pruebas de inactivación termal, el virus fué inactivado entre 65 y 70C. Se realizaron ademas inoculaciones mecánicas en 21 especies de leguminosas de diferentes géneros. Los estudios realizados hasta el momento parecen indicar que se trata del virus del mosaico del "cowpea."

Chrysanthemum stunt, a viroid disease. T. O. DIENER & R. H. LAWSON (ARS, USDA, Beltsville, Md.). RNA extracted and purified from stunt-diseased chrysanthemum leaves by a method developed for the purification of the potato spindle tuber viroid (PSTV) consists mainly of host cell RNA, but contains the infectious agent (CSV) as evidenced by bioassay on *Senecio cruentus* plants. Preparations were further purified by adsorption of the RNA onto hydroxyapatite and elution with phosphate buffer, followed by ethanol precipitation and resuspension in electrophoresis buffer. Analysis of infectivity distribution after isokinetic rate-zonal density-gradient centrifugation and polyacrylamide gel electrophoresis revealed that CSV is a low molecular-weight RNA. Coelectrophoresis of CSV and PSTV in 20% polyacrylamide gels showed that CSV moves significantly faster than PSTV, indicating that its molecular weight is somewhat less than that of PSTV (ca. 5×10^4 daltons). CSV, therefore, is a viroid similar to, but distinct from, PSTV.

Characterization of green fluorescent pseudomonads isolated from apparently healthy, dormant peach trees. W. M. DOWLER & D. J. WEAVER (USDA, Clemson, S.C., Byron, Ga.). Fluorescent, gram-negative bacteria were readily isolated from twig and trunk tissue samples obtained at monthly intervals during dormancy from 2- to 4-year-old apparently healthy peach trees in commercial orchards. The isolates were characterized by oxidase reaction, utilization of organic acids, aesculin hydrolysis, arbutin hydrolysis, and tyrosinase activity. The bacteria were found regularly in some orchards, rarely in others. The isolates were about equally divided into two groups, both of which appear to be closely related to *Pseudomonas syringae*. Group A was negative for oxidase, utilization of tartrate, and tyrosinase activity; positive for utilization of lactate and for aesculin and arbutin hydrolysis. Group B was similar, but was negative for lactate utilization and positive for tartrate utilization. A few isolates similar to *P. mors-prunorum* also were found. Representative

isolates from all groups produced a hypersensitive reaction in tobacco leaves and produced toxin on potato-dextrose agar supplemented with glucose and casamino acids, but few isolates were virulent in actively growing peach seedlings. These studies indicate that heterogeneous populations of pseudomonads may exist in dormant peach trees in the southeastern United States.

Diurnal heat predisposition of Pinto bean to virus transmission. T. DUAFALA (Univ. Calif., Berkeley). One primary leaf of each Pinto bean plant (*Phaseolus vulgaris*) was heated for 6 sec at 50 C in a water bath, and the opposite leaf served as a control. Immediately after heating and air drying, both leaves were inoculated by means of a brush with tobacco mosaic virus (TMV)-infected tissue (0.05 g of infected tissue ground in 1 ml of water and diluted to 100 ml) or tobacco necrosis virus (TNV) (0.5 g ground tissue in 100 ml), plus Celite. Heat treatments and inoculations were done every 3 hr over an 18- to 24-hr period. The predisposition index (PI) is given as the number of lesions on the heated leaf divided by the number of lesions on the control. Highest PI for both TMV and TNV (12 and 4.5, respectively) were obtained at 0900 and 1800-2100. The diurnal cycle was most pronounced with plants seeded for 9 days and decreased with older plants. Also, higher PI (12-fold) were observed with plants seeded for 9 days than with older plants. Plants appeared to be most responsive to heat predisposition just prior to the age of optimum susceptibility (10- to 12-day-old plants).

Effect of Difolatan and basic copper sulfate on Nectria galligena in relation to the control of European canker of apple. H. J. DUBIN & H. ENGLISH (Univ. Calif., Davis). Oil enhanced the toxicity of basic copper sulfate (BCS) to *Nectria galligena* conidia at copper concentrations between 10^0 and 10^2 $\mu\text{g/ml}$, but reduced toxicity at 10^3 $\mu\text{g/ml}$ as measured by germination inhibition. The effect was not observed with respect to inhibition of germ tube elongation. Difolatan [N-(1,1,2,2-tetrachloroethyl)sulfonyl]-*cis*-4-cyclohexene-1,2-dicarboximide] was fungitoxic in vitro at 1 $\mu\text{g/ml}$. In field spray tests, Difolatan 4 Flowable (3 qt/100 gal) reduced conidial and perithecial production, but BCS (5 lb. + 2 quarts Supreme oil/100 gal) decreased only perithecial production. Both fungicides were retained on the trees for the 12-week leaf fall period, but only Difolatan was redistributed in fungitoxic amounts. Three weeks after spraying, Difolatan had increased on twigs, whereas it had decreased on leaves. The less retentive leaves served as a temporary reservoir for Difolatan. BCS decreased linearly with time irrespective of the amount of rain. Bioassays showed that Difolatan totally inhibited conidial germination during the normal infection period, but inhibition on BCS-sprayed trees decreased rapidly with time.

Tumor development and reaction of cultivars of sweet potato plants to false broomrape. P. D. DUKES (ARS, USDA, U.S. Veg. Breeding Lab., Charleston, S.C.). Rooted sweet potato (SP) vine cuttings of Centennial, Jersey Orange, and Tintian cultivars were inoculated with an isolate of false broomrape (FBR) highly infectious to tobacco. Inoculated and control SP and tobacco plants were grown in soil at 38 C. After 4 months of growth, the underground parts were examined for tumor development. Centennial plants had the most infection and the largest tumors (up to 106 g). Tintian had small tumors with the largest weighing 2.3 g; and a few minute tumors developed on Jersey Orange. Tumors on SP were similar in gross morphology to those on tobacco. Tumor differentiation into shoots was not as advanced as on tobacco, but some rudimentary shoots were found on large

tumors. Internally, the tumorous tissues on Centennial were firm and dense, and had the orange flesh color of storage roots. In contrast, tobacco FBR tissues were succulent and white. Nearly all SP tumors developed on underground stems; whereas tobacco tumors developed on all underground parts. Centennial was the most susceptible SP cultivar to FBR, but not nearly so susceptible as tobacco. This disease does not appear to be a threat to SP production, although further studies on the causal agent and storage root infections are warranted.

The effect of tentoxin on transpiration. R. D. DURBIN & T. F. UCHYTIL (Univ. Wis., Madison). The transpiration rate ($\text{g dm}^{-2} \text{ hr}^{-1}$) of mung bean leaves was 2.2, 1.1, 0.9, 0.6, and 0.5 for seedlings treated with 0, 4, 20, 40, and 200 μM , respectively, of tentoxin, a cyclic tetrapeptide produced by *Alternaria tenuis*. Stomatal closure occurred within 1 hr when stems of bean, mung bean, and cucumber seedlings were immersed in 200 μM tentoxin. Conversely, stomatal aperture and transpiration rate of cabbage, turnip, and radish seedlings were little affected by tentoxin at this concentration. In CO_2 -free air in the dark, the opening of stomata of bean and broad bean plants treated with 200 μM tentoxin, but not that of control plants, was significantly inhibited. The chlorophyll content of mung bean leaves treated initially for 6 hr with 200 μM tentoxin declined at the same rate over a 5-day period (12 hr photoperiod) as leaves of control and tentoxin-treated plants held in the dark.

Effects of uranyl salts on the ultrastructure of victorin-treated leaves. CAROL Z. EASTON & PENELOPE HANCHEY (Colo. State Univ., Fort Collins). Previous studies have shown that uranyl salts suppress both electrolyte leakage and ultrastructural damage in oat leaves treated simultaneously with victorin, the *Helminthosporium victoriae* pathotoxin. Further ultrastructure investigations on victorin-treated leaves given suppressive pretreatments and noneffective posttreatments with uranyl have been conducted. The deposition pattern of uranium crystals was identical when uranyl-treated leaves were subsequently treated with either water or victorin. Crystals occurred only in cell walls and near the plasmalemma. Plasmalemma invaginations and cytoplasmic vesicles of unknown origin were frequently present, but no evidence of pinocytosis was found. Intracellular crystals were found in necrotic cells of victorin-treated leaves posttreated with uranyl. This treatment resulted in more severe ultrastructural damage than water posttreatment. These results suggest that victorin treatment facilitates uranyl entry, which then contributes to cellular damage. The mechanism of the suppressive effects of uranyl appears to be complex. Both partial inactivation of victorin and its binding sites within the tissue may be involved.

Bacteriophages for the detection of Corynebacterium michiganense in North Carolina. E. ECHANDI (N.C. State Univ., Raleigh). Bacteriophages specific to *Corynebacterium michiganense* were isolated in North Carolina from tomato stems infected with *C. michiganense*. Phage isolates differed in host range when tested on virulent *C. michiganense* isolates obtained from the trellised tomato area of western North Carolina and other parts of the country. Phage CMPI lysed all 22 *C. michiganense* isolates tested, whereas the other phage isolates lysed 19 or less. None of the phages lysed species in *Agrobacterium*, *Bacillus*, *Erwinia*, *Pseudomonas*, *Xanthomonas*, and *Corynebacterium* other than *C. michiganense*. Since CMPI has a wide host range and can be a useful tool in diagnostic and ecological studies of the bacterial pathogen, it was characterized in more detail.

Plaques of CMPI are about 0.5 to 1 mm in diam, clear, round, and entire. They are formed at 16, 20, and 24 C with an optimum of ca. 24 C. No plaques were formed at 28 C or above. CMPI is gradually inactivated over 45 C, and completely inactivated between 55 and 60 C for 10 min. Preliminary electron micrographs revealed that CMPI has a polyhedral head about 624 Å wide and a thin tail about 3,137 Å long.

Fungitoxicity of o-phenylphenol and derivatives. J. W. ECKERT, O. F. ESURUOSO, & JUNE CAREY (Univ. Calif., Riverside). Several isolates of *Penicillium digitatum* and *Diplodia natalensis* were inhibited 40-50% by 3 ppm *o*-phenylphenol (OPP) or 2-biphenyl acetate (BA) in potato-dextrose agar (PDA). *D. natalensis* isolates were inhibited more than 50% by 15 ppm *o*-phenylanisole (OPA), whereas *P. digitatum* isolates were not affected significantly by 250 ppm in PDA. Both fungi in liquid cultures hydrolyzed 80% of the BA to OPP in 8 hr, but did not metabolize the latter during the 24-hr incubation period. *D. natalensis* cleaved 10% of the OPA to OPP in 24 hr, whereas *P. digitatum* converted less than 1% of the ether to the phenol. OPP was identified in OPA cultures by chromatography, absorption spectra, and mixed melting point. *D. natalensis* did not take up significantly more OPA than *P. digitatum*, nor was OPP formed from OPA in sufficient quantities to explain the selective inhibition of *D. natalensis* by OPA.

Induced resistance to bean anthracnose in varieties susceptible to all four races of Colletotrichum lindemuthianum. J. E. ELLISTON & E. B. WILLIAMS (Purdue Univ., Lafayette, Ind.). Etiolated hypocotyls of Stringless Green Pod and Bountiful, cultivars of *Phaseolus vulgaris* susceptible to alpha, beta, gamma, and delta races of *Colletotrichum lindemuthianum*, were protected against anthracnose using *Colletotrichum* spp. nonpathogenic to bean. Organisms used included *C. gossypii*, *C. fragariae*, *C. lagenarium*, races 1, 2, and 3, *C. phomoides*, *C. trifolii*, and an isolate of *C. truncatum* nonpathogenic to bean. Suspensions of conidia of the nonpathogens containing $1-2 \times 10^6$ conidia/ml were applied to nonelongating regions of hypocotyls of 8- to 10-cm plants as 1.5- μ l drops. The drops were evaporated 0, 6, 12, 18, or 24 hr later, and the sites reinoculated with drops of a suspension of *C. lindemuthianum* conidia ($1-2 \times 10^6$ conidia/ml). Plants were observed 7-8 days later. The organisms varied in the rate and extent of protection induced; however, in all cases a lag period of 24 hr between inoculation with the nonpathogen and *C. lindemuthianum* resulted in reduction of symptoms. Many of the organisms induced complete protection, some with lag periods as short as 0 hr.

Evaluation of resistance of H2990, a new tomato cultivar, to bacterial canker by different inoculation methods. D. A. EMMATTY & C. A. JOHN (Heinz USA, Bowling Green, Ohio). Tomato seedlings of H2990 (resistant) and H1350 (susceptible) determinant cultivars were used to evaluate the resistance of H2990 to bacterial canker by comparison with different methods of inoculation with *Corynebacterium michiganense*. Root-dip (RD) at transplanting (cotyledonary stage), leaf-cut (LC) 2 weeks after transplanting using a knife dipped in inoculum, and a combination of the two methods (RD + LC) were used. The inoculum was either the bacterium suspended in water or the suspension thickened by a bleeding with solidified nutrient agar at a 2:1 ratio. Readings were taken 4, 6, and 8 weeks after inoculation, with the dead seedlings compared to survivors (healthy and partially wilted) as the criterion for infection. Eventually, the wilted plants either died or recovered. Irrespective of the method used, a

statistically significant greater number of H1350 plants was killed than H2990. The RD and the RD + LC methods, using bacterium suspended either in water or nutrient agar, gave comparable results with a statistically significant greater number of dead plants as compared to the noninoculated checks for the respective tomato lines. However, response with the LC technique was significantly different only with the susceptible line H1350.

Complete control of Fusarium wilt of chrysanthemum with chemotherapeutants combined with a high lime and nitrate-nitrogen culture regime. A. W. ENGELHARD & S. S. WOLTZ (Univ. Fla., Bradenton). Benomyl and BAS 3201 F 50W [methyl 1-(methylthioethylcarbamoyl)-2-benzimidazole carbamate] drenched on potted chrysanthemums grown on a high lime-all nitrate nitrogen cultural regime provided complete control of Fusarium wilt (*Fusarium oxysporum* f. sp. *chrysanthemi*) in the highly susceptible cultivar Yellow Delaware. The soil was methyl bromide-fumigated Leon fine sand amended with peat. Chemical rates as low as 0.125 lb. active ingredient/100 gal drenched twice at 200 ml/application to 6-inch pots were effective and were not phytotoxic. $\text{Ca}(\text{OH})_2$ at 2 g/kg of soil medium, and NaNO_3 as the only nitrogen source contributed additionally to the control of the disease. Prior to planting, the soil pH was 7.9; and at harvest, 8.6. Iron, manganese, and zinc were added to the foliage and/or soil as required to produce good-quality plants. Ratings of foliage symptoms and vascular discoloration, development of the flowers, weights of the plants, and laboratory tissue isolations confirmed the control of Fusarium wilt. This chemotherapeutant-soil amendment procedure appears to be the first effective method for the complete control of Fusarium wilt in chrysanthemum.

Dynamics of infection by Pseudomonas phaseolicola in partially resistant populations of bean. G. L. ERCOLANI (Univ. Bari, Italy). The mean probability (p) per inoculated bacterium of multiplying in leaves to cause visible infection did not vary with inoculum dose (d). A stochastic interpretation of p , as provided by the birth-death model, accounted for, several but not, all features of infection. Observed incubation periods (t) within each dose group were compatible with normal distributions of either $1/t$ or $\log t$, but not of t . The mean incubation period (RT50) and the reciprocal of the harmonic mean incubation period for each dose group regressed linearly on $\log d$, except at extreme dosages. The observed regressions of the standard deviation and the skewness of the distributions of t on d mimicked those expected in a birth-death process. Doubling times of bacteria in vivo and viable counts (n) in each visible infective center at RT50 were invariant to d from 0.05 to 500 ED^{50} 's. Increments of n in vivo were invariant to d up to 5 ED^{50} 's, then declined. Growth curves of challenging bacteria were parabolic, indicating that infection might be a birth-death process with linear time-dependence. A deterministic interpretation of p based on host site heterogeneity accounted for the observed standard deviation of $\log n$ in infections caused by 2 to 2,000 ED^{50} 's, but not in those initiated by 0.125 to 1 ED^{50} .

Gamma radiation detects defects in trees and logs. T. H. FILER, JR. (USDA Forest Serv., Stoneville, Miss.). Three isotopes with different energies of radiation were used with a portable scintillation counter to detect rots and defects in trees and logs. Defects greater than 2.5 cm^3 could be detected in trees or logs of 10 to 37.5 cm with gamma rays from 1 mc of Ba^{133} . Gamma radiation from 1 mc of SN^{113} detected rot in trees less than 25 cm in diam; and for diameters less than 15 cm, Co^{57} was effective. Theoretical

and observed attenuation curves discriminate between brown and white rot. Voids larger than 1.5 cm are detectable. The scintillation counter and allied equipment weigh ca. 18.2 kg, and can be manually transported to examine logs or mounted on a movable platform on the front bumper of a four-wheel-drive vehicle for examination of trees.

Chemical control of melanose on citrus in Florida. F. E. FISHER (Univ. Fla., Agr. Res. & Educ. Center, Lake Alfred). Melanose (*Diaporthe medusaea*), causes severe damage to leaves, twigs, and fruits of *Citrus* spp., particularly under conditions of prolonged high moisture. Until recently, copper sprays were the only recommended means of chemical control in Florida. Tests during the past 4 years on grapefruit (*C. paradisi*) and orange (*C. sinensis*) have shown that chorothalonil 1.5 lb./100 gal applied at petal-fall and repeated 6 weeks later was significantly more effective in controlling melanose on fruit than basic copper sulfate, 0.75 lb./100 gal. In 1971, MBR 6866 (3M Co.) 0.5 lb./100 gal was also more effective than basic copper sulfate. Benomyl, 0.5 lb./100 gal, was as effective as basic copper sulfate in reducing melanose on fruit, and resulted in greener foliage. With a single petal-fall application, chlorothalonil or basic copper sulfate gave comparable control of fruit melanose, but was less effective than the dual applications indicated above; benomyl was ineffective.

Lack of specificity of the Colletotrichum lindemuthianum endopolygalacturonase inhibitor. MINA FISHER, ANNE ANDERSON, & P. ALBERSHEIM (Univ. Colo., Boulder). A protein from Red Kidney bean hypocotyls, which was purified for its ability to inhibit an endopolygalacturonase (endo-PG) secreted by *Colletotrichum lindemuthianum*, has no demonstrable activity against the polygalacturonase secreted by *Sclerotium rolfsii*. However, this protein inhibits a mixture of polygalacturonases secreted by *Aspergillus niger* (Pectinol-R-10). One of the inhibited enzymes, an endo-PG, was purified by a combination of ion exchange chromatography and gel filtration chromatography. One unit of the purified *A. niger* endo-PG is inhibited 50% by 0.36 μ g of inhibitor, whereas one unit of the *C. lindemuthianum* endo-PG is inhibited 50% by 0.24 μ g of this inhibitor. Thus, this highly purified plant protein is clearly capable of efficiently inhibiting the endo-PG's secreted by both *C. lindemuthianum* and *A. niger*. This lack of specificity with regard to the source of the endo-PG demonstrates that a plant may have a few partially specific inhibitors of pathogen-secreted endo-PG's rather than a unique inhibitor for each endo-PG secreted by every pathogen.

Oospore germination of Pythium aphanidermatum as affected by casein, gallic acid, and pH levels in a selective agar medium. R. A. FLOWERS & R. H. LITRELL (Univ. Ga. Coastal Plain Sta., Tifton). Oospores of *Pythium aphanidermatum* were obtained from cultures grown at 30 C for 21 days in 40 ml of Schmitthenner's synthetic liquid medium and incubated on the test media at 30 C for 10 hr. Casein and gallic acid were separately incorporated at varying concentrations into a selective medium containing 7.5 g sucrose, 0.5 g $MgSO_4 \cdot 7H_2O$, 1 g KH_2PO_4 , 0.5 g yeast extract, 2 mg thiamine-HCl, 80,000 units penicillin G, 100,000 units nystatin, and 15 g agar/liter. The pH was adjusted to 4.0-7.0. Casein at 2 g/liter, gallic acid at 400 mg/liter, and pH 6.0 in the selective medium were optimal for oospore germination (\sim 95%) and colony growth. To obtain satisfactory germination of other species of *Pythium* and *Phytophthora*, it was necessary to lower the pH to various levels down to 5.0 depending upon the species involved. Using this technique, it is possible to obtain large

numbers of germinating oospores for genetic studies within 3 weeks after inoculating the synthetic liquid medium for oospore production. In addition, the propagule density of certain species of *Pythium* and *Phytophthora* in soil may be ascertained in 1 day.

Effect of age, temperature, and inoculum concentration on bacterial canker development in resistant and susceptible Lycopersicon spp. R. L. FORSTER & E. ECHANDI (N.C. State Univ., Raleigh). Plants of bacterial canker-resistant *Lycopersicon esculentum* (P.I. 340905), *L. hirsutum* (P.I. 251305), *L. esculentum* \times *L. pimpinellifolium* (MR 4 and Bulgaria 12), and susceptible *L. esculentum* (manapal) and *L. peruvianum* (P.I. 251306) were grown from seed in a Phytotron at 26/18 C day/night temperatures superimposed on a photoperiod of 9 hr of high intensity light (4,000 ft-c) plus 1 hr of incandescent light. Three days before inoculation, plants were transferred to the different temperature regimes. We inoculated plants by stabbing the stem above the cotyledonary leaves with a dental root canal file dipped in 10^7 cells/ml of *Corynebacterium michiganense*. Differences in disease indices between resistant and susceptible cultivars were greater in 5-week-old plants (16-24 cm tall) than in 4-, 6-, and 7-week-old plants. The age \times cultivar interaction was significant. Five-week-old plants at 24/18 C showed greater differences in disease indices than those at 20/18, 28/18, or 32/18 C, and the temperature \times cultivar interaction was significant. Inoculum that contained 10^9 cells/ml induced larger differences in disease indices in 5-week-old plants at 24/18 C than inoculum with 10^5 and 10^7 cells/ml, but inoculum with 10^7 cells/ml induced differences almost as great as 10^9 cells/ml.

Sorghum rust, a naturally stabilized disease in North America. R. A. FREDERIKSEN & D. T. ROSENOW (Tex. A&M Univ., College Station, Lubbock, Tex.). Sorghum rust caused by *Puccinia purpurea* develops in *Sorghum bicolor* in certain areas of North America each year. In our tests, all commercially grown grain sorghum hybrids are susceptible but rust very slowly; and in most years, losses are slight. Sorghum entries selected as candidate differentials grown in Texas, Louisiana, and Mexico, naturally and artificially infected, reacted similarly to naturally occurring rust at these locations. Consequently, we have no evidence for physiological races of *P. purpurea*. Host reactions may be divided into three categories: (i) freedom from visible infection; (ii) restricted pustule size; and (iii) reduced number of pustule per unit area. The lack of specific resistance conditioning genes in either the perennial host reservoir, *S. halepense*, or grain sorghum hybrids probably accounts for the absence of detectable races of *P. purpurea*, whereas observed levels of generalized resistance in hybrids may be correlated with environmental factors.

Evaluación de la resistencia de clones de papa a Pseudomonas solanacearum. E. R. FRENCH (N.C. State Univ., Raleigh). Los híbridos de papa Wisconsin V-7, 7-6, y BR-6014 derivados de *Solanum phureja* y *S. phureja* 1386.26, que se comportaron resistentes a la marchitez bacteriana causada por *Pseudomonas solanacearum* en campos infestados del trópico americano, fueron sometidos a pruebas de resistencia 30 días después de sembrados. Las plantas fueron inoculadas con la cepa PI de *P. solanacearum* raza 3, punzando el tallo en la antepenúltima axila a través de una gota de suspensión bacteriana que contenía 2×10^{10} células/ml o infestando el suelo de cada maceta (1500 ml) con 80 ml de una suspensión que contenía 5×10^9 células/ml. Se inocularon 6 plantas por tratamiento y se mantuvieron en dos regímenes de temperatura;

diurna/nocturna de 28/16 C (templado) y 20/8 C (frío). Se evaluaron las plantas 22 días después, siendo todas susceptibles cuando inoculadas al tallo. En suelo infestado BR-6014 fue susceptible en el régimen templado, pero tolerante en el frío, mientras que los cultivares restantes fueron tolerantes en el templado y resistentes en el frío. Los niveles de resistencia fueron expresados en suelo infestado, pero no por inoculación al tallo. La resistencia fue mayor en el régimen de menor temperatura.

Experimental transmission of Cristulariella pyramidalis from silver maple to pecan and other hosts. W. J. FRENCH (Univ. Fla., Monticello). In 1970, zonate leaf spot (*Cristulariella pyramidalis*) was observed on silver maple (*Acer saccharinum*) and London planetree (*Platanus acerifolia*) in Florida. This was the first appearance on record of the disease in Florida. The appearance of the disease, which also occurs on pecan in Georgia and Alabama, caused concern for the pecan nursery industry centered in northern Florida. Plants of five genera were inoculated with conidiophore suspensions, diseased leaf pieces, and mycelium from isolates of *C. pyramidalis* from silver maple. The inoculated leaves were enclosed in plastic bags and incubated at 21-24 C for 48 hr. Brown zonate leaf spots appeared within 48 hr, followed in 2-3 days by the production of conidiophores on the infected tissue. The amount of leaf damage and conidiophore production varied with host species. The experiment demonstrated that isolates of the fungus from silver maple were pathogenic to pecan seedlings (*Carya illinoensis*), Stuart pecan, London planetree, kenaf (*Hibiscus cannabinus*), okra (*H. esculentus*), cotton (*Gossypium hirsutum*), and tomato (*Lycopersicon esculentum*).

Formamide hydro-lyase, an adaptive enzyme from Stemphylium loti. W. E. FRY & R. L. MILLAR (Cornell Univ., Ithaca, N.Y.). Leaves of cyanogenic birdsfoot trefoil (*Lotus corniculatus*) release hydrogen cyanide (HCN) when infected by *Stemphylium loti*. *S. loti* spores incubated in low levels of HCN (ca. 1.0 mM) develop a tolerance to HCN, and have been designated adapted spores. Adapted spores but not nonadapted spores convert HCN to formamide (HCONH₂). Experiments involving H¹⁴CN, column chromatography, colorimetric analyses, and infrared spectrophotometry demonstrated that cell-free preparations from adapted spores also convert HCN to formamide (HCONH₂), whereas cell-free preparations from nonadapted spores do not. We believe that the conversion of HCN to HCONH₂ is effected by formamide hydro-lyase. This enzyme(s) obtained from adapted spores was retained upon dialysis, was inactivated when incubated 5 min in boiling water, remained in the supernatant fraction after centrifugation at 100,000 g for 90 min, and was excluded from Sephadex G-200. The enzyme(s) apparently does not require cofactors, is maximally active at pH 7-9, and loses 5-10% of its activity per week of storage at 4 C.

Effects of sugars, tryptone, PPLO broth, yeast extract, and horse serum on growth of the mycoplasma-like organism associated with stubborn of citrus. A. E. A. FUDL-ALLAH & E. C. CALAVAN (Univ. Calif., Riverside). The mycoplasma-like organism cultured frequently from stubborn-diseased citrus, but not from healthy plants, grew well in media containing 0.1% glucose, 0.1% fructose, 0.1% sucrose, 2% PPLO broth, 10% of 25% yeast extract, 20% horse serum, distilled water and, for agar media, 1% Bacto-agar. Concentrations of certain ingredients in the media were varied individually to determine effects on growth. No growth occurred without horse serum and PPLO broth. Growth was optimal with 2%, less with 4%, and very

slight with 6% PPLO broth. Colonies were small on media with 1.5% tryptone, but were normal at 0.0, 0.5, and 1% tryptone. No growth occurred on media with 1.5% fructose. Only a few small colonies grew on media with 1.5% glucose. Small, dense colonies without central spots developed at 1%, and optimal growth occurred at 0.0-0.5% glucose or fructose. Sucrose concentrations of 2-4%, 8% in the absence of other added sugars, were optimal; 30% sucrose prevented growth. Growth was progressively stimulated by the addition of yeast extract up to 8-10%.

Pudrición bacteriana del capítulo del girasol (Helianthus annuus). L. FUCIKOVSKY (Colegio de Postgraduados, Chapingo, México). En 1970 fue observada por primera vez en Chapingo, una pudrición del capítulo gel girasol. Los daños estimados, en porcentaje de plantas atacadas, varió de 2 a 51%, dependiendo de la variedad y la fertilización. La mosca, *Neotephritis finalis*, fue identificada como el vector de la enfermedad. Este insecto deposita los huevos en las brácteas, y las larvas al penetrar introducen la bacteria a los tejidos del capítulo, el cual bajo condiciones favorables se pudre en 4 a 6 días. De los excrementos de la mosca se aisló una bacteria y se comprobó su patogenecidad mediante pruebas de laboratorio y de campo. *Tithonia tubaeformis* es también hospedante de *N. finalis*, sin embargo, la pudrición bacteriana no se desarrolla en esta planta. Las semillas de ambas plantas pueden ser portadoras de la bacteria externamente. El patógeno se identificó como una especie de *Pseudomonas* del grupo fluorescente, cuyas características morfológicas y fisiológicas difieren de las del agente causal de la enfermedad designada como bacteriosis foliar en el mismo hospedante, en el Estado de México.

Control of a Fusarium storage rot of Hubbard squash with 2-(4-Thiazolyl)-benzimidazole. R. L. GABRIELSON, R. C. MAXWELL, & O. O. KIENHOLZ (Wash. State Univ., Puyallup, Pullman). *Fusarium roseum* 'Sambucinum' causes serious storage losses of Hubbard squash on Whidbey Island, Wash. Neither curing nor controlled temperature storage controls this pathogen. Eighty-eight fungicides were tested in vitro; forty-seven prevented spore germination at 10 µg/ml or less. Of 39 of these subsequently tested on wound-inoculated squash, only Thiabendazole, [2-(4-Thiazolyl)-benzimidazole], benomyl, and 2-(2-furyl)-benzimidazole provided enough control to warrant further testing. In further comparison on wound-inoculated squash, Thiabendazole gave the most consistent and effective control. The sequence of wounding, inoculation of spores, and Thiabendazole treatment at 1,000 µg/ml were interacted, and all combinations provided effective control except fungicide-wounding spores. Residue analysis of peeled squash treated at 1,078 µg/ml (0.9 lb./100 gal) showed a maximum residue of 0.2 µg/g. This, plus the lack of inhibition of spore germination by Thiabendazole at 1 µg/ml, indicates that the control is surface-protective rather than systemic. Over a 3-year period, Thiabendazole has consistently and significantly reduced *Fusarium* storage rots and increased the numbers of healthy squash in properly maintained commercial storages.

Aplicación de fungicidas al suelo y al follaje para el control de la piricularia (Pyricularia oryzae) en arroz. G. E. GALVEZ-E. y JAIRO CASTAÑO-Z. (Centro Int. de Agr. Tropical, Cali, Colombia). Se hicieron tratamientos al suelo con 34 fungicidas en dosis de 2 a 240 kg i. a./ha usando las variedades susceptibles IR-532, Canilla, y Fanny. Los productos más promisorios fueron benomyl (Metil 1-(butilcarbamoil)-2- benzimidazolecarbomato), NF-44 (1,2 bis (3, metoxicarbomil-2-tioureido) benzeno, y

Kitazingranulado (0,0-diisopropil-5-benzil tiofosfato). Benomyl y NF-44 en polvo mojable protegieron el arroz con aplicaciones al suelo de 20 Kg i.a./ha, observándose un efecto residual hasta después de 5 meses de su aplicación. Kitazin granulada se comportó bien a 7.65 Kg i.a./ha bajo condiciones de riego pero no fué efectivo en condiciones de secano. Con benomyl (115 gr. i.a./ha), Kasumin (kasugamycin) (30 ml i.a./ha), Blastin (pentaclorobencilol) (700 ml i.a./ha), Hinosan (0-etil-S-S-difenil-ditiofosfato) (350 ml i.a./ha) y Kitazin (650 ml i.a./ha), aplicados a la variedad Bluebonnet 50 en tres épocas, una antes de la emersión de la panícula seguida de otras dos a intervalos de 8 días se obtuvo hasta 60% de control. El incremento promedio en producción fué de 1500 Kg/ha. En mezcla benomyl (100 gr i.a./ha) y Kasumin (20 ml i.a./ha) aumentaron la efectividad y redujeron los costos de aplicación. Este control fué efectivo en regiones propicias al desarrollo del hongo pero con un régimen de lluvias intermitentes. Los mismos tratamientos bajo condiciones de lluvias fuertes y continuas no fueron efectivos.

Some properties and beetle transmission of bean yellow stipple virus. R. GAMEZ (Univ. de Costa Rica, San José). A mild disease of beans (*Phaseolus vulgaris*) in Central American, characterized by a distinctive yellow stippling and slight malformation of the infected leaves, is caused by bean yellow stipple virus (BYSV). BYSV infected 14 or 24 mechanically inoculated species of Leguminosae. All of 542 cultivars of *P. vulgaris* tested were shown to be susceptible. No species outside Leguminosae were infected. Properties in unbuffered sap were: thermal inactivation point between 74 and 76 C; longevity in vitro at 20 C between 24 and 48 hr; dilution end point between 10^{-2} and 10^{-3} . Virus was purified from infected beans by chloroform-butanol clarification and differential centrifugation in 0.01 M phosphate buffer, pH 6.9. BYSV separated in a single component after centrifugation in sucrose gradients. Particles were isometric and ca. 30 nm in diam. The virus was not related serologically to members of the cowpea mosaic virus group. BYSV was not transmitted through seeds of infected plants, but was transmitted by the chrysomelid beetles *Ceratoma ruficornis* and *Diabrotica balteata*. Differences in the efficiency of transmission of the virus by these species were observed.

Deoxyribonucleic acid homologies between Erwinia species and between other members of the Enterobacteriaceae as determined by molecular hybridization and renaturation techniques. J. M. GARDNER & C. I. KADO (Univ. Calif., Davis). DNA-DNA hybridizations among typical *Erwinia* species were below 50% in all cases under moderately stringent conditions. In many instances, relatedness values (degree of homology) between *Erwinia* spp. and other *Enterobacteriaceae* (*Escherichia*, *Salmonella*, and *Klebsiella*) were as high or higher than values obtained within the genus *Erwinia*. The hybridization data were supported by optical DNA renaturation experiments. Furthermore, the artificial grouping within *Erwinia* (*Carotovora*, *Amylovora*, and *Herbicola* groupings) is not supported by the hybridization data. We therefore propose that *Erwinia* is a heterogeneous and somewhat artificial genus whose members should be distributed within the general family of enterobacteria. The data are consistent with published evidence showing efficient genetic transfer between *Erwinia* and other enterobacteria.

Induction of the hypersensitive reaction in tobacco with specific high-molecular weight substances derived from the osmotic shock fluid of Erwinia rubrifaciens. J. M. GARDNER & C. I. KADO (Univ. Calif., Davis). When

Erwinia rubrifaciens cells are osmotically shocked, substances are released that produce a typical hypersensitive reaction (HR) when sterile solutions are infiltrated into leaves of tobacco (*Nicotiana tabacum* 'Havana'). The reaction is induced within 6-12 hr which mimics the HR induced by the bacterial cells. Activity is destroyed by heat (80 C, 10 min), trypsin (30 μ g/ml), and pronase (30 μ g/ml), but not by ribonuclease, deoxyribonuclease, lipase, neomycin, or streptomycin, indicating that the substances are proteins. The active substances are of high molecular weight since they are excluded from Sephadex G-100. Similar "shock protein" preparations from the nonpathogen *E. herbicola* failed to induce any HR. The osmotic shock procedure has been previously shown to selectively release proteins from surface layers of enterobacteria. Increased electrolyte losses within 1 hr from tobacco leaf tissue treated with *E. rubrifaciens* "shock protein" (< 1 mg/ml) are consistent with published results with HR induced by bacterial cells, suggesting that loss of membrane integrity is a primary consideration in HR.

Ozone injury to tobacco and alfalfa in South Dakota. W. S. GARDNER & JOANN SAFFORD (S. D. State Univ., Brookings). Ozone injury symptoms appeared on leaves of *Nicotiana tabacum* 'Turkish' and 'Wisconsin 38' in 1970-1971; and on 'Bel-W3', 'Bel-C', and 'Bel-B' in 1971 at Brookings, S. D. Bel-B showed mild injury on lower leaves, and Bel-C showed moderate injury on midplant leaves at Sioux Falls and Beresford in 1971. Statewide field plots of Bel-W3 produced injury that was greater at Sioux Falls > Beresford > Brookings > Milbank > Pierre > Rapid City > Highmore > Tuthill, S. D. Air pollution episodes were monitored by continuous production and observation of Bel-W3 in a Brookings greenhouse. Injury periodically appeared on newly matured leaves as dark-gray flecks that whitened with age. During a period of heavy fleck on Bel-W3, ozone injury was found on alfalfa (*Medicago sativa*) at Brookings. Bioindicators for peroxyacetyl nitrate (*Petunia hybrida* 'White Cascade' and *N. glutinosa*); fluoride (*Gладиолус* sp. 'Snow Princess'); and sulfur dioxide (*Gossypium hirsutum*) showed no injury in South Dakota. On 11 Feb. 1972, the day after an air pollution alert was sounded for the Minneapolis-St. Paul, Minn. area, ozone fleck appeared on Bel-W3 in the greenhouse at Brookings.

Organic matter, major elements, and peanut pod breakdown. K. H. GARREN & D. M. PORTER (USDA, ARS, Va. Polytech. Inst., Va. State Univ., Holland, Va.). A 2-year study was conducted to determine the role of cellulose from cow manure, peanut hay, and rye straw in relationship to N and K in the enhancement of peanut (*Arachis hypogaea*) pod breakdown caused by *Pythium myriotylum* and *Rhizoctonia solani*. Cow manure was applied in 1970 at ca. 13.4 or 26.8 MT/hectare and 10-10-10 fertilizer or KCl were used to supply N and K or K alone at rates equivalent to that of the manure. In 1970, pod breakdown was significantly greater in plots receiving the high rate of manure or both rates of fertilizer than in untreated plots. The effect of the low rate of cow manure and both rates of KCl on pod breakdown was not significantly different from untreated plots. *R. solani* was the predominant fungus isolated from rotted fruits. In 1971, rye straw and peanut hay at ca. 11.2 or 22.4 MT/hectare were included with previous treatments in a field infested with *P. myriotylum*. Pod breakdown was significantly greater in plots receiving both rates of cow manure and peanut hay and the high fertilizer rate than in untreated plots. Pod breakdown in plots receiving both rates of rye straw and KCl and the low fertilizer treatment was not greater than in untreated plots. These data indicate that pod breakdown may be increased by high levels of N with or without the cellulosic components of organic matter.

Stabilization and separation of sugarcane mosaic virus and maize dwarf mosaic virus in mixed infections of sugarcane and other grasses. A. G. GILLASPIE, JR., & H. KOIKE (ARS, USDA, Houma, La.). Mixed infections with one or more strains of sugarcane mosaic virus (SCMV-A, -B, -D, -H, or -I) and maize dwarf mosaic virus strain-A (MDMV-A) were established on itchgrass, and the mixtures were inoculated to Rio sorghum and to Johnson grass. After repeated passages through Rio, the symptoms became more like those of the SCMV strains, and after repeated passages through Johnson grass, the symptoms produced on Rio became more like those of MDMV-A. These mixtures became stabilized after three to four passages through Rio or Johnson grass so that single passages through Johnson grass or Rio, respectively, and through itchgrass or Chunnee sugarcane did not change the symptom expression on Rio. The components were apparently separated by passage through certain sugarcane cultivars. The Rio-to-Rio mixtures inoculated to Louisiana Striped sugarcane yielded the SCMV component, and the Johnson grass-to-Johnson grass mixtures inoculated to L 60-25 sugarcane occasionally yielded the MDMV-A component. The results indicate that in these mixed infections maintained on one host, a stable balance of components is obtained, but that the balance differs in different hosts.

Seed transmission of banana viruses. A. H. GOLD (Univ. Calif., Berkeley). Virus infection of commercial banana cultivars, plantains, *Musa balbisiana*, and *M. acuminata* seems to be widespread. In most cases, infection is symptomless. A seedling lot of *M. balbisiana*, germinated under Berkeley greenhouse conditions, showed many plants with vivid, viruslike symptoms. Transmission assay to *Chenopodium quinoa* demonstrated that the seedlings indeed were virus-infected. Assays of other seedling lots of *M. balbisiana*, *M. acuminata*, and hybrids demonstrate that virus transmission through true seed may occur in high percentage, even when symptoms are weak or absent. The seed transmission of banana viruses is an important factor which should be taken into consideration in banana breeding.

Phytophthora-alfalfa mosaic disease synergism in alfalfa. A. H. GOLD & G. ASHCRAFT (Univ. Calif., Berkeley). Combined infection of alfalfa by alfalfa mosaic virus (AMV) and *Phytophthora megasperma* under the low light conditions of Berkeley winter weather causes top dwarfing, greatly exceeding the effect of either pathogen alone. However, in sunny weather the dwarfing effect of *P. megasperma* alone is so great that the additional impact of AMV is not so apparent. Main root growth of alfalfa practically ceases within 2 days of infection by *P. megasperma*. Although healthy secondary roots grow from infected primary roots within 1 week, the dwarfing effect persists. Wilting behavior of *P. megasperma*-infected plants in sunny weather suggests that water stress is an important factor in dwarfing. Combined AMV and *P. megasperma* infection of alfalfa is so widespread in California that the interaction may be an important factor in winter survival of the crop.

Deoxyribonucleic acid polymerase from Rhizopus stolonifer. C. GONG, L. D. DUNKLE, & J. L. VAN ETTEN (Univ. Nebr., Lincoln). DNA polymerase activity was detected in cell-free extracts of ungerminated and germinated spores of *Rhizopus stolonifer*. The enzyme from germinated spores was prepared by centrifuging the 105,000 g supernatant fraction through a glycerol density gradient (GDG). Enzyme activity was dependent upon Mg^{++} and the simultaneous presence of all four deoxynucleoside triphosphates, and was stimulated by K^+ . Activity was

independent of added template DNA. However, after DNase treatment of the enzyme fraction and recentrifugation on GDG, the enzyme required a template DNA for activity and exhibited a preference for heat-denatured or activated DNA. Other nucleic acids such as single- and double-stranded RNA's were inactive as templates. The reaction was inhibited by actinomycin D, pyrophosphate, and DNase, whereas RNase and orthophosphate had no effect. The reaction product was degraded by DNase but resistant to RNase.

Field resistance to bacterial wilt in hybrid potato progenies. L. C. GONZALEZ, L. SEQUEIRA, P. R. ROWE, & R. BIANCHINI (Univ. Costa Rica, San José, Univ. Wisc., Madison). Resistance to *Pseudomonas solanacearum* derived from *Solanum phureja* was effective under tropical field conditions. Progenies from crosses of *S. phureja* × *S. tuberosum* were grown in naturally infested soils in Costa Rica, and clones were selected on the basis of tuber type and resistance to bacterial wilt. Progenies were reduced from 3,270 to 22 clones after five successive plantings. Screening was most effective when initial selection was done in infested highland soils (Cot, temperature range 11-21 C), and survivors were then planted at lower elevations in uninfested soil (Alajuela, 17-30 C). Commercial varieties of *S. tuberosum* exhibited 5 to 6 times more wilt than clones selected in the last plantings. Resistance of nine of the surviving clones was later evaluated at the University of Wisconsin, Madison, by stem inoculation in a growth chamber under conditions highly favorable for disease development; all clones exhibited from moderate to high resistance to a virulent isolate (S-206); and three showed moderate resistance to a highly virulent isolate (S-213). Field-screened material is now being used for further breeding with commercial varieties.

Enhancement by potato virus Y of potato virus X synthesis in tobacco leaves depends on the timing of systemic invasion by the viruses. R. M. GOODMAN & A. F. ROSS (Cornell Univ., Ithaca, N.Y.). The time between inoculation of a lower leaf of *Nicotiana tabacum* 'Samsun NN' with potato virus X or Y (PVX, PVY) and entry of the virus into the fourth leaf above was determined. Fourth leaves, selected because they can support enhanced PVX synthesis when doubly infected, were removed from different lots of plants at 6- or 12-hr intervals after inoculation of lower leaves, held in moist chambers for 6 days, then indexed for virus. Under controlled conditions, the time required by each virus to reach the fourth leaf was reproducible and was the same in singly and doubly inoculated plants. These data provided a guide for inoculation of lower leaves with the two viruses at intervals predicted to result in entry of the viruses into fourth leaves at desired intervals. These leaves, and comparable leaves of PVX-inoculated control plants, were later assayed for PVX by ultraviolet scanning after rate-zonal centrifugation. Enhancement of PVX synthesis was small when PVY preceded PVX by 36 hr or more, maximal when PVY preceded PVX by 12 hr, and nearly nil when PVX preceded PVY by 60 hr. We believe our data support the hypothesis that enhancement of PVX synthesis by PVY occurs only in cells which when invaded by PVX are still supporting the rapid PVY synthesis normal to recent infections.

Purification of maize dwarf mosaic virus by continuous-flow centrifugation. D. T. GORDON & R. E. GINGERY (Ohio Agr. Res. Development Center, & ARS, USDA, Wooster). The application of continuous-flow centrifugation was extended to the purification of a slowly sedimenting plant virus, maize dwarf mosaic virus (MDMV). By this technique, MDMV strains A, D, and E and 13B virus

were concentrated from large volumes of clarified extract and separated from most of the ultraviolet-absorbing contaminants. Further purification was achieved by isopycnic-zonal centrifugation in CaCl gradients. The efficiency of the continuous-flow rotor was greatest at high flow rates at which only a fraction of the virus sedimented into the gradient during a single passage of extract. Multiple passages through the rotor allowed nearly complete removal of virus from the extract. Yields of 15-30 μg virus/g of tissue were obtained with MDMV-A, and 5-20 μg /g were obtained for MDMV-D, -E, and 13B. No detectable impurities were present in final virus preparations as judged by electron microscopy, sedimentation properties, and serology.

Prolonged control of Fusarium nivale on bentgrass turf by single applications of benomyl. C. J. GOULD, R. L. GOSS, & V. L. MILLER (Wash. State Univ., Puyallup). Applications of benomyl every 2 weeks at 1 oz active/1,000 ft² controls Fusarium Patch on putting turf in Washington State. Although such frequent applications during "Fusarium weather" have been standard procedure, they are laborious, and schedules cannot always be followed because of inclement weather, conflicts with tournaments, etc. Occasional heavy applications provide another approach. Benomyl was applied once on 23 Sept. 1971 at 1, 2, 4, and 8 oz active or at 1 oz every 3 weeks in 10 gal of water/1,000 ft² on Highland bentgrass turf (*Agrostis tenuis*). A severe epiphytic occurred in October of 1971. On 15 Oct., only 0.006 diseased spots/ft² were present in the five benomyl-treated areas as compared to 4.06 diseased spots in the untreated areas. A severe attack occurred again in February, following repeated snowstorms. An average of 2.03 spots was present on 11 Feb. 1972 in the untreated area as compared with 1.16, 0.40, 0.30, and 0.08 for single applications of 1, 2, 4, and 8 oz of benomyl, respectively; and 0.11 spots for the 1 oz repeated treatment (total of 5 oz to 9 Feb). Color and density of all benomyl-treated plots were superior to the untreated. Thus, one application of 8 oz and perhaps 6 oz benomyl/1,000 ft² prevents severe attacks by *Fusarium nivale* for at least 141 days.

An unidentified isometric virus from Plantago. A. L. GRANETT (N.Y. State Agr. Exp. Sta., Geneva). A virus isolated from a *Plantago major* plant with chlorotic mottle was very restricted in host range. It was mechanically transmitted only to *Pisum sativum* 'Ranger', *Plantago*, *Tetragonia expansa*, *Antirrhinum majus*, and *Proboscidea jussieui*, although 37 other species were inoculated. In Ranger pea, terminal epinasty and systemic mottle appeared 10 days after inoculation, with gradual recovery of new growth. In *A. majus*, symptoms appeared in 12 days as expanding necrotic ringspots without systemic invasion. On *Proboscidea*, the virus incited tiny primary necrotic lesions and systemic mottle. Detached *Proboscidea* leaves were useful for local lesion assays. The virus was increased in Ranger pea and purified by butanol clarification and differential centrifugation. Electron microscopy of negatively stained leaf dips and partially purified infectious preparations revealed isometric particles, both "filled" and "empty", about 30 nm diam. Spectrophotometric analysis yielded a typical isometric virus curve with a 260:280 ratio of 1.9.

Effectiveness of benomyl as a protectant and therapeutant for oak wilt disease in red oak seedlings. G. F. GREGORY, P. MCWAIN, & T. W. JONES (USDA Northeastern Forest Exp. Sta., Delaware, Ohio). Solubilized benomyl in 0.05 N HCl was fed into the xylem of greenhouse-grown red oak seedlings. One set of seedlings received 4 ml of a 1 g/liter and another set, 4 ml of a 10 g/liter solution. One day later, the plants were inoculated

with 2 ml of a *Ceratocystis fagacearum* spore suspension containing 2×10^6 conidia/ml. Two other sets of untreated plants were similarly inoculated; 12 days later, leaf wilt symptoms were observed on some plants, then one set was given 4 ml of a 1-g/liter solution and the other, 4 ml of a 10-g/liter solution. No plants treated with solutions before inoculation developed symptoms in 2 months after inoculation. Of plants treated after inoculation, 28.5% remained symptomless, 43% were dead above the inoculation site 2 months after inoculation, and 28.5% developed symptoms but then resumed growth above the inoculation site. Half of the plants that resumed growth above the inoculation site did so from the terminal buds of symptomatic branches. No difference in effectiveness of protection or therapy was observed between the two concentrations of fungitoxicant. Protection from natural oak wilt infection would appear attainable and therapy a distinct possibility.

Infection of alfalfa by Ditylenchus dipsaci as affected by thermal acclimatization of the nematode. G. D. GRIFFIN (USDA, ARS, Utah State Univ., Logan). Six-month-old Ranger alfalfa plants were each inoculated with a suspension of *Ditylenchus dipsaci*, extracted from infected Ranger alfalfa plants. After 60 days in growth chambers at temperatures of 15, 20, and 25 C and a greenhouse moisture chamber (80-100% RH) at 22 ± 2 C, nematodes were extracted and placed on germinated alfalfa seed. Seeds inoculated with 50 nematodes each from each of the plant growth temperatures were grown for 14 days at 15, 20, 25, and 30 C, and infection was determined. Nematodes showed an affinity, in relation to infection, to the temperature they had previously been subjected to or at which they reproduced. Numbers of nematodes infecting plants at 15, 20, 25 and 30 C, respectively, were: 31, 25, 23, and 8 from plants previously grown at 15 C; 21, 33, 20, and 9 from plants previously grown at 20 C; and 17, 21, 26, and 12 from plants previously grown at 25 C. However, plants inoculated with nematodes from a greenhouse moisture chamber temperature of 22 ± 2 C were infected with 18, 29, 24, and 16 nematodes/infected plant at 15, 20, 25, and 30 C, respectively.

Xanthomonas phaseoli nonsystemic in some Phaseolus vulgaris cultivars. J. H. HAAS (Can. Dep. Agr., Harrow, Ont.). *Xanthomonas phaseoli* was not detected in stems of *P. vulgaris* 'Sanilac', 'Seaway', or 'Seafarer'. Seeds from 10 infected crops were germinated in soil in the greenhouse. The soil was watered carefully, and no free water was permitted on leaves. No leaf infection was found during 30 days. Stems were taken from 100 of these plants, dipped in 0.5% NaClO, and blended for 30 sec in saline. Four aliquots of each suspension were spread on nutrient broth agar and incubated. *X. phaseoli* was not present. The pathogen also was not recovered from 800 stems of field-grown 1- to 5-week-old seedlings. Surface-sterilized cotyledons from 20% of these seedlings contained the pathogen. It also was present on the surfaces of cotyledons from 5- to 10-day-old seedlings grown in a moist chamber. Symptoms were not observed on primary leaves where they would be expected if the bacteria moved through the stele. Cotyledons unfolded about 5 days and dropped 15 days after planting. Symptoms on trifoliate leaves occurred about 25 days after planting. The 10-15 days between the availability of inoculum on cotyledons and the appearance of leaf symptoms corresponds with the incubation period of the disease. The location and time of the first leaf symptoms corroborates the hypothesis that the pathogen is not systemic.

Phytoalexin production induced by heavy metals: a

proposed mode of action. L. A. HADWIGER (Wash. State Univ., Pullman). The mode by which CuCl_2 induces phytoalexin production in various plant species is not known. The action of copper and other heavy metals is usually assumed to result from selective cellular destruction or protein alterations. Recent information on metal-DNA complexes indicates that Cu^{2+} , Cd^{2+} , and Hg^{2+} bind preferentially and selectively to heterocyclic bases of DNA, thus disrupting hydrogen bonding and lowering the temperature of thermal denaturation (Tm). Other ions such as Na^+ , Mn^{2+} , and Co^{2+} appear to bind to the sugar-phosphate backbone, thus stabilizing the double-helical conformation and raising the Tm. CuCl_2 , CdCl_2 , or HgCl_2 (1×10^{-3} to 3×10^{-3} M solutions) induce the production of pisatin and increase the activity of phenylalanine ammonia lyase (PAL) in pea pod tissue. CdCl_2 induced 12-fold increases in PAL activity within 18 hr. MnCl_2 , NaCl , or CoCl_2 do not induce either pisatin or PAL formation. However, MnCl_2 partially modifies the induction potential of HgCl_2 . These results are compatible with our previous hypothesis that phytoalexin production can be triggered by a specific changing of the conformation of chromosomal DNA.

Survival of Pseudomonas syringae on hairy vetch in relation to epidemiology of bacterial brown spot of bean. D. J. HAGEDORN, R. E. RAND, & G. L. ERCOLANI (Univ. Wis., Madison). Isolates of *Pseudomonas syringae*, highly virulent (Vi) for bean, were obtained throughout the year (except late July to mid-August) from leaf surfaces of healthy hairy vetch (*Vicia villosa*) plants collected monthly in bean-growing areas of central Wisconsin. In May, epiphytic Vi populations were largest (10^7 viable cells/g fresh wt) on vetch collected in or near fields where brown spot of bean was severe the previous year. In June, Vi isolates were also detected on vetch in areas where brown spot was not evident before. During the early bean-growing season, samples of rain splash containing the highest number of Vi bacteria were collected near vetch. Several outbreaks of brown spot occurred later near patches of vetch adjacent to bean fields. In September, Vi isolates were prevalent on vetch in or near bean fields affected with brown spot, but were rarely detected in other fields. Numbers of Vi bacteria recoverable from vetch remained constant during the winter, but declined in April. In the greenhouse, Vi bacteria never infected vetch, although they could establish epiphytic populations of ca. 3×10^6 viable cells/g fresh wt on 30- and 60-day-old plants sprayed 2 weeks earlier with 10^3 to 10^7 viable cells/ml.

Etiology of the pecan shuck disease. R. S. HALLIWELL & J. JOHNSON (Tex. A&M Univ., College Station). Premature ripening of pecan shucks, a disease of the cultivar Success and frequently those cultivars with a Success parent, is not caused by a pathogen or poor cultural practices. The premature ripening of the shucks is caused by the apparent premature development and early necrosis of the cells of the abscission zone of the pecan bearing shoot. The disease was experimentally reproduced by treatment of shucks with ethylene or the shoot abscission zones with ethrel. Likewise, premature ripening was prevented by the ethylene suppressants, CO_2 and auxins. Copious amounts of ethylene generated by early ripening shucks were recorded. The disease has been controlled using auxin-type growth regulators.

Temperature effects on penetration and reproduction of soybean-cyst nematode. M. L. HAMBLEEN, D. A. SLACK, & R. D. RIGGS (Univ. Ark., Fayetteville). Penetration and reproduction of *Heterodera glycines* was determined in Lee soybean and other plants grown in sand in plastic cups in

controlled temperature tanks. Inoculations were made with eggs and larvae prior to the placing of plants in the tanks, or after 48 hr at the desired temperature in the penetration studies. One day after inoculation, 4 times as many nematodes were found in roots at 28 C as at 22 C, which was second highest, and after 14 days there were 12 times as many at 28 C. White females were recovered earliest (14 days) and 28 and 31 C, but not until 58 days at 14 C. Little penetration and no maturation occurred at 35 C. Males were recovered first (14 days) at 22, 24, 28, and 31 C, and were never recovered at 14 and 35 C. New larvae were first found (22 days) at 28 C, and were never found at 14, 33, and 35 C. Varying the temperature reduced the number of *H. glycines* maturing in 42 days on Lee soybean. Temperature did not change the resistance of Peking soybean or Doark vetch. The optimum temperature for reproduction on *Lupinus albus* and *Vigna wilmsii* was 22 C.

Ultrastructure of virus-infected Cattleya flowers. PENELOPE HANCHEY, C. LIVINGSTON, & F. B. REEVES (Colo. State Univ., Fort Collins). An orchard hybrid, *Cattleya* 'Claesiana,' infected with *Cymbidium* mosaic virus, developed necrotic streaks in the perianth ca. 2 weeks after flowering. Sepals, petals, and pollinia from both symptomless and necrotic flowers were examined. Electron-microscopic observations demonstrated cytoplasmic paracrystalline viral inclusions in both vascular and parenchymatous tissues. No virus was found in pollinia. Inclusions were most prevalent in parenchymatous tissue where visible necrosis and ultrastructural degeneration were first evident. Using the triphenyl tetrazolium chloride-reaction, the greatest amount of stain reduction occurred adjacent to necrotic streaks. This area was also characterized by a disappearance of starch from plastids, followed by plastid degeneration. Degeneration of other organelles coincided with later stages of necrosis, although the paracrystalline inclusions remained intact.

Influence of squash mosaic on squash hypocotyl permeability. J. G. HANCOCK & A. MAGYAROSY (Univ. Calif., Berkeley). As shown previously, exudation of ^{14}C -assimilates from hypocotyls of intact squash mosaic virus (SMV)-infected squash seedlings was normally higher than from healthy ones; yet tissue sections from subterranean portions of hypocotyls from healthy and SMV-infected plants exhibited no differences in electrolyte leakage and efflux of 3-O-methylglucose (MeG) or α -aminoisobutyric acid (AIB). No consistent differences in urea uptake or efflux of urea from cytoplasm or vacuoles were noted between hypocotyl sections from healthy and virus-infected plants. Uptake (V_{max}) of MeG in hypocotyl tissues was unaffected by SMV infection. AIB uptake was greater by SMV-infected than healthy hypocotyl tissues at low solute concentrations (0.1-2.5 mM). Cell permeability changes in subterranean hypocotyl tissues would be unlikely to account for the higher hypocotyl exudation and the increased microbial populations in soil surrounding hypocotyls of SMV-infected squash seedlings.

Ultrastructural studies of Fomes annosus. R. T. HANLIN (Univ. Ga., Athens). *Fomes annosus*, the cause of a serious root rot of pine, produces both basidiospores and conidia. The conidial state is assigned to the imperfect genus *Oedocephalum*, as *O. lineatum*. Because *Oedocephalum* also contains the conidial states of certain Ascomycetes, some method of separating the conidial states of the two classes would be useful. Since modern classification schemes for the imperfect fungi emphasize conidial ontogeny, electron-microscopic studies of conidial development were undertaken on *O. lineatum*. Large, erect hyphae swell at the

apex to form a rounded ampulla. All over the surface of the ampulla, small areas of the protoplast and associated wall begin to bulge out synchronously. The outer layers of the wall soon break, leaving the thin inner layers which push out to form the slender denticle. The apex of the denticle swells greatly, forming the conidium, which is cut off by a septum. Conidia produced by tissue-culture isolates were all dikaryotic. The mycelium of *Fomes annosus* contains typical basidiomycetous septa with a septal swelling and a perforate septal cap.

Host range and identity of cherry raspleaf virus in British Columbia. A. J. HANSEN (Can. Dep. Agr. Res. Sta., Summerland, B.C., Can.). A virus has been repeatedly isolated from cherry trees with raspleaf symptoms in four British Columbia orchards. After inoculation, young plants of *Chenopodium amaranticolor*, *C. murale*, *C. quinoa*, *Cucumis sativus*, *Gomphrena globosa*, *Nicotiana* (three species, four cultivars), *Momordica balsaminea*, *Sesbania exaltata*, *Solanum sisymbriifolium*, and *Zinnia hybrida* became systemically infected; *Petunia* cultivars, some *Vigna sinensis* cultivars, and *Cyamopsis tetragonoloba* reacted with local lesions, whereas *Solanum melongena* and *Spinacea oleracea* were immune. Thermal inactivation point was 55 C; dilution end point was 1:500. The virus was easily distinguished from other stone fruit viruses by the typically faint and temporary mottle induced on *C. quinoa*. *Prunus mahaleb* seedlings were successfully sap-inoculated. Buds from these seedlings induced the original raspleaf symptoms in Bing, Lambert, and Van cherry, but did not infect *P. armeniaca* or *P. persica*. Virus movement within and between infected trees was slow, and symptom distribution was incomplete. The local isolates were serologically related to the cherry raspleaf virus of California, but not to tobacco ringspot virus. The pattern of natural occurrence indicates that the virus may be indigenous.

Effect of ozone and sulfur dioxide on injury, growth, and yield of soybeans. A. S. HEAGLE (ARS, USDA, Environmental Protection Agency, Research Triangle Park, N.C.). Soybeans (*Glycine max* "Dare") were grown in field chambers and exposed to pollutants for 6 hr/day beginning 14 days after emergence. The treatments were 0 pphm O₃ (control), 5 pphm O₃ (low O₃), 10 pphm O₃ (high O₃), 10 pphm SO₂ (SO₂), and 10 pphm O₃ + 10 pphm SO₂ (mix). Various plant responses of injury, growth, and yield were evaluated 43, 92, and 133 days after exposures began. SO₂ alone, or in the mixture, did not significantly affect these responses. Low O₃ significantly increased injury and defoliation, but did not significantly reduce growth or yield. The high O₃ and mix caused significant increases in injury and defoliation and a significant reduction in all yield responses. Defoliation after 92 days in the SO₂, low O₃, high O₃, and mix treatments was 112, 122, 153, and 168%, respectively, of the control; the field weight of seeds after 133 days was 99, 97, 45, and 36%, respectively, of the control. The results show that injury can occur without yield losses. The results suggest that, unless acute episodes occur, soybean yield will not be reduced in areas with seasonal daily 6-hr averages of less than 5 pphm O₃ and 10 pphm SO₂.

Degradation of phaseollin and pisatin by Stemphylium botryosum. MICHELE C. HEATH & VERNA J. HIGGINS (Univ. Toronto, Ontario, Can.). The ability of the alfalfa pathogen, *Stemphylium botryosum*, to degrade pterocarpanoid phytoalexins from nonhost plants was investigated by incubation of growing mycelium with purified phaseollin or pisatin. Recovery of both phytoalexins decreased with time, and this loss was accompanied by the

appearance and increase in compounds detectable with Silica Gel thin-layer chromatography (TLC). The first recognizable degradation product of each phytoalexin was isolated by preparative Silica Gel-TLC. In neutral and acidic ethanol, both compounds had an ultraviolet absorption spectrum similar to that of the original phytoalexin. Under alkaline conditions, however, both showed a more pronounced shift of absorption maxima. In mycelial growth bioassays, both compounds were as fungistatic towards *S. botryosum* as their parent phytoalexin (ED₅₀ less than 50 µg/ml). Rates of degradation of both phytoalexins depended on their initial concentration and on the size of fungal inoculum. Maxima were recorded of 50 µg/day for phaseollin and 10 µg/day for pisatin. This is in contrast to previous studies on the breakdown of the alfalfa phytoalexin, medicarpin, where degradation rates of about 350 µg/day were observed.

Steroid structure in relation to induction and inhibition of oospore formation in Pythium periplocum and P. prolatum. J. W. HENDRIX & SHELLEY-MARIE GUTTMAN (Univ. Ky., Lexington). Oospore formation was induced in *Pythium periplocum* and *P. prolatum* by 3-β-hydroxysterols commonly occurring in plants, animals, and fungi. Cholesterol and sitosterol esters induced oospore production. Cholestanol induced formation of abortive oogonia in both fungi; and lanosterol, 24-dihydrolanosterol, and coprosterol induced formation of abortive oogonia in *P. prolatum* only. β-Estradiol and 16-epiestriol prevented oospore formation by *P. periplocum* in medium containing sitosterol; estrone, estriol, and 4-estren-17β-ol-3-one reduced oospore production. The 3-acetate of β-estradiol was completely inhibitive, whereas the 17-acetate and the diacetate were inactive. α-Estradiol was inactive. Some estrogens reduced growth of *P. prolatum*, but none affected reproduction. A number of androstane compounds reduced reproduction in *P. periplocum*, but pregnane compounds did not affect either fungus. Other compounds inhibiting reproduction in both fungi included digitonin, tomatine, tomatidine, and solanidine, but not solanine.

Characteristics of the rust flora of Mexico. J. F. HENNEN (Purdue Univ., Lafayette, Ind.). The Uredinales (rusts), all plant parasites, comprise the largest natural order of fungi, estimated at 130 genera and 5,000 species worldwide. Much of Mexico remains to be sampled for rusts (Uredinales), but field and herbarium studies during the past 5 years show that currently there are 44 genera and 524 known species—as compared with 35 genera and 710 species for North America north of Mexico, an area 10 times larger, and 42 genera and 384 species for Guatemala, an area 10 times smaller. There are 67 vascular plant families that serve as hosts for rusts in Mexico as compared to about 160 worldwide. Families in Mexico with the largest number of rust species are the Compositae-123, Gramineae-95, and Leguminosae-38. The rust genera *Puccinia* and *Uromyces* combined comprise 55% of the total species. About 20 species are economically important plant pathogens. An earlier report that the aecial stage of *P. graminis* occurs in Mexico is doubtful because it has not been found recently, and the specimen upon which the report is based could be a stage of *P. berberidis-trifoliae*, a correlated microcyclic species.

Isolation of a toxic factor from Pyrenochaeta terrestris and ultrastructure of toxin-treated onion seedlings. W. M. HESS, S. L. HESS, & D. J. WEBER (Brigham Young Univ., Provo, Utah). *Pyrenochaeta terrestris* (C-44-1), the onion pink root fungus, was grown on Czapek's solution medium with powdered cellulose on a reciprocal shaker at 22 C for 30-40 days. Filtrate was collected and concentrated on a

rotary flash evaporator. The concentrated filtrate was precipitated with two volumes of methanol. The precipitate was discarded and the supernatant liquid was then partitioned with butanol. The aqueous layer was evaporated to dryness in a flash evaporator. Further purification steps involved column chromatography and recrystallization with butanol-methanol (1:1, v/v). Infrared spectra were obtained on the purified fractions. The results indicate that the toxic product was a carbohydrate compound. Ultrastructural comparisons were made between onion roots infected with *Pyrenochaeta terrestris* and those treated with the toxin. The results indicate that toxin-treated roots were affected in like manner, but much more severely than roots of susceptible varieties infected by the fungus.

Sources and inheritance of Peru tomato virus tolerance in tomato. H. R. HIKIDA & W. B. RAYMER (Campbell Inst. Agr. Res., Cinnaminson, N.J.); Fifty-five tomato cultivars and wild species selected for tolerance to tobacco etch virus were tested for tolerance to the Peru tomato virus (PTV). Tolerance was evaluated by means of a 0-5 rating system based on severity of mottling, rugosity, and leaf distortion. No resistance to infection by sap-inoculation was found. The most tolerant lines were P.I.'s 128653 (*Lycopersicon peruvianum*), 247087 (*L. hirsutum*), and 126955 (*L. esculentum*). P.I. 126955 and P.I. 127827 (*L. hirsutum*), with moderate tolerance to PTV and excellent tolerance to tobacco etch virus, were used in crosses with Campbell 25, a susceptible variety. When P.I. 126955 was used, all F_1 plants were susceptible; F_2 plants segregated to fit a 3 susceptible:1 tolerant ratio; backcross to P.I. 126955, a 1:1 ratio; and all plants backcrossed to Campbell 25 were susceptible. When P.I. 127827 was used, all F_1 plants were tolerant; F_2 plants segregated to fit a 3 tolerant:1 susceptible ratio; and backcross to Campbell 25, a 1:1 ratio. Tolerance to PTV in P.I. 126955 is controlled by a single recessive gene; and in P.I. 127827, by a single dominant gene.

Characterization of the maize dwarf mosaic virus strain B protein. J. H. HILL, R. E. FORD, & HELEN I. BENNER (Iowa State Univ., Ames). We purified maize dwarf mosaic virus strain B (MDMV-B) by grinding infected corn in 0.5 M phosphate buffer, pH 7.1 containing 1% mercaptoethanol, squeezing through gauze, clarifying the filtrate by pH adjustment to 4.7, readjusting to pH 7.0 30 min later, and emulsifying with CHCl_3 (25/100, v/v). After low-speed centrifugation, we dissolved polyethylene glycol 6,000 to 8% in the supernatant. We collected precipitate 12-18 hr later and resuspended in 0.05 M phosphate buffer, pH 7.4, containing 0.5 M deionized urea and 0.1% mercaptoethanol. Two differential centrifugations, with the second high speed through 30% sucrose, and equilibrium centrifugation in CsCl completed purification. Dialyzed MDMV with low nucleic acid content characteristic of the potato virus Y group was infectious. Virus was degraded by guanidine HCl in the presence of LiCl. Virus protein was multibanded in polyacrylamide gels, but reduction and carboxymethylation yielded a single homogeneous band. This suggested that protein aggregates from spontaneous oxidation and formation of intermolecular disulfide bridges. Subunit molecular weight estimates by sodium dodecylsulfate gel electrophoresis are 35-36,000 daltons.

Losses due to Verticillium wilt in sunflower. J. A. HOES (Can. Dep. Agr., Morden, Man.). The effects of wilt (*Verticillium dahliae*) on sunflower (*Helianthus annuus*) cropped in 1971 in Manitoba, Can., were determined for two fields of the large-seeded cultivar Commander, which is highly susceptible to wilt. Yield was reduced by 66% in the field with

50% diseased plants, and by 59% in the field with 60% disease incidence. Plants with severe, moderate, and mild leaf symptoms showed average yield reductions of 80%, 55% and 18%, respectively, and head diameter reductions of 42%, 28% and 16%, respectively. Data indicate that the most important factor in reduction of yield is reduction in head size. Bushel weight of achenes or seeds averaged 11.2 kg for healthy plants and 10.1 kg for diseased plants. Seeds of diseased plants were smaller. Hull content was not appreciably affected, but the oil content of the meat fraction was on the average reduced from 51.4 to 46.2% as a result of wilt.

A new nitroheterocyclic bactericidal chemical controlling bacterial wilt of chrysanthemum. H. A. J. HOITINK & S. M. ALCORN (Ohio Agr. Res. Development Center, Wooster, Univ. Ariz., Tucson). Bioassay discs saturated with 100 $\mu\text{g/ml}$ 2-amino-5-(1-methyl-5-nitro-2-imidazolyl)-1,3,4-thiadiazole (American Cyanamid Compound 64,855) in agar diffusion tests inhibited soft-rot *Erwinia* spp., *E. quercina*, *E. stewartii*, *Agrobacterium rhizogenes*, *A. tumefaciens*, and five *Bacillus* spp., but not *E. amylovora*, *E. tracheiphila*, *A. pseudotsugae*, six *Pseudomonas* spp., six *Xanthomonas* spp., and three *Corynebacterium* spp. Cells ($10^4/\text{ml}$) of 22 isolates of *E. carotovora* var. *chrysanthemi* were killed in 30 min when exposed to 50 $\mu\text{g/ml}$. The minimal bactericidal concentration was 0.6 $\mu\text{g/ml}$. Roots of tomato seedlings 8-15 cm tall were immersed in 125 $\mu\text{g/ml}$ of the chemical for 4 hr and washed, and different plant tissues were bioassayed. Apical stem sections of these plants produced zones of inhibition on seeded agar plates indicating systemic uptake. Chrysanthemum cuttings soaked in *E. carotovora* var. *chrysanthemi* (10^7 cells/ml) for 4 hr were placed in various concentrations of bactericide for 16 hr, rooted, and grown to maturity. All treated plants remained visibly healthy. The pathogen was isolated from 36, 25, 8, 0, and 0% of plants grown from cuttings treated with 5, 10, 25, 50, and 100 $\mu\text{g/ml}$ of the bactericide, respectively. The high concentrations apparently eradicated the pathogen.

Competition between rice and weeds in nematode control tests. J. P. HOLLIS (La. State Univ., Baton Rouge). Gulf Coast rice fields infested with ring nematodes, *Criconeoides onensis* 1959, and an undescribed species of *Criconeoides* have been investigated since 1966 with respect to geographic distribution and pathology. Control tests in rice fields with organophosphatic compounds, principally Nematicur [Ethyl 4-(methylthio)-*m*-tolyl isopropylphosphoramidate], revealed a nematicide-weeds interaction which partially masks the beneficial effects of nematode control on rice. Grass development in chemically treated rice plots infested with *Criconeoides* and the demonstration that the common rice field weeds *Echinochloa colona*, *Cyperus iria*, *C. haspan*, *C. articulatus*, and *Fuirena* sp. are hosts for the ring nematode *C. onensis* support the view that nematicide treatments stimulate grassy weed development in competition with rice. For this reason, complete grassy weed control is necessary when evaluating a nematicide on rice.

Sulfide diseases of rice. J. P. HOLLIS, A. I. ALLAM, & G. PITTS (La. State Univ., Baton Rouge). Our evidence suggests that three types of sulfide disease occur in Gulf Coast rice fields: (i) Autumn decline (akiochi, in Japan), which links plant symptom expression with free soluble soil sulfide, occurring in the Grand Marais area of southwest Louisiana; (ii) straighthead, with characteristic symptoms only in susceptible rice cultivars, found mainly in soils predominating in kaolin clays that do not sorb H_2S from the soil solution [higher H_2S values (0.1-0.9 mg/ml) are obtained from Texas kaolin (straighthead) soils than from diverse Louisiana

soils]; and (iii) symptomless or mild sulfide disease (MSD), associated with possible yield reduction, visible late-season plant decline, and other factors, widespread in Gulf Coast rice areas. The etiology of MSD lies in the significant inhibition of seedling enzymatic and respiratory activities, including cytochrome oxidase activity, by common rice field levels of H_2S and inhibition of nutrient uptake in rice plants reported from Japan. Highest soil concentrations of H_2S occur at rice plant flowering stages, and are correlated with late-season symptom expression in sulfide diseases.

Formation of oospores by Phytophthora parasitica in liquid medium. R. C. HONOUR & P. H. TSAO (Univ. Calif., Riverside). Oospores of *Phytophthora parasitica* free from chlamydospores and sporangia can be produced in abundance in liquid medium. Approximately 2×10^6 oospores/25 ml medium were formed in clear V8-CaCO₃ broth in a 12-oz prescription bottle 10 days after it was seeded with suspensions of mycelial fragments of two compatible mating types (citrus isolates T131 \times T515) and incubated at 25 C in the dark. Under these conditions, formation of chlamydospores and sporangia was prevented or greatly reduced. Formation of oogonia and oospores commenced at day 3, and continued at a constant rate until day 10. Although almost all oogonia contained oospores, the number with mature oospores was less than 1% at day 10; it increased to 45% at day 21. Sexual reproduction occurred at 18-30 C with the optimum at about 24 C, and was greatly inhibited by exposure to light. Application of this technique to *P. palmivora*, *P. drechsleri*, *P. cinnamomi*, *P. megasperma* var. *sojiae*, and *P. cactorum* resulted in oospore formation with yields in the range of 4×10^4 to 9×10^5 /culture.

Influence of volatile inhibitor from soil on seed germination. T. S. HORA & R. BAKER (Colo. State Univ., Fort Collins). We have previously demonstrated that volatiles from soils inhibit germination of fungal propagules. Pathogenic effects of these volatiles from field soils of various textures and pH on the germination of wheat, golden millet, alfalfa, cress, lettuce, radish, cucumber, calendula, dianthus, and saliva seeds were determined. Surface-sterilized seeds were placed on moist, sterile filter papers, suspended over soil samples in closed petri dishes, and incubated for 4 days at 25 C. Seed germination was delayed and root development was retarded (up to 90%) with clay loam soils of pH 8.6. Less inhibition was apparent with clay loam soils of pH 7.5. No inhibition was detectable in loam soils of pH 5.6 and 6.5. The amount of inhibition observed when seeds were in contact with soil was positively correlated with inhibition following exposure only to volatiles. Thus, volatiles have a profound influence on seed germination; the inhibition observed in alkaline soils is the result of the presence of volatiles rather than the direct influence of pH.

Effect of light on symptoms of chrysanthemum chlorotic mottle. R. K. HORST, A. W. DIMOCK, & C. M. GEISSINGER (Cornell Univ., Ithaca, N.Y.). The intensity of chlorotic mottle symptoms of *Chrysanthemum morifolium* 'Deep Ridge' subsequent to tissue implant inoculations can be controlled by light intensity and photoperiod. Effect of light on symptoms was based on numbers of plants with symptoms after given time intervals and on the length of time for symptoms to appear on each plant. For all experiments, temperature was maintained at 30 C, and relative humidity at ca. 50%. In one series of experiments, plants received 270 or 2,700 ft-c with a 14-hr photoperiod. In another series, plants received 500 or 2,000 ft-c for 8-, 12-, or 16-hr photoperiods. The optimum light intensity for symptom expression was 2,700 ft-c with 14-hr photoperiod, or 2,000 ft-c with 16-hr

photoperiod; however, good symptom development was obtained in the latter tests with light intensities of 2,000 ft-c with 12-hr photoperiod. The mean length of time for symptoms to appear on 15 plants with 8-, 12-, and 16-hr photoperiods at 2,000 ft-c was 11, 9, and 9 days, respectively; whereas with 8-, 12-, and 16-hr photoperiods at 500 ft-c, mean symptom development time was 20, 14, and 13 days, respectively. These results agreed with observations of symptom expression under natural light conditions in greenhouses.

Pyrenophora trichostoma and Leptosphaeria avenaria f. sp. triticea, a major leaf spot complex on wheat. R. M. HOSFORD, JR. (N. D. State Univ., Fargo). *Pyrenophora trichostoma* has been reported to damage wheat in Africa, Australia, Europe, India, Japan, North America, and the Middle East. It has caused severe leaf spotting (alone) and in complexes with *Septoria nodorum*, *S. tritici*, and *Helminthosporium sativum*. In North Africa, it contributed to the very severe leaf and glume spotting attributed to Septoria in 1968-1970. *Leptosphaeria avenaria f. sp. triticea* has been reported on wheat only in Canada and the United States. In 1970 and 1971 field trials at four North Dakota experiment stations, a complex of similar leaf spots caused by *P. trichostoma* and *L. avenaria f. sp. triticea* was related to spring and durum wheat losses averaging 12.9% in yield and 1.0% in test weight. Host resistance related to duration of moisture on the leaves was overcome, and the natural spread of inoculum was enhanced in 1971, a wet year. The spring wheat ND487 maintained its previously reported resistance to *L. avenaria f. sp. triticea*. In the greenhouse, winter wheat line WW8 displayed resistance to *P. trichostoma* isolate PyW17. This isolate had been virulent on all tested wheats and had overcome previously detected resistance in *Triticum turanicum* P.I. 184526.

Effect of herbicides on soil-borne pathogens. L. D. HOUSEWORTH & B. G. TWEEDY (Univ. Mo., Columbia). The effect of atrazine, fluorodifen, alachlor, and flumeturon on radial growth of *Fusarium oxysporum*, *Gibberella zeae*, and *Diplodia maydis* on potato-dextrose agar was determined. At 50 μ g/ml, radial growth of *F. oxysporum* was inhibited 4 and 37% by fluorodifen and alachlor, respectively. *G. zeae* was inhibited 10, 21, and 26% by atrazine, fluorodifen, and flumeturon, respectively. *D. maydis* was inhibited 46, 57, 37, and 83% by atrazine, fluorodifen, alachlor, and flumeturon, respectively. Other treatments were the same as the controls. To determine the effect of the pesticides on disease incidence caused by the test organisms, soybeans or corn were planted in pesticide-treated soil containing a high level of inoculum of these pathogens. Disease severity was determined as the per cent of plants emerged at 10 days. Soybean emergence in sterile soil averaged 95% regardless of pesticide treatment; in *F. oxysporum* infested, nontreated soil, it was 56%. Emergence in infested soil treated with fluorodifen was 29%. Alachlor had no effect. Atrazine increased the per cent of surviving corn plants 23 and 22% in soil infested with *G. zeae* and *D. maydis*, but had no effect on emergence in soil inoculated with *F. oxysporum*. Alachlor decreased injury by *D. maydis* 15%.

Comparison of lesions in vector cell monolayers and in leaves for assay of potato yellow dwarf virus. H. T. HSU & L. M. BLACK (Univ. Ill., Urbana). The *sanguinolenta* variety of potato yellow dwarf virus (SYDV) is unique in that it is possible to compare the effectiveness of its infectivity assays on vector cell monolayers and on leaves. The monolayers consisted of AS2 cells derived from *Aceratagallia*

sanguinolenta, the vector of SYDV; the leaves were those of *Nicotiana rustica*, on which SYDV forms primary local lesions. Comparisons were made under conditions that were optimal for inoculation of monolayers in one case and leaves in the other. Dilution end points on monolayers grown on coverslips 1.77 cm² in area were about 10^{-2.5} times those on leaves about 200 cm² in area. The monolayers yielded about 10^{4.5} and about 10^{3.7} more infections per unit area and per unit number of cells, respectively, than did the leaves. Leaf cells required wounding; monolayer cells did not. The coefficient of variation of representative assays on 10 coverslip monolayers was about 6%, whereas it was about 73% for 22 half-leaves; and for leaf inoculation it was necessary to use virus concentrations that were 250 times greater than those used for cell monolayers. Furthermore, lesions can be counted in monolayers about 40 hr after inoculation, whereas about 2 weeks must elapse before lesions on leaves can be counted readily.

Chlamydospore formation by Fusarium in model systems. S. C. HSU & J. L. LOCKWOOD (Mich. State Univ., East Lansing). Simple systems were sought which would induce chlamydospore formation in *Fusarium oxysporum* f. sp. *lycopersici*, *F. oxysporum* f. sp. *melonis*, *F. roseum* f. sp. *cerealis* 'Culmorum'; *F. solani* f. sp. *phaseoli* (two isolates), and *F. solani* f. sp. *pisi*. Chlamydospores were formed from washed macroconidia of all species incubated on Nuclepore filters floating on 0.03 M Na₂SO₄ solution (pH 5.0-7.0). Numbers and rate of formation were equivalent to those on soil, except for reduced production by *F. solani* f. sp. *pisi*. Other salts (e.g., NaCl and Na₂CO₃) showed greater species and clonal specificity. Water was ineffective. Germlings, obtained by incubating macroconidia on Nuclepore filters on potato-dextrose agar, produced as many chlamydospores during incubation on acid-washed, sterilized sand leached with water, 0.03 M Na₂SO₄, or 0.025 M phosphate buffer (pH 6.9) as on soil. Fewer chlamydospores were formed in nonleaching conditions. Washed germlings produced in potato-dextrose broth and incubated directly in 0.03 M Na₂SO₄ gave chlamydospore production equivalent to that in a sterilized extract of soil. Distilled water was slightly less effective. The results indicate that chlamydospore production requires an environment deficient in utilizable energy sources, but containing appropriate mineral salts.

Reaggregation properties of structural protein from chloroplast membranes of bacterially induced hypersensitive tobacco tissues. J. S. HUANG, P. Y. HUANG, & R. N. GOODMAN (Univ. Mo., Columbia). Structural protein (SP) prepared from chloroplast membranes of tobacco leaf tissues 6 hr after infiltration with either H₂O or 10⁸ cells/ml of *Erwinia amylovora* was solubilized with sodium dodecyl sulfate. SP from water-infiltrated tissues formed spherical clumps, whereas the SP from bacteria-infiltrated tissues formed irregular aggregates upon dialysis against a Tris [tris(hydroxymethyl)amino methane] buffer containing 20 mM Mg⁺⁺. In the presence of chloroplast lipid (from water infiltrated tissue) extracted with a chloroform-methanol mixture, solubilized SP from water-infiltrated tissue reaggregated to form membranelike structures closely approximating those of chloroplast membrane in thickness and orientation. The lipid (from water-infiltrated tissue) and SP from bacteria-infiltrated tissue were less competent in forming these membranelike structures. The reaggregation properties of SP from bacteria-infiltrated tissue were attributed to changes in surface charge indicated by the presence of two new bands in electrophoretic profile and by the change in amino acid composition. These changes may be responsible for the alteration in membrane permeability and

integrity in bacterially induced hypersensitivity in plant tissues.

A strain of tobacco mosaic virus with defective coat protein and its reversion to a functional form. J. J. HUBERT & M. ZAITLIN (Univ. of Ariz., Tucson). A defective strain of tobacco mosaic virus (TMV) PM6, was isolated after treatment of the common (U1) strain with nitrous acid. PM6 viral coat protein aggregates in an aberrant, nonfunctional manner, and does not coat the viral RNA, resulting in a labile infectious principle and a distinctive symptomatology. The defective protein aggregates into a "stacked disc" configuration rather than the normal helical pattern known for TMV protein. However, when plants infected with PM6 are grown in the greenhouse during summer, they frequently develop U1 symptoms; virus particles with functional coat protein may be extracted from these plants. Coat protein analyses of 19 from 20 such isolates examined revealed one less residue of alanine and one additional threonine when compared with U1; coat protein from the 20th isolate had the U1 composition. These results suggest that conversion from a nonfunctional to a functional coat protein is a real phenomenon, and does not result from chance contamination by the U1 strain.

The fate of mercury in vegetative parts of barley grown from seed treated with mercury fungicides. D. HUISINGH & D. M. KLINE (N.C. State Univ., USDA, ARS, Raleigh). Seed treatment with mercury-containing fungicides, extensively used for the control of many seed-borne and seedling diseases, has been restricted because of concerns about heavy metal pollution. The fate of mercury in barley was studied in seedlings grown from Hg-treated seed in the greenhouse and field. Treatments 0.5, 1, 2, and 5 times the recommended rates included ethyl mercury *p*-toluene sulfonanilide, ethyl mercury chloride, cyano(methylmercuri)guanidine, phenyl mercuric acetate, phenyl mercuri triethanol ammonium lactate, methoxyethylmercury chloride, hydroxymercurichlorophenols/hydroxymercurinitrophenols, mercuric chloride, and two controls, water and 5, 6 dihydro-2-methyl-1, 4 oxathiin-3-carboxanilide. Flameless atomic absorption analyses of the mercury content of barley tissues revealed absorption and translocation of Hg from treated seeds to seedling leaves and roots. Mercury uptake and translocation in barley leaves and roots showed a definite dosage-concentration dependence over the range of rates tested. The anionic moiety to which Hg is attached influenced absorption and translocation. Mercury levels in plants decreased with time; the levels in leaves decreased more rapidly than that in roots.

Separation of microsclerotia of Verticillium albo-atrum from soil residues by density flotation. O. C. HUISMAN & L. J. ASHWORTH, JR. (Univ. Calif., Berkeley). A technique consisting of wet sieving and flotation centrifugation was used to determine population levels of *Verticillium albo-atrum* in field soils. This modification of Ashworth's sieving method improves the ease of enumerating by removing obscuring sand and silt. Soil samples (15 g dry wt) were washed through 125- μ and 37- μ sieves. Residues retained by the 37- μ sieve were suspended in a 70% sucrose (w/v) solution and centrifuged for 15 min at 1,700 g. Materials floating on the sucrose solution were collected and washed with sterile water by centrifugation. Final preparations (about 3 mg) were essentially free from sand and silt, and consisted mainly of organic material. Microsclerotia of *Verticillium* could be readily seen upon microscopic examination. Identification was confirmed, and quantification was performed by a plating of the residue on an agar medium (Czapek's salts plus 1% sodium polypectate

and 0.1% Tergitol-NPX) and observing germination and fungal colony development.

A foliar disease of Anthurium seedlings caused by Aphelenchoides fragariae. J. E. HUNTER, W. H. KO, R. K. KUNIMOTO, & T. HIGAKI (Univ. Hawaii, Hilo). A foliar disease caused by *Aphelenchoides fragariae* was discovered recently on seedlings of *Anthurium andraeanum*. The disease was found only on plants propagated from seeds germinated on shredded logs of the Hawaiian tree fern, *Cibotium chamissoi*. Disease incidence and severity was greatest during rainy periods. *A. fragariae* was constantly associated with necrotic lesions on the leaves. Lesions were temporarily restricted by large veins, but eventually the entire leaf became diseased; infected seedlings frequently died. Pathogenicity was demonstrated by attaching pieces of diseased leaves to healthy leaf tissue. Transmission also occurred when infested filter paper discs or sponges were attached to the leaves. In a moist environment, symptoms developed on immature leaves within 5-7 days; mature leaves remained healthy. *Anthurium* seeds were invaded and destroyed when germinated on tree fern medium previously used to grow seedlings which had been infected. Seedlings propagated on other media or on hot water-treated tree fern medium remained free of the disease. Hot-water therapy at 46.7 C for 12 min controlled nematodes, but caused injury to roots and succulent leaves. Chemotherapy with Nematicur [ethyl-4-(methylthio)-m-totyl isopropyl phosphoramidate] was successful at high rates.

Characterization of multiple RNA components from purified barley stripe mosaic virus. A. O. JACKSON, L. C. LANE, & M. K. BRAKKE (ARS, USDA, Univ. Neb., Lincoln, John Innes Institute, Norwich, England). Barley stripe mosaic virus (BSMV) was purified by a series of differential centrifugations, and RNA was extracted by phenol treatment, by a bentonite-SDS procedure, or by an urea-SDS-mercaptoethanol method. Analysis by polyacrylamide gel electrophoresis revealed the presence of more than one RNA species in all virus strains examined, but the number, size, and relative proportions of the RNA species differed with different virus strains. RNA species were resolved which were common to Argentine Mild, ND18, Canadian Severe, ATCC-Type, and Rothamsted strains. However, Argentine Mild and ND18 contained RNA components which were not present in the Canadian Severe and ATCC-Type strains. Infectivity studies showed that more than one species of the separated RNA's were necessary for establishment of an infection, although the role of individual components has not been established. Comparison of the polyacrylamide gel electrophoretic profiles and sedimentation profiles of brome mosaic virus (BMV) RNA and BSMV-RNA suggests that the RNA's of BSMV differ in molecular configuration from those of BMV. The RNA's of BSMV migrate more slowly in gels than might be expected from their sedimentation rates, based on BMV-RNA's as standards.

Interaction of Pseudomonas solanacearum and Meloidogyne incognita on bacterial wilt-resistant and susceptible cultivars of tomato. S. F. JENKINS, JR. (N.C. State Univ., Raleigh). The split-root technique was used in the greenhouse to test the interaction of *Pseudomonas solanacearum* and *Meloidogyne incognita* on southern bacterial wilt-resistant cultivars Venus and Saturn and wilt-susceptible Manapal. Treatments included inoculation of roots with bacteria through root wounding, or plastic tubes placed in soil on plants previously noninoculated with *M. incognita* or previously inoculated in the same root system or

the opposite split system. Using the symptomatic criteria of wilting and death on a time-scaled index, Manapal seemed to be slightly more susceptible to bacterial wilt when previously infected with *M. incognita*. Venus and Saturn, however, remained equally resistant in the presence or absence of the nematodes. The inheritance of resistance in Venus and Saturn to southern bacterial wilt is thought to be polygenic, and it is concluded that the root knot nematode is not ordinarily able to predispose the resistant cultivars to infection by the bacterium, *P. solanacearum*. This confirms similar observations in field wilt nurseries during the years 1969-1971.

A comparison of Cronartium fusiforme and C. quercuum in pine. F. F. JEWELL (La. Tech. Univ., Ruston). A quantitative study was made of *Cronartium fusiforme* in slash (*Pinus elliotii*) and loblolly (*P. taeda*) pine and *C. quercuum* in jack (*P. banksiana*) and Virginia (*P. virginiana*) pine to determine whether measurable differences existed between the two rust species. Hyphal width and haustorial length and width were each measured 60 times in galls of unknown ages from each of 10 slash, loblolly, and jack pine, and in galls from six Virginia pine. *C. fusiforme* hyphae averaged 3.2 μ in slash and 3.7 μ in loblolly, and haustoria averaged 16.3 \times 3.3 μ in slash, and 14.1 \times 3.8 μ in loblolly. *C. quercuum* averaged 3.5 and 3.2 μ for hyphae in jack and Virginia pine with haustoria of 14.1 \times 3.5 μ and 15.7 \times 3.3 μ , respectively. Analysis revealed no significant difference between the two rusts, but significant differences were revealed within each rust species in its respective pine, indicating an effect on the rust by the individual pine. The general morphological features of *C. fusiforme* and *C. quercuum* and the reaction of the various hosts to the rusts were considered to be nearly identical, and no microscopic separation of the rust species was possible. Evidence indicates that *C. fusiforme* and *C. quercuum* are so similar, other than in gall shape, that possibly they should not be considered separate species.

Effects of organic mulches upon the incidence of root knot in potted tomato plants. L. F. JOHNSON (Univ. Tenn. Agr. Exp. Sta., Knoxville). Pots of soil infested with eggs and larvae of *Meloidogyne incognita* were mulched to a depth of 1 inch with dried crop residues chopped to 2-mm particle size. The pots were placed in the greenhouse and watered periodically to prevent drying. After 10 weeks of incubation, assays of root knot severity were made by the transplanting of tomato seedlings to the mulched pots. Roots were examined after the tomatoes had grown for 6 weeks. Many of the mulches stimulated growth and resulted in larger root systems, but tomatoes grown in soil mulched with oat straw were stunted and had smaller root systems. This apparent toxic effect could not be overcome with applications of fertilizers. All the residues tested produced significant reductions in root knot severity when compared with unmulched pots and pots mulched with peat moss or sand. Numbers of galls per plant and per gram of root were reduced 75-99% in pots mulched with flax, alfalfa, or orchard grass. Mulches of fescue or oat straw resulted in significant but smaller reductions. The use of crop residues as mulches was only slightly less effective for controlling root knot than the incorporating of the residues into soil.

Galactosidase production by Sclerotium rolfsii. T. M. JONES & D. F. BATEMAN (Cornell Univ., Ithaca, N.Y.). When grown in culture on autoclaved bean hypocotyls, *Sclerotium rolfsii* (isolate 14) produces a number of polysaccharide-degrading enzymes. These include enzymes which catalyze the hydrolysis of both α - and β -p-nitrophenyl galactosides, and which release reducing groups and

monomeric galactose from Lupin galactan, a β -1,4-linked galactose polymer. The presence of at least two β -galactosidases in culture extracts is indicated by different pH optima for the enzymatic hydrolysis of *p*-nitrophenyl- β -galactoside (pH 2.0-2.5) and of Lupin galactan (pH 4.0-4.5). D-galactal, a competitive inhibitor of many β -galactosidases, is a more effective inhibitor of the *p*-nitrophenyl- β -galactosidase activity than is the galactan-degrading activity. This observation is consistent with the presence of multiple β -galactosidases. These phenomena have also been observed with enzyme preparations from *S. rolfii*-infected bean hypocotyls. Monomeric galactose is released from hypocotyl cell walls of bean plants upon infection with *S. rolfii*. The galactan-degrading enzyme from culture extracts has been partially purified and partially separated from the *p*-nitrophenyl- β -galactosidase activity by precipitation with ammonium sulfate, DEAE-cellulose chromatography, and gel filtration on Sephadex G-75.

Variation between southern and northern isolates of Scirrhia acicola. A. G. KAIS (USDA [Forest Serv.], Gulfport, Miss.). Significant differences in cultural characteristics and pathogenicity have been demonstrated between northern and southern conidial isolates of *Scirrhia acicola*. Three isolates collected from Scotch pine, two in Wisconsin and one in Minnesota, were compared to three isolates collected from longleaf pine in Mississippi. Isolates from the two sources could be differentiated in culture by growth rate and by formation of pigments in growth medium. The southern isolates grew faster in shake liquid culture, and, in all tests, had a higher per cent germination. Optimum temperature for spore germination of the southern isolates was 25 C as compared with 20 C for the northern isolates. I determined differences in pathogenicity between the geographic sources by measuring disease severity on five species of pine. Southern isolates were most pathogenic to jack, sand, and longleaf pine, and least pathogenic to Scotch and loblolly pine; northern isolates were most pathogenic to Scotch, jack, and sand pine, and least pathogenic to loblolly and longleaf pine. With the exception of Scotch pine, the southern isolates were more virulent on all species of pine. The extent of geographic variation demonstrated by these studies indicates the existence of races of *Scirrhia acicola*.

Seed transmission of bean yellow mosaic virus in broad beans in Iran. W. J. KAISER (USDA, Mayaguez, Puerto Rico). Bean yellow mosaic virus (BYMV) is an important disease of broad beans (*Vicia faba*) in Iran. Several pathogenic strains of the virus were isolated from broad beans in different areas of that country. One from Khuzestan Province was sometimes seed-borne in *V. faba*, but not in other food legumes. The virus was transmitted in seed of inoculated and naturally infected broad beans. Mosaic symptoms were usually discernible in the simple primary leaves, and became more pronounced later in the compound leaves. Virus transmission occurred in 0.19 to 0.95% of the seed from plants inoculated at different stages of growth, and was highest in plants infected at or prior to flowering. In a planting of 56 broad bean types from 14 countries, BYMV was seed-borne in 80% of the lines, and incidence ranged from 0.1 to 2.4%. Seed transmission of BYMV was usually less than 0.5% in broad bean plantings in southwestern Iran, but was an important factor in subsequent spread and survival of the virus. BYMV was spread rapidly from diseased seedlings by several aphid vectors, resulting in a high incidence of virus infection annually.

Studies on the nature of soybean resistance to seed rot

caused by Pythium. B. L. KEELING (ARS, USDA, Stoneville, Miss.). Seed rot of soybeans, *Glycine max*, caused by *Pythium* sp. was studied in relation to the soluble carbohydrates exuded from the seed during the first hours of germination. Comparative studies using the soybean cultivars Hood (susceptible) and Semmes (resistant) showed that germinating seed of Hood stimulated the growth of *Pythium* more than germinating seed of Semmes. Paper chromatographs of exudates from germinating seed of both cultivars revealed at least 10 different carbohydrates. However, no qualitative differences between the two cultivars were indicated. Quantitative determinations show that germinating seed of the susceptible cultivar exude as much as twice the amount of soluble carbohydrates as do the resistant cultivar. Coating seed of the resistant cultivar with glucose increased its susceptibility. A direct relationship between the amount of soluble carbohydrates exuded by germinating seed and seed rot caused by *Pythium* is indicated.

Pathogen-produced elicitor of a chemical defense mechanism in soybeans monogenically resistant to Phytophthora megasperma var. sojae. N. T. KEEN, J. E. PARTRIDGE, & A. I. ZAKI (Univ. Calif., Riverside). Hypocotyls of Harosoy 63 (H63) soybeans (monogenically resistant to *P. megasperma* var. *sojae* [Pms]) accumulated hydroxyphaseollin (HP), coumestrol, daidzein, and an unidentified yellow, fluorescent substance (PA_k) 10-100 times more rapidly following inoculation with Pms than the near-isogenic susceptible cultivar Harosoy (H). Wounded cotyledons produced HP and PA_k only when surface-inoculated with Pms, but no difference was observed in production by the two cultivars. Cell-free culture fluids from Pms on natural and synthetic media also contained a substance(s) which elicited production of PA_k and HP in cotyledons. Elicitor production was not enhanced by exposure of the fungus to extracts from H or H63 soybean hypocotyls. Fungus-inoculated and elicitor-treated cotyledons accumulated HP and PA_k at similar rates, beginning 8-10 hr after treatment and increasing rapidly to a maximum at 24-48 hr. Treatment with partially purified (deionization and gel filtration) elicitor preparations caused similar levels of HP and PA_k in both H and H63 cotyledons, but in hypocotyls elicited 5-10 times more HP in H63 than in H. The data indicated that the H63 resistance gene is expressed only in hypocotyls, and that a constitutive fungus metabolite elicits gene activation.

Influence of nitrogen and potassium on fire blight severity in Bartlett pear trees in sand culture. H. L. KEIL & C. B. SHEAR (ARS, USDA, Beltsville, Md.). Bartlett pear trees were grown outdoors for 2 years in washed quartz sand in 5-gal pails. Each pail received 2 liters of nutrient solution supplying either high (225 ppm) or low (56 ppm) nitrogen (N) combined with either high (100 ppm) or low (19 ppm) potassium (K) once or twice weekly from early May to mid-September. All other essential nutrients were supplied at adequate levels. No symptoms of nutrient deficiency developed at any combination of levels of N and K. In August of each year, one succulent shoot tip on each of 10 plants of every treatment was inoculated by hypodermic needle with an aqueous suspension (5×10^6 cells/ml) of *Erwinia amylovora*. The first year, 26 days after inoculation, the mean extent of blight and leaf K (% dry wt) were, respectively: low N + high K, 15 and 1.56; high N + high K, 19 and 1.48; low N + low K, 24 and 1.26; and high N + low K, 28 and 1.28. The second year, 20 days after inoculation, these values were: low N + high K, 18 and 1.58; high N + high K, 41 and 1.59; low N + low K, 30 and 1.36; and high N + low K, 43 and 1.27. Spread of fire blight was least in trees

receiving low N and high K and greatest in trees receiving high N and low K.

Genetic study of resistance to corn rust in Hawaii. S. K. KIM, J. L. BREWBAKER, & F. F. LAEMMLEN (Univ. Hawaii, Honolulu). Rust caused by *Puccinia sorghi* is one of the common corn diseases in Hawaii together with maize mosaic virus 1 and *Helminthosporium turcicum* blight. A single report of *P. polysora* in Oahu in 1970 has not been verified. Rp_1 alleles *a, b, c(2), c-k, d, d(2), f, g, j,* and *k* have been determined as sources of monogenic resistance to *P. sorghi* in Hawaii. Rp_1 alleles *c, l,* and *m* also confer partial resistance. No resistance was conferred by the following genes: Rp_1 allele *h; Rp_2; Rp_3 alleles *a, b, c, d, e,* and *f; Rp_4* alleles *a* and *b;* and Rp_5 . Rp_1 from variety Cuzco confers excellent monogenic resistance which is being introduced into all Hawaiian maize stocks. Another type of resistance obtained from inbred CM105 is under genetic study, and is being introduced into several lines. Inheritance appears to be polygenic, and the resistance effective to a wide variation in rusts. Studies will be reported of the increasing peroxidase activity of infected leaf tissue in resistant versus susceptible germ plasm.*

Species, cultivars, and lines of Phaseolus resistant to Pythium aphanidermatum. S. H. KIM & J. G. KANTZES (Univ. Md., College Park). Nine species of *Phaseolus* (*P. acutifolius, P. acutifolius, P. angularis, P. aureus, P. calcaratus, P. coccineus, P. vulgaris, P. lunatus,* and *P. mungo*), including 172 cultivars and lines, were tested for resistance to *Pythium aphanidermatum*. Two- to 6-week-old plants were spray-inoculated with zoospores and incubated in a moist chamber. None of the plants was immune. *P. lunatus* (P.I. 164893 Guat. Kumasi, P.I. 234256 N. Hamp., and P.I. G-12355 Arizona Hopi Grey Lima) and *P. mungo* (P.I. 288600 India) were more resistant than 138 cultivars and lines of *P. vulgaris* tested. Among the cultivars and lines of *P. vulgaris*, P.I. 180466 India and P.I. 201389 Mexico gave evidence of resistance, whereas P.I. 203958 Mexico N 203, which had been reported resistant, was found to be susceptible in this study.

Relationship of endomycorrhizae to citrus stunting in fumigated soils. G. D. KLEINSCHMIDT & J. W. GERDEMANN (Univ. Ill., Urbana). Soils sterilized with methyl bromide or heat have been reported to be toxic to certain plants. This problem has occurred in fumigated citrus nurseries of California and Florida. Although toxins have been isolated from some heat-treated soils, no toxins have been found in these citrus soils, but their "effects can be partially alleviated by high rates of phosphate fertilizer. Citrus plants growing normally in scattered areas of fumigated nurseries were mycorrhizal, and *Endogone* spores were found in the soil near their roots. Stunted and chlorotic plants were nonmycorrhizal. These plants grew normally after inoculation with *Endogone mosseae*, an endomycorrhizal fungus. Citrus grew poorly in steamed, autoclaved, or methyl bromide-treated soil in greenhouse experiments. Plants produced excellent growth in these treated soils when inoculated with *E. mosseae*. Inoculation also improved the growth of citrus plants in an Illinois field that had been fumigated with methyl bromide. All mycorrhizal plants had a greater dry weight and a higher per cent phosphorus than did nonmycorrhizal plants. Citrus appears to be highly dependent upon mycorrhizal infection for adequate nutrition.

Control of Rhizoctonia cutting rot of two ornamental foliage plant species. J. F. KNAUSS (Univ. Fla. Agr. Res.

Center, Apopka). During cutting propagation and growth of many foliage plants, extensive disease losses result from infection by *Rhizoctonia* spp. *Gynura aurantiaca* and *Hoya carnosa* 'Compacta', susceptible to *Rhizoctonia* under Florida growing conditions, were selected as test species for fungicide control evaluations. An isolate of the pathogen was obtained from naturally infected specimens of each host and employed on its host throughout the test period. Infestation was effected by pouring 25 ml of a mycelial suspension prepared from 7-day-old potato-dextrose broth cultures over the soil surface of each pot. The fungicides, applied as a drench (200 ml/4-inch pot) under glasshouse conditions 1 to 3 days after infestation, were: Banrot, combination of ethazol-Thiabendazole [2-(4-Thiazolyl)-benzimidazole]; benomyl; chloroneb; chlorothalonil; Cleary's 3336, diethyl 4, 4'-0-phenylenebis (3-thioallophanate); ferbam, pentachloronitrobenzene (PCNB). In three experiments on *G. aurantiaca*, benomyl and PCNB proved both nonphytotoxic and effective in disease control. In two experiments on *H. carnosa* 'Compacta', benomyl and PCNB were both effective in controlling the pathogen, but PCNB proved to be phytotoxic.

Growth response of dwarf mistletoe-infected ponderosa pine seedling. D. M. KNUTSON (Forest Serv., USDA, Corvallis, Ore.). Height growth of 2-year-old ponderosa pine seedlings was compared in uninfected plots and plots with 66% of seedlings infected with *Arceuthobium campylopodum*. For a 2-year period, uninfected trees in infected plots have been significantly taller than either the infected trees or trees in uninfected control plots. Mean heights (from cotyledons to the shoot apex) in March 1972 were 75 mm for infected trees, 84 mm for trees in control plots, and 100 mm for uninfected trees in infected plots. Differential root competition is the probable reason: infected trees have less-developed root systems, and are poor competitors for soil nutrients. Differences in growth of paired infected and uninfected trees were significant at high, but not at low, temperature regimes. In other studies with trees grown at two light intensities (750 and 1,500 ft-c), host vigor was a better predictor of dwarf mistletoe growth (aerial shoot elongation) than was the level of light reaching the dwarf mistletoe plants.

Characteristics of a volatile inhibitor from certain alkaline soils. W. H. KO & FRANCES K. HORA (Univ. Hawaii, Hilo). A volatile substance released from the remoistened air-dried alkaline soils inhibited germination of conidia of *Aspergillus fumigatus, Mucor remannianus, Penicillium frequentans,* and *Trichoderma viride*. It also inhibited germination of ascospores of *Neurospora tetrasperma* which is not sensitive to widespread soil fungistasis. The inhibitory effect of soil and the volatile inhibitor released from it were not destroyed by autoclaving or by gas sterilization. Nutrients neither nullified the inhibitory effects of soil nor prevented the release of the volatile inhibitor from it. The only similarity between soil fungistasis and inhibition by the volatile inhibitor is that both are fungistatic. Therefore, it is suggested that the inhibition of spore germination due to the volatile inhibitor is different from that of widespread soil fungistasis.

Occurrence and symptoms of sugarcane mosaic virus and maize dwarf mosaic virus in mixed infections of sugarcane and other grasses. H. KOIKE & A. G. GILLASPIE, JR. (ARS, USDA, Houma, La.). Sugarcane mosaic virus (SCMV) infects sweet sorghum (*Sorghum bicolor* 'Rio'), but does not readily infect Johnson grass (*S. halepense*), whereas maize dwarf mosaic virus, strain A (MDMV-A) readily infects both.

Symptoms on Rio seedlings are distinguishable: a green-on-green mosaic for strains SCMV-A, -B, -D, or -H; reddening of midvein and sheath for SCMV-I; and severe mosaic with red flecking and spindle necrosis for MDMV-A. In one experiment, Johnson grass, itchgrass (*Rottboellia exaltata*), and sugarcane (*Saccharum* hybrids) were inoculated with SCMV-H preceding or after inoculation with MDMV-A. In a second experiment, itchgrass and Rio were inoculated with MDMV-A together with one or more of five SCMV strains. Inoculum taken from singly inoculated control plants elicited the typical response on Johnson grass and Rio; inoculum from doubly inoculated plants infected Johnson grass and Rio, but the symptoms on Rio were milder than with MDMV-A and distinct from SCMV. The results indicate that mixed infections may be initiated on several hosts, whether the viruses are inoculated together or separately. In terms of host range and symptoms, these mixtures could be construed to be several new strains of MDMV. Similar "strains" have been isolated from field-infected Rio and itchgrass.

Temperature effects on seedling wilt from corn kernels infected with Helminthosporium maydis. T. KOMMEDAHL & D. S. LANG (Univ. Minn., St. Paul). Corn (*Zea mays*) planted 13 May in the field when the mean temperature for 3 weeks afterward was 13 C yielded 0-2% (average 1%) wilted seedlings, but kernels sown 28 May when the mean temperature for 3 weeks afterward was 22 C yielded 0-30% (average 8%) wilted seedlings, based on 2,100 and 5,100 kernels sown, respectively. Kernels of the same seed lot, sown in metal flats in the greenhouse, at 18 C yielded 14-21% wilted plants but at 24 C yielded 27-30% wilted plants, based on 1,200 kernels each trial. Seedlings started and kept at 18 C for 2 weeks, then moved to 24 C for 2 more weeks, yielded 14% wilted plants, but those started at 24 C for 2 weeks before transfer to 18 C for 2 more weeks yielded 28% wilted plants. Nonwilted seedlings with root necrosis from infected kernels amounted to 2-8% for plants started at 18 C, but 7-12% for those started at 24 C. Thus, the higher temperature led to greater seedling loss from infected kernels, and early planting would most likely reduce loss from sowing kernels infected with *H. maydis* by minimizing wilt and root rot.

Electron microscopy of sowthistle yellow vein virus-infected Sonchus leaves utilizing conventional thin-sectioning and freeze-etching techniques. C. J. KRASS & D. E. SCHLEGEL (Univ. Calif., Berkeley). Membrane-limited clusters of randomly oriented virions were found in the nuclei and cytoplasm of cells surrounding the vascular bundles. In later stages, the virus had spread to most cells, including the epidermis. Results of the freeze-etching in general paralleled those of the thin sectioning, with some additional information obtained about viral and host membranes. Samples were taken from leaf areas showing typical vein-clearing symptoms in young *Sonchus* plants that had previously been inoculated with sowthistle yellow vein virus by means of the aphid vector. Material for thin sectioning was fixed and processed by conventional procedures. Fresh material, frozen in liquid Freon, was used for freeze-etching, thereby providing a comparative basis for the fixed samples.

Effects of Aspergillus niger on epidermal nuclei of onion. F. B. KULFINSKI & A. J. PAPPALIS (Southern Ill. Univ., Carbondale). *Aspergillus niger* (avirulent) was found to decrease nuclear area and nuclear dry mass of onion epidermal nuclei by approximately 10%, whereas a virulent isolate from onion decreased these characteristics by approximately 40%, ahead of the mycelium. Nuclei in close

proximity to the mycelium were destroyed. The decreases in nuclear area were comparable to those observed in naturally occurring neck rot. Culture filtrate from the virulent form caused killing in 15 sec and a reduction in nuclear size and mass comparable to those caused by infection. Living host cells ahead of the mycelium, in infected onions, were observed to orient their nuclei very markedly toward the advancing mycelium.

Decline of Ohia and Koa Forests in Hawaii. F. F. LAEMMLEN & R. V. BEGA (Univ. Hawaii, Pacific S.W. Forest Range Exp. Sta., Berkeley, Calif). *Metrosideros collins* and *Acacia koa* forests occupy ca. 600,000 acres on the island of Hawaii. According to aerial surveys in 1954, 67,000 acres of this area were in a slight (less than 20% dead trees) to severe (60% or more dead trees) decline. In 1965, the affected areas had increased by 10,000 acres, and the severely affected portion had increased from 5,000 acres (1954) to 14,000 acres (1965). Recent serial and ground surveys indicate a steadily deteriorating situation. The forest decline is seen as a rapid wilt and death of trees and/or a slow progressive decline causing a thinning of foliage, many dead twigs, and general unthrifty appearance followed by complete defoliation and death. The most severely affected forest in on the slopes of Mauna Kea at an elevation of 750-1,700 m. The region receives 250-750 cm median annual rainfall. Possible causal and contributing agents include *Armillaria mellea*, *Phytophthora cinnamomi*, *Diatrype princeps*, *Xylosandrus compactus*, and *Plagithmysus bilineatus*, as well as drainage changes in the substratum, wild pigs, and other mammals, microorganisms, and insects. Quantitative and qualitative studies of the decline have been initiated.

Reaction of cowpea seedling to phytopathogenic bacteria. M. LAI & BARBARA HASS (Calif. Dep. Agr., Sacramento). Testing of bacterial cultures by mechanical inoculation on primary leaves of cowpea (*Vigna sinensis*) seedlings has proved to be a valuable quick method for identification of *Pseudomonas syringae*. Tests have been completed on 397 bacterial cultures representing 68 isolates of 4 *Agrobacterium* spp., 23 of 7 *Corynebacterium* spp., 55 of 10 *Erwinia* spp., 199 of 35 *Pseudomonas* spp., and 52 of 12 *Xanthomonas* spp. on the primary leaves of cowpea seedlings. The positive results consisted of formation of brown necrotic lesions on leaves 24 hr after inoculation when the inoculated plants were maintained in the moist chamber. Bacteria which yielded positive tests on the cowpea variety California Blackeye 3, included eight isolates of *E. amylovora*, two isolates of *P. eriobotryae*, one isolate of *P. fluorescens*, and all tested isolates of *P. allicola*, *P. cichorii*, *P. pisi*, *P. syringae*, and *X. vesicatoria*. Ten cowpea varieties reflected either positive or negative responses to different isolates of *P. syringae*, and California Blackeye 3 is a "universal" test variety for this pathogen.

Light microscope preselection of flat-embedded specimen for electron microscopy. W. G. LANGENBERG & HELEN F. SCHROEDER (ARS, USDA, Univ. Nebr., Lincoln). Tobacco tissue infected with tobacco mosaic virus (TMV), embryonic bovine cells, and spores of *Helminthosporium maydis* race T were fixed, dehydrated, and infiltrated with plastic via conventional means. Tobacco tissue pieces were macerated in a few drops of plastic to release TMV crystalline inclusions from the cells, the plastic was transferred to a carbon-coated slide and covered with a carbon-coated cover slip, and the resin was polymerized at 65 C. Bovine cells grown on carbon-coated cover slips and fungal spores, germinated on a thin layer of water agar on similar cover slips, were infiltrated

with 100% resin and inverted on carbon-coated slides, and the resin was polymerized. We easily separated the thin layer of hardened plastic from the glass surface by slipping a razor blade between resin and glass. Light microscopy at all magnifications could be used to study and select desired cells or cellular organelles for electron microscopy. A small area containing the selected cell, organelle, or crystalline inclusion could be cut out and mounted at the desired angle for thin sectioning and subsequent electron microscopy.

Mechanical resistance of selected genotypes of dried peanuts to colonization by strains of aflatoxin-producing Aspergillus sp. J. C. LA PRADE & J. A. BARTZ (Univ. Fla., Gainesville). Data from inoculations of whole, shelled peanuts from 165 Florida breeding lines with conidia from three isolates of the *Aspergillus flavus-oryzae* group (NRRL 3794, NRRL 2999, and one Florida isolate) revealed statistically significant differences in tolerance to colonization by the fungus. When seed coats were punctured with a needle or abraded with Carborundum before inoculation, there were no significant differences in tolerance among the varieties. When intact seeds were soaked in an aqueous solution of 1.0% 2, 3, 5-triphenyl-2H-tetrazolium chloride (TZC), a red stain occurred in the cotyledons of the susceptible lines, but not those of the tolerant lines. This suggested that the seed coats of the tolerant lines were not as permeable as those of the susceptible lines. Aqueous extracts of intact seeds of both tolerant and susceptible lines stimulated germination of *Aspergillus* conidia when compared to conidia incubated in distilled water. Diethyl ether extracts of intact seeds of tolerant peanut lines revealed no germination inhibition when compared with conidia incubated in distilled water. Thus, an intact testa was required for tolerance and appeared to function as a mechanical barrier to penetration by the fungus.

Viral protein synthesis of Vicia faba inoculated with broad bean mottle virus. J. R. LASTRA (Univ. Calif., Berkeley). In the early stages of infection of *Vicia faba* with broad bean mottle virus (BBMV), viral protein was found to build up first in the cell nucleus and then move into the cytoplasm, in agreement with early findings for tobacco mosaic virus but in contrast to results obtained with clover yellow mosaic virus. The events were followed using immunoradioautography. The first pair of fully expanded leaves of *V. faba* plants was inoculated with BBMV and held in a growth chamber at 75 F with a 15-hr light regime. Samples were taken from the third leaf pair above the inoculated leaves. Light microscopy immunoradioautography of these leaves showed strong nuclear labeling by the 3rd day; track counts showed a high percentage of the grains localized over the cell nucleus. As infection advanced, the proportion of the silver grains over the nucleus declined steadily and at the same time increased over the cytoplasm, indicating movement of viral protein from the nucleus to the cytoplasm. The nucleolus and the chloroplasts were free of labeling, which indicated the absence of viral protein in these organelles.

Two views of pathogenic stability in Pyricularia oryzae. FRANCES M. LATTERELL (ARS, USDA, Frederick, Md.). During 20 years' study of the rice blast pathogen, we have developed techniques for producing dry spore inocula containing as many as 2×10^9 spores/g, >80% viable, with half-life as long as 12 years. It has been possible to conduct hundreds of tests with inoculum originating from a single spore. Many cultural variants have appeared, some associated with pathogenic changes, but incidence of observable changes has been low, considering the numbers of spores involved. Type cultures of most races, once being selected for

sporulating capacity, have retained their characteristic growth habit through 10 to 20 years of periodic transfer, as well as their pathogenic pattern toward differential rice varieties. Our findings are in contrast to those reported recently by several workers whose data indicate the fungus to be extremely labile with respect to pathogenic specialization. In attempting to resolve this perplexing discrepancy, we conducted tests involving 600 single-spore isolates from cultures and lesions. Among these, cultural incompatibilities were common, but changes in race pattern were rare. We believe that the degree of instability reported by these workers is exaggerated, and can lead to serious misconceptions regarding the value of breeding for specific resistance. Suggestions for cooperative exchanges that may explain this dichotomy are offered.

Cercospora kikuchii infection of soybean as affected by stage of plant development. F. A. LAVIOLETTE & K. L. ATHOW (Purdue Univ., Lafayette, Ind.). Purple seed stain of soybean caused by the fungus *Cercospora kikuchii* occurs in all soybean-producing areas, and is one of the causes of lowered seed quality. Seed germination is not greatly reduced, but infected seed frequently produces weak seedlings and less productive plants. The discoloration of the seed coat also lowers seed grade and is objectionable in the export market. Inoculum was grown on V8 juice agar plates, and a spore suspension sprayed to thoroughly wet the plants. To determine at what stage of plant growth infection occurred, field plots were inoculated 1-9 times at weekly intervals, beginning at early bloom. Results indicate that one or two well-timed inoculations during the full flower period gave the maximum infection. More than 90% of the seed from the most susceptible cultivars were infected, whereas less than 1% of the seed from the most resistant lines were infected.

Heterokaryosis of Fusarium oxysporum Schlecht. causing crown rot of tomato. J. V. LEARY (Univ. Calif., Riverside). Single-spore isolations from original cultures of the *Fusarium oxysporum* strain which causes crown rot of tomato yielded at least two distinct segregant types. The first type, designated orange, segregated further into two subclasses. The second type, designated purple, remained stable. These two types differ in pigment production, mycelial morphology, numbers of macro- and microconidia produced, chlamydospore formation by cells of the macroconidium, severity of symptoms, and host tissues attacked. Differences in temperature effects on growth and conidial production were also characterized. Experiments on infections with mixed cultures and artificially induced heterokaryons indicate that the original isolates are fairly stable heterokaryons comprised of two distinct genomes.

A pathogenic bacterium from healthy soybean plants. CURT LEBEN & T. D. MILLER (Ohio Agr. Res. Development Center, Wooster). Colonies resembling those of *Pseudomonas glycinea* grew on a H_3BO_3 selective medium developed for *P. glycinea* when bud macerates from three cultivars of healthy field plants were cultured. Seedling soybean and other bean leaves were wound-inoculated with ca. 10^8 cells/ml derived from *P. glycinea*-like colonies. All isolates produced a nonprogressive necrosis without grossly evident water-soaking in 2 to 7 days. A progressive tissue collapse prevented or reduced seedling emergence when seeds of soybean cultivars, red kidney bean, and cucumber were germinated 2 to 5 days, inoculated by piercing the cotyledons or the hypocotyl, and replanted. Emerged seedlings often were small or malformed. Isolates were hypersensitive in tobacco leaves. Common biochemical tests

were run. On the basis of gelatin liquefaction and potato degradation, isolates appeared to resemble *P. syringae* more than *P. glycinea* or *P. viridiflava*. Results suggest that in nature, in Ohio, the bacterium may multiply in buds and perhaps on other aerial parts of healthy soybean plants. A progressive disease probably would occur only when the organism entered seedling wounds.

St. Augustine decline disease development on millet species. T. A. LEE, JR. & R. W. TOLER (Tex. A&M Univ., College Station). St. Augustine decline (SAD), a mosaic disease of St. Augustine grass (*Stenotaphrum secundatum*) is epiphytic on St. Augustine grass in areas of Texas where millet is grown as a forage. Accessions and cultivars of Pearl millet (*Pennisetum typhoides*), Proso millet (*Panicum miliaceum*), and Foxtail millet (*Setaria italica*) were inoculated with extract from SAD-virus infected leaves. Inoculum consisted of infected St. Augustine grass (common cultivar) macerated in a Waring Blendor at 1:1 by weight with 0.01 M phosphate buffer pH 7.0 and 1% 600 mesh Carborundum. Inoculations in the greenhouse were made 21 and 31 days after seeding, and disease was rated 15 days later. All inoculated plants were reassayed on (*Setaria italica*) German strain R. In the Proso accessions, a severe mosaic was followed by death. The symptom on susceptible Pearl millets was terminal dieback. The symptom on Foxtail millet was a uniform mottling. Susceptible Proso accessions were Common and Turgahi. Both Common and German strain R Foxtail millets were susceptible. Susceptible Pearl accessions included Starr, Tift 23B, and Gahi-2. Pearl accessions Tift 239, Tiflate, Gahi-1, and Common failed to develop SAD. In these experiments, resistance to the SAD virus was apparent in Pearl millets.

Formation of perithecia by Cochliobolus heterostrophus on infected corn leaves. K. J. LEONARD (ARS, USDA, N.C. State Univ., Raleigh). The occurrence of *Cochliobolus heterostrophus*, the perfect stage of *Helminthosporium maydis*, has rarely been reported in nature. The factors which limit the development of perithecia in nature are not known. Although perithecia of *C. heterostrophus* were not found on infected corn leaves in field collections, fertile perithecia did form on field samples incubated on moist sand in the laboratory. Fertile perithecia also formed in the laboratory on detached leaves from corn plants inoculated in the greenhouse with a mixture of compatible isolates of race T. The infected leaves were collected 2 weeks after inoculation and air-dried. Leaves were incubated on moist sand for 3 to 4 weeks at room temperature. Samples were removed at 2-day intervals, dried for 3 days, and returned to the moist chambers. In another test, samples were exposed to alternate days of moist and dry conditions for the full period of perithecial development. Drying at any stage during development did not prevent later maturation of the perithecia, although drying during the first 8 days reduced the numbers of ascospores in perithecia.

Different specific toxins produced by different isolates of Helminthosporium maydis. G. D. LINDBERG (La. State Univ., Baton Rouge). A black mutant, BlspHM, was isolated from a colony of race T of *Helminthosporium maydis* and regularly sectored in colonies of race O of *H. maydis*. Conidia of races O and T sprayed on the leaves of normal cytoplasm (La-9211) and T-cytoplasm (Fla-200A) corn produced lesions readily, but BlspHM conidia failed to produce lesions on either corn line. An isolate of *H. carbonum* produced large, necrotic lesions on Pr × K61 corn, but only tiny white flecks on Fla-200A corn. Conidia of BlspHM produced only an occasional lesion on Pr × K61; reisolates (BlspHM-61) from

such rare lesions, however, produced large necrotic lesions on Pr × K61 and tiny white flecks on Fla-200A. Culture filtrates of *H. carbonum* and BlspHM-61 were about 10 times more inhibitory to the roots of Pr × K61 corn seedlings than to Fla-200A corn seedlings, whereas culture filtrates of strain T of *H. maydis* were >10 times more inhibitory to the roots of Fla-200A than to the roots of Pr × K61. Thus isolate BlspHM-61 of *H. maydis* produced a toxin with specific activity unlike that of the toxin produced by strain T of *H. maydis*. The study also indicated that the toxins can change as isolates of *H. maydis* change.

Formation of microsclerotia of Cylandrocladium in infected azalea leaves, flowers, and roots. R. G. LINDERMAN (USDA, ARS, Beltsville, Md.). Azalea leaves, flowers, and roots, infected with *Cylandrocladium scoparium*, *C. theae*, or *C. floridanum*, were cleared for examination with NaOH (leaves and roots) or methanol (flowers). No pigmented microsclerotia were observed in leaf lesions until after the leaves abscised. Saprophytic growth of each fungus in infected leaves, held at a high relative humidity following abscission, resulted in the formation on many microsclerotia by *C. scoparium* and *C. floridanum*, but relatively few by *C. theae*. Microsclerotia occurred randomly in interveinal and vascular bundle parenchyma, and were not specifically associated with stomata. Both *C. theae* and *C. floridanum* produced perithecia of their *Calonectria* stages on the leaf surface on sclerotiumlike stromata embedded in the leaf. Microsclerotia and smaller, pigmented cell aggregates formed in flower tissues infected with all 3 *Cylandrocladium* spp. Perithecia of *C. theae* and *C. floridanum* generally occurred only on the reproductive flower parts. Microsclerotia occurred in the cortex of all sizes of azalea roots infected with *C. scoparium* or *C. floridanum*, but were fewer and somewhat smaller than those in leaves. Microsclerotia were absent from roots infected with *C. theae*.

Relationship of stripe rust infection types on wheat seedlings at controlled temperatures to resistance in the field. R. F. LINE (ARS, USDA, Pullman, Wash.). Resistance of several hundred wheat selections to *Puccinia striiformis* was studied on seedlings at controlled temperatures, either programmed to gradually alternate between a low night and a high daylight temperature, or at a constant temperature, and on plants in all stages of growth at several field locations in the Pacific Northwest. The selections could be separated into four groups based on the stripe rust infection type (IT) that occurred at the various environmental conditions and on the change in infection type with duration of infection (plants with low IT are resistant; with high IT, susceptible): (i) IT remained stable throughout duration of infection at all controlled temperatures in the field; (ii) IT was affected by temperature at controlled temperature and in the field (in most cases IT was high at low temperatures and low at high temperatures); (iii) IT changed with duration of infection at controlled temperatures (initially a low IT, eventually a moderately high to high IT) but remained low in the field; (iv) IT was high at all controlled temperatures but lower in the field, especially at higher temperature and in later stages of growth. Thus, field evaluation of selection is important. Group iv can only be identified in the field. Some types of resistance can only be identified by use of several temperatures and recording of disease data several times.

Epidemiology and survival of Helminthosporium maydis race T in mixed populations of normal and Texas male sterile cytoplasm corn. R. H. LITRELL & D. R. SUMNER (Univ. Ga. Coastal Plain Exp. Sta., Tifton). Normal and Texas male sterile (TMS) cytoplasm seed of a commercial corn hybrid

were blended into six lots of 0, 20, 40, 60, 80, and 100% TMS and planted in three randomized complete blocks in 1971. Five TMS seeds were planted in the center of each plot, and when the plants were 10-36 cm high, they were dusted with corn residue naturally infested with *Helminthosporium maydis* race T. There was significantly more leaf blight in the center of the plots, and very few lesions were noted on the edges until 75 days after planting. There was a highly significant positive correlation between leaf blight ratings and percentage TMS and a highly significant negative correlation between leaf blight severity and grain yields. As the TMS increased, damaged kernels and stalk rot significantly increased and test weight significantly decreased. Residue was chopped and left on the surface in September 1971 and assayed for survival and infectivity of the fungus in November 1971 and February 1972. The fungus survived the winter in residue from all treatments. In February, however, there were significantly more total and viable conidia of *Helminthosporium* spp. on residue from 100% TMS and significantly more lesions were produced on TMS plants inoculated with residue from 100% TMS plants than from other treatments.

Strains of sugarcane mosaic virus in Puerto Rico. L. J. LIU (Agr. Exp. Sta., Univ. Puerto Rico, Río Piedras). Sugarcane mosaic virus isolates from Puerto Rican fields were identified as strains A, B, and D on the basis of symptoms induced on differential hosts; C.P. 31-294, C.P. 29291, Co. 281, Black Cheribon, and C.P. 31-588. Strain A caused little reduction in growth and sucrose content of C.P. 31-294, whereas strain D caused severe stunting and reduced sucrose content of the same variety. Strain A was most frequently collected in sugarcane fields in the Central Rufina area, where B. 34-104 was grown. Strain D was most frequently obtained from fields in the Central Aguirre area where B. 37161 was grown. Puerto Rican varieties differed greatly in their susceptibility to local mosaic virus strains. Forty per cent mosaic infection was obtained when P.R. 980 was mechanically inoculated with strain A. However, P.R. 980 did not develop symptoms when inoculated with strain B. Canes "recovered" from infection with either strain A or strain B can be reinfected by the same strains of the virus.

Sexual compatibility, morphology, physiology, and pathogenicity of Thielaviopsis paradoxa infecting sugarcane and pineapple in Puerto Rico. L. J. LIU & A. RODRIGUEZ (Agr. Exp. Sta., Univ. Puerto Rico, Río Piedras). A previously unreported race of *Thielaviopsis paradoxa* was isolated from pineapple fruits with symptoms of black rot disease. Macro- and microconidia resemble those of *T. paradoxa*, the causal agent of the pineapple disease of sugarcane. Perithecia were produced in potato-dextrose agar (24-28 C) when the pineapple isolate was crossed with the light strain of *T. paradoxa* from sugarcane. Perithecia are characteristic of *Ceratocystis paradoxa* (hornlike appendages on the base of the perithecia and long, pointed ostiolar hyphae) and are morphologically indistinguishable from those obtained by crossing two sexually compatible strains from sugarcane. However, the rate of growth of the pineapple isolate, under various temperatures and cultural media, was consistently different from those characterizing other strains of the fungus. The pineapple strain attacks sugarcane seedpieces more virulently than isolates obtained from sugarcane. This constitutes the first report on sexual compatibility between isolates of *T. paradoxa* from sugarcane and from pineapple.

Some physical properties of oat blue dwarf virus determined by membrane feeding. DONNA LONG & R. G. TIMIAN (N.D. State Univ., ARS, USDA, Fargo). Physical

properties are known for only a few plant viruses that are solely insect transmitted. Properties of oat blue dwarf virus (OBDV) were determined by the feeding of adult aster leafhoppers, *Macrostelus fascifrons*, on sap from virus infected barley, *Hordeum vulgare*, plants through membranes. OBDV transmission in wild leafhopper populations is low; thus, leafhoppers were chosen from a line selected and bred for high transmission efficiency. Sap from virus-infected plants was extracted in 0.01 M phosphate buffer pH 7.0 and used for the physical property examinations. The virus suspension was clarified by cycle centrifugation, and its activity determined after each treatment to determine physical properties. Five per cent sucrose was added to the virus suspension, and leafhoppers were allowed a 24-hr acquisition feeding through Parafilm M membranes. The virus suspension was maintained in a microcooling system at 9 C to prevent inactivation during acquisition. The dilution end point of OBDV was found to be greater than 1:256 but less than 1:512. It remained viable for 16 days in buffered plant sap at room temperature, and retained infectivity after treatment at 60 C for 10 min.

Caracterización de la roya del tallo de triticale. A. LÓPEZ, S. RAJARAM, y L. I. DE BAUER (CIMMYT y Colegio de Postgraduados, Chapingo, Mexico). En un estudio comparativo de las royas del tallo de triticale, trigo y centeno, 11 de 19 aislamientos de *Puccinia graminis* de triticale indujeron reacción de susceptibilidad en algunas líneas de triticale y en ciertas variedades de trigo y centeno. Los aislamientos restantes provocaron reacción de resistencia en triticale y centeno. En trigo, las reacciones varietales indicaron tanto resistencia como susceptibilidad. A pesar de las diferencias en habilidad patogénica, parece probable que la roya del tallo de triticale pertenece a la forma especial *tritici* ya que los aislamientos de trigo, exhibieron un comportamiento similar a los aislamientos de triticale. Los aislamientos de centeno mostraron un patrón de virulencia diferente; 9 de los 10 utilizados no produjeron reacción de susceptibilidad en las variedades de trigo ni en las líneas de triticale, pero sí indujeron reacciones diferenciales en centeno. Sólo uno de ellos se comportó como *P. graminis secalis* al ser inoculado a centeno y como *P. graminis tritici* al ser inoculado en trigo; además mostró una patogenicidad similar sobre las líneas de triticale. Este aislamiento es difícil de clasificar ya que aún cuando podría considerarse dentro de la forma *tritici*, su alta virulencia tanto en trigo como en centeno, sugiere la posibilidad de que se trate de un combinante genético entre las formas *tritici* y *secalis*.

Control of Botrytis leaf blight of onion by protective and systemic fungicides and their combinations. J. W. LORBEER (Cornell Univ., Ithaca, N.Y.). In one set of field trials under heavy natural disease pressure, chlorothalonil (Bravo), maneb (Dithane M-45), Thiabendazole ([2-(4-Thiazolyl)-benzimidazole]) (TBZ), and benomyl (Benlate) controlled *Botrytis* leaf blight of onion caused by *Botrytis squamosa* in decreasing order of efficacy as measured by lesion counts. Yield was highest for chlorothalonil, approximately equal for maneb and benomyl, and lowest for TBZ. Benomyl in combination with maneb increased both disease control, yield, and late-season leaf greenness above that achieved by all individual fungicides. Increased leaf greenness resulted from potentiated synergism, whereas increased disease control and yield resulted from supplementary synergism. In other sets of field trials under low levels of disease pressure, a combination of maneb (Manzate-D) and benomyl increased disease control over that achieved by maneb or benomyl alone. Combinations of maneb (Dithane M-45) plus TBZ or

benomyl increased disease control over that achieved by each fungicide alone. A number of fungicide combinations (2-3 components) with each material used at half normal dosage or less also effectively controlled leaf blight.

Host variation to maize dwarf mosaic virus infection. R. LOUIE, J. K. KNOKE, & W. R. FINDLEY (ARS, USDA, Ohio Agr. Res. & Development Center, Wooster). In greenhouse tests, responses of corn inbreds rub-inoculated with maize dwarf mosaic virus (MDMV) included local lesions and/or systemic infection, symptomless infection, and no infection. Inbreds Mp412, Tx601, Mo18W, Mp339, Ga209, Ga203, and Ab28A reported as resistant to a MDMV-A isolate from Mississippi were susceptible to infection to an Ohio MDMV-A isolate. Reactions of inbreds I11.A, Oh07, and others from different seed sources varied after inoculation with different strains of MDMV. Corn inbred I11.A from Illinois, Iowa, and Ohio, but not from Indiana was resistant to systemic infection by MDMV-D and MDMV-F. Inbred Oh07 from Illinois, Ohio, Mississippi, and Wisconsin, but not from Indiana was susceptible to systemic infection by MDMV-D. Variations in response of near-isogenic inbreds to infection have been useful for characterization of MDMV strains. Similarly, MDMV strains may be used to detect genetic differences in response of inbreds to virus and to differentiate among inbred populations.

Development of stem cankers in peach trees inoculated with ring nematodes, Pythium spp., and Pseudomonas syringae. B. F. LOWNSBERY, W. H. ENGLISH, E. H. MOODY, & F. J. SCHICK (Univ. Calif., Davis). In March 1970, Carolyn peach trees on Lovell root were planted in 11-liter cans of steamed sandy soil infested with (i) 35,000 ring nematodes (*Criconeimoides xenoplax*); (ii) three *Pythium* spp. (*P. debaryanum*, *P. irregulare*, and *P. ultimum*); (iii) *C. xenoplax* and *Pythium* spp.; or (iv) nothing. Eight of 16 replicates were inoculated with *P. syringae* by stem-injection in January 1971, and again by spraying leaf scars in November 1971. *C. xenoplax* increased 6 times in 1 year and 60 times in 2 years, and caused a small (13%), but significant, decrease in peach growth. In spring 1971 and spring 1972, length of cankers resulting from inoculation with *P. syringae* was not affected by soil infestation. In 1972, more cankers developed on trees growing in soil infested with *C. xenoplax* or *Pythium* than on trees in uninfested soil. In this experiment, there was little of the rapid canker development which kills trees in the field. We conclude that *C. xenoplax* or *Pythium* spp. may predispose trees to infection by *P. syringae*, but other factors limit canker development.

Control de una bacteriosis en yuca (Manihot esculenta) en Colombia. J. C. LOZANO (Centro Int. de Agr. Tropical, Cali, Colombia). Control de una bacteriosis en yuca, prevalente en varios países de Latinoamérica, se logró por poda y destrucción de la parte aérea afectada, por obtención de plantas sanas a partir de plantas afectadas y por uso de variedades resistentes. Plantaciones con cultivares resistentes, moderadamente susceptibles y susceptibles se liberaron de la afección por poda masiva de todas las plantas. La poda se hizo a una altura máxima de 40 cm arriba del suelo. Todo residuo vegetal fué sacado de las plantaciones y quemado. Mediante observaciones semanales, entresaque y eliminación de plantas reinfectadas, se aseguró la exclusión del patógeno. Aproximadamente 21 cultivares mostraron cierto grado de resistencia de cerca de 1500 inoculados por aspersión y por punción con una suspensión acuosa que contenía 1×10^9 células/ml. Ningún cultivar fué inmune, pero tres de ellos fueron altamente resistentes a la invasión foliar y a la invasión del tallo.

Bacteriosis de la yuca (Manihot esculenta) en Colombia. J. C. LOZANO y L. SEQUEIRA (Centro Int. de Agr. Tropical, Cali, Colombia; Univ. Wisc., Madison). Se llevaron a cabo estudios con 15 aislamientos de la bacteria causante de una enfermedad de la yuca caracterizada por manchas y quemazones foliares, marchitamiento, muerte descendente de las ramas y exudación de goma. El patógeno no mostró ninguna relación serológica con 15 especies pertenecientes a *Erwinia* (3), *Pseudomonas* (2) y *Xanthomonas* (10), incluyendo cinco especies no pigmentadas de este último género. Sólo hubo una reacción muy tenue cuando se usó *X. manihotis* como antígeno por el método de doble difusión en agar. Bacteriófagos y *Bdellovibrio* sp. aislados de la bacteria causante no indujeron lisis de ninguna de las bacterias anotadas arriba, incluyendo *X. manihotis*. Los aislamientos de yuca pertenecen a dos grupos serológicos claramente distinguibles por la formación de uno a dos anillos de precipitado en agar. La bacteria penetra el hospedante por los estomas y por heridas de la epidermis; una vez dentro del hospedante, se mueve a través de los tejidos vasculares. La dispersión local es principalmente por medio de la salpicadura de exudado bacteriano por acción de la lluvia. De un sitio a otro, la bacteria se disemina por medio de estacas infectadas que se usan como semilla vegetativa.

Localization of pH changes in bean hypocotyls infected with Sclerotinia sclerotiorum. R. D. LUMSDEN (ARS, USDA, Beltsville, Md.). The pH of liquids from specific areas of water-soaked hypocotyl tissue infected with *Sclerotinia sclerotiorum* was measured in microcapillary needles using aqueous bromophenol blue indicator. The pH was determined from the ratio of optical densities at 435 and 590 nm compared to standard ratios obtained from dye and buffered solutions. The pH decreased from pH 5.0, in healthy tissue and immediately adjacent to the lesion in infected tissue, to pH 4.07 (range 3.30-4.30) at the margin of the water-soaked lesion. The pH was similar in liquids drawn from increasingly older portions of the lesion, or from the advancing water-soaked margin of older lesions. The pH values of triturates from infected tissue decreased from pH 6.0 for healthy tissue to pH 5.6 and 4.3 for 1- and 2-day-old water-soaked lesions, respectively. A gradual increase occurred from pH 4.4 at day 3 to 5.8 at day 7 as the lesion developed. Changes in pH were concomitant with appearance of fungal mycelium on the host surface and the collapse of infected tissue. Differences between pH values obtained by microdetermination versus crude extraction techniques may result from disrupted superficial mycelium (pH 6.5) which could increase the pH of crude extracts.

The histochemistry of vascular browning associated with Fusarium wilt of susceptible and resistant tomato isolines. M. E. MACE & J. A. VEECH (ARS, USDA, Beltsville, Md.). The substrate for initial vascular browning in wilt-resistant or susceptible tomato isolines infected with *Fusarium oxysporum* f. sp. *lycopersici*, race 1 or 2, is localized in scattered xylem parenchyma cells. Brown products diffuse from these localized sites into surrounding xylem tissues. Phenols were detected histochemically in the xylem parenchyma cells during initial or early stages of browning. The histochemical data indicate that the major type of phenol localized at the sites of vascular browning is an *o*-dihydric phenol with an unsubstituted position para to one of the hydroxyl groups. Phenols were not detected histochemically in the healthy, nonbrowned xylem parenchyma. These observations suggest that the phenols may occur in the healthy stem xylem in conjugated forms

from which free, oxidizable phenols are released after infection. Where localized vascular browning occurred, no differences in the histochemical reactions were noted between the two tomato cultivars inoculated with race 1 or 2 of *F. oxysporum* f. sp. *lycopersici*.

Evaluation of two epidemiological models to the identification of slow stem rusting in wheat. D. R. MACKENZIE (CIMMYT, Londres 40, Mexico). The plant epidemic models of van der Plank (rate of increase) and Gregory (dispersal) were studied for practical application for use in identifying cultivars of wheat which, when subjected to epidemics of *Puccinia graminis tritici*, rust more slowly than do other cultivars. Five 14.4 × 14.4 m plots (isolated by nonsusceptible oats) of Pitic 62 (2 plots), Bonza 55 (2 plots), and Penjamo 62 (1 plot) were "point source" inoculated with a mixture of *P. graminis tritici* races on the up-wind corner. Analyses of the resulting stem rust epidemic suggested that the cultivars differed in disease dispersal patterns and in their rates of disease increase. The rate of disease increase (i.e., "r") was found to be a complex function of the distance from the point source of inoculum. These techniques may prove useful for screening new genetic material for broad-based rust resistance before multilocal international testing.

Effect of squash mosaic on microbial activity in soil around squash hypocotyls and exudation. A. MAGYAROSY & J. G. HANCOCK (Univ. Calif., Berkeley). Previous investigations suggest that the cross protection afforded squash seedlings to *Fusarium* stem rot after virus infection occurs in the inoculation phase. Penetration by *F. solani* f. sp. *cucurbitae* of hypocotyls and initial lesion development is not significantly changed by squash mosaic virus (SMV) infection. Chlamyospore germination in soil contiguous to hypocotyls is not affected by SMV infection, although higher (ca. 15%) amounts of ¹⁴C-assimilates (Photosynthetic) exude from hypocotyls. However, the soil microbial population is 7- to 8-fold higher around SMV-infected than healthy plants in soil 0-3 mm from subterranean hypocotyls. Fluctuations were observed in exudation patterns which may be related to variations in photosynthesis and translocation caused by virus infection. Translocation was often reduced during early stages of virus infection. Although photosynthesis was depressed 5 days after SMV-inoculation, it was higher or the same as healthy thereafter. The influence of these physiological changes on exudation are difficult to evaluate. Nevertheless, increased resistance to stem rot induced by virus infection may be related to increased microbial competition in the soil in the vicinity of the hypocotyl.

Degradation of bean cell walls during early stages of halo blight infections caused by Pseudomonas phaseolicola and interactions with Achromobacter sp. A. L. MAINO (Univ. Calif., Berkeley). *Pseudomonas phaseolicola* and *Achromobacter* sp. produced hemicelluloses which degraded isolated cell wall materials from *Phaseolus vulgaris* 'Red Kidney' and purified xylan preparations. Under identical conditions, β-glucosidase, β-galactosidase, β-xylosidase, and β-mannosidase activities were higher in *Achromobacter* than *P. phaseolicola*. Galactosidase activity was 42% lower in diseased than healthy tissues 12 hr after inoculation, but rose after 24 hr, terminating in a 4-fold increase over healthy tissues. β-Glucosidase activity was 27% higher in diseased tissues 7 hr after inoculation, then increased 2- to 3-fold. Xylosidase and mannosidase activities were similar in diseased and healthy tissues. Diseased tissues contained 49% less ¹⁴C-hemicellulose than healthy 48 hr after inoculation, and kinetic studies of changes in this component during

pathogenesis indicated that hemicellulose degradation occurred. Depending on the degree of infection, decreases of xylan (31-59%), araban (22-44%), glucan (64-76%), and galactan (24-40%) occurred in diseased tissues. *Achromobacter* rendered hemicellulose more susceptible to acid hydrolysis and perhaps more susceptible to enzymatic attack by the pathogen in dual infections.

Increased severity of halo blight by Achromobacter sp. A. L. MAINO & M. N. SCHROTH (Univ. Calif., Berkeley). A bacterium found frequently in cultures of *Pseudomonas phaseolicola* caused a mottled chlorosis when inoculated onto the primary leaves of *Phaseolus vulgaris* 'Red Kidney', 'Pinto', and 'Tendercrop'. Physiological and morphological characteristics indicated that this was an *Achromobacter* sp. The number of water-soaked lesions caused by *P. phaseolicola* increased two- to nearly fourfold when mixed with *Achromobacter* sp. The number of lesions increased linearly within the range of 10⁴ to 10⁶ cells/ml of *Achromobacter* added to a constant inoculum (10⁵ cells/ml) of *P. phaseolicola*. Enhancement was observed regardless of the inoculation method used: infiltration, mechanical abrasion, or spray. It occurred when *Achromobacter* was inoculated simultaneously, or 5 hr prior to inoculation with the pathogen. All *Achromobacter* strains tested and one strain of *P. syringae* enhanced infections of *P. phaseolicola*. A *Pseudomonas* soft-rot organism and a saprophyte, *P. fluorescens*, decreased the severity of symptoms when present in mixed inoculations. *P. phaseolicola* multiplied logarithmically in bean leaves following a 12- to 15-hr lag phase, whereas *P. fluorescens* declined. Populations of *Achromobacter* remained steady throughout a 144-hr test period after inoculation.

Mancha zonada del maíz en Venezuela. G. MALAGUTI (Centro de Investigaciones Agronómicas, Maracay, Venezuela). En los últimos tres años la mancha zonada, causada por *Gloeocercospora sorghi*, se ha presentado en forma epifitótica en siembras comerciales de maíz en los Estados de Aragua y Portuguesa, especialmente en siembras tempranas cuando la floración coincide con períodos frecuentes de lluvia. La enfermedad no ha sido observada hasta el momento en siembras efectuadas bajo riego en la época seca. El promedio de plantas atacadas en siembras evaluadas al azar, fue de 13 a 54%. *G. sorghi* se observa frecuentemente atacando sorgo en Venezuela, pero no había sido reportada hasta el momento en el país atacando cultivos de maíz. Morfológicamente los aislamientos de *G. sorghi* procedentes de maíz son similares a los de sorgo; sin embargo, en maíz los esclerocios son más abundantes y de mayor tamaño que en sorgo. En inoculaciones cruzadas, los aislamientos procedentes de maíz fueron más virulentos en maíz que en sorgo y los de sorgo fueron más virulentos en sorgo que en maíz. Los híbridos de maíz Obregon, Arichuna y Tunapuy aparecieron más afectados que los cultivares de maíz Sicarigua y Venezuela 1; así como algunos maíces criollos cultivados en pequeña escala. El notable incremento de la mancha zonada se ha relacionado con el aumento del área cultivada de sorgo y con el probable desarrollo de un biotipo de *G. sorghi* más virulento en maíz.

La roya causada por Phakospora gossypii en siembras comerciales de algodón en Venezuela. G. MALAGUTI y O. LOPEZ P. (Centro de Investigaciones Agronómicas Maracay, Venezuela). La roya del algodón causada por *Phakospora gossypii*, ha sido observada esporádicamente en Venezuela atacando algodón silvestre de tipo perenne, especialmente una especie aún no determinada de *Gossypium* presente en ciertos lugares altos del país. Desde el año 1969 esta

enfermedad se ha notado en campos comerciales en los Estados de Aragón y Portuguesa tanto en *G. hirsutum* como en *G. barbadense* en siembras efectuadas bajo riego en la época seca. No se ha observado hasta el momento en siembras normales efectuadas durante la época lluviosa. La fuerte incidencia de esta enfermedad ha sido asociada con factores ambientales tales como temperatura y humedad. En ensayos preliminares realizados en Maracay utilizando 30 cultivares de algodón; Pima 53, Pima 54, y Tanguis C.I.A. fueron los menos atacados.

Apple mildew on peach. B. T. MANJI (Univ. Calif., Davis). Apple mildew (*Podosphaera* sp.) inoculations were made weekly, beginning at shuck-split, on Rio Oso Gem peach fruit in an orchard interplanted with Jonathan apples. The orange-brown spots that developed were identical to the symptoms of the peach disease called rusty spot. Peaches adjacent to apples had the highest incidence of rusty spot resulting from natural infection. Fruit on trees sprayed with sulfur, triarimol (EL 273), or benomyl had 48.5, 47.8, and 16.8% rusty spot, respectively, as compared to 80.3% for the control. Both apple and rose mildew inoculations were made weekly, beginning at shuck-split, on Rio Oso Gem peaches in an orchard isolated from apples. Typical peach mildew (*Sphaerotheca pannosa*) symptoms developed in 23 of 123 rose mildew inoculations. Symptoms similar but not identical to rusty spot developed in 43 of 174 apple mildew inoculations.

Aerial photography of apple diseases. F. E. MANZER, R. C. MC CRUM, M. T. HILBORN, & G. R. COOPER (Univ. Maine, Orono). Aerial photographic techniques used successfully to detect certain potato and citrus diseases have proved in repeated attempts poorly adaptable to detection of apple diseases in Maine. Apple scab was readily detectable with infrared photography in a solid canopy of foliage from the ground. Commercial pruning practices, however, allow sunlight penetration through the foliage, producing shadows which appear similar to scab-affected foliage in aerial photographs. Other diseases such as those caused by viruses and root pathogens were not detectable by either ground or aerial photography. It is suggested that some of these latter disorders may be detectable by varying film-filter combinations and/or sun and camera angles.

Etiology of rice yellow dwarf in India. K. MARAMOROSCH, BILJANA PLAVSIC-BANJAC, V. T. JOHN, & S. P. RAYCHAUDHURI (Boyce Thompson Inst., Yonkers, N.Y., Univ. Sarajevo, Yugoslavia; All India Coordinated Rice Improvement Program, Hyderabad; & Indian Agr. Res. Inst., New Delhi). For several years, rice yellow dwarf has been diagnosed by Hyderabad and New Delhi on the basis of symptomatology and *Nephotettix impicticeps* transmission. In February 1971, samples from diseased and healthy rice plants at the above localities and at Cuttack, Orissa State, where only symptomatology was used for diagnosis, were fixed for electron microscopy. Postfixation, sectioning, staining, and electron microscopy observations were performed at Boyce Thompson Institute a few weeks later. Typical mycoplasma-like microorganisms (MLO) were detected in phloem elements of all samples from diseased plants but not from healthy ones. Seedlings infected experimentally by vectors in Hyderabad contained very few MLO as compared with the field-infected older plants from the same area and from other areas of India. No virus particles, bacteria, or fungi were found in the rice plants. The characteristic symptoms, vectors, and MLO in the phloem of diseased rice plants from widely separated areas of the Indian subcontinent suggest that the disease is similar in etiology to the rice yellow dwarf of Japan and The Philippines.

Influence of soil management practices on numbers of the root-lesion nematode, Pratylenchus penetrans, in peach orchard soils. C. F. MARKS, W. J. SAIDAK, & P. W. JOHNSON (Can. Dep. Agr., Vineland Station, Harrow, Ont.). The effects of herbicides and cover crops in peach orchards on the numbers of *Pratylenchus penetrans* in Fox sandy loam were studied at 7 and 8 years after planting. Plots treated with paraquat (1-1'-dimethyl-4-4'-bipyridinium ion) + linuron [3-(3,4-dichlorophenyl)-1-methoxy-1-methylurea] had the smallest numbers of *P. penetrans* in the soil. The paraquat + linuron treatment had no nematocidal effects on *P. penetrans* in the soil. Clean cultivation until 1 July, followed by a mowed weed cover, resulted in the largest numbers of nematodes. A creeping red fescue (*Festuca rubra*) cover retarded the increase in numbers of *P. penetrans*, but Sudan grass (*Sorghum vulgare* var. sudanense) did not. Periodic use of paraquat throughout the season resulted in nematode numbers equal to those in nonweeded plots; thus, it is important to use herbicide combinations that prevent even temporary establishment of weed cover. A nematode control program utilizing preplant nematicides, application of the proper herbicides and use of poor- or nonhost cover crops, should not only reduce replant problems but also prolong the control period of a preplant nematicide.

Effect of Thiabendazole on development of stem rot of sweet potato. W. J. MARTIN (La. State Univ., Baton Rouge). Centennial and Porto Rico sweet potato terminal cuttings were treated with Thiabendazole [2-(4-Thiazolyl)-benzimidazole] (TBZ) at different intervals after inoculation of the cuttings in a mycelial-spore suspension of *Fusarium oxysporum* f. sp. *batatas*. Stem rot development was completely arrested in the moderately resistant Centennial plants inoculated 24 hr before dipping for 1 min in a suspension of 6.7 lb. TBZ/100 gal water; and stem rot severity was greatly reduced in Centennial plants dipped in the fungicide 96 hr after inoculation. In the susceptible Porto Rico, stem rot was completely arrested in plants treated in the TBZ suspension 48 hr after inoculation, and stem rot severity was reduced in plants treated 72 hr after inoculation. TBZ completely inhibited stem rot development in Porto Rico plants treated within 1 hr after inoculation in 3.3 lb. TBZ/100 gal water. Severity of stem rot was greatly reduced in Porto Rico plants treated in 0.83 lb./100 gal water, but not in 0.41 lb./100 gal water.

Improvements in a culture medium for the growth of agallian leafhopper cell monolayers. G. MARTINEZ-LOPEZ & L. M. BLACK (Univ. Ill., Urbana). The medium described by Chiu & Black in 1967 was made less expensive with no reduction of quality by reducing the content of foetal bovine serum (FBS) to 10%, and was markedly improved by buffering at pH 6.40 with 0.05 M histidine (4.72 g L-histidine-HCl, and 4.27 g L-histidine-free base/liter). During storage in clean serum bottles, the pH of unbuffered medium increased, whereas that of buffered medium remained constant. Cell growth was adversely affected by pH values other than the optimum. A concentration of 0.005 M histidine buffered the medium at the optimum pH, but 0.05 M gave faster growth, and the appearance of cells was improved. Whether the pH of the medium was too high or too low, the insect cells changed it towards the optimum during 4 or 5 days' growth at 28 C. After a few more days at 28 C without a change of medium, the pH begins to rise and the cells begin to deteriorate. Cultures held at 13 C undergo similar changes but much more slowly. All such changes occur more rapidly in unbuffered medium. Different batches

of FBS, histidine, and lactalbumin hydrolysate, even from the same company, varied in their suitability for the medium; some were toxic. The modified medium gave improved growth of cell monolayers from three species of agallian leafhoppers in continuous culture.

Use of Malus sargentii seedlings as dwarfing rootstocks and sensitive indicators for apple stem-pitting virus. R. C. MC CRUM & M. T. HILBORN (Univ. Me., Orono). Forty seedling rootstocks of *Malus sargentii* budded with a clone of McIntosh, positive for apple stem-pitting virus (ASPV) but negative for other latent virus entities (chlorotic leaf spot, flat limb, rubbery wood, brown line, and mosaic), produced 1-year-old McIntosh whips. Thirty-nine of the 40 plants, however, died the second season after budding, and all exhibited severe stem-pitting symptoms. Extreme sensitivity to ASPV was further confirmed when eight *M. sargentii* clonal seedling selections, 40 buds of each, were double-budded with three different McIntosh clones. *M. sargentii* budded with apple cultivars Cortland, Golden Delicious, Red Delicious, and McIntosh, indexed free of the above latent viruses, continues to show promise as size-reducing seedling rootstocks for the maintenance of indexed budwood. In addition, any spread of ASPV in the mother block planting, if it occurs, would be self-eliminating.

Racial variation of Cronartium ribicola on Pinus monticola. G. I. MCDONALD & R. J. HOFF (U.S. Forest Serv., Moscow, Idaho). Two colors of needle spots, red and yellow, were found 9 months after artificial inoculation of 2-year-old, nursery-grown seedlings with field-run inoculum. Verification of the causal agent was obtained by histological examination of samples of the two spot types. The typical pseudosclerotium of the rust was present in all specimens of both types. When 2,413 seedlings were classed according to spot type, 68.5% were found to exhibit only yellow spots; 5.7%, only red spots; and 25.9%, both red and yellow spots. The mean spot frequency for each seedling class was: yellow alone, 0.40 spots/cm² of needle surface; red alone, 0.34 spots/cm²; and both, 0.69/cm². The existence of distinct seedling classes combined with evidence that the sum of the means of the 2 single-type classes (0.40 + 0.34 = 0.74) was nearly equal to the mean of the double-type class (0.69) led the authors to conclude that field-run inoculum was composed of at least two races and that pine seedlings exhibited differential resistance to the races.

Protection of Bartlett pear by avirulent Erwinia spp. and Pseudomonas tabaci. J. L. MC INTYRE & E. B. WILLIAMS (Purdue Univ., Lafayette, Ind.). Fire blight symptoms were delayed when actively growing Bartlett pear shoots were inoculated with 10⁶ cells of inducer (avirulent *Erwinia amylovora*, *E. herbicola*, or *Pseudomonas tabaci*) 24 hr before inoculation with 10⁶ cells challenge (virulent *E. amylovora*). The delay lasted at least 3 days, and in a few cases appeared permanent. Symptoms were not delayed when challenge was applied 0.5 hour after inducer. A technique was developed using etiolated pear seedlings to study protection. This technique provided large quantities of uniform, highly susceptible tissue. Symptom expression was delayed when etiolated seedlings were inoculated with 10⁶ cells of inducer either 0.5 or 24 hr before inoculation with 10⁴, 10³, or 10² cells of challenge. Delay lasted from 1 to at least 14 days, with maximum delay occurring when challenge was inoculated 24 hr after the inducer. The delay in symptoms was lengthened as the challenge concentration was decreased.

Mitosis and nuclear movement in Erysiphe graminis hordei. W. E. MC KEEN (Univ. Western Ont., London, Can.). A central body was always present at a specific site on the nuclear membrane in the interphase nucleus and was connected to chromatic spherical bodies in the nucleoplasm. A microtubular spindle oblique to the nuclear and fungal-cell axes formed within the nuclear membrane. Typical prophase, metaphase, anaphase, and telophase were observed. A narrow corridor connecting daughter nuclei for 3 to 10 min was filled mainly with microtubules. At least six chromosomes were present in each nucleus. Occasionally, the nucleus was surrounded by several membranes. Also, microtubules which originated in the spindle plaque occasionally fanned out into the cytoplasm. Long straight or undulating strands, about 0.5 - 1 μ in width and up to 40 μ in length, composed of microtubules or laminated membranes, were observed in *Erysiphe graminis* cells in which nuclei move but were not observed in conidia, where nuclei remain stationary. The end of the strand was attached to the nuclear membranes or occasionally extended past the nucleus. It is postulated that the strands, astral rays, and spindle plaques are associated with controlled nuclear movement independent of cytoplasmic streaming.

Characteristics of an electrophoretic mutant of cowpea mosaic virus. G. D. MC LEAN, J. B. DIXON, & J. S. SEMANCIK (Univ. Neb., Lincoln). Evidence has shown that cowpea mosaic virus (CPMV) exists as slow and fast electrophoretic forms. Studies of the in vivo and in vitro conversion of slow to fast forms suggest that at least seven amino acids are cleaved from the C-terminal end of the protein subunits. A nitrous acid mutant of CPMV was obtained in which the slow form electrophoresed in a way intermediate to that of the slow and fast forms of wild type in polyacrylamide gel electrophoresis. Carboxypeptidase A and B treatment of both the mutant and the wild type, followed by coelectrophoresis, showed that both were converted to the fast electrophoretic form. The mutant exhibited milder symptoms on cowpea than the wild type, and also yielded less than 50% of the yield of the wild type when both were purified under similar conditions.

Effectiveness of copper when combined with Nu-Film-17 for control of mango anthracnose. R. T. MC MILLAN, JR. (Agr. Res. Educ. Center, Univ. Fla., Homestead). Tribasic copper sulfate (3 lb./100 gal) and Kocide 101 (cupric hydroxide) (2 lb./100 gal) were compared alone and in combination with Nu-Film-17 (Di-1-p-Menthene) (1 pint/100 gal) and Triton B-1956 (modified phthalic glycerol alkyl resin) (2 oz/100 gal) for control of anthracnose (*Colletotrichum gloeosporioides*) on Irwin mango. Sprays were applied at 300 psi with a hand-operated gun. Sprays were started when panicles were 2 inches long (March 1970) and applied weekly until fruit set, after which sprays were applied monthly until harvest for a total of 12 applications. Fruits were harvested in July 1970 and rated anthracnose-free, mild, or severe. All fungicidal treatments gave good control. Kocide 101 alone or combined with Nu-Film-17 or Triton B-1956 were significantly superior to Tribasic copper sulfate alone or combined with Triton B-1956. No treatment significantly affected total yield, but all Kocide 101 sprays and Tribasic copper sulfate plus Nu-Film-17 provided significantly more marketable fruit (disease free plus mild anthracnose) than other treatments.

Fatty acids and sterols of some mycorrhizal fungi. J. H. MELHUISE, JR., G. A. BEAN, & E. HACSKAYLO (Univ. Md., College Park, USDA Forest Serv., Beltsville, Md.). The sterols and fatty acids of three mycorrhizal fungi, *Amanita*

rubescens, *Pisolithus tinctorius*, and *Hebeloma sarcophyllum*, were investigated. Hyphae were transferred from stock cultures to petri dishes containing ammonium tartrate nutrient agar containing, per liter: 1.0 g KH_2PO_4 , 0.5 g $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$, 5.0 g NH_4 tartrate, 0.5 ml Zn SO_4 (1:500 aqueous solution), 50 mg thiamin, 20.0 g glucose, 0.5 ml of a 1% solution of ferric citrate, and 15 g agar. After 4 to 6 weeks, plugs were cut from the edges of the colonies and transferred to fresh NH_4 tartrate agar medium. After the hyphal fans on the edges of the plugs developed sufficiently to enable the plugs to float, they were transferred to NH_4 tartrate liquid medium. The mycelium was collected and lyophilized, and the lipids were extracted with chloroform/methanol after 1 month of growth on the liquid medium. Fatty acids were identified by gas liquid chromatography (GLC) and thin-layer chromatography; sterols were tentatively identified by GLC. The major fatty acids were 16:0 and 18:2, with lesser amounts of 18:0 and 18:1; trace amounts of 14:0, 15:0, 16:1, 16:2, and 17:0 were also detected. The three fungi could be distinguished from one another based on fatty acid patterns. The major sterol in the fungi was ergosterol.

Occurrence and significance of soft-rotting bacteria in healthy vegetables. J. C. MENELEY & M. E. STANGHELLINI (Univ. Ariz., Tucson). Healthy vegetables have long been known to harbor a mixed, internally borne bacterial flora. However, the significance of this flora has not been elucidated. Bacteria were isolated from the internal tissues of healthy, surface-sterilized potatoes (68%) and cucumbers (78%). Soft-rotting bacteria, identified by their ability to hydrolyze sodium polypectate and macerate uniformly cut potato or cucumber slices, were isolated from 17% of the potatoes and 56% of the cucumbers. The soft-rotting bacteria from cucumbers belong to the following genera in decreasing order of occurrence: *Pseudomonas* spp., *Erwinia* spp., *Bacillus* spp., and *Xanthomonas* spp. Activation of the bacteria, resulting in the production of a bacterial soft-rot, occurred only after high-temperature (37 C) incubation of the cucumbers or inoculation with *Pythium aphanidermatum*. Soft-rotting bacteria were also isolated from the internal tissue of healthy carrots, celery, string beans, cauliflower, bell peppers, tomatoes, broccoli, cabbage, and onions. The activation of the resident soft-rotting bacterial flora within healthy vegetable tissue, either acting alone or in conjunction with pathogenic fungi, may account for many of the storage rots previously attributed to external bacterial contamination.

Genetic relationship of resistance to Heterodera solanacearum in dark-fired and burley tobacco. L. I. MILLER, J. A. FOX, & L. SPASOFF (Va. Polytech. Inst. & State Univ., Blacksburg). Inheritance of resistance to *Heterodera solanacearum* was studied in the following reciprocal crosses of tobacco: DVA 606 (D) (a resistant dark-fired breeding line) \times Hicks (H) (a susceptible flue-cured variety); BVA 523 (B) (a multigenic resistant burley breeding line) \times H, and D \times B. An isolate of the nematode from Scott's Fork, Va., was tested to determine its ability to develop egg-bearing females on 10 plants of each parent and 20 plants of each F_1 . One hundred cysts containing 130-196 eggs/cyst were introduced into cyst-free soil in 15-cm pots. A single 2-month-old seedling was transplanted to each pot and grown at air temperatures of 23-27 C. After 5 weeks, the soil was screened and the means and ranges of the females counted were: D, 5.3 (2-7); B, 7.5 (3-15); H, 290.0 (195-435); D \times H, 119.4 (25-255); B \times H, 157.7 (0-265); D \times B, 12.1 (0-44). Counts of the reciprocal crosses were combined because no differences in the

resistance reaction were noted within a combination of parents. The resistant reaction of the D \times B hybrids as compared to the intermediate reaction of D \times H and B \times H hybrids, and the continuous range from resistant to susceptible reactions of the F_2 (0-535, $n=530$) of the D \times H cross, suggests that resistance in DVA 606 is also multigenic.

A selective medium for isolating and identifying Erwinias. T. D. MILLER (Ohio Agr. Res. Development Center, Wooster). A medium selective for *Erwinias* (MSE) was prepared by adding the following ingredients serially: 970 ml distilled H_2O ; mannitol, 10 g; nicotinic acid, 0.5 g; L-asparagine, 3 g; K_2HPO_4 , 2 g; $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$, 0.2 g; Na taurocholate (Difco), 2.5 g; Tergitol 7 (Na heptadecyl sulfate), 0.1 ml; nitriolotriacetic acid, 0.2 g (as a 2% aqueous solution neutralized with 0.73 g KOH/g NTA); bromothymol blue, 45 mg (as a 0.5% aqueous solution); neutral red 12.5 mg (as a 0.5% aqueous solution); agar, 20 g. The pH was adjusted to 7.3 with NaOH. After autoclaving cycloheximide, 50 mg (as 1% aqueous solution) and thallium nitrate (K&K), 17.5 mg (as a 1% aqueous solution) were added. Orange-colored colonies are indicative of the *Erwinia* genus. Colony inspection with a dissecting microscope further identified *Erwinia* "groups" or species. Greatest differentiation was evident when the periphery of transmitted parallel light is directed on the colonies. The percentage recovery of 12 *Erwinia* spp. (34 isolates) on MSE and on nutrient glucose media were similar. Only 7% of 200 isolates (79 species from *Pseudomonas*, *Xanthomonas*, *Corynebacterium*, and *Agrobacterium*) tested on MSE produced significant growth. In contrast to the orange *Erwinias*, these colonies were green.

Phenolase and ascorbic acid oxidase activities in apple leaves after grafting with healthy and virus infected Golden Delicious buds. D. F. MILLIKAN & MARIAN SANIEWSKI (Univ. Mo., Columbia). Two virus-sensitive cultivars, Spy 227 and Russian, and a tolerant one, MM 104, were chip-budded with buds from a healthy (4-4) and a virus-infected (E-1) clone of Golden Delicious. Fully differentiated tip leaves were collected at 3-week intervals after bud break until the cessation of growth. Three-g samples were macerated 0.1 M Phosphate buffer, pH 7.0, and free phenols eliminated by 48-hr dialysis against a large volume of buffer. Phenolase activity in leaf extracts was measured with an oxygen electrode. Activity was highest in Spy-227 and lowest in MM-104 leaves. Three weeks after budding with buds of E-1, the phenolase activity was reduced in the sensitive cultivars but not in the tolerant one. Three weeks later, Spy 227 and Russian had increased phenolase activity, whereas those budded with 4-4 had reduced activity. In the third and final collection, all three cultivars had increased activity as a result of budding with E-1. Ascorbic acid oxidase was relatively unaffected in the three cultivars budded with virus-free buds, but was increased in all the varieties budded with E-1.

Mycotoxins from Stachybotrys alternans grown on oats. C. J. MIROCHA, M. PALYUSIK, S. PATHRE, & BETH SCHAUERHAMER (Univ. Minn., St. Paul). *Stachybotrys alternans* colonizes substrates rich in cellulose such as hay or straw of wheat, barley, and oats. Infected straw eaten by animals causes a condition called stachybotryotoxicosis characterized by cutaneous and mucosal necrosis, leukopenia, agranulocytosis, and arrest of systolic contractions in the heart. Aerosols of toxic substrates also affect man. The disease caused by this fungus is a classic in mycotoxicology

and was extremely important in the Soviet Union. Two major components were extracted from toxic oats colonized by *S. alternans* and purified by thin-layer chromatography. A major component (coded b and 3 by us) has a mass of 386 ($C_{23}H_{30}O_5$) whereas the second has a mass of 486 ($C_{27}H_{34}O_8$) as determined by mass spectroscopy. Further analyses suggest that these components contain a steroid nucleus, an unsaturated lactone at C-17, two phenolic hydroxyl groups, and an aldehyde. Both components are readily separated by gas-liquid chromatography as the trimethylsilyl ether derivatives. These toxic metabolites closely resemble the toxins originally described in the early Russian literature.

Cell wall-degrading extracellular enzymes produced by Xanthomonas oryzae and X. translucens f. sp. oryzae. S. K. MOHAN & A. NOVACKY (Univ. Mo., Columbia). A comparison of the extracellular enzyme activities of the two bacterial pathogens of rice revealed that *Xanthomonas oryzae* produced pectin methylesterase, pectin methyl-trans-eliminase, polygalacturonase, and cellulase (Cx), but no protease in vitro, whereas culture filtrates of *X. translucens f. sp. oryzae* showed neither pectolytic nor cellulolytic activity, but did show considerable proteolytic activity. *X. oryzae* is a vascular pathogen causing leaf blight and/or wilt, and *X. translucens f. sp. oryzae* attacks the parenchyma causing leaf streak. Differences in their ability to elaborate various cell wall-degrading enzymes in vitro are interpreted as reflections of their different mechanisms of pathogenesis.

Fireblight resistance to streptomycin in California. W. J. MOLLER, J. A. BEUTEL, W. O. REIL, & B. G. ZOLLER (Univ. Calif., Davis). During May 1971, fireblight (*Erwinia amylovora*) became epidemic in a number of Sacramento Valley pear orchards despite the use of a streptomycin program which had been effective for years. The prevalent strain of the bacterium was resistant to 200 ppm streptomycin, and the population of sensitive strains was at an extremely low level. The resistant strain progressively adapted to 1,000 ppm. Previous history of streptomycin usage was unrelated to the outbreak; in some affected orchards the chemical had been used for less than 2 years. The streptomycin-resistant strain exhibited greater virulence than the normal strain in controlled inoculations in the field. Holdover cankers tested during the remainder of 1971 and in the spring of 1972 revealed that the resistance was stable. All evidence points to overwintering in holdover cankers and not as resistant epiphytic microflora. The organism readily multiplies as an epiphyte in healthy blossoms, with occasional flowers becoming diseased.

Bacteriosis foliar en girasol (Helianthus annuus). J. MONTAÑO, E. OLIVAS, L. FUCIKOVSKY (Colegio de Postgraduados, Chapingo, México). En 1970 se observó una enfermedad en el girasol cultivado en Chapingo. Los síntomas aparecieron en forma de manchas necróticas angulares que llegaron a destruir hasta un 60% de la lámina foliar. El agente causal fue aislado e inoculado en 5 variedades de girasol. Posteriormente fue identificado como una especie de *Pseudomonas* fluorescente. En plantas fertilizadas y vigorosas las lesiones aparecen necróticas, irregulares y angulares; mientras que en plantas mal nutridas las lesiones necróticas presentan un halo clorótico. Las observaciones indicaron que

la bacteria puede penetrar por los estomas. Pero también se han observado lesiones sistémicas a lo largo del tejido vascular de la hoja, producidas por la misma bacteria, la cuál es introducida al tejido vascular por la larva minadora de un díptero del Suborden Brachycera, Familia Clusidae. La mosca deposita sus huevos en la hoja, y la larva al emerger, se alimenta de los tejidos, haciendo galerías alrededor de las venas hasta que pupa en el envés. En tanto que las galerías se necrosan a causa de la bacteria. Dado que la bacteria fue aislada del interior de la larva, se concluye que la mosca actúa como vector.

Specificity of in vitro reconstitution in tobacco rattle virus. T. J. MORRIS & J. S. SEMANCIK (Univ. Neb., Lincoln). Nucleoprotein particles were assembled in vitro from acetic acid-extracted protein and phenol-extracted RNA. Optimal conditions for assembly involved dialysis of a protein and RNA mixture (5:1) against 0.25 M Glycine-OH buffer, pH 8.0, at 9 C. Reconstituted nucleoprotein particles and indeterminate length protein tubes composed of stacked disc structures were detected at 2 hr, and were complete after 12 hr of incubation. Maximum reconstitution efficiencies of nucleoprotein particles approached 60%. Reconstituted nucleoprotein and native virus sedimented at the same rate after sucrose density-gradient centrifugation. Samples isolated from the density gradient were infectious after pancreatic ribonuclease treatment. Encapsulation of short rod RNA, which contains the coat protein message, was favored; however, long rod assembly was enhanced in higher molarity glycine-OH buffer. The specificity of reconstitution as related to the intrinsic stability of the functionally-distinct RNA's of this multiple component virus will be discussed.

Ultrastructure of phenol-containing cells in banana and tomato. W. C. MUELLER & C. H. BECKMAN (Univ. Rhode Island, Kingston). Phenols are manufactured and stored in specialized cells in a large number of plants, but the function of these cells and their possible contribution to disease resistance are speculative. The phenol-containing cells of banana and tomato were examined by electron microscopy. In banana, phenol-containing cells in the vascular cylinder of the root are characterized by a dense amorphous inclusion of phenol in the vacuole. Mature cells have a limited cytoplasm, whereas immature cells possess a dense and apparently active cytoplasm with a well-developed network of endoplasmic reticulum. In tomato, phenol is present as amorphous deposits in the vacuoles in cells of capitate hairs. There is an inverse correlation between the amount of phenol and the amount of starch in the leucoplasts of these cells: as the cells mature, the amount of starch decreases and the amount of phenol increases. Cells similar to those found in banana are also found in the vascular system of tomato after infection with *Fusarium oxysporum f. lycopersici*.

Peroxidase activity and phenol content in Wando peas infected with two populations of Ditylenchus dipsaci. B. D. MUSE & R. R. MUSE (Kent State Univ., Stark Regional Campus, Canton, Ohio; Ohio Agr. Res. Development Center, Wooster). It has been reported that the Raleigh, N.C. (RNC), population of *Ditylenchus dipsaci* causes conspicuous gall formation on pea seedling shoots (*Pisum sativum* 'Wando'), whereas the Waynesville, N.C. (WNC), population induces a

necrotic reaction. Peroxidase activities, total phenols, and ortho-dihydroxyphenols were studied in RNC-infected, WNC-infected, and control (noninoculated) pea seedlings. Seedlings were harvested 24, 48, 72, and 120 hr after inoculation with suspensions of nematodes. We assayed spectrophotometrically peroxidase activity by following the oxidation of pyrogallol. At 120 hr after inoculation, the level of peroxidase activity in RNC-infected tissue was 3 times greater than the control. Peroxidase activity in WNC-infected tissue, however, was 30 times greater than the control. Successively higher total phenol and ortho-dihydroxyphenol contents were found at each harvest. Total phenol and ortho-dihydroxyphenol contents were similar in control and RNC-infected tissues, but were reduced in WNC-infected tissues. This study indicates that high peroxidase activity and decreased total phenol and ortho-dihydroxyphenol contents accompanied the necrotic reaction in WNC-infected tissue, but their roles have not been determined.

Influence of nutrition on the development of Helminthosporium red leaf spot on Seaside bentgrass. R. R. MUSE (Ohio Agr. Res. Development Center, Wooster). Seaside bentgrass was grown in silica sand and subjected to different nutritional regimes by modifying standard Hoagland's solution (1.0 H). Thirty-five-day-old plants were inoculated with an aqueous spore suspension of *Helminthosporium erythrospilum*. Disease severity ratings were based on the percentage of blighted plants per container. Disease severity for bentgrass plants grown under 0.1 H and low nitrogen was significantly lower than controls (1.0 H). Plants grown under 3.0 H, high nitrogen, low and high phosphorus, low potassium, or low calcium nutrition were more prone to foliar blighting than were controls. Foliar analyses were made prior to inoculation for N, P, K, Ca, Mg, reducing and total carbohydrates, total phenols, ortho-dihydroxyphenols, and tannins. There was a significant correlation (1% level) between nitrogen content and blighting. Significant inverse correlations (1% level) were found between disease severity and foliar content of reducing and total carbohydrates, total phenols, ortho-dihydroxyphenols, and tannins. This study indicates, that of the nutrient elements studied, nitrogen appears to exert the most influence on disease severity.

Increased susceptibility of dicofol-treated cotton to Verticillium albo-atrum. H. W. MUSSELL & ANGELA RAMFTL (Boyce Thompson Inst., Yonkers, N.Y.). The acaricide, dicofol (Kelthane EC 18.5), when applied to greenhouse-grown cotton plants 4 and 6 weeks after germination, shortened the time to first symptom expression and increased the severity of symptoms after inoculation with *Verticillium albo-atrum*. In the *Gossypium hirsutum*/severe pathotype combination, this resulted in earlier defoliation and tip necrosis, whereas in the *G. hirsutum*/mild pathotype combination, more extensive stunting and leaf damage were observed. Resistant *G. barbadense* types did not usually develop symptoms when inoculated with mild pathotypes, but slight stunting and mature leaf damage occurred when the plants had been sprayed with dicofol. *Verticillium*-induced defoliation of *G. barbadense* types was not accelerated by treatment with dicofol, but stunting and damage to immature leaves was

more pronounced in dicofol-treated plants. Leaves from *G. hirsutum* plants which had been treated with dicofol exhibited a more pronounced sensitivity to damage induced by an endopolygalacturonase produced by *V. albo-atrum*.

Events leading to the expressing of the hypersensitive reaction of Malus to Venturia inaequalis. R. L. NICHOLSON & J. KUC (Purdue Univ., Lafayette, Ind.). Etiolated hypersensitive and susceptible apple and etiolated bean (*Phaseolus vulgaris*) seedlings were simultaneously inoculated with single isolates of *Venturia inaequalis*. Germination and appressorium formation occurred at the same rate on each host. Penetration pegs (easily observed on bean) either occurred before or to a greater extent than primary hyphae on apple. Within a 24-hr period after inoculation, primary hyphae were consistently observed on susceptible apple. On hypersensitive apple, primary hyphae were either absent, present and appearing at the same rate as on susceptible apple, or present and appearing at a slower rate than on susceptible apple. If penetration occurred at the same rate on each host, then primary hyphae should have formed at the same rate on both apple hosts. That this did not occur suggests that, in the hypersensitive interaction, fungal growth is inhibited at, or close to, the time of penetration. Phenolic oxidation, known to result in browning characteristic of the hypersensitive reaction, has been suggested as responsible for containment of the fungus. However, for each isolate studied, cellular browning of the host was not observed until late in the host-parasite interaction. This suggests that fungal containment is not initiated by phenolic oxidation.

Resistance of selected alfalfa clones to the root knot nematode, Meloidogyne incognita. E. L. NIGH, JR. (Univ. Ariz., Tucson). Two clones of *Medicago sativa* 'Mesa Sirsa' were selected for resistance and susceptibility to infection by *Meloidogyne incognita*. Approximately 80% more larvae penetrated the susceptible clone (ED-7) than the resistant clone (ED-9). Infection and larval development in ED-9 was inversely proportional to plant age; the susceptible clone was uniformly attacked from germination to 12 months of age by approximately the same number of larvae. Temperatures ranging from 22 to 37 C did not influence resistance or susceptibility of the clones. Grafting scions of ED-9 to ED-7 roots or the reciprocal did not influence host susceptibility. Larvae developed normally if successful in penetrating ED-9 roots. Syncytia developed in both clones with no visual difference in size. Hypersensitive responses were observed only in the resistant clone (ED-9) beginning 7 days after infection as evidenced by degeneration. These reactions, progressively more apparent 10-17 days after infection, resulted in death of the nematode before maturation.

Prevention of the bacterially induced hypersensitive reaction by low concentration of living incompatible bacteria. A. NOVACKY, G. ACEDO, & R. N. GOODMAN (Univ. Mo., Columbia). Rapid development of the hypersensitive reaction (HR) occurs in tobacco leaves after infiltration with 10^6 - 5×10^6 cells/ml of the incompatible species, *Pseudomonas pisi*. Although several procedures have been described which prevent the development of HR, low numbers of living, incompatible bacteria have not been previously reported to prevent subsequent development of HR when treated tobacco leaves are challenged with

HR-inducing populations of incompatible bacteria. When 5×10^5 cells/ml of *P. pisi* were infiltrated into the tissue, the bacteria multiplied to 2×10^6 cells/ml after 24 hr. HR did not occur in these tissues, nor did it occur after challenging with higher concentrations of the same species (5×10^6 - 10^7 cells/ml). This protective effect was light-dependent, and occurred as early as 3 hr after first inoculation.

Ground and aircraft applications of thiophanate-methyl on control of stone fruit brown rot blossom blight. J. M. OGAWA, W. E. YATES, B. T. MANJI, & R. E. COWDEN (Univ. Calif., Davis). Systemicity and high fungicidal activity of 1,2-bis(3-methoxycarbonyl-2-thioureido) benzene (70% thiophanate-methyl; Topsin M) make possible low dosage and low gallonage applications for protection from *Monilinia blossom blight*. Two pounds of Topsin M applied in 400 gal of water/acre before the showing of anthers reduced *Monilinia fructicola* blossom blight on Dixon peach from 21 to 3.2%; and number of blossoms blighted by *M. laxa* on Fay Elberta peach, from 14 to 1/tree. A 30-acre Blenheim apricot orchard, with blossoms varying from red bud to 50% open, was used to compare ground airstream and fixed-wing aircraft applications of Topsin M (2 lb./acre) for control of *M. laxa* blossom blight. The aircraft plot using 9.2 gal/acre had 9.5% blossom blight and the ground plot using 400 gal/acre 2.4%, whereas the control had 31%. No significant difference was shown other than from the control. Aircraft foam (Accutrol, Velsicol Chem. Corp.) produced with special nozzles did not significantly increase the effectiveness of blossom blight control (9.0% blight) from the conventional aircraft spray application. Spray distribution was assessed by hanging double microscope slides horizontally on trees and using $MnSO_4$ in the spray mix as an indicator.

Correlation of incidence of Trichoderma and Armillaria for 29 days following treatment of Armillaria-infected roots with methyl bromide. H. D. OHR & D. E. MUNNECKE (Univ. Calif., Riverside). Succession of *Armillaria* and *Trichoderma* in roots naturally or artificially infected with *Armillaria* was determined after methyl bromide (MB) fumigation. Citrus roots 2.5×7.6 cm were exposed for 3 hr to flowing air (control) or to a mixture of MB in air (35 ml MB/1 liter air). All *Armillaria* propagules tested were viable immediately after fumigation. The fumigated roots were buried in a sandy loam soil, and sampled to determine viability of *Armillaria* and subsequent colonization by *Trichoderma*. Root pieces were plated on selective media daily for 8 days, then at 3-day intervals for 21 days. There was a positive correlation between the decrease of *Armillaria* and the increase of *Trichoderma*, as determined by the frequency of isolations of the fungi. As isolation of *Armillaria* from infected roots declined to near zero, *Trichoderma* also decreased, perhaps indicating a decrease of substrate for *Trichoderma*. In one experiment, *Armillaria* infection remained low; conversely, *Trichoderma* did not decline. The incidence of *Fusarium* spp. or bacteria also increased during the incubation, but no correlation was found between them and *Armillaria*. This is the first evidence that shows a direct correlation between the decline of *Armillaria* and the increase of *Trichoderma*.

Natural occurrence of Fusarium moniliforme in corn kernels with opaque endosperm. J. J. OOKA, T. KOMMEDAHL, & R. E. STUCKER (Univ. Minn., St. Paul). Seed lots of 7-9 near-isogenic hybrids (O_2 versus normal endosperm) of corn (*Zea mays*) were compared for infected kernels in the 1969 and 1971 crop seasons. *Fusarium moniliforme* was the predominant *Fusarium* sp. isolated when surface-treated kernels (1% NaOCl for 1 min) were cut

in halves, placed on PCNB-peptone agar, and incubated at 24 C. Thirty-two seed lots in 1971 yielded 34% infected kernels, but 18 seed lots of the same hybrids assayed in 1969 yielded only 10% infected kernels. In 1969, there were five of nine pairs in which the incidence of infected kernels in the O_2 hybrid was more than twice that in the normal hybrid of each pair. In 1971, the incidence of infected kernels of the same hybrids was higher in only three of seven O_2 hybrids (average 60%) when corn was grown on low nitrogen (N) plots and higher in only two of seven (average 38%) in the high N plots. Thus, kernel infection by *F. moniliforme* and O_2 endosperm may be associated with certain hybrids only, and the incidence might be altered by N application to soil. Counts of viable propagules of *F. moniliforme* in air oven corn fields, using the Andersen Air Sampler, suggested that ears might have become infected from air-borne inoculum and not necessarily from the planting of infected kernels.

Purification and characterization of a virus causing a latent infection in Gynura aurantiaca. F. M. OSMAN, L. G. WEATHERS, & D. J. GUMPF (Univ. Calif., Riverside). A virus causing a latent infection in *Gynura aurantiaca* was purified by differential ultracentrifugation. Infected leaves were extracted in 0.5 M pH 9.0 orthoborate containing 0.02 M sodium sulfite, expressed through two layers of cheese cloth and centrifuged for 10 min at 3,000 g. The supernatant fluid was centrifuged for 2.5 hr at 23,000 rpm in a Spinco No. 30 rotor. The drained pellet was resuspended in 0.05 M pH 9.0 orthoborate buffer, then centrifuged for 10 min at 3,000 g. The supernatant fluid was centrifuged for 1 hr at 36,000 rpm in the Spinco No. 40 rotor, and the final pellet resuspended in 0.05 M orthoborate buffer. Electron micrographs revealed flexuous, rodlike particles 20 nm in width having a mean length of 700 nm. A prominent central core was evident in negatively stained preparations. High yields of particles were obtained from *Gynura* plants concurrently infected with citrus exocortis virus. Crude extracts as well as purified virus produced necrotic lesions when inoculated to leaves of petunia. Thermal inactivation point of the virus was 70-75 C; longevity in vitro was 5 days; and dilution end point, 1:10,000. Frozen crude extract from infected plants remained infective up to 8 months.

Chemotactic migration and diauxic growth patterns in Pseudomonas phaseolicola and effect of 3':5'-cyclic AMP. N. J. PANOPOULOS & M. N. SCHROTH (Univ. Calif., Berkeley). *Pseudomonas phaseolicola* exhibited chemotaxis towards a variety of carbohydrates including arabinose, glucose, K^+ -gluconate, glycerol, fructose, mannose, and organic acids such as pyruvate, citrate, and succinate. Formation of migrating rings ("ringing out") in semisolid plates (0.2% agar) containing ca. 1 mM arabinose, glucose, fructose, mannose, or pyruvate occurred 1 to 4 hr earlier in the presence of ca. 5 mM 3':5'-cAMP. Typical diauxic growth patterns were obtained when the bacterium was grown on combinations of single carbon sources, such as glucose plus arabinose, K^+ -gluconate plus glucose, or succinate (or citrate) plus glucose (or arabinose), the preference patterns being glucose over arabinose, K^+ -gluconate over glucose, and the TCA acids over the sugars. In the case of glucose-arabinose diauxie, addition of 5 to 8 mM cAMP abolished the diauxic lag, suggesting that the enzymes for arabinose utilization were subject to catabolite repression.

Enzymatic responses of soybean hypocotyls to wounding and inoculation with Phytophthora megasperma var. sojae. J. E. PARTRIDGE & N. T. KEEN (Univ. Calif., Riverside). Phenylalanine ammonia-lyase (PAL), chalcone-flavanone isomerase (CFI) and peroxidase (PO) were presumed by us to

be involved in the biosynthesis by soybeans of the induced antifungal compound, 6a-hydroxyphaseollin (HP). Six-day-old hypocotyls of susceptible Harosoy (H) and near-isogenic resistant Harosoy 63 (H63) plants were assayed at various times after wounding and/or inoculation with *Phytophthora megasperma* var. *sojae*. The specific activities of all three enzymes increased markedly by 6 hr after treatment, and attained specific activities 30-125 times that of uninjured hypocotyls (basal level) at 12 to 15 hr after treatment. The specific activities then decreased rapidly. PAL returned to its basal level in 30 to 36 hr, but CFI and PO specific activities remained 10- to 50-fold higher than the basal level to 48 hr. Wounded tissues of H and H63 responded similarly, except that enzyme activities in inoculated H63 increased more rapidly than in H. Control of HP biosynthesis and expression of the H63 resistance gene (Rps) does not appear to involve activation of these enzymes because the enzymes responded similarly in wounded or inoculated H and H63 plants; they increased well before the appearance of HP at 10 to 12 hr; and they decreased during rapid HP biosynthesis.

Purification of the phytotoxin from Pseudomonas phaseolicola. S. S. PATIL (Univ. Hawaii, Honolulu). DEAE cellulose gradient chromatography resolved the partially purified *Pseudomonas phaseolicola* toxin into two distinct anionic species. Physicochemical and biological properties of the two species were similar except for the charge. Sephadex gel chromatography of the toxin samples which contained various amounts of toxin and NaCl indicated that species a, but not b, was capable of reversible molecular association. Species b was hydrolyzed in 6 N HCl, and the resulting amino acids were converted to N-trifluoroacetyl n-butyl esters. The GLC of derivatives showed that the toxin contained three known and two unknown amino acid residues. One of the known amino acids labeled uniformly with ^{14}C was provided to a *P. phaseolicola* culture. When the culture filtrate was purified and fractionated using gel chromatography, complete correspondence between labeling and biological activity was found. Both species of the toxin are capable of incorporating the ^{14}C label.

Deoxyribonucleic acid homologies among phytopathogenic and saprophytic fluorescent pseudomonads. P. C. PECKNOLD & R. G. GROGAN (Univ. Calif., Davis). Genetic relatedness among 53 strains of fluorescent pseudomonads comprising representative isolates of 18 plant-pathogenic nomenclatures and two saprophytic fluorescent species was assessed by means of DNA-DNA hybridization. Most phytopathogenic nomenclatures used as references had interstrain DNA homology values of >90%, indicating homogeneous DNA clusters within a nomenclature. With *Pseudomonas syringae* and *P. mors-prunorum* as references, all plant-pathogenic pseudomonads tested (except *P. cichorii*, *P. viridiflava*, and *P. marginalis*) fell into one genetically distinct group (>50% homology). Three subgroups were recognized within this group and are provisionally referred to as "syringae", "mors-prunorum", and "tomato". *P. cichorii* was distinct from the other phytopathogens. It showed a closer relationship to the phytopathogenic than to the saprophytic pseudomonads; however, it was more closely related to the saprophytes than were other phytopathogens except *P. marginalis*. *P. viridiflava* was most closely related to cytochrome oxidase-negative pseudomonads isolated from plant roots, and appeared to be a heterogeneous nomenclature distinct from the other phytopathogens. *P. marginalis* had homology values similar to those of the saprophytes.

Histopathology of carnation infected with Fusarium oxysporum f. sp. dianthi. BARBARA W. PENNYPACKER & P. E. NELSON (Pa. State Univ., University Park). Carnations, cultivar Improved White Sim, were inoculated with three isolates of *Fusarium oxysporum* f. sp. *dianthi* and examined histologically to determine the anatomical effects of the pathogen on the host and to determine if there was any anatomical basis for the success of culture indexing as a control measure for Fusarium wilt of carnation. *Fusarium oxysporum* f. sp. *dianthi* isolates from Pennsylvania (A-31), California (A-15), and Denmark (A-80) were used. Isolates A-31 and A-15 caused similar anatomical responses in carnation stems. Histological examinations of infected carnation stems revealed vascular plugging, hypertrophy and hyperplasia of xylem parenchyma cells, xylem parenchyma cell disintegration, and the formation of vascular cavities. Isolate A-80 incited more cell proliferation in the xylem parenchyma, and less vascular cavity formation. No tyloses were seen in the xylem vessel elements of infected carnations regardless of the isolate used in inoculation. No conidia were observed in advance of the mycelium in xylem vessel elements. The absence of conidia in advance of mycelium in the xylem vessel elements is the primary reason for the success of culture indexing as a control measure for Fusarium wilt of carnation.

Influence of photoperiod on brown stem rot of soybean. D. V. PHILLIPS (Univ. Ga. Exp. Sta., Experiment). Plants of Lee soybean (*Glycine max*) of different ages and developmental stages were obtained by a varying of planting dates and length of photoperiod. Plants were inoculated with *Cephalosporium gregatum*, and symptom ratings based on the length of internal stem browning were made 28 days later. Brown stem rot symptom ratings were lower in young than in old vegetative plants. After floral induction, symptom ratings were similar regardless of age. An increase in symptom rating occurred when young plants were changed from a long-day to a short-day photoperiod. Plants exposed to a short-day photoperiod with a light-interrupted dark period responded similarly to those exposed to a long-day photoperiod.

Acquisition factor required for aphid transmission of purified cauliflower mosaic virus. T. P. PIRONE & M. C. Y. LUNG (Univ. Ky., Lexington). Aphids (*Myzus persicae*), which have probed or fed on healthy mustard leaves and which subsequently probe through Parafilm membranes into suspensions of purified cauliflower mosaic virus (CIMV), are unable to transmit the virus. Aphids which first probe leaves infected with CIMV, then probe into purified virus, are able to transmit the purified virus; they are, however, unable to transmit the purified CIMV when the sequence of probing is reversed (i.e., purified virus, then infected leaves, then test plants). The origin of the virus being transmitted is determined by the use of purified virus of a strain different than that with which the leaves are infected. The data suggest that aphids acquire, from infected leaves, a factor which allows them to acquire and subsequently transmit purified CIMV.

Reaction of Venturia inaequalis ascospore progeny to dodine. F. J. POLACH (N.Y. State Agr. Exp. Sta., Geneva). Isolates of *Venturia inaequalis* from New York apple orchards where dodine did not control scab were compared in vitro with isolates from orchards in which dodine performed satisfactorily. Single-spore isolates from orchards where dodine controlled scab satisfactorily did not grow on potato-dextrose agar supplemented with 0.25 ppm dodine. Isolates from orchards where tolerance was observed grew at

0.5 ppm dodine. A total of 243 random ascospore progeny from tolerant by nontolerant crossings were analyzed for reaction to dodine at concentrations of 0, 0.25, 0.5, and 1.0 ppm. In four of five crosses, involving 171 progeny, the genetic ratios closely approximated a 1:1 segregation for tolerance at 0.25 ppm and a 1:3 segregation for tolerance at 0.5 ppm dodine. These data suggest that two genes control tolerance to dodine. The segregation ratios for the fifth cross, consisting of 72 progeny, did not fit this interpretation. Inheritance of tolerance to dodine in this cross appeared to be more complex. These results confirm that tolerance to dodine is genetically controlled in *V. inaequalis*.

Virulencia de grupos subespecíficos de Rhizoctonia solani en cultivares de frijol. CARLOS DIAZ POLANCO (Centro de Investigaciones Agronómicas, Maracay, Venezuela). Ciento cuarenta y nueve aislamientos de *Rhizoctonia solani* patógenos a *Phaseolus vulgaris*, *Vigna sinensis*, *Glycine max*, *P. aureus* y otras leguminosas, fueron colectados en diferentes regiones agrícolas de Venezuela. Mediante características morfológicas que han demostrado un alto grado de estabilidad fenotípica en cultivos monocelulares, se determinó la existencia de 6 grupos subespecíficos de *R. solani*. Se observó además, compatibilidad por anastomosis entre los grupos y se estudió su virulencia en los cultivares de *P. vulgaris* Cubagua, Coche y Tacarigua. Cuatro de los aislamientos correspondientes al grupo RS-3 fueron los más virulentos; mientras que los menos virulentos correspondieron a los grupos RS-2, RS-5 y RS-6. El grupo RS-6 fue el que presentó el mayor número de aislamientos de baja virulencia. El cultivar Tacarigua resultó ser el más tolerante a los aislamientos más virulentos del hongo.

Endogone species in roots of Virginia type peanuts. D. M. PORTER & M. K. BEUTE (USDA, ARS, Holland, Va., N.C. State Univ., Raleigh). Peanut (*Arachis hypogaea*) plants grown in the greenhouse were colonized by a zygosporic species of *Endogone*, presumably *E. gigantea*, recovered from the root zone of field-grown peanuts. Two months after inoculation, numerous zygospores and echinulate vesicles, typical of *E. gigantea*, were produced on roots of peanut seedlings, but measurable growth responses were negligible. A chlamydosporic species obtained from soybean roots, presumably *E. macrocarpa*, was not observed to have colonized peanut roots, but aboveground portions of inoculated plants were larger in size and higher in dry weight than noninoculated plants. Zygospores of *E. gigantea* were abundant in peanut and corn field soils, but rarely found in soils growing fescue. *Endogone* zygospores obtained from soil surrounding corn roots germinated readily, colonized, and produced more zygospores on peanut roots. Isolates of *E. gigantea* from peanuts and corn reproduced equally on peanuts. Peanuts grown in soils maintained at low nitrogen levels had higher zygospore populations of *E. gigantea* than did peanuts grown in soil maintained at high nitrogen levels.

Characterization of oat blue dwarf virus and its nucleic acid. D. R. PRING, R. J. ZEYEN, & E. E. BANTTARI (Univ. Neb., Lincoln, Univ. Minn., St. Paul). The oat blue dwarf virus (OBDV) is a spherical virus (28-30 nm) which is phloem-limited in its plant hosts and is transmitted by the aster leafhopper, *Macrostelus fascifrons*. Infectious, purified preparations of OBDV were centrifuged in linear-log sucrose gradients, using brome mosaic virus and southern bean mosaic virus as sedimentation markers. A single zone was evident with a sedimentation coefficient of about 119 S. A single nucleic acid was easily released from the particles in each of five buffers at pH 9.0 containing bentonite. The nucleic acid was resistant to deoxyribonuclease and

susceptible to ribonuclease and to alkaline degradation, and displayed a sedimentation coefficient of 31.9 S. The sedimentation coefficient of formaldehyde-denatured OBDV-RNA was 21.1 S, suggesting a molecular weight of about 2.13×10^6 daltons.

Armillaria mellea in crown gall tissues. R. D. RAABE (Univ. Calif., Berkeley). Naturally occurring crown gall tissues, resulting from infection by *Agrobacterium tumefaciens* on peach and almond, occasionally have been found to be infected by *Armillaria mellea*. Infected gall tissues were more permeated by the hyphal fans of the fungus than nongalled tissues of the same plants. In the greenhouse, peach and tomato seedlings were inoculated with *A. tumefaciens*. After galls had developed, *A. mellea* was added to the soil. Within 6 months, the gall tissue on the peaches was invaded by the fungus, whereas invasion of the plants not infected with crown gall was just starting. In the tomatoes, the gall tissue was invaded by the fungus, but stem tissue of noncrown gall-infected plants was not. Warm water extracts were made from gall tissues from peach and from similar areas of noninfected peach. When added to dextrose agar, *A. mellea* did not grow on agar with extracts from noninfected peach, but the fungus produced more hyphae and rhizomorphs on media to which extracts from crown gall tissues had been added than on potato-dextrose agar.

Active phytoalexin response in Phaseolus vulgaris associated with both induced resistance and subsequent protection against anthracnose. J. E. RAHE & GILLIAN THORSON (Simon Fraser Univ., Burnaby, B.C., Can.). Heat treatment of anthracnose-infected bean plants prevented subsequent pathogenesis and the associated large increases of phenylalanine ammonia lyase (PAL) and phenolic substances. Smaller increases of PAL and the appearance of qualitatively distinct phenolic substances and phaseollin were induced by heat treatment of infected plants. The appearance of phaseollin preceded the development of flecks within which the pathogen was contained after heat treatment. Comparison of fluorescing and phenolic substances associated with natural resistant and susceptible interactions indicated that a shift from susceptibility to resistance was induced by heat treatment. Plants that were reinoculated (challenged) after a heat-induced resistant interaction were protected against subsequent parasitism. Increase in PAL, phaseollin, and phenolic substances characteristic of resistant interaction occurred in protected plants in response to infection by the challenge.

A medium for the selective isolation of Cephalosporium gramineum from soil. A. V. RAVENSCROFT & M. V. WIESE (Mich. State Univ., East Lansing). A medium was developed which permitted the detection and enumeration of propagules of *Cephalosporium gramineum* in soil. The medium sufficiently suppressed faster-growing soil microorganisms so that entrapment of *C. gramineum* in host plants or in host debris is no longer prerequisite for its selective recovery from soil. The medium contained a hot water extract of 100 g of green seedling wheat leaves, 1 g $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ (added after autoclaving), and 20 g Difco Bacto agar/liter of distilled water. Optional additions of 1 to 10 mg of pentachloronitrobenzene/liter of autoclaved medium were used to restrict contaminant organisms in certain soils. Colonies of *Cephalosporium* developing on the medium were distinguishable at low magnifications within 1 week after plating soil suspensions, and recovery from soils amended with known quantities of the fungus exceeded 50%. *Cephalosporium* populations in field soils varied in proportion to the distribution and frequency of susceptible

host plants and to the prevalence of host plant residues in the soil. The highest populations of *C. gramineum* were recovered from wheat field soils in the fall after infected straw was turned under by disking or shallow plowing. Such soils contained up to 30,000 propagules of *C. gramineum*/g dry wt.

Penetration of peach fruit by benomyl and 2,6-Dichloro-4-nitroaniline fungicides. D. J. RAVETTO & J. M. OGAWA (Univ. California, Davis). Benomyl (Benlate) 50W and Botran (2,6-Dichloro-4-nitroaniline) 75W, either alone or in combination with light summer oil, were used at the rates of 0.50, 1.33 lb., and 1 gal/100 gal of water, respectively, as a 20-min dip for harvest-mature Halloween peaches. Injection of 1 ml of spore suspension (2×10^4 /ml) of *Monilinia fructicola* into pit cavities of fruit 2 hr after dipping, followed by 96-hr incubation at 20 C and 90 relative humidity, resulted in the following zones (mm) of healthy outer mesocarp tissue: control, 0; benomyl, 5.9; Botran, 6.7; benomyl + oil, 7.0; Botran + oil, 11.6; and benomyl + Botran + oil, 9.8. Chemical analysis of the healthy tissues showed the following ppm residues of Methyl 1-(butylcarbamoyl)-2-benzimidazolecarbamate (MBC breakdown product of benomyl) and/or Botran: Control, 0; MBC (benomyl), 2.7; Botran, 7.5; benomyl + oil, 3.4; Botran + oil, 8.0; and benomyl + Botran + oil, 2.0 ppm MBC and 12.7 ppm Botran. Fruit sprayed 1 day before harvest with the above fungicides or fruit receiving the preharvest spray and a commercial postharvest Botran + wax treatment were injected with *M. fructicola*, as per the dipped fruit, 24 hr after treatment or following 2 weeks' storage at 32 F. Penetration of the fungicides increased with storage. Similar results were obtained utilizing *Rhizopus stolonifer* as the test organism.

A new tomato virus from Peru. W. B. RAYMER, R. P. KAHN, H. R. HIKIDA, & H. E. WATERWORTH (Campbell Inst. Agr. Res., Cinnaminson, N.J., APHS, ARS, USDA, Glenn Dale, Md.). An unknown virus causing leaf distortion, stunting, and severe yield reduction was isolated from tomato plants near Trujillo, Peru. The virus is designated as the Peru tomato virus (PTV) until its relationship to other viruses is established. Electron micrographs of leaf dips from infected tissue contained flexuous rods 750 nm in length. Campbell 25 tomato plants inoculated with PTV developed chlorotic mottling, rugosity, twisting, and deformation of the leaflets, and were severely stunted. Dilution end point of the virus in sap was 1:1,000-1:10,000, and the thermal inactivation point was 60-70 C. The virus was transmitted by *Myzus persicae* in a nonpersistent manner. *Datura stramonium* was immune to PTV. Serological tests with potato virus Y and other members of that group failed to indicate any relationship with PTV. Cross protection tests with PVY were also negative. The virus was extracted by comminution in 0.025 M phosphate buffer, pH 7.5, containing 0.5 M urea and 0.1% mercaptoethanol and mixed with chloroform in the proportions 1:1.2:0.6, tissue:buffer:chloroform. After low-speed clarification, the virus was given two cycles of high and low speed followed by density-gradient centrifugation. Virus concentrated from gradients was used to prepare an antiserum.

Increase of wound tumor virus in leafhoppers as assayed on vector cell monolayers. D. V. R. REDDY (Univ. Ill., Urbana). Measurements of wound tumor virus (WTV) concentration in extracts from insect vectors (*Agallia constricta*) were made by inoculating monolayer cultures of the vector and subsequently counting the foci of infection. Optimum conditions for performing such an assay were

determined. Brief exposure of insects to 50% ethanol and to certain antibiotics was necessary to avoid contaminations from the insect extracts. The optimum pH value for the extraction of virus was 7.0, and for inoculation was about 5.3 when a solution of 0.1 M histidine and 0.01 M MgCl₂ was used. The focus counts were converted to absolute concentrations of virions by a comparison of the numbers of foci with those obtained from tumor extracts of known absolute virus concentration. After a 1-day acquisition period, virion concentrations in leafhoppers were determined at various times, from the 8th to the 49th day. The virus concentration increased from 10^4 infectious particles/leafhopper to 10^8 or 10^9 between day 8 and days 23-28, when a plateau level was reached. Later, the apparent concentration of virions was markedly affected by the solution used for extraction. The decline in virion concentration after a maximum concentration was reached, reported in previous experiments, was shown to be an artifact of extraction.

Reduction of decay and chilling injury of avocados in a controlled atmosphere. W. F. REEDER & D. H. SPALDING (ARS, USDA, Miami, Fla.). All Lula and 97% of Fuerte avocados were marketable after storage in a controlled atmosphere (CA) of 2% O₂ + 10% CO₂ at 7.2 C for 20 days, followed by ripening at 21 C. Eighty per cent of Lula and 60% of Fuerte avocados stored in air were marketable after ripening. After storage for 40 days, followed by ripening, 97% of Lula and 90% of Fuerte avocados from the CA were marketable compared with 3% or less of the avocados stored in air. Fuerte losses were due primarily to anthracnose (*Colletotrichum gloeosporioides*) and stem-end rot (usually *Diplodia natalensis*). External chilling injury in the rind of Fuerte avocados stored in air for 20 days was slight. Any external chilling injury developing in Fuerte avocados after 40 days in air was masked by severe decay. Only slight external chilling injury developed in Fuerte avocados, even after 40 days in CA. Up to 20% of the losses in Lula avocados stored in air were due to anthracnose combined with chilling injury. The remaining losses were due entirely to the dark discolored rind caused by chilling. Only trace external chilling injury developed in Lula avocados from the CA. Internal chilling injury usually did not occur in either variety.

Decay resistance of six wood species from the Amazon Basin of Brazil. M. S. REIS (Universidade Federal de Vicosa, Vicosa, Minas Gerais, Brasil). The soil-block test was used to determine the comparative decay resistance of outer heartwood and sapwood in single boards of six wood species from the Amazon Basin of Brazil. *Polyporus versicolor* and *Poria monticola* were used as representative white-rot and brown-rot test fungi. Outer heartwood of Freijo [*Cordia goeldiana* Huber], Pau-roxo [*Peltogyne maranhensis* Ducke], and Massaranduba [*Manilkara huberi* (Ducke) A. Chev.] were considered at least resistant; Andiroba [*Carapa guianensis* Aubl.], at least moderately resistant; and Ucuuba [*Virola sp.*] and Ucuuba [*Virola surinamensis* Warb.], definitely nonresistant to decay by both test fungi. *P. versicolor* caused more decay than *P. monticola* in heartwood and sapwood of all species except Pau-roxo. Sapwood of all species was nonresistant to decay. The natural decay resistance of heartwood of Freijo and Pau-roxo was due to decay-inhibiting extractives which could be removed, at least partly, by extraction with acetone, methanol, and hot water. The extractives removed with acetone, methanol, and hot water were very inhibitory to decay by both test fungi. Within each of the six wood species, the decay-susceptibility of sapwood was related directly to its N content; the greater the N content, the greater the susceptibility to decay by both test fungi.

Purification of a protease secreted by Colletotrichum lindemuthianum. S. RIES (Univ. Colo., Boulder). Plant cell walls possess a hydroxyproline-rich structural protein. One of the initial enzymes secreted by *Colletotrichum lindemuthianum*, when grown on Red Kidney bean cell walls, is a protease. This protease may aid in pathogenesis by degrading the structural protein of the cell wall. Maximal enzyme secretion occurs after 6 days of growth on minimal salts medium containing 5% glucose and 0.5% collagen as the nitrogen source. Protease activity has been assayed using a collagen-red dye compound (Azocoll) as substrate. The protease has been purified 300-fold with an 8% yield by a combination of $(\text{NH}_4)_2\text{SO}_4$ fractionation; ion-exchange chromatography on carboxylic acrylic acid polymer, DEAE-cellulose, and sulfoethyl Sephadex resins; and by passage through polyacrylamide gel columns. The enzyme appears to selectively adsorb to polyacrylamide, possibly due to substrate affinity for the amide groups of the polymer. The enzyme has a pH optimum of 7.6 and is stable for several months at 4 C. This is the first proteolytic enzyme from a plant pathogen to be highly purified and characterized.

Inheritance of resistance to culture 111-SS2 of Puccinia graminis f. sp. tritici in the powdery mildew differential C.I. 14115. L. REYES, F. J. GOUGH, & O. G. MERKLE (USDA, Tex. A&M Univ., College Station). Seedling resistance of C.I. 14115 (C.I. 13836/8 Chancellor) to culture 111-SS2 of *Puccinia graminis f. sp. tritici* was conditioned by two independent dominant genes in the F_3 from crosses with susceptible cultivar Little Club. Singly, one gene conditioned a 0; 1 infection type and the other a 3⁻. Infection types in C.I. 14115, Chancellor, and Little Club were 0, 0; 1, and 4, respectively. Neither gene for resistance to culture 111-SS2 was associated with the gene, *Pm1* (derived from C.I. 13836), for resistance to powdery mildew. Others have reported that in several cultivars *Pm1* was closely linked on chromosome 7A with a gene for resistance (designated *Sr15*) to some cultures of race 21 of *P. graminis f. sp. tritici* and to one for resistance to several cultures of *P. recondita f. sp. tritici*. Our data indicate that C.I. 14115 either does not possess *Sr15*, or culture 111-SS2 lacks the corresponding gene for avirulence necessary for its detection.

Histopathology of Pinus ponderosa ectomycorrhizae infected with a Meloidogyne species. J. W. RIFFLE (U.S. Forest Serv., Rocky Mountain Forest Range Exp. Sta., Albuquerque, N. Mex.). An undescribed *Meloidogyne* sp. was found infecting ectomycorrhizae of mature *Pinus ponderosa* in southwestern New Mexico in 1963. Larvae penetrated the branches of the ectomycorrhizae, migrated to the vascular tissue, and developed into mature egg-laying females with their heads buried in the plerome. Multiple infections of ectomycorrhizae were common. As the female developed, cortical cells and the Hartig net at or near its body were pushed aside and crushed. The fungal mantles were usually ruptured, and the gelatinous matrix containing egg masses protruded from the roots. The feeding activities of the nematode caused hypertrophy of the vascular tissue and proliferation of branch rootlets. Syncytia developed as a cluster of multinucleate cells in the vascular tissue immediately adjacent to the head of the nematode. Cytoplasm of the syncytia was granular in texture, and nuclei were greatly enlarged, with irregularly shaped membranes. Nucleoli were also enlarged. Xylem tracheids were disorganized and crushed by the formation of the syncytia.

Populations of root knot nematode and root

development in cotton after a Bahia grass sod. R. RODRIGUEZ-KABANA & R. W. PEARSON (Auburn Univ., Auburn, Ala.). A 3-year study was conducted to compare changes in populations of root knot nematodes (*Meloidogyne incognita*) and root densities in plots under cotton (McNair 1032B, tolerant) monoculture (CM) and plots planted to cotton for 1 (C1), 2 (C2), 3 (C3), and 4 (C4) years following Bahia grass (*Paspalum notatum*) sod. Each season, plots were sampled every 3-4 months to represent various stages of cotton growth. Numbers of root knot larvae in C1 and C2 were very low, reflecting the previous growth of Bahia grass, a nonhost; significant numbers of larvae were recorded only in CM, C3, and C4. C3 and C4 showed consistently higher numbers of larvae than CM. Differences in larval population density between C3 and C4 were greatest at harvest time and during winter months, when numbers of larvae in C4 were 2-3 times those in C3. Root development in CM was restricted to a depth of 30 cm, whereas cotton roots in other plots penetrated to depths greater than 60 cm. Cotton yields in CM were half those of other plots. Differences in yields between any of the post-Bahia grass plots were not significant. The higher numbers of root knot larvae recorded for C3 and C4 are interpreted to be the result of the deeper penetration and higher total number of roots in post-Bahia grass cotton than in CM.

Characterization of cellular RNA of Phytophthora spp. by disc-gel electrophoresis. J. R. ROHEIM, J. V. LEARY, & G. A. ZENTMYER (Univ. Calif., Riverside). Ribosomes were isolated from mycelium, encysted zoospores, and germinating cysts of several *Phytophthora* spp., but ribosomes could not be isolated from ungerminated oospores. Large amounts of rRNA and 4-5 S RNA were detected in mycelium and encysted zoospores by disc-gel electrophoresis of total RNA prepared by phenol-sodium dodecyl sulfate extraction. RNA concentrations in mycelium changed with age. Extraction of equivalent dry weights of dormant oospores produced by both old and young mycelium yielded only very small amounts of low molecular-weight RNA. Small amounts of high molecular weight RNA were obtained when 5-6 times greater amounts of dormant oospores were extracted. Sedimentation coefficients for these RNA's were estimated to be 16 to 18 S and 28 S which are similar to values reported for rRNA. These results support the hypothesis that the dormant *Phytophthora* oospore does not contain necessary components for protein biosynthesis as do other morphogenetic stages.

Herbicide predisposition of snapbeans to Rhizoctonia solani. W. R. ROMIG & M. SASSER (Univ. Delaware, Newark). Field observations indicate that two herbicides, Trifluralin (trifluoro-2,6-dinitro-N,N-dipropyl-p-toluidine) and Dinoseb (dinitro-o-sec-butylphenol) increase the incidence and severity of *Rhizoctonia solani* damage on snapbeans. In vitro studies indicated that herbicide concentrations of 10 to 80 ppm did not enhance fungal growth or levels of cellulolytic or pectolytic enzyme activity of *R. solani*. No differences in amount of exudation of carbohydrates or amino acids were attributable to herbicide treatment. Hypocotyl cellulose content, methylated pectin content, and resistance to mechanical penetration were significantly reduced by the herbicide treatments. Insoluble calcium and magnesium levels were unchanged in the hypocotyls of treated plants. Snapbeans grown in soil treated with Trifluralin or Dinoseb had more numerous and larger lesions on hypocotyls infected with *R. solani* than did the plants grown without herbicide. Levels of phytoalexin and

phaseollin were reduced in direct relation to the herbicide concentration applied. Trifluralin and Dinoseb herbicide applications appear to reduce snapbean structural and biochemical defenses, leading to increased disease losses.

A simulated epiphytotic of corn stunt. E. ROSENKRANZ (ARS, USDA, Miss. State Univ., State College). Work on host resistance to corn stunt (CS) is hampered by annual fluctuations in the incidence of the disease and by the impracticality of artificial inoculation in the field. An epiphytotic was simulated under screen on 100 widely diverse corn genotypes. When infective *Dalbulus maidis* were released at the rate of two leafhoppers per seedling, all susceptible plants became infected with corn stunt agent (CSA). Based on the incidence of CS among 10 key cultivars, the inoculum pressure in the screenhouse was 8, 15, and 3.5 times as great as it was in nature in 1969, 1970, and 1971, respectively. Incidence of CS among all entries was greater under screen than in the field in any year. The inoculation period of CSA proved to be a function of the genotype's level of susceptibility. Intensity of leaf discoloration depends on the relative number of genes for plant color a cultivar possesses. To measure the effectiveness of the simulated epiphytotic to unmask potentially susceptible genotypes, plants of the inbred Mp412 were inoculated individually, subjected to released infective leafhoppers in the screenhouse, and exposed to natural infection. Under these conditions, there were 89.7, 82.1, and 27.5% diseased plants, respectively.

Local and systemic effects of ethylene on tobacco mosaic virus lesions in tobacco. A. F. ROSS & D. W. PRITCHARD (Cornell Univ., Ithaca, N.Y.). Attached leaves (two/plant) on 10- to 12-inch tobacco (*Nicotiana tabacum* 'Samsun NN') plants were sealed in polyethylene bags and subjected for 5-6 days to a flowing air stream (ca. 1 liter/min per leaf) with or without added ethylene. Ethylene at 2-6 ppm (continuous) or 6-80 ppm (intermittent) caused trace to moderate epinasty, wilting, yellowing, and spot necrosis (higher dosages only) in treated leaves. Postinoculation treatments, started 1 day after inoculation with tobacco mosaic virus, effected up to a 70% increase in final lesion size (diameter) in treated bagged leaves. Preinoculation treatments, ended 1-2 days before inoculation, inhibited lesion growth and enhanced peroxidase activity both in treated bagged leaves and in untreated unbagged leaves on the same plants. Final lesion size in treated leaves was usually about half that in control plants on which the comparable leaves were either unbagged or bagged and subjected to the air stream minus ethylene. In general, inhibition tended to increase as ethylene dosage was increased. Restriction of lesion size in unbagged untreated leaves on treated plants was generally 20-30%, but occasionally was as great as 50%. The systemic effect on lesion size was variable, however, and was not consistently related to dosage or to extent of injury to treated leaves.

*Pathological histology of resistant and susceptible sugar beet roots inoculated with *Rhizoctonia solani*.* E. G. RUPPEL (ARS, USDA, Colo. State Univ., Fort Collins). Resistant and susceptible sugar beet cultivars were penetrated by *Rhizoctonia solani* directly by individual hyphae or by infection cushions. Lesion diameter and depth were greater in susceptible roots than in resistant roots. Necrosis and some tissue degeneration preceded hyphal advance in all roots after penetration. Hyphae in resistant roots usually were observed only in the periderm or secondary cortex, whereas in susceptible roots, the hyphae often transected several vascular rings. Hyperplasia of cortical cells occurred at the margin of necrotic and healthy tissue, but no wound periderm

or cicatricelike cell layers were evident in resistant roots. Resistance in sugar beet to *R. solani* was not found to be due to mechanical barriers to the pathogen.

Nematicidal properties of potassium and sodium azides under laboratory, greenhouse, and field conditions. M. C. RUSH, C. W. AVERRE, K. R. BARKER, & J. N. SASSER (N.C. State Univ., Raleigh). Eggs of *Meloidogyne incognita* were killed in solutions of KN_3 at 15 $\mu\text{g/ml}$. NaN_3 (15 $\mu\text{g/ml}$) and KN_3 (25 $\mu\text{g/ml}$) solutions prevented emergence of larvae of soybean cyst nematode, *Heterodera glycines*. More than 95% of the nematode eggs in cysts were killed by solutions of KN_3 and NaN_3 at concentrations above 25 $\mu\text{g/ml}$. *Belonolaimus longicaudatus*, *Helicotylenchus dihystera*, *H. glycines*, *M. arenaria*, *M. incognita*, *M. javanica*, *Scutellonema brachyurum*, *Trichodorus christiei*, *Tylenchorhynchus claytoni*, and *Xiphinema americanum* in naturally infested soil were killed by fumigation in capped glass jars with KN_3 mixed in soil at 5 to 20 $\mu\text{g/g}$. Bioassay tests and Baermann funnel extractions were used to determine per cent kill and survival; *Pratylenchus zaeae* and *T. christiei* were controlled at 15 and 40 $\mu\text{g/g}$ KN_3 , respectively. In field trials conducted at various locations in North Carolina on cotton, peanut, and soybean, KN_3 at 11, 22, and 45 kg/hectare in the row was usually ineffective for most nematodes; however, *B. longicaudatus*, *M. incognita*, *S. brachyurum*, and *T. claytoni* were controlled in one or more of these field tests.

*Response of cultured cells of *Nicotiana tabacum* 'Samsun' and 'Samsun NN' to microinjection with tobacco mosaic virus.* T. E. RUSSELL & R. S. HALLIWELL (Univ. Ariz., Tucson, Tex. A&M Univ., College Station). Cultured cell chains derived from two *Nicotiana tabacum* cultivars were microinjected with tobacco mosaic virus (TMV) for comparison of the effects of the virus on cultured cells of a systemic host (Samsun) with those of a local lesion host (Samsun NN). Eight Samsun NN cell chains injected with infectious TMV became infected and contained inclusions; two showed no response. Five Samsun NN chains injected with heat-treated TMV produced neither symptoms nor inclusions. Eight Samsun chains injected with infectious TMV showed symptoms with inclusion formation; five chains injected with heat-treated TMV were symptomless. Evidence of infection in the Samsun NN cells included rapid loss of transvacuolar streaming, apparent infection of some cells and not others in a cell chain, cytoplasmic vesiculation, and infrequent formation of small virus inclusions. Response of Samsun cells in chains was milder and more predictable than that in infected Samsun NN cells. Infected Samsun cells maintained their integrity longer, and produced larger and more numerous inclusions, and symptoms appeared in all cells of each chain showing virus spread from cell to cell. Bioassay of inclusion bearing cell chains of both cultivars verified virus multiplication from injected TMV.

Influence of host nutrition on bacterial brown spot of bean. S. M. SAAD & D. J. HAGEDORN (Univ. Wis., Washington County, Madison). The relationship of host nutrition to lesion type and disease severity with bean bacterial brown spot, *Pseudomonas syringae* van Hall, was studied in greenhouse sand culture. Molar concentrations of reagents were used to obtain 2-fold, standard, and nil concentrations of N, P, and K, and 0.75, 0.50, 0.25 levels of K. Tenderwhite bean plants were started in perlite, transplanted into silica sand in 5-inch plastic pots, and inoculated at the third trifoliate leaf stage with *P. syringae* inoculum dosage of 10^7 cells/ml applied with a mist sprayer at 15 psi. With minus N, the lesions were larger than normal,

translucent, and red brown. With minus P, lesions were discrete dark purple, sunken, smaller than normal, and without water-soaking and chlorosis. With the different levels of K and 2-fold N and P, lesions were quite typical of bacterial brown spot: oval; brown, with water-soaked edges; and surrounded by a halo with puckering of the surrounding tissues. Disease severity also increased under those treatments when compared to the control; i.e., 1 times Hoagland. The over-all effect on the host was greatest with minus K and least with minus P. These studies suggest that symptom development and disease severity may be influenced by soil fertility.

Changes in some oxidative enzyme activities and auxin content of apple leaves after a grafting with healthy and virus-infected buds of Golden Delicious. MARIAN SANIEWSKI & D. F. MILLIKAN (Univ. Mo., Columbia). Leaf tissue of Spy 227, Russian, and MM 104 grafted with buds of healthy (4-4) and virus infected (E-1) clones of Golden Delicious were lyophilized and examined for phosphatase, catalase and peroxidase activities, and auxin content. Three collections were made which showed that phosphatase activity was greatest in MM 104 but was reduced by budding with E-1 in the first and final collections. Phosphatase activity initially was unchanged in Spy 227 and Russian, but was increased in MM 104 by budding with 4-4 and decreased with E-1. In the second collection, phosphatase activity of Spy 227 was increased by E-1 and reduced in the third. Budding with 4-4, however, increased the phosphatase activity of Russian in both collections. Catalase activity initially was increased in Spy 227 and Russian by budding with 4-4, but reduced in MM 104. In the second collection, budding with E-1 decreased the catalase activity in Spy 227, increased it in Russian, but did not affect MM 104. In the final collection, catalase activity was reduced in Spy 227 and Russian by budding with E-1 and increased in MM 104. Budding with 4-4 had no effect on Spy 227, but increased the catalase activity in Russian and reduced in MM 104. Peroxidase activity of all cultivars was affected in a manner similar to catalase, but the auxin content appeared to be inversely correlated.

Control of grain storage fungi with propionic and acetic acids. D. B. SAUER, R. BURROUGHS, & J. A. SIMON (ARS, USDA, Kansas State Univ., Manhattan). Yellow corn harvested at 22% moisture content and treated with 0.4% propionic acid or 0.8% acetic acid had no detectable mold growth in 6 months at 25 C. Lower levels of acid prevented mold growth for shorter periods. Grain sorghum and wheat harvested and stored at 24-25% moisture content and 25 C were kept free of storage fungi by application of 0.4-0.5% propionic acid. Some tests indicated that slightly higher application rates were required for grain in which moderate fungal invasion had occurred prior to treatment. Fungi which grew in grain treated with insufficient amounts of acid were the same as those in untreated grain, but the species diversity was not as great in treated grain. Fat acidity values of treated corn and grain sorghum increased continuously during storage in the absence of detectable microorganisms. Increases in fat acidity were slower with increasing application rates of acetic or propionic acid. All treatments which effectively controlled fungal growth destroyed the germinability of the seeds.

A new medium for the isolation of Xanthomonas campestris. N. W. SCHAAD (Univ. Ga. Exp. Sta., Experiment). *Xanthomonas* grows readily on potato-dextrose agar (PDA), yeast-dextrose calcium carbonate agar (YDC), and nutrient agar + 0.5% glucose (NA). A selective medium

(D-5) for the isolation of *Xanthomonas* species is also available. These media are, however, either too inhibitory to *X. campestris* or not sufficiently inhibitory to other yellow, mucoid colonies. A differential medium which proved somewhat selective contained: K_2HPO_4 , 2 g; NH_4Cl , 5 g; beef extract, 1 g; soluble-potato starch, 10 g; methyl violet B, 10 mg; methyl green, 20 mg; cyclohexamide, 250 mg; agar, 15 g; and distilled water, 1,000 ml. *X. campestris* colonies were observable after 48 hr at 30 C, and were ca. 7 mm in diam after 7 days. The colonies were mucoid, convex, transparent with a purple center, and surrounded by a clear zone indicative of starch hydrolysis. When a standard suspension of *X. campestris* was plated, the number of colonies on the new medium was ca. 50% of that on NA (optimum recovery medium), whereas no colonies appeared on D-5. When a 1:10 dilution of a Cecil, sandy clay loam soil containing 1×10^3 added viable cells/g was plated, the percentage recovery was 29% for the new medium and 0.0% for NA and D-5. These results illustrate the superiority of the new medium for isolation of *X. campestris*.

Interaction of Pseudomonas glycinea and yellow bacteria in development of bacterial blight of soybean. R. H. SCHERFF (ARS, USDA, Univ. Mo., Columbia). A group of yellow bacteria were isolated from bacterial blight-infected leaves of Clark 63 soybeans. These bacteria were small, gram-negative rods, highly motile, and oxidase positive. They possessed polar flagella, and produced a light-yellow, water-insoluble pigment. When *Pseudomonas glycinea* was mixed with yellow bacteria isolate YB-3 at a 1:9 ratio and immediately rubbed onto Carborundum-dusted soybean leaves, lesion development of bacterial blight was inhibited. At a ratio of 1:4, a 50% reduction resulted and at a 1:1 ratio there was only slight lesion inhibition. The initial cell concentrations were 10^8 cells/ml. By incubating the *P. glycinea*-YB-3 mixture 24-48 hr prior to inoculation, nearly complete lesion inhibition resulted with the 1:4 and 1:9 ratios; however, there was no reduction of symptoms at the 1:1 ratio. Population levels of *P. glycinea* and YB-3 were assayed 1, 2, 3, 7, and 14 days after a careful atomizing of bacteria onto soybean leaves so as not to injure the leaf surface. At the 1:9 ratio of *P. glycinea* to YB-3, a sharp reduction in the number of *P. glycinea* cells resulted after day 7; whereas in the 1:1 ratio or *P. glycinea* alone, the number of *P. glycinea* cells continued to increase. In all ratios, populations of YB-3 increased through day 3, then dropped sharply.

Changes in glucose-6 phosphate dehydrogenase, glyceraldehyde-3 phosphate dehydrogenase, and peroxidase activity in Populus tremuloides after inoculation with Hypoxylon mammatum or wounding. A. L. SCHIPPER, JR. (USDA Forest Service, St. Paul, Minn.). Aspen (*Populus tremuloides*) trees were inoculated with *Hypoxylon mammatum* (143 trees), wounded or untreated (43 trees each). Treated areas from separate groups of 15 inoculated, 10 wounded, and 10 untreated trees were sampled at 2-week intervals for 10 weeks, and the specific activities (units enzyme (U)/g dry wood or bark) of glyceraldehyde-3-phosphate dehydrogenase (G13PD), glucose-6-phosphate dehydrogenase (G6PD), and peroxidase (PR) determined. At 6 weeks G13PD (ca. 1 μ g at 0 week) had increased 11% in wounded areas but declined 55% in inoculated areas. At 4 weeks, G6PD (ca. 0.05 μ g at 0 week) increased 366% in wounded areas and 154% in inoculated areas, but at 6 weeks, wounded and inoculated areas had the same activity (186% of 0 week). At 4 weeks, PR (ca. 70 μ g at 0 week) increased 26-fold in inoculated areas, but only 471% in wounded areas. The high PR activity in inoculated areas

persisted for 2 weeks, then declined. Lower activity of G6PD in inoculated than in wounded areas may indicate that fungal interference with wound healing is a factor in pathogenesis because the G6PD peak in wounded areas coincided with maximum callus activity. PR activity was highest as mycelium reached the sampling point (4 weeks), but in aspen, high PR activity apparently does not indicate resistance to *H. mammatum*.

Effect of N, P, and K on the incidence of fusiform rust galls on greenhouse-grown seedlings of slash pine. R. A. SCHMIDT, M. J. FOXE, C. A. HOLLIS, & W. H. SMITH (Univ. Fla., Gainesville). In independent experiments conducted in subsequent years, greenhouse-grown slash pine (*Pinus elliottii* var. *elliottii*) seedlings, in pots containing 1 kg of Leon fine sand, were fertilized when 3 or 6 weeks old with 0, 50, and 100 ppm N (H_2HCONH_2 or NH_4NO_3), 0 and 50 ppm P [$Ca(H_2PO_4)_2 \cdot H_2O$ or $NaH_2PO_4 \cdot H_2O$], and 0 and 50 ppm K (KCl). Two weeks after fertilization, pines were inoculated with basidiospores of *Cronartium fusiforme* by the foliar deposition or stem injection technique. Seedling height increased with increasing levels of P and K, but decreased with increasing levels of N. Seedling height and total amount of nutrients were positively correlated with numbers of rust galls in one foliar deposition experiment. A significant increase in rust was associated with increased levels of P. The effect of P on rust was greater than its effect on height growth, suggesting a relationship between P metabolism and fungus growth. Suggestions of an interaction between inoculation technique and fertilizer were evident. In general, these data support the hypothesis that increased rust is associated with increased host vigor, but differ with respect to published field data where increased levels of N stimulated plant growth.

Effect of light and calcium on germination of oospores of Pythium aphanidermatum. A. F. SCHMITTHENNER (Ohio Agr. Res. Development Center, Wooster). Media for oospore production consisted of 2.4 g sucrose, 0.27 g asparagine, 0.15 g KH_2PO_4 , 0.15 g K_2HPO_4 , 0.10 g $MgSO_4 \cdot 7H_2O$, 4.4 mg $ZnSO_4 \cdot 7H_2O$, 1.0 mg $FeSO_4 \cdot 7H_2O$, 0.07 mg $MnCl_2 \cdot 4H_2O$, 2.0 mg thiamine, 10.0 mg ascorbic acid, and 10 mg cholesterol/liter; with agar and calcium level and source varied. Germination of oospores produced on media with agar and $CaCl_2$ germinated less than 12% when flooded and incubated in the dark increased significantly (up to 31%) by exposing oospores for 12 hr prior to flooding to 300-350 ft-c produced by white and near-ultraviolet fluorescent lamps. Exposure of 36 hr produced optimum germination (up to 94%). Oospores produced in media with $CaCl_2$ but without agar required 12 hr light for optimum germination (up to 85%) when tested in intact culture mats. However, 50% or more of oospores separated from liquid culture mycelium or the surface growth on agar cultures germinated without light treatment. Oospores produced on agar media containing greater than 20 ppm Ca required more light for optimum germination than those produced on an agar medium with less than 10 ppm Ca. The type of Ca source added for oospore formation affected the percentage of germination in liquid media.

Tobacco ringspot virus codes for the coat protein of its satellite. I. R. SCHNEIDER (ARS, USDA, Beltsville, Md.). Two isolates of tobacco ringspot virus (TRSV) and two isolates of its satellite virus (S-TRSV) were used to determine whether S-TRSV or TRSV codes for the coat protein of S-TRSV. The two TRSV isolates (TRSV-WS, from Maryland, and TRSV-NC 87, from North Carolina), when used as antigens in Ouchterlony double-diffusion plates, were

distinguishable serologically because, when the antigens were placed in adjacent wells, their reaction with TRSV antisera caused spur formation. Similarly, the two satellite isolates (S-TRSV-WS and S-TRSV-NC 87), when placed in adjacent wells, formed spurs in reactions with TRSV antisera. Four groups of bean plants (*Phaseolus vulgaris* 'Black Valentine') were inoculated with mixtures of (i) TRSV-WS + S-TRSV-WS; (ii) TRSV-WS + S-TRSV-NC 87; (iii) TRSV-NC 87 + S-TRSV-WS; and (iv) TRSV-NC 87 + S-TRSV-NC 87. The partially purified virus preparations from the infections were >90% S-TRSV, and were serologically compared using antiserum to TRSV-WS or TRSV-NC 87. The presence or absence of spurs, as affected by the inoculum composition, showed that each product was serologically determined by the TRSV isolate and not by the S-TRSV isolate in the inoculum.

Sporulation of Aphanomyces euteiches. C. L. SCHOULTIES & C. Y. YANG (Univ. Ky., Lexington). Numbers of zoospores produced by *Aphanomyces euteiches* in unit time were studied under varying ionic conditions. Sporulation was achieved by replacing the nutrient medium with a solution containing $CaCl_2$, KCl, and $MgSO_4$. Zoospore release from sporangia began 5 to 6 hr later, and continued for 24 to 60 hr. Duration of sporulation depended on culture age at time of induction. Sporulation occurred in two phases, with maxima at 12 hr and 18 to 24 hr after replacement. The extent of the initial response diminished with culture age, whereas the extent of the second response increased. Calcium seemed essential for the second response, and also facilitated motility of zoospores. If calcium was absent in the replacement medium, only the initial response occurred and the motility of zoospores was impaired. Corroborative data are available with other isolates of *A. euteiches*.

An outbreak of lethal yellowing of coconut palms in Miami, Florida. C. P. SEYMOUR, J. W. MILLER, & D. A. ROBERTS (Fla. Dep. Agr., Univ. Fla., Gainesville). Lethal yellowing of coconut palms was found for the first time on the Florida mainland (Miami and Coral Gables) in September 1971. A ground survey showed that the primary concentration of 57 diseased trees occurred in a one-half-square-mile area. Helicopter surveys made in December 1971 and March 1972 revealed no other outbreaks on the mainland. Continued ground and helicopter surveys to determine the rate and direction of spread showed that the highest incidence of disease has been restricted to the primary infested area, with the most distant occurrence from the center being 3.5 miles away. Thus far, the disease has progressed in all directions, with the greatest spread toward the southwest. Numbers of diseased trees detected were: Sept.-Nov., 151; Dec., 40; Jan., 27; Feb., 45; and March, 62. Because of the importance of the ca. 400,000 coconut palms in this urban area, an effort was made by the Florida Department of Agriculture and Consumer Services to combat lethal yellowing by (i) continuing ground and helicopter surveys to locate newly diseased trees; (ii) reducing inoculum through the cutting of diseased trees; and (iii) encouraging the replanting with resistant Malayan Dwarf varieties.

The form and composition of laminate inclusion components in potato virus X-infected cells. T. A. SHALLA & J. F. SHEPARD (Univ. Calif., Davis, Montana State Univ., Bozeman). Cells of tobacco leaves (*Nicotiana tabacum* 'Burley 21') infected with potato virus X (PVX) contained amorphous inclusion bodies which, in addition to normal cytoplasmic materials and viral aggregates, were composed of

conspicuous laminate structures. Examination of serially sectioned tissue with the electron-microscope revealed that these were collateral bundles of smooth or beaded sheets which were either lightly curved or rolled into scrolls, usually with virus particles interspersed between alternating layers. The beaded sheets were destroyed when tissues were exposed to KMnO_4 and were susceptible to digestion by the proteolytic enzyme subtilisin. Tagging experiments with ferritin-labeled antibodies showed that neither the beads nor sheets were antigenically related to PVX or to PVX-depolymerized structural protein. The sheets often appeared as single electron-dense layers ca. 3 nm thick. The ribosomelike beads, modal diam 11-14 nm, were smaller than cytoplasmic ribosomes. It was concluded that the laminate components were virus-specific infection products similar in several respects to the intracellular cylindrical inclusions induced by viruses of the PVY group.

Effect of surface charge of virus particles on their attachment to leaves during mechanical inoculation. J. G. SHAW (Univ. Ky., Lexington). The possibility that an interaction between charged substances in leaf surface layers and the surface charges of virus particles may be a factor in the attachment of virions during mechanical inoculation was investigated. Since leaf surfaces carry a net negative charge, it was postulated that a preponderance of negative charges on the surfaces of virus particles would inhibit virion attachment. Two viruses of very similar morphology but with quite different surface charges were used. Radioactive preparations of cowpea chlorotic mottle virus (CCMV) and brome mosaic virus (BMV), which are isoelectric at pH 3.6 and 7.9, respectively, were applied to corn and cowpea leaves. About 5 times more BMV than CCMV was retained by both hosts after the leaves were rinsed. Furthermore, the proportion of retained labeled material remaining in the tissue residues after extraction was greater with BMV than with CCMV. It thus appears that negative charges in the surface areas of leaves may interfere with the attachment of the more electronegative CCMV to a greater extent than it hinders the attachment of BMV.

Decay associated with paraformaldehyde-treated tapholes in sugar maple. A. L. SHIGO & R. S. WALTERS (USDA Northeastern Forest Exp. Sta., Durham, N.H.). Pills of paraformaldehyde increase the period of sap flow when placed in tapholes of sugar maple, *Acer saccharum*. The effect of the pills on wood surrounding tapholes was determined. In February 1970, 250 mature trees in six locations in Vermont were drilled for two tapholes, each 1.1 cm-diam and 8 cm deep. A 250-mg paraformaldehyde pill was placed in one hole on each tree. In April 1970, one tree from each location was cut; no Hymenomyces were isolated from wood above and below the holes. In October 1970, another tree from each location was cut. Wood above and below all six treated holes yielded Hymenomyces: 51 of 144 chips. Three chips from one control hole yielded a Hymenomyces. In October 1971, two trees were cut from each location. Hymenomyces were isolated from wood above and below every treated hole: 179 of 288 chips. Wood above and below three control holes yielded Hymenomyces: 27 of 288 chips. There was no significant difference in length of 24 columns of discolored wood associated with all holes, but the frequency of decay and the length of decay columns associated with treated holes were significantly greater than those of the controls.

Relationships between steroid glycoalkaloid and rishitin accumulation in potato tuber slices. M. SHIH & J. KUC (Purdue Univ., Lafayette, Ind.). Steroid glycoalkaloids

accumulate in the top two mm of noninoculated tuber slices. Accumulation is markedly suppressed by sonicates of *Phytophthora infestans* and inoculation with *P. infestans* or *Helminthosporium carbonum*. Incompatible races of *P. infestans* are more effective suppressers than are compatible races. Rishitin accumulation in incompatible interactions or after treatment with sonicates approximately coincides with the time when suppression is evident. Isotope incorporation experiments using labeled acetate and mevalonate indicate that rishitin and steroid glycoalkaloids are synthesized de novo. Slices aged for 72 hr before inoculation with *P. infestans* do not accumulate rishitin, whereas sonicates are effective inducers. It appears that injury activates the acetate-mevalonate pathway for the synthesis of steroid glycoalkaloids, and inoculation or treatment with fungal sonicates diverts the pathway to rishitin synthesis.

Further characterization of nucleic acid components associated with tobacco mosaic virus infection. A. SIEGEL, M. ZAITLIN, & C. DUDA (Univ. Ariz., Tucson). Several components associated with virus infection can be identified when RNA extracted from ^3H -uridine labeled, infected, separated leaf cells is subjected to gel electrophoresis. These are RI, RF, tobacco mosaic virus (TMV) RNA and a low-molecular weight (ca. 350,000 d.) component (LMC). In order to characterize these components further, the labeled RNA extracted from gel slices was heated and quick-cooled in order to melt double-stranded RNA, and then either self-annealed or annealed in the presence of either TMV RNA or double-stranded (DS) RNA isolated from infected leaf tissue. It was found that the labeled RF component annealed with DS RNA to a considerably greater extent than it did with TMV RNA. This was interpreted as indicating an asynchrony in replication of RF during the labeling period in such a way that more TMV RNA strand was synthesized than its complement. DS RNA annealed well with the TMV RNA from the gel, as expected, but surprisingly, it also hybridized to a lesser extent with RNA from the ribosomal RNA and LMC regions of the gel. No such annealing was noted with similarly treated RNA extracted from uninfected cells. Several interpretations for this phenomenon are considered.

Distribution and Metabolic fate of methyl 2-benzimidazolecarbamate in strawberry. M. R. SIEGEL (Univ. Ky., Lexington). Strawberry plants (cultivar Superfection) were grown in Hoagland's solution and root-treated with C^{14} labeled MBC (methyl 2-benzimidazolecarbamate) at 4-day intervals during an 88-day period. Plants were extracted with organic solvents, and the distribution and metabolic fate of the labeled fungicide in the various extracts and plant residue determined. The per cent of total label in compounds soluble in organic solvents, water fraction, and those bound to the residue was 46, 31, and 23, respectively. Labeled compounds were identified as MBC, 2 aminobenzimidazole (2-AB), and unknowns. The organic solvent extracts contained 66, 18, and 15%, respectively, of these compounds; the water soluble fraction contained 3, 24, and 74%, respectively. Fruit harvested from treated plants, representing ca. 10% of the total dry weight, contained less than 0.001% of the total label. MBC is metabolized in strawberry to a much greater extent than in other plants. The metabolites (2-AB and unknowns) are primarily water-soluble or bound to the residue. The lack of accumulation of labeled products in the fruit is, however, similar to that reported for other species.

*Anatomical marker for resistance to Dutch elm disease in *Ulmus americana*.* W. A. SINCLAIR, J. P. ZAHAND, & J. B. MELCHING (Cornell Univ., Ithaca, N.Y.). Xylem characters

of American elms (*Ulmus americana*) judged resistant (R) or susceptible (S) to Dutch elm disease (DED) on the basis of reaction to inoculations with *Ceratocystis ulmi* were examined. The mean radial diameters of vessels measured in the outer xylem sheath of 2-year-old branches from 10 R and 10 S trees were 39.5 and 49.6 μ , respectively (difference significant at 1% level). A vessel diameter index (VDI) was developed to facilitate measurements. VDI equals mean percentage of vessels of diameter $\geq 50 \mu$ among vessels in the second and third xylem rings outward from the pith along an arbitrary radius in each of three cross-sections from each of three branches/tree. VDI was not related to apical or radial growth rates of branches. Trees with VDI ≤ 12 may be resistant to DED. Five of 11 trees and clones that recovered from at least two systemic DED infections and five of 11 trees free of DED symptoms growing in areas where nearly all other elms had died had a VDI ≤ 12 . None of seven trees and clones selected for DED susceptibility and one of 18 nonselected field trees had a VDI ≤ 12 . VDI may be useful for rapid assessment of possible resistance in American elms that survive DED epidemics.

Adhesion of zoospores of Phytophthora palmivora to solid surfaces. V. O. SING & S. BARTNICKI-GARCIA (Univ. Calif., Riverside). Fungal zoospores adhere to solid surfaces after cessation of motility. Firm adhesion of some pathogens to their host tissue is an important step in the infection sequence. Zoospores of *P. palmivora*, papaya strain P113, were incubated at 1 C for 20 min (5 ml zoospore suspension in a 30-ml beaker), and the per cent adhesion was determined by counting the zoospores left in the suspension with a Coulter counter. Under such conditions, 50-60% of the zoospores adhered to the bottom of the vessel and remained adhered even after application of a strong shearing force. Less than 10% of the nonadhered zoospores had formed an alkali-resistant cyst wall. Zoospore suspensions, agitated in a Vortex mixer from 15 to 180 sec, underwent rapid encystment. In this time period, the per cent encystment increased from 4 to 82, while the per cent adhesion decreased from 54 to 10. Evidently, zoospores are not able to adhere to solid surfaces after they are encysted, although they retain the ability to adhere to one another. Seemingly, zoospore adhesion occurs during, or immediately before, an alkali-resistant cyst wall is formed. This adhesion is nonspecific, since zoospores adhere equally well to glass, plastic, or leaf surfaces.

Similarity of host symptoms induced by citrus exocortis and potato spindle tuber causal agents. R. P. SINGH, M. C. CLARK, & L. G. WEATHERS (Can. Dep. Agr., Fredericton, N.B., Univ. Calif., Riverside). A comparative study of host-range was made with citrus exocortis (CE) and potato spindle tuber (PST) agents. Potato, tomato, and six species of *Scopolia* were tested. Leaves of citron (*Citrus medica*) and tomato (*Lycopersicon esculentum* "Sheyenne") infected with CE and PST, respectively, were ground in phosphate buffer containing bentonite. The resulting homogenate was inoculated on leaves of the test plants. In potato (*Solanum tuberosum*), both CE and PST agents caused stunting, reduction in leaf size, and necrotic speckling on the stem and petioles. Similarly, in tomato, both agents produced severe stunting, rugosity of leaves, veinal necrosis, and clustering of top leaves. Inoculation of *Scopolia corniolica*, *S. lurida*, *S. sinensis*, *S. stramonifolia*, and *S. tangutica* with both agents separately resulted in local, circular, necrotic lesions, attended by a progressive systemic veinal necrosis. Despite their similarity with both agents, lesions appeared a few days earlier in response to PST. In *Scopolia physaloides*, both agents were carried without symptoms. The infectious agent

was recovered from all test plants, as demonstrated by the ability of nucleic acid extract to produce the same symptoms in *Scopolia sinensis* plants.

Control of physiological and pathological breakdown of peaches and nectarines during storage. W. L. SMITH, JR., & R. E. ANDERSON (USDA, Beltsville, Md.). Physiological breakdown of peaches and nectarines during holding at 18 C after storage for 6 weeks at 0 C was less when fruit was stored in an atmosphere of 1% O₂, 5% CO₂, and 94% N₂ rather than air. Pathological breakdown did not differ in the two atmospheres. Development of brown rot during storage and holding after storage of naturally inoculated fruit, or those artificially inoculated with spores of *Monilinia fruticola* (Wint) Honey, depended on the treatment applied before storage. Fruit treated for 2.5 min in suspensions of 100 ppm benomyl at 46 C or 21 C developed significantly less brown rot after 6 weeks' storage at 0 C and after holding in air at 18 C than untreated fruit. Fruit treated in 52 C water developed less decay than untreated fruit, but often were injured severely. Treatment at 46 C avoided injury, but only partially controlled decay. Decay usually was higher on fruit held at 18 C after storage than on fruit held at 18 C immediately after harvest.

Further evidence of geographic variation in Cronartium fusiforme. G. A. SNOW, A. G. KAIS, & R. J. DINUS (U.S. Forest Serv., USDA, Gulfport, Miss.). Fourteen open-pollinated families of slash pine were inoculated with *Cronartium fusiforme* derived from aeciospores collected along an east-west transect and two north-south transects in the southern United States. *Quercus nigra* seedlings (southern Mississippi seed source) were employed as the alternate host for maintenance of rust cultures. In the first experiment, rust cultures were established from Texas, Louisiana, Mississippi, Georgia, and Florida. Ten pine families were inoculated with cultures from each state. Two families were resistant to all cultures; three were uniformly susceptible; and the remaining five families were resistant to cultures from some states but susceptible to those from others. In the second experiment, one north-south transect was in Mississippi and the other was in Florida and Georgia. Aeciospores were collected near the mouth of the Pearl River, near Saucier and at Laurel, Miss. To the east, spores were collected near Perry, Fla., and Bainbridge and Macon, Ga. The response of four pine families to these cultures of *C. fusiforme* revealed pathogenic differences between the eastern and western sources of the fungus and between rust cultures within both north-south transects. This is further evidence that source of inoculum must be considered in determining resistance to fusiform rust in slash pine.

Efecto del tratamiento de cobre y nutrientes foliares en la calidad del café. CARLOS A. SOTO y RICARDO A. RODRÍGUEZ (Min. de Agr. y Ganadería, San José, Costa Rica y Kennecott Copper Corp., Houston, Tex.). Por espacio de 5 años se ha venido investigando en Costa Rica un fungicida a base de cobre (Kocide 101, hidróxido de cobre 83%) y aplicaciones foliares de nutrientes, tales como urea, Nu-Z y Polyboron, en sus efectos sobre el combate de la chasparria o mancha de hierro, causada por *Cercospora coffeicola*. En 1971, estas mezclas también se evaluaron en su efecto sobre la calidad del licor y los rendimientos de cosecha. La calidad, medida en base a cataciones por aroma, cuerpo y acidez fue significativamente mejor que el testigo para Kocide y algunas mezclas comunes. Resultados similares fueron observados en cuanto a control y a rendimientos de cosecha. Se considera que la acción benéfica del fungicida en la calidad, reside en el control de la chasparria, lo cual

permite el desarrollo y maduración completa del fruto. A esta acción, debe sumarse el efecto nutricional de algunos elementos incluidos en las mezclas.

Properties of tomato aspermy virus. R. STACE-SMITH & J. H. TREMAINE (Can. Dep. Agr., Vancouver, B.C.). The yields of tomato aspermy virus in sap from *Nicotiana tabacum*, *N. clevelandii*, and *N. glutinosa* were estimated at 2 mg/ml, 2 mg/ml, and 1 mg/ml, respectively. We purified virus by homogenizing leaves in 0.5 M citrate buffer, pH 6.5, and chloroform in the proportion of 1:1:1, followed by alternate low- and high-speed centrifugation and sucrose-gradient centrifugation. The purified virus exhibited a single peak when examined in the analytical ultracentrifuge, with a sedimentation coefficient of 97 S. The nucleic acid constituted 17.7% of the virus weight. The molar per cent nucleotide composition of the viral RNA was G=23.7, A=26.4, C=21.2, and U=28.7. The molecular weight of the virus particle was 5.3 million. Electrophoresis on polyacrylamide gel indicated a protein subunit molecular weight of 26,000, and the number of protein subunits per virus particle was calculated to be 167.

Induced susceptibility in pepper to Xanthomonas vesicatoria. R. E. STALL, J. A. BARTZ, & A. A. COOK (Univ. Fla., Gainesville). A macroscopic lesion (hypersensitivity) occurs in pepper leaves after inoculation with an avirulent strain of *Xanthomonas vesicatoria* at concentrations of 10^6 - 10^9 cells/ml. Rate of lesion development was estimated from electrolyte leakage from inoculated tissues. Hypersensitivity was dominant when equal numbers of avirulent and virulent cells were mixed and used as inoculum. Hypersensitivity was also dominant when virulent cells exceeded avirulent cells by as much as 100 times. However, inoculation with virulent cells at least 6 hr before inoculation with avirulent cells resulted in a delay of hypersensitivity. Varying amounts of delay occurred, depending upon the concentrations of the two kinds of cells and the period between the two inoculations. A similar phenomenon was noted when avirulent cells were inoculated into leaves that had received heat-killed virulent cells at least 24 hr earlier. However, the latter results were inconsistent. Results could be interpreted to mean that virulent cells block hypersensitivity and therefore induce susceptibility.

Exogenous nutrient requirements for germination of Pythium aphanidermatum oospores. M. E. STANGHELLINI (Univ. Ariz., Tucson). In vitro germination of snail-ingested oospores of *Pythium aphanidermatum* consisted of two distinct stages: (i) absorption of the endospore wall which was dependent upon an exogenous source of calcium (2-200 μ g/ml); and (ii) production of a germ tube which was dependent upon an exogenous carbon source. Percentage germination in sterile distilled water (SDW) containing 100 μ g/ml calcium plus 1% of the following were: dextrin (94%); levulose (93%); maltose (95%); starch (94%); sucrose (92%); galactose (86%); arabinose (75%); dextrose (74%); and lactose (61%). No increase in percentage germination over the controls (SDW containing 100 μ g/ml calcium) (6%) occurred in 1% solutions of mannitol, mannose, raffinose, rhamnose, or sorbitol. Ninety-four per cent germination occurred in 1% solutions of sucrose at pH 6.0 to 8.0 (0.05 Tris[tris(hydroxymethyl) amino methane]-malate buffer). Fifty-four and 22% germination occurred at pH 5.2 and 8.6, respectively.

³⁵S uptake by Triticum aestivum and transfer to Erysiphe graminis f. sp. tritici during primary infection. R. E. STUCKEY & A. H. ELLINGBOE (Mich. State Univ., East

Lansing). Six-day-old wheat plants with no known dominant *Pm* genes for resistance were inoculated with conidia of *Erysiphe graminis* f. sp. *tritici* (Strain MS-1). At various times after inoculation, seedlings were severed at the base of the primary leaf and the cut ends immersed in a solution of 100 μ g/ml $H_2^{35}SO_4$ in 0.1 M Na-K- PO_4 buffered at pH 6.9. Leaves were incubated 5 hr in the presence of the ^{35}S in light (1.1×10^4 ergs $cm^{-2} sec^{-1}$) or in darkness at 22 C. Radioactivity was determined using 1-cm leaf sections, epidermal strips, and the fungus on the leaf surface. Leaf sections and epidermal strips incubated in light yielded 20 and 4.5×10^4 CPM, respectively, more than twice the CPM when incubated in darkness. This difference was correlated with a greater transpirational uptake of labeled solution under light conditions. CPM in the fungus on the leaf surface ranged from background to 2,500. The rates of ^{35}S transfer from host to parasite were affected by the amount of tracer absorbed by the plant, the stage of development of the parasite during primary infection, and by host genes for compatibility. There were differences in ^{35}S transfer between four different parasite/host genotypes for incompatibility (*PL/Pml*, *P2/Pm2*, *P3/Pm3*, and *P4/Pm4*) after correction for tracer uptake by the plant.

The effect of nematodes and crop sequence on Fusarium wilt of watermelon. D. R. SUMNER & A. W. JOHNSON (Univ. Ga. Coastal Plain Exp. Sta., Tifton). Eight cultivars of watermelon were grown in a greenhouse in soils from 22 fields planted to watermelons at least once during the previous 10 years. Soils were assayed for plant-parasitic nematodes and *Fusarium oxysporum* propagules before planting. When plants were 49-54 days old, roots were indexed for galls and each soil was assayed for nematodes. There was a significant positive correlation of wilt with root knot nematode, inoculum density (ID) of *F. oxysporum*, and the cultivar previously grown. In 10 soils not cropped to watermelons during the preceding 3-10 years, no root galls were observed on plants, and only 5% of the plants wilted in four soils, whereas in six soils where plants were galled, 29% of the plants wilted. In seven soils with a *F. oxysporum* ID of 200-500 propagules/g (PG), 11% wilt was noted as compared to 26% in 15 soils with an ID of 522-6542 PG. In eight soils where plants were free of root galls and no larvae of *Meloidogyne* spp. were recovered, an average of 10% wilt occurred in three cultivars resistant to wilt in contrast to 23% wilt in 14 soils containing root knot nematodes. In methyl bromide-treated soil artificially infested with root knot nematodes, all cultivars were susceptible to *M. incognita*, *M. arenaria*, and *M. javanica*.

After infection control of apple scab with phenylmercuric acetate and triarimol. M. SZKOLNIK (N.Y. State Agr. Exp. Sta., Geneva). Unsprayed greenhouse-grown Rome Beauty apple trees were inoculated with conidia of *Venturia inaequalis* at 65,000 spores/ml. With an infection period of 1 day at 18 C, 38 scab lesions developed/leaf with 53,000 spores/lesion; within a period of 2 days there were 82 lesions with 129,000 spores/lesion. Phenylmercuric acetate (PMA), 30 ppm, applied 1 and 2 days after inoculation allowed no scab development. PMA, 60 ppm, applied 3, 4, and 7 days after inoculation allowed 0, 21, and 80 lesions/leaf, respectively, but these were chlorotic only, and produced no spores. PMA, 60 ppm, applied 8 and 9 days after inoculation, allowed 88 lesions/leaf which were chlorotic or burned out by the PMA, and which produced only 45 spores/lesion. Lesion burning with PMA was evident within 6 hr of spraying. Triarimol, 40 ppm, applied (as EL 273 25W), 1 and 2 days after inoculation allowed 1 lesion/leaf with 468 and 3,348 spores/lesion, respectively. Triarimol applied 3 and 4

days after inoculation allowed 6 or less lesions/leaf which produced no spores; that applied 7, 8, and 9 days after inoculation allowed 46, 48, and 60 lesions/leaf with 85, 4,299, and 2,429 spores/lesion, respectively. These lesions were mostly chlorotic; triarimol did not burn out scab lesions as did PMA.

Relationship of brown stem rot resistance to soybean mosaic virus infection in soybeans. H. TACHIBANA & L. C. CARD (ARS, USDA, Iowa State Univ., Ames). Five lines of soybeans (P.I. lines 95.769, 90.138, 88.820N, 86.150, and 84.946-2) known to be resistant to brown stem rot (BSR), caused by *Cephalosporium gregatum*, were compared in the greenhouse for reaction to BSR with the susceptible cultivar Clark 63 and two susceptible P.I. lines (171.434 and 69.507). The BSR-resistant lines all developed dark green mottled and ruffled leaves typical of plants infected with soybean mosaic virus (SMV), whereas the BSR-susceptible lines showed no such symptoms. The seed stocks from which the BSR-resistant lines had been taken all showed high percentages of mottled seed, an indication of SMV infection, whereas seeds of BSR-susceptible lines had few or no mottled seeds. Leaf extracts from BSR-resistant lines induced a positive SMV reaction in the local lesion host Kentucky Wonder Wax Pole beans; extracts from BSR-susceptible lines did not. Plants grown from mottled seed of a given line developed about one-half as much BSR as plants from nonmottled seed of the same line when artificially inoculated. The highly BSR-susceptible Ontario soybeans developed only one-half as much BSR when inoculated with both SMV and *C. gregatum* as when inoculated with *C. gregatum* alone.

Natural biological control of oak wilt in Arkansas. F. H. TAINTER (Univ. Ark., Fayetteville). Of 900 trees of five oak species inoculated in April 1971 with *Ceratocystis fagacearum* and harvested at monthly intervals, isolations from trunk sapwood were positive from only seven trees. Normal pressure pads were produced in November-December on three trees, and partially disintegrated pads were found in two other trees in February. Within 2 months after inoculation, several species of *Hypoxylon* colonized the trunk sapwood of wilting southern red and black oak trees (*Erythrobalanus*), producing a yellow decay. *C. fagacearum* was apparently unable to compete with the *Hypoxylon* spp. Isolations were much higher from twigs than trunk sapwood, but decreased as *Hypoxylon* colonized the twigs. Twig isolations were successful from white and post oak (*Leucobalanus*), but not from trunk sapwood. Scattered branches of many trees of the *Leucobalanus* group showed wilt, but few trees expressed general wilt. Neither pads nor *Hypoxylon* were observed on wilted stems of the *Leucobalanus* group. In blackjack oak (*Erythrobalanus*), twig isolations were highly successful, trunk isolations were not, and the stems were not colonized by *Hypoxylon*. Local spread of oak wilt through root grafts was not observed from the inoculated trees.

Light and electron microscopy of resistant and susceptible alfalfa roots infected by Meloidogyne hapla. B. A. TAIT & W. M. HESS (Brigham Young Univ., Provo, Utah). Resistant (65-410 and 65-392) and susceptible (breeders Lahontan) alfalfa seedlings were infected with the northern root knot nematode, *Meloidogyne hapla*. Infected roots were fixed, stained, and embedded in plastic for ultrastructural investigations. Sections of plastic embedded tissue were stained with 0.6% toluidine blue for 10-15 min for light microscopy investigations. In the susceptible cultivar, significant proplastid changes and abnormal mitochondrial

complexes were observed in the giant cells. The resistant plant responded to the nematode by a hypersensitivity reaction followed by extensive wall buildup and diffuse sclerotization, and nematodes were walled off by the sclerotized cells.

Relationship between the degree of resistance to a pulsed electric current and wood in progressive stages of discoloration and decay in living trees. T. A. TATTAR & A. L. SHIGO (U.S. Forest Serv., Portsmouth, N.H.). Twenty-seven living trees containing columns of wood in progressive stages of discoloration and decay were subjected to a pulsed electric current. The degree of electrical resistance of the wood was correlated primarily with its moisture and mineral contents, and thus, its stage of deterioration. Below the fiber saturation point, the resistance was correlated primarily with the amount of moisture; above the fiber saturation point, resistance was correlated primarily with the concentration of mobile potassium and calcium ions. The apparatus used was accurate in detecting and in indicating the stage of deterioration of discolored and decayed wood. The apparatus was also found to be accurate electrically within the range of resistances encountered in the trees studied.

Effect of field fungicide programs on mycoflora incidence in pecan kernels. J. TAYLOR & R. E. WORLEY (Univ. Ga., Athens). Interest in mycoflora associated with pecans has intensified due to emphasis on nut quality as related to kernel color and mycotoxins. Preliminary investigations during 1970 indicated that nuts from sprayed orchards were of better quality and showed lower mold incidence than did nuts from unsprayed trees. Kernel halves from pecans produced during 1971 were surface-sterilized in 0.5% sodium hypochlorite and plated on rose bengal agar and on water agar. Fungal counts were essentially the same for each medium. Nuts harvested during November from unsprayed trees and from trees sprayed with Cypres (dodine), Duter (Triphenyltin hydroxide 47.5), and Benlate (benomyl) produced fungi in 100, 58, 28, and 18% of platings, respectively. *Pestalotia gracilis*, the predominating species, was isolated from 86, 26, 4, and 6 of the platings, respectively. *Aspergillus flavus* developed from less than 1% of the platings. Nuts from the same treatments harvested during the following January showed one or more species of fungi in all platings. These investigations indicate that mold incidence is affected by the fungicide used in the field disease control program, and that delay in harvest may allow mold contamination regardless of the previous fungicide program.

Survival and germination of zoospores of Phytophthora parasitica in aquatic environments. S. V. THOMSON & R. M. ALLEN (Univ. Ariz., Tucson). Zoospores of *Phytophthora parasitica* are spread in irrigation water in Arizona. They retained motility for 20 hr in nontreated or sterilized waste water from citrus groves, but only 7 hr in sterile distilled water (SDW). Agitation in SDW, glucose (5-1,000 mg/liter), or orange peel in SDW, caused zoospore encystment and sedimentation. Cysts eventually germinated in most media, but in low nutrient (<5 mg/liter glucose), germling growth ceased, protoplasm contracted, and pseudo-septa formed. Empty cysts or germ tubes often lysed; remaining parts containing protoplasm survived 40 days at 25 C in nontreated waste water and grew normally if placed on selective media. Cyst germlings grown in high nutrient (10-1,000 mg glucose/liter SDW) often produced appressoriumlike structures when in contact with the container surface. When nutrient solution was replaced by nontreated waste water, appressoriumlike structures usually germinated to produce microsporangia-yielding single zoospores. Mycelial inoculum

from these zoospores was pathogenic to roots of citrus seedlings. Microsporangia survived in nontreated waste water at 25 C for 60 days.

Efficacy of low concentrations of benomyl for control of postharvest decays of oranges. L. W. TIMMER (Tex. A&M Univ. Citrus Center, Weslaco). Benomyl at 50, 100, and 250 ppm and TBZ [2-(4-Thiazolyl)-benzimidazole] at 100 and 250 ppm were applied to Marrs Early oranges 13 and 60 days preharvest. Biofilm (0.5 ml/liter) was added to all treatments. One control was sprayed with Biofilm, and another left unsprayed. Harvested fruit was stored under ambient conditions (average temperature 20 C) for 4 weeks, and per cent loss to *Penicillium* decays and stem-end rot assessed. All benomyl treatments and TBZ (250 ppm, 13 days preharvest) were equally effective and reduced decay significantly below that of the Biofilm control. Benomyl (50 ppm) and TBZ (250 ppm, 13 days preharvest), however, did not reduce decay significantly below that of the unsprayed control. TBZ (100 ppm and 250 ppm, 60 days preharvest) did not reduce decay. Benomyl was also applied as a postharvest dip to Hamlin oranges at 0, 10, 25, 50, and 100 ppm. Fruit was stored and decay evaluated as above. Rates of 10-100 ppm were equally effective, and significantly reduced decay. While low rates of benomyl might be less effective under other conditions, currently recommended rates of 600-1,200 ppm seem unnecessarily high.

Spectral reflectance measurements of St. Augustine decline disease. R. W. TOLER, N. K. SHANKAR, & W. C. ODLE (Tex. A&M Univ., College Station, Lockheed Electronics Co./NASA, Houston). St. Augustine grass (*Stenotaphrum secundatum*, a host for St. Augustine decline (SAD) virus, was used to study spectral reflectance properties to differentiate healthy and virus-infected grass by remote sensing techniques. Laboratory measurements of spectral reflectance using a Cary-14 spectrophotometer showed higher reflectance variance around 4400A and 6900A. Green:blue and green:red ratio was higher for SAD-infected grass as compared to the healthy grass. This data was verified by multiband photographic techniques using an array of 4 Nikon cameras and various film/filter combinations. Polarized spectral reflectance characteristics were also measured. Strong polarization differences were observed in the blue, red, and near-IR regions, whereas relatively no polarization differences were observed in the green band. Analysis of polarization and reflectance data revealed a good correlation differentiating the healthy from SAD-infected grass.

The serological relationship between peanut stunt virus and cucumber mosaic virus. S. A. TOLIN & S. BOATMAN (Va. Polytech. Inst. & State Univ., Blacksburg, & Hollins College, Va.). Peanut stunt virus from Virginia (PSV-V), North Carolina (PSV-NC), and Washington (PSV-W), a virus from crown vetch (CVV), and cucumber mosaic virus (CMV) strains S and Y were compared by gel diffusion and density-gradient serology. In gels, precipitin bands were sharpest using 0.7% Ionagar with 0.01-0.10% Na₂S₂O₃ and 0.10-0.50 mg/ml of virus. Density-gradient serology was done by the incubating of antigen-antibody mixtures for 5 min in the cold prior to centrifugation and analysis. The highest antiserum dilution removing all of the homologous antigen was used in heterologous reactions. Antisera to PSV isolates and to CVV reacted with PSV-V to form bands that fused completely, but did not react with CMS-S. In density-gradient analyses, incubation of each of the viruses with anti-CMV-Y resulted in complete removal of PSV-V and CMV-S, but no removal of PSV-NC, PSV-W, or CVV. PSV-V

was related closely to PSV-NC, PSV-W, and CVV as shown by complete removal of heterologous antigen in reciprocal tests, but only partially to CMV-Y and distantly to CMV-S. PSV-NC and PSV-W were related closely to each other and to CVV, but showed little or no relationship to CMV-Y and CMV-S. Thus, although not all PSV isolates are related to CMV-S, the virus should be included as a member of the CMV group.

Efficacy of fungicidal drenches in inhibition of Phytophthora at various soil depths. J. L. TROUTMAN & J. C. MATEJKA (Univ. Ariz., Tucson). *Phytophthora parasitica* and *Phytophthora citrophthora* cause severe root rot of citrus in Arizona. Since most of this citrus is grown on Superstition sand, penetration by fungicidal solutions seemed likely. Downward movement of 11 selected fungicides through *Phytophthora*-infested soil was determined using columns, 45 cm long, of 2.5-cm-diam polyvinylchloride tubing arranged in 5-cm segments. Fungicidal solutions (180 ml each; concentration 100 µg/ml) or sterile water were percolated through firmly packed columns. After 48 hr of infiltration the columns were disassembled by segment and soil from each segment was removed and mixed with 800 ml of water in a beaker. Six citrus leaf-pieces ca. 1 cm² were floated in each beaker as bait for *Phytophthora* to determine efficacy of each fungicide at various soil depths. Periodically thereafter, a leaf disc was removed and examined microscopically for presence of sponangia. This test showed that water and three fungicides failed to inhibit the fungus at any level, four fungicides were effective through the first 5-cm segment, two were effective through the second segment (10 cm), and two were effective through 45 cm of the column.

The biological control of Cassia surattensis brush weed of pastures in Hawaii with Cephalosporium sp. E. E. TRUJILLO & F. P. OBRERO (Univ. Hawaii, Honolulu). A severe vascular wilt of kolomona (*C. surattensis*) on Kauai is caused by *Cephalosporium* sp. Typical symptoms of the disease appeared in wound-inoculated young plants in less than 4 weeks. Chlorosis and wilting resulting in plant death were characteristic. Xylem discoloration and plugging of vessels were indicative of the vascular nature of the disease. The time required for the pathogen to kill the host depended on age of plant and moisture availability. In the presence of adequate soil moisture, 3-month-old seedlings usually succumbed in less than 10 weeks, and large bushes 4 to 5 years old died, in 3 to 4 months. Prolonged dry periods hastened plant death. A host range study involving six *Cassia* spp. and one *Leucaena* sp. demonstrated pathogenic specificity of this *Cephalosporium* sp. to (*C. surattensis*) Introduction of disease to three localities of Kauai by wound inoculations of healthy host with a spore suspension of the pathogen demonstrated that effective eradication of *C. surattensis* is possible in 1 year. Once the pathogen is introduced into a new area, slow natural spread occurs.

The effect of benomyl, oxycarboxin, captan, and triarimol and the metabolism of ¹⁴C-labeled glucose and acetate by Fusarium oxysporum. B. G. TWEEDY & MARY QUADE (Univ. Mo., Columbia). Microconidia of *Fusarium oxysporum* were incubated with ¹⁴C-glucose labeled in the 1, 2, 3, and 4 or 6 positions with ¹⁴C-acetate labeled in the 1 or 2 position. The fungicides were added at the beginning of the incubation period. The carbon dioxide evolved from the cultures was collected and quantitatively assayed for radioactivity at 30-min intervals for 5 hr. Benomyl, oxycarboxin, and triarimol had no significant effect upon the rate of metabolism of the selectively labeled glucose or

acetate substrates. Depending upon the position of the labeling, captan caused a 64 to 78% reduction in ^{14}C -carbon dioxide evolution as compared to the controls. Captan was equally inhibitory to the glycolytic and pentose pathways. With the exception of benomyl, the effects of the four fungicides on glucose and acetate metabolism correlated with their respective effects on germination.

Intraspecific transformation of Pseudomonas syringae. W. TWIDDY and S. C. Y. LIU (Mich. Univ., Ypsilanti). Prototrophic *Pseudomonas syringae* was grown in broth with mutagenic nitrosoguanidine ($100\ \mu\text{g/ml}$) for 20 min at 24 C. Treated cells were plated on Czapek's medium, to which penicillin (2,000 units/ml) was added. Plates showing growth were discarded. To plates showing no growth, amino acids were added to induce colony formation of auxotrophs. Five complementary auxotrophic mutants ($\text{Leu}^+\text{Val}^+\text{Thr}^-\text{Met}^-$), ($\text{Leu}^-\text{Val}^-\text{Thr}^+\text{Met}^+$), ($\text{Asp}^-\text{Pro}^+\text{Cys}^+$), and ($\text{Asp}^+\text{Pro}^+\text{Cys}^-$) were obtained. To washed cells ($1 \times 10^6/\text{ml}$) in 5 ml of Na-citrate saline (SCS) at pH 7.3, 0.5 ml of Na lauryl sulfate (20% in SCS), 1.25 ml of Na-perchlorate (5 M) and 6 ml of CHCl_3 -*n*-butanol were added for deproteinization. The mixture was allowed to react for 20 min and treated with CHCl_3 -*n*-butanol for phase separation. To the aqueous phase, 5 ml of phenol-saturated SCS were added. Denatured protein was extracted into phenol, whereas nucleic acid remained in the aqueous layer. DNA in aqueous phase was precipitated by 95% alcohol at cold, and collected. Auxotrophic cells were grown on Czapek's agar with amino acids added. To young colonies, extracted DNA of complementary auxotroph-donors was added. They were incubated for 2 days. Harvested cells were plated on Czapek's agar without amino acids. This was to check for transformants. Results demonstrated that transformants ($\text{Leu}^+\text{Val}^+\text{Thr}^-\text{Met}^+$) or ($\text{Asp}^+\text{Pro}^+\text{Cys}^+$) were effected by DNA of complementary auxotrophs.

A re-examination of the relationship between Helminthosporium turcicum infection and frost sensitivity of maize. C. D. UPPER, D. C. ARNY, & F. C. VOJTIK (ARS, USDA, Univ. Wis., Madison). It has been reported from field observation that inoculation with *Helminthosporium turcicum* increased the susceptibility of maize to frost damage as compared to controls. In our work, three-leaf-stage maize seedlings were inoculated and held in a mist chamber 24 hr in the dark before they were frozen in a controlled environmental chamber. Plants inoculated with powdered, dried, infected corn leaves, as was done in the field, were significantly more susceptible to frost than noninoculated controls. Seedlings inoculated with washed spores of *H. turcicum* were not more frost sensitive than controls. Corn, sham-inoculated with noninfected corn powder, was more sensitive to frost than were controls, but not different from plants inoculated with noninfected corn powder and spores simultaneously. Corn powder also increased the frost sensitivity of lettuce. Application of corn powder 0-1 hr before placing the plants at low temperature made both corn and lettuce plants even less sensitive than controls. Thus, infection does not change the frost sensitivity of corn, but corn powder can change the frost sensitivity of both corn and lettuce.

Distribution of Entyloma smut of beans in Central America. N. G. VAKILI (USDA, Mayaguez, Puerto Rico). Smut of beans (*Phaseolus vulgaris*) caused by an *Entyloma* sp., occurs in all bean-growing areas of Guatemala, El Salvador, Honduras, Nicaragua, and Costa Rica surveyed for bean diseases during 1971. No smut lesions were observed on beans cultivated in the Cauca and Bogota valleys of Colombia

or bean farms in the highlands of Chiriqui province in northern Panama. This observation suggests that Costa Rica is the southern limit for the spread of this disease. Bean smut has not been reported from as far north as Mexico. The 1971 survey revealed that smut and common bacterial blight were two diseases occurring with greatest frequency in Central America. Smut lesions were found in association with bacterial, viral, and fungal diseases encountered on beans. Usually, only the primary leaves or first and second trifoliate leaves were infected. However, 40-60% total foliage infection was not uncommon. The disease was observed on all but one of 81 Central American selected bean cultivars observed at El Paraiso, Honduras. Smut occurred on all three bean crops grown during the year in Central America. This fungus should be studied in greater detail, and its potential for future epiphytotic should be investigated.

Microbial metabolism of the fungicide, 2,6-dichloro-4-nitroaniline. N. K. VAN ALFEN & T. KOSUGE (Univ. Calif., Davis). Unidentified bacteria isolated from the surface of ripe peaches were screened for their ability to metabolize the fungicide 1,6-dichloro-4-nitroaniline (DCNA) in liquid culture. After 48-hr incubation, 24% of the isolates completely metabolized $2\ \mu\text{g/ml}$ ^{14}C -DCNA, and accumulated one major metabolite in the culture fluid. The metabolite was isolated and identified as 4-amino-3,5-dichloroacetanilide (ADCAA) by mass spectrometry, proton magnetic resonance spectroscopy, and infrared spectroscopy. The structure of the metabolite was confirmed by spectroscopic comparison with chemically synthesized ADCAA. At least four additional metabolites were accumulated to a lesser degree by certain of the microorganisms. A number of known plant-pathogenic and saprophytic bacteria and fungi were screened for their ability to metabolize DCNA. These experiments indicated that this pathway of metabolism is common in both bacteria and fungi. Conditions of low aeration enhanced metabolism of DCNA and accumulation of ADCAA by the bacteria tested.

Possible origin of peanut rust epidemics in Texas. E. P. VAN ARSDEL & A. L. HARRISON (Tex. A&M Univ., College Station). Since 1965, peanut rust has been important in south Texas; 1971 had the greatest epidemic. Since no overwintering hosts have been found, rust overwintered on standing dead peanuts could not infect volunteer or greenhouse peanuts, and 1 April through 30 June night temperatures were ideal for infection (15-25 C) with adequate moisture, but rust was not present; we decided to explore possible external sources of the rust. During July and August 1970, rust spores were collected with rainwater twice (22-31 July and 1-6 August) in first 4 and then 3 of 17 traps set along a 240-km line. The finding of pustules 10-15 days later (incubation period) only in those fields adjacent to the rust spore catches confirmed that the spores were probably peanut rust. The periods in which the spores were collected matched windflows at 0-600 m from Merida, Yucatan, Mexico, 27-31 July and 3-4 August with rain in Texas to bring the spores down. These winds exceeded the 40 hr at 9 m/sec required for transport over the 1,290 km. Of the five peanut-growing regions in Mexico, rust was abundant only in Yucatan where peanuts are grown throughout the year. Spore traps were not set in 1971, but the 2 July sighting of the rust matched the 21-23 June continuous wind from Merida with rain in Texas. Thus, the 1971 rust introduction data supported the 1970 transport theories. The rust is probably reintroduced annually.

Alteration of phaseollin by Fusarium solani f. sp. phaseoli. J. VAN DEN HEUVEL & H. D. VAN ETEN

(Cornell Univ., Ithaca, N.Y.). The response of the bean pathogen *Fusarium solani* f. sp. *phaseoli* to the phytoalexin phaseollin was determined in shake culture by exposing ca. 6-mg (dry wt) portions of actively growing mycelium to a 2% glucose medium containing phaseollin at 16 or 44 $\mu\text{g}/\text{ml}$. Metabolism of phaseollin ($\text{C}_{20}\text{H}_{18}\text{O}_4$) was followed by thin-layer chromatography (TLC) and/or gas-liquid chromatography. After 2- to 3-hr incubation, no phaseollin could be detected in cultures exposed to 16 $\mu\text{g}/\text{ml}$, and *Fusarium* growth was only slightly affected. After 3 hr, most of the phaseollin was still present in cultures treated with 44 $\mu\text{g}/\text{ml}$, and growth was inhibited. However, if cultures were preincubated with 16 μg of phaseollin/ml for 1.5 hr, then treated with an additional 44 $\mu\text{g}/\text{ml}$, almost no phaseollin was detected 1 hr after the second addition. Coincident with the disappearance of phaseollin, a new compound appeared which was detected by TLC ($R_F = 0.5$ on silica gel plates developed with chloroform:methanol, 25:1). This compound has an ultraviolet absorption spectrum resembling that of phaseollin, with λ max at 280 and 290 nm. Mass spectral analysis revealed an empirical formula of $\text{C}_{20}\text{H}_{18}\text{O}_5$. The compound seems to be less fungitoxic than phaseollin. We believe that the fungus detoxifies phaseollin, possibly by an inducible oxidase system.

Relationship between severity of fire blight in pear cultivars and virulence of Erwinia amylovora isolates. T. VAN DER ZWET (ARS, USDA, Beltsville, Md.). Severity of natural fire blight was measured in four trees in each of nine cultivars of *Pyrus communis*. Maximum length of blight in shoots was 1 cm in Richard Peters; 10-50 cm in Ree Carlo di Wurtemberg (RCW), Moonglow, and Waite; 1-1.5 m in Magness and Old Home; 2-3 m in Dawn and Maxine; and over 5 m in Bartlett. To determine if differences in blight resulted from strains with different virulence, isolates of *Erwinia amylovora* were obtained from these cultivars. Single colony cultures, producing typical small, white, glistening colonies on nutrient yeast-dextrose agar (NYDA), were morphologically similar. Aqueous cell suspensions (5×10^6 cells/ml) were equally pathogenic to immature Bartlett and Kieffer fruit slices and to succulent shoots of potted Bartlett trees. The original single-colony isolates were transferred weekly for 3 times on NYDA slants, then injected into Bartlett shoots. Three weeks after inoculation, the isolate from Richard Peters induced blight in the shoots measuring 15-30 cm; isolates from RCW, Moonglow, Waite, Magness, and Old Home, 31-40 cm; and isolates from Dawn, Maxine, and Bartlett, 41-50 cm. The small difference in virulence of *E. amylovora* appears to be inadequate to explain the large differences in severity of natural blight infection in the above cultivars.

Antifungal and hemolytic activities of four pterocarpan phytoalexins. H. D. VAN ETEN (Cornell Univ., Ithaca, N.Y.). The ED_{50} value for inhibition of linear mycelial growth (zero inhibition = ca. 25 mm) of *Rhizoctonia solani* by medicarpin was 0.15 mM, of *Phytophthora megasperma* var. *sojae* by hydroxyphaseollin was 0.21 mM, of *Fusarium solani* f. sp. *curvibitae* by phaseollin was 0.05 mM, and of *Helminthosporium turcicum* and *F. solani* f. sp. *curvibitae* by pisatin were, respectively, 0.19 and 0.10 mM. Shake cultures containing ca. 6 mg (dry wt) of each of the above mycelia in 4.0 ml of media, exposed to twice their ED_{50} concentrations of phaseollin, hydroxyphaseollin, and medicarpin (phenolic pterocarpan), underwent a loss in dry weight. At 2-3 times the ED_{50} values, pisatin (a nonphenolic pterocarpan) failed to induce a loss in mycelial dry weight. Phaseollin, hydroxyphaseollin, and medicarpin, at concentrations ≤ 0.35 mM, caused lysis of bovine

erythrocytes, whereas pisatin was not hemolytic at concentrations up to 0.5 mM. Dehydrophaseollin, a phenol, was hemolytic, whereas phaseollin derivatives in which the phenol group had been methylated or acetylated caused little hemolysis. The membrane-damaging potential previously suggested for phaseollin may be due to the presence of the phenol group and, by analogy, a portion of the antifungal activity of the phenolic pterocarpan may be attributed to phenol toxicity.

Differential synthesis of proteins during the germination of Botryodiplodia theobromae spores. J. L. VAN ETEN, H. R. ROKER, & E. DAVIS (Univ. Neb., Lincoln). During the initial stages of *Botryodiplodia theobromae* spore germination, protein synthesis occurs in the absence of detectable RNA synthesis. The nature of this early protein synthesis, which is presumed to be directed by a preformed messenger RNA (mRNA) in the spore, and which is apparently essential for germination, is not known. A spore suspension was incubated with ^3H -amino acids from 0 to 1 hr, and a separate sample was incubated with ^{14}C -amino acids from 4 to 5 hr (90% germination). Soluble proteins were isolated from the combined labeled spores and separated by polyacrylamide gel electrophoresis. To determine whether the proteins synthesized during the first hour of germination were stable or rapidly turned over, a pulse-chase experiment was conducted. Three conclusions are apparent from the results: (i) spore mRNA codes for the synthesis of several proteins; (ii) spore mRNA directs the synthesis of at least one protein which is not synthesized at 4 to 5 hr; and (iii) most proteins synthesized early in germination are stable.

A benzimidazole-resistant strain of Erysiphe graminis. J. M. VARGAS, JR. (Mich. State Univ., East Lansing). Systemic benzimidazole fungicides normally give excellent control of powdery mildew, *Erysiphe graminis*, on Kentucky bluegrass (*Poa pratensis* 'Merion'). During 1971, however, powdery mildew began to occur on turf plots treated with benzimidazole fungicides. Infected plants from the treated plots were transferred to a growth chamber, and the inoculum was allowed to increase. Healthy Merion plants which received two 50-ml, 100 $\mu\text{g}/\text{ml}$, drench applications at 2-week intervals of benomyl, thiabendazole [2-(4-Thiazolyl)-benzimidazole], thiophanate-methyl [1,2-bis(3-methoxycarbonyl-2-thiouredido)-benzene], EL-273 [α -(2,4-Dichlorophenyl)- α -phenyl-5-pyrimidinemethanol], and water were inoculated after the first application with the suspected benzimidazole-resistant powdery mildew. Another set of plants was inoculated after the first application with common (nonbenzimidazole-resistant) strains of *E. graminis*. All plants inoculated with the benzimidazole-resistant strain of *E. graminis* became 100% infected except those treated with EL-273, which remained disease-free. All fungicide-treated plants exposed to common (nonbenzimidazole-resistant) strains of *E. graminis* remained disease-free, whereas untreated controls became 100% infected.

An air-quality indicator plant network in Georgia. J. T. WALKER & J. C. BARLOW (Univ. Ga. Agr. Exp. Sta., Experiment, Ga. Dep. Public Health, Atlanta). To evaluate the effect of ambient ozone on plants in Georgia, a biological monitoring system was established in 1971 at seven locations. At monthly intervals, nine different greenhouse-grown indicator plants were placed in once-treated (4,4'-dioctyl diphenylamine) and nontreated 56- x 50- x 50-cm cheesecloth chambers. After exposure, height and fresh and dry weight were recorded. Bel W3 tobacco, Show Princess gladiolus, and Snowstorm, petunia were the only plants in

the group with consistent growth differences in the two chambers. Average seasonal height and weight of tobacco from the treated chambers exceeded those from the nontreated chambers at six and four locations, respectively. Gladiolus height and weight were greater from treated chambers at five locations; petunia height and weight were greater at three locations. Typical ozone injury occurred on tobacco in untreated chambers but not in treated chambers until the latter half of the season. Fluoride injury occurred in gladiolus, particularly at one location. Information obtained is being correlated where possible with sampling data from mobile units of the Air Quality Evaluation Service, Ga. Dep. Health.

β -Glucosidase in vascular browning in Fusarium wilt of susceptible and resistant tomato isolines. J. V. VEECH & M. E. MACE (ARS, USDA, Beltsville, Md.). The sites of localization and the relative activity of β -glucosidase were studied in the stems of noninoculated and inoculated, susceptible or resistant, isolines of tomato. The enzyme could not be demonstrated histochemically in the vascular tissue of noninoculated stems of either cultivar; however, in inoculated stems of both cultivars, the enzyme was detected in xylem vessels infected with the fungus. Localization at these sites suggested that β -glucosidase was an extracellular enzyme of the fungus. Spectrophotometric assay of cell-free culture filtrates of the fungus supports this observation. The absence of free oxidizable phenols in noninoculated tomato stems, and the presence of *o*-dihydric phenols in *Fusarium*-infected stems, suggest that the enzymatic hydrolysis of phenolic glycosides is involved as a preliminary step in vascular browning. Our data supports the hypothesis that phenols involved in vascular browning are derived from pre-existing glycosides. Furthermore, in *Fusarium* wilt of tomato, we maintain that the hydrolysis of the phenolic glycosides is accomplished largely or entirely by an extracellular enzyme of the pathogen.

Properties of cherry leaf roll virus. D. G. A. WALKER & R. STACE-SMITH (Can. Dep. Agr., Vancouver, B.C.). Four isolates of cherry leaf roll virus originating from cherry, rhubarb, elderberry, and elm were purified and their properties compared. The isolates were propagated in *Nicotiana tabacum*, *N. clevelandii*, or *N. rustica*. The best propagation host was *N. clevelandii*, with optimal harvest dates 11-14 days after inoculation. Preliminary purification was by a homogenizing of infected leaves in 0.5 M citrate buffer, pH 6.4, and chloroform at the rate of 1:1:1. The clarified extract was concentrated by differential centrifugation or by the addition of 6% polyethylene glycol. Purified virus preparations of each isolate exhibited two major peaks in the analytical centrifuge, with sedimentation coefficients of 114 and 132 S. A minor peak sedimenting at 50 S was observed when highly concentrated preparations were examined. The components of each isolate were separated on a sucrose density gradient and were found to be serologically identical. All four isolates were serologically related; spur reactions indicated varying degrees of relationship. The virus protein of each strain had a molecular weight of 54,000 as determined by polyacrylamide gel electrophoresis.

Translocation of benomyl through the carpel wall and bracts of cotton bolls. S.-Y. C. WANG & J. A. PINCKARD (La. Agr. Exp. Sta., Baton Rouge). The apical half of detached cotton bolls, with and without bracts, were immersed in a suspension of 100 μ g/ml (active) of benomyl for 24 hr at room temperature. Significant quantities of benomyl were detected in the extracts of washed immersed

and unimmersed parts of carpel walls and in attached bracts and calyx tissues. We determined presence of benomyl in the extracts by noting the fungistatic action of the extracts on freshly formed spores of *Diplodia gossypina* which are sensitive to benomyl at 0.2 μ g/ml (active) and upward. Additionally, the presence of benomyl in boll tissues was confirmed by disc bioassay and thin-layer chromatography. Extracts of the lint and peduncle showed no fungistatic activity. Extending the immersion time to 4 days resulted in slight inhibition of spore germination on the surface of endocarp tissue and lint. Fungistatic activity of the calyx extract was greatly reduced when the bracts were removed before immersion, suggesting a functional relationship. The significance of these results lies in the potential use of systemic fungicidal formulations applied to boll and plant surfaces at the onset of dew formation.

Anaerobic bacteria associated with honeycomb and ring failure in red and black oak lumber. J. C. WARD (U.S. Forest Products Lab., Madison, Wis.). Lumber from northern red oak (*Quercus rubra*) and black oak (*Q. velutina*) trees infected with anaerobic heartwood bacteria is highly susceptible to costly internal checking (cracking) when dried. These checks occur mostly as honeycomb (HC) or internal wood separations along the rays, and as ring failure (RF) or separations tangential to the annual growth rings. Varying strains of strictly anaerobic, gram-positive to gram-variable *Clostridium* spp. having terminal to subterminal spores were consistently isolated from both clear and discolored heartwood that subsequently developed either HC or RF. Anaerobic, gram-positive bacteria commonly isolated along with *Clostridium* from clear and discolored heartwood were nonsporulating, and produced both rod and coccoid cells. *Bacillus cereus*, a facultatively anaerobic bacterium, and the nonhymenocetous fungus *Paecilomyces varioti* were frequently isolated from discolored heartwood. Only green (unseasoned) heartwood infected with *Clostridium* alone or in association with other micro-organisms appears to form HC or RF during normal drying operations. Neither HC nor RF developed in clear, noninfected heartwood, even with relatively severe or accelerated drying.

Effect of preservatives, storage temperatures, and freezing-thawing on titer of southern bean mosaic virus antiserum. H. E. WATERWORTH, J. W. BLIZZARD, & R. H. LAWSON (Plant Introduction Sta., Glenn Dale, ATCC, Rockville, Plant Ind. Sta., Beltsville, Md.). Effect of methods of storing southern bean mosaic virus (SBMV) antiserum (titer = 1:4096) on antibody titer were compared. All treatments were assayed after 1 year with 2-fold dilutions of purified SBMV in microprecipitin tests. Treatments and final antibody titers were: (i) continuous freezing at -70 and -20 C (1:1024) or thawed for 2 hr 260 times (1:1024); (ii) stored with 0.02% Na₂S₂O₃ at -20 C undiluted, at 1:16 and 1:64 (1:1024), and diluted 1:512 and 1:1024 (1:4096); (iii) stored diluted 1:2 with Na₂S₂O₃ or 50% glycerine at -70, -20, +4, and +26 C (1:1024), and at +37 C with Na₂S₂O₃ (1:128) or glycerine (1:32); (iv) frozen or freeze-dried with 0.02% Na₂S₂O₃ at -20, +4, and +26 C (1:1024); and (v) freeze-dried with 0.02% Na₂S₂O₃, 5% peptone, or no additive and stored at -20 and +4 C (1:1024) and at +26 C (1:512). The antiserum titer dropped (1:32 to 1:128) in all frozen and freeze-dried treatments stored at +37 C except those containing peptone (1:1024). The titer of antiserum stored in glycerine or Na₂S₂O₃ was about the same when stored frozen; however, 0.02% Na₂S₂O₃ was better than no additive in freeze-dried antiserum in maintaining antibody titer when stored at -20 C.

Detection and identification of pathogenic strains of

Verticillium. R. D. WATSON & J. J. RENK (Univ. Idaho, Moscow). Several methods were tested to evaluate strains and virulence of *Verticillium* isolates. The host from which the fungus was isolated was of little value in determining strain or virulence to other plants. Fungus morphology; i.e., microsclerotia or dark mycelial types, were not correlated with host reaction or pathogenicity. Seedling and/or cutting pathogenicity tests were made using 20 *Verticillium* isolates representative of six species selected for their geographic and isolated host diversity. These isolates were evaluated on 8-10 host plants. All of the isolates tested were highly pathogenic to the host from which they were isolated. The isolates showed wide and often unexpected differences in pathogenicity to the other host plants used in tests. Northern or southern isolates showed no geographic specificity to hosts normally grown on a regional basis. Host reaction varied from infection with no evident symptoms, to severe necrosis, wilt, and death of the plant. The seedling or cutting host reaction seems a valid criteria useful in *Verticillium* strain differentiation.

Effect of tryptophan and dimethyl sulfoxide on sporulation of Rhizopus arrhizus. D. J. WEBER & M. GUNASEKARAN (Brigham Young Univ., Provo, Utah). Twenty amino acids (0.002 M) were added individually to Fothergill's medium to determine their ability to stimulate growth and sporulation of *Rhizopus arrhizus*. Maximum growth (dry wt) was obtained with tryptophan. Sporulation was stimulated in the dark by the addition of tryptophan or methionine. Indole acetic acid was detected in the culture medium of *R. arrhizus* grown in tryptophan. The addition of indoleacetic acid to the medium did not stimulate sporulation in the dark. In contrast to tryptophan, dimethyl sulfoxide (DMSO) (0.64 M) inhibited sporulation and spore germination. Light (white fluorescent) enhanced the inhibitory effect of DMSO on sporulation. Tryptophan provides a system for investigating the mechanism of light and nutrition stimulation, and DMSO provides a system for investigating the inhibition of sporulation in *R. arrhizus*.

The identification and metabolism of sterols of Rhizopus arrhizus. J. D. WEETE, J. L. LASETER, & G. C. LAWLER (Lunar Sci. Inst., Houston, Tex., La. State Univ., New Orleans). The lipid components previously reported for *Rhizopus arrhizus* include the isoprenoid hydrocarbon squalene, fatty acids, glycerides, and phospholipids. In addition, methyl and ethyl esters of fatty acids isolated from this organism were reported for the first time as fungal products. The 4-desmethyl sterols have now been identified by GLC/MS combination as ergosterol ($\Delta 5,7,22$ -ergostatrienol), 5-dihydroergosterol ($\Delta 7,22$ -ergostadienol), fungisterol ($\Delta 7$ -ergostenol), and $\Delta 5,7,14(15)$ -ergostatrienol. Using both $2\text{-}^{14}\text{C}$ -acetate and $2\text{-}^{14}\text{C}$ -mevalonate, the metabolism of lipophilic isoprenoid compounds was followed through a complete life cycle of the fungus. Log phase growth was completed after 72 hr, whereas the maximum rate of total lipid synthesis appeared after 45 hr. The rate of $2\text{-}^{14}\text{C}$ -acetate incorporation paralleled the rate of lipid synthesis. In contrast, the rate of $2\text{-}^{14}\text{C}$ -mevalonate incorporation and the rate of sterol synthesis was minimal during log phase growth, but reached a maximum after 140 hr of growth. Sterol synthesis appears to be associated with the onset of the sporulation process and sterol accumulation associated with aging tissues.

Initial host responses in cotton to infection by Rhizoctonia solani. A. R. WEINHOLD & J. MOTTA (Univ. Calif., Berkeley). A histochemical and morphological study was made of the initial processes of infection of cotton

hypocotyls by *Rhizoctonia solani*. Emphasis was placed on the relationship between pathogen location and host damage, and the stage of infection at which the effects of cell wall degrading enzymes could be detected. *R. solani* used in this study formed dome-shaped infection cushions on the surface of the hypocotyls. The first host response that could be detected was removal of ruthenium red positive substances, presumably pectic materials, from the cell walls. This occurred as early as 18 hr after the pathogen came in contact with the host plant. At this stage there was extensive hyphal branching, and infection cushions were beginning to form. Observation of cells below newly formed cushions, using polarized light, revealed a loss of birefringence in the cell walls, indicating damage to the crystalline structure of the cellulose. Twenty-four to 30 hr after host contact, an extensive cavity had formed beneath the cushions, but penetration had not occurred and the cuticle could be observed tightly appressed to the cushions. Later, penetration occurred by growth of numerous hyphae directly from the base of the infection cushions into the tissue macerated by the pathogen.

Aerial invasion of peanut flower tissues by Aspergillus flavus under gnotobiotic conditions. T. R. WELLS & W. A. KREUTZER (Colo. State Univ., Fort Collins). Peanut plants (cultivar Tenn. Red) were grown gnotobiotically within flexifilm isolators in pots especially designed to separate plant aerial from subterranean zones. We accomplished this by creating effective physical barriers of synthetic filter wool to maintain microbial isolation of each zone. Flowers inoculated with washed conidia of *Aspergillus flavus* and held at high relative humidities (>90%) were readily colonized by the fungus without apparent damage to the developing ovules. The fungus could be cultured from surface-sterilized pegs (1 min in 0.1% HgCl_2) prior to their penetration through the synthetic barrier, as well as from all stages of pod development after penetration into the sterile rooting zone. Based on this work we hypothesize that, under favorable environmental field conditions, *A. flavus* can colonize peanut flowers nonpathogenically during the blossom period, remaining associated with the sound, developing pod tissue until harvest.

Ultrastructural investigation of the ontogeny of Woronin bodies in Fusarium oxysporum. W. P. WERGIN (ARS, USDA, Beltsville, Md.). Woronin bodies are cytoplasmic organelles which commonly lie near septa in ascomycetous fungi. Although they were described 100 years ago, nothing is known about their origin and development. To establish the ontogeny of Woronin bodies, an ultrastructural investigation was undertaken using hyphae from *Fusarium oxysporum* f. sp. *lycopersici* race 1. In this fungus, Woronin bodies originated within microbodies. Development begins with the appearance of an electron-dense material which forms at a localized site adjacent to the membrane of the microbody. This material aggregates and condenses into a single spherical or hexagonal structure. Further aggregation and compaction of additional material results in the formation of a dense paracrystalline inclusion. During this development, the portion of the microbody containing the inclusion gradually becomes extruded, and eventually is separated from the parent microbody by an exopinocytotic mechanism. After its separation, the paracrystalline inclusion migrates to the septal pore. Although many recent electron-microscopic studies have referred to these membrane-bound inclusions as lipid granules, spherosomes, or lysosomes, in *Fusarium* these inclusions are believed to correspond to the Woronin bodies initially described by light microscopists. Hopefully, this ontogenetic study will help

clarify the origin, development, and ambiguous terminology which currently surround these organelles.

Cell wall and plasma membrane alterations in diseased and injured plants. H. WHEELER (Univ. Ky., Lexington). Alterations at the cell wall-plasma membrane interface are common in plants infected with fungal and viral pathogens. These alterations vary from small, localized invaginations of the protoplast to very large, irregularly contoured elaborations which may envelop the tip of a penetrating hypha. Cell wall modifications, similar to those in diseased plants, can be induced in plant roots exposed to uranyl acetate at moderately toxic concentrations (10^{-4} M). In such roots, the plasma membrane can be seen clearly because its over-all width and stainability are greatly increased. In cells with highly modified walls, the plasma membrane showed distinct discontinuities, whereas in adjacent cells with little or no wall modification the plasma membrane was always continuous. In both types of cells, internal cytoplasmic structures and organelles appeared normal. In untreated roots, apparent discontinuities in the plasma membrane have been seen only in secretory outer cap cells with masses of mucilaginous material between the wall and protoplast. These observations support suggestions that cell wall alterations play a role in the response of plants to infectious or toxic agents.

Specificity of the toxin(s) from Helminthosporium maydis race T to mitochondria from various plant cytoplasms. J. E. WHEELER, R. G. MC DANIEL, & R. B. HINE (Univ. Ariz., Tucson). The relationship between susceptibility of corn lines and other plants to *Helminthosporium maydis* race T, and mitochondrial sensitivity to the fungal toxin(s) were investigated as a possible method of detecting resistant plants. Toxic extracts from race T were bioassayed for potency using Pioneer 3306 corn line incorporating male sterile cytoplasm (cms-T). Mitochondrial sensitivity was determined by addition of toxic extracts to suspensions of plant mitochondria and the measuring of O_2 uptake. Mitochondria from four susceptible corn lines (B37 × H84, B37, Wf9, Pioneer 3306, all with cms-T) were highly sensitive to the toxin(s), as revealed by a marked decrease in ADP/O ratios and respiratory control ratios (RCR). Mitochondria from resistant Pioneer 3306 corn (normal cytoplasm) were less sensitive, but nevertheless, a sharp decline in ADP/O ratios and RCR occurred. In addition, the toxic extracts were found to affect the density of mitochondria from susceptible corn lines as determined on sucrose density gradients. Mitochondria from wheat with male sterile or normal cytoplasm were relatively insensitive to additions of the toxic extracts.

Ultrastructural changes in corn mesophyll cells induced by Helminthosporium maydis race T. J. A. WHITE, O. H. CALVERT, & M. F. BROWN (Univ. Missouri, Columbia). Corn seedlings, of a strain having Texas male-sterile cytoplasm, were inoculated with conidia of *H. maydis*, race T. Infected leaf tissues were sampled at 6, 12, 24, and 48 hr after inoculation. Electron-microscopic examination of these tissues revealed that the first detectable change, rupture of the tonoplast, occurred within 6 hr. By 12 hr, chloroplasts had become spherical in shape, and numerous vacuolelike bodies were present in the plastid stroma. At 24 hr, the plasma membrane had ruptured, chloroplast envelopes had become disorganized, and the mitochondrial matrix was gone. In many cells, the plastid envelopes had disintegrated, the stroma was gone, and numerous irregular, osmiophilic bodies were interspersed within the lamellar systems. By 48 hr, the chloroplast lamellae had clumped together and had

undergone extensive structural breakdown. Tissue samples taken from 48-hr necrotic lesions revealed the most advanced stages of cell breakdown. We postulate that the pathotoxin produced by race T of *H. maydis* functions by altering the structural integrity of cell membranes and membrane systems.

Electron microscopy of alfalfa mosaic virus in alfalfa leaves. R. D. WILCOXSON, F. I. FROSHEISER, & LOIS B. JOHNSON (Univ. Minn., St. Paul). Eight strains of alfalfa mosaic virus (AMV) were studied in expanded alfalfa (*Medicago sativa*) leaves. AMV was found only in mesophyll cells in cytoplasm, and occasionally in vacuoles and intercellular spaces. It was not associated with organelles. The strains were placed in five groups based on appearance of virus particles in vivo: (i) Particles were aggregated in short rafts surrounded by nonaggregated particles that made the cytoplasm extremely electron-dense. (ii) Similar to group one except that aggregation of virus was rare and sometimes groups of particles were membrane-bound. (iii) Particles were in cytoplasm and intercellular spaces packed side by side in long bands that in cross section formed a hexagonal lattice. (iv) Particles were sometimes membrane-bound in vacuoles and cytoplasm or aggregated in short rafts that were hexagonally packed in cross section. Infected cells were vacuolated and the tonoplast was damaged. (v) Regularly sized particles were aggregated in short rafts in cytoplasm and vacuoles. There were also groups of extremely long particles that were occasionally membrane-bound, but were packed into interlocking rings that formed herringbone patterns and swirls. The tonoplast was often absent or damaged near long particles.

Effects of 2-picolinic acid and copper on indoleacetic acid oxidase activity of peroxidase. D. M. WILSON (Univ. Vt., Burlington). The experiments reported here were done to test for possible effects of Cu^{2+} and chelating agents, including the fungal metabolite picolinic acid on indoleacetic acid oxidase (IAA oxidase) activity of peroxidase. Salt-free horseradish peroxidase in 0.2 M Na acetate buffer pH 4.2 was used as the IAA oxidase with $MnCl_2$ and *p*-coumaric acid as cofactors. The effects of different concentrations of $CuSO_4$, picolinic acid and Na EDTA on the rate of IAA oxidation were monitored at 261 nm. Cu^{2+} slowed the reaction rate at concentrations above 1×10^{-5} M. Picolinic acid from 1×10^{-4} to 10^{-6} M and Cu^{2+} from 1×10^{-4} to 10^{-6} M when added together in several combinations further decreased the rate. However, picolinic acid at 1×10^{-7} M reversed the inhibition by Cu^{2+} . Picolinic acid alone at 1×10^{-4} M increased the reaction rate above the control. EDTA had no effect on the rate. EDTA and Cu^{2+} when added together increased the rate of reaction except at high concentrations of both. Data indicate that the Cu^{2+} inhibition of IAA oxidase can be increased, decreased, or abolished by chelation with different concentrations of picolinic acid. Data suggest that picolinic acid may play a role in the regulation of IAA oxidation in some disease syndromes. EDTA is able to reverse inhibition by Cu^{2+} except at high concentrations of Cu^{2+} and EDTA.

Resistance to Verticillium wilt transferred from Gossypium barbadense to upland cotton phenotype. S. WILHELM, J. E. SAGEN, & H. TIETZ (Univ. Calif., Berkeley). Upland cottons, resistant to *Verticillium* wilt caused by *Verticillium albo-atrum* (microsclerotial type), which approach limited usefulness for local areas, have been obtained from a few progenies of susceptible Upland cotton × resistant *Gossypium barbadense*. Upland parents giving high-percentage fertile offspring beyond F_1 were Rex,

Alabama H257, and a Hartsville derivative; low-percentage fertile offspring were Hopi Acala 6-1-5 and several New Mexico Acalas. These all, no doubt, show recent *G. barbadense* introgression. Resistance was derived primarily from the varieties Seabrook 12-B-2 and Ida Corrizal (Peru). The reactions of F_1 of 250 different crosses showed that resistance was clearly dominant over susceptibility. Resistant hybrids, now in F_6 and highly uniform, exhibit slight traits of *G. barbadense* including faint petal spot, glabrous leaves, orange pollen, and 30- to 35-mm fibers. Select resistant hybrid individuals maintained as clones provide genetically stable parents of known pedigree. Crossed among themselves, these pedigree hybrids yield predominantly resistant offspring and offer possibilities of synthesizing families of closely related wilt-resistant cottons.

Control of Fusarium wilt of tomato by varying the nutrient regimes in soils. S. S. WOLTZ & J. P. JONES (Agr. Res. Educ. Center, Univ. Fla., Bradenton). A practical degree of control of Fusarium wilt was obtained under conditions of heavy inoculum potential by the establishment of soil chemical environments unfavorable to fungus growth and pathogenesis without material interference with growth and productivity of the tomato plant. Reduction in disease severity was accomplished by the maintenance of lower levels of phosphorus, iron, manganese, and zinc. Field plots of Leon fine sand that received no supplemental applications of these nutrients had 11% disease incidence, whereas those receiving 40, 5, 6, and 4 kg/hectare of phosphorus, iron, manganese, and zinc, respectively, in a single application had 45% disease incidence. This increased incidence, found at slightly acid soil pH values, did not occur in neutral or slightly alkaline soil. Race 2 tomato Fusarium inoculum grown on ammonium as the sole nitrogen source was twice as virulent in terms of disease incidence as the same amount of inoculum grown solely on nitrate nitrogen. Soils fertilized solely with nitrate produced only 25% as much disease based on vascular browning as did soil receiving ammonium nitrogen. Soil-agar culture techniques were evaluated for testing the growth, sporulation, and subsequent virulence of Fusarium. Supplementation of Leon fine sand with phosphorus or a combination of EDTA compounds of iron, manganese, and zinc increased virulence of the inoculum over that of inoculum produced without supplemental nutrients. Growth and sporulation were also enhanced.

Powdery mildews favored by agriculture. C. E. YARWOOD & M. W. GARDNER (Univ. Calif., Berkeley). The following Erysiphaceae have been found in habitats disturbed by man, and have not been found where the hosts occur naturally, relatively undisturbed by man: *Erysiphe cichoracearum* (conidiophore type) on *Baccharis pilularis*; *E. cichoracearum* on 11 *Eucalyptus* species; *E. polygoni* on *Eschscholtzia californica*; *Sphaerotheca fuliginea* on *Navaretia squarrose*; *S. fuliginea* on *Prunella vulgaris*, and *S. lanestris*; *E. polygoni*, *Microsphaera alni*, and *Phyllactinia corylea* on 30 species of *Quercus* and *Lithocarpus*. Only three host:pathogen associations (*M. alni* on *Alnus oregana*, *P. corylea* on *Diplacis aurantiacus*, and *E. cichoracearum* on *Myosotis sylvatica*) appeared to be as abundant on undisturbed wild plants as on those manipulated by man. Most of the some 1,400 collections of Erysiphaceae on some 536 host species from 1934 to 1972 have been on cultivated plants, or in botanic gardens, greenhouses, parks, campuses, and roadsides, and it is not known whether the mildew also occurred as abundantly in the native habitat of the host. The relative role of tillage, fertilization, watering, pruning, greenhouse culture, or some unknown factor in favoring powdery mildews is not clear. Of these, only tillage and

pruning have been demonstrated to favor disease in this study.

Evidence for inheritance of tolerance to dodine in Venturia inaequalis. K. S. YODER & E. J. KLOS (Mich. State Univ., East Lansing). In 1969, monoconidial isolates of *Venturia inaequalis* from sprayed and unsprayed apple trees in Michigan, New York, Maryland, and Ohio were tested for tolerance to dodine by disc assay, spore germination, and growth rate in liquid culture. The correlated results showed a fairly wide range in tolerance among the isolates. The most tolerant isolate was crossed in vitro with normal isolates, and tetrad analysis was conducted on monoascospore cultures of all the spores from asci from these crosses. Frequently, the tetrads analyzed were found to be tetratype asci in which one pair of spores had a level of tolerance similar to that of the most tolerant parent, one pair similar to the least tolerant parent, and two pairs with intermediate levels of tolerance. The presence of these tetratype asci suggests that at least two genes may be involved in conditioning the level of tolerance to dodine in the most tolerant isolate.

A host-specific toxic metabolite produced by Phyllosticta maydis. O. C. YODER & D. M. MUKUNYA (Cornell Univ., Ithaca, N.Y.). Both nuclear and cytoplasmic factors condition susceptibility of corn to *Phyllosticta maydis*. The pathogen in liquid culture produced a metabolite(s) which apparently interacted with both nuclear and cytoplasmic products. Sensitivity of corn lines was demonstrated by (i) inhibition of root growth of seedlings in culture filtrate; (ii) necrosis of cuttings in culture filtrate; and (iii) necrosis of plants injected with culture filtrate. Corn cultivars Pa33, W182B, NY821, CO109, Pa83Rf, WF9, Cornell M-4, and Cornell M-3 with Texas cytoplasmic male sterility (Tcms) and NY821, CO109, and Cornell M-3 with normal (N) cytoplasm were susceptible to the fungus and were sensitive to culture filtrate. Corn cultivars Pa33, W182B, Pa33 × D410, and C153 × CO109 with N cytoplasm and B14A with Tcms were resistant to the fungus and insensitive to culture filtrate. The filtrate caused a 2- to 3-fold increase in respiration rate of mitochondria isolated from Pa33 with Tcms, but had no effect on respiration of mitochondria from Pa33 with N cytoplasm. The toxic principle was dialyzable, soluble in methanol, partially soluble in butanol, and nearly insoluble in chloroform and benzene. A nonpathogenic *Phyllosticta* sp. and a saprophytic *Mucor* sp. did not produce the host-specific metabolite.

Persea americana, a new host for Phytophthora citricola. G. A. ZENTMYER, C. J. HICKMAN, & LAURA JEFFERSON (Univ. Calif., Riverside, Univ. Western Ontario, London, Can.). *Phytophthora citricola* was isolated from avocado (*Persea americana*) roots and from cankers on avocado trunks in several groves in southern California. An isolate from avocado fruit in Mexico, originally described by Fucikovsky as *P. cactorum*, is similar to the *P. citricola* isolates from California. All of the avocado isolates are homothallic, with paragynous (rarely amphigynous) antheridia, and form oospores (15-28 μ in diam) readily on V-8 juice agar. Sporangia with shallow apical thickenings are formed only occasionally on solid media, but are abundant in water or soil extract. Sporangia are typically varied in shape and size, and may be distorted, elongated, bifurcate, or more irregular with several apices. Irregular sporangia are more commonly produced by some avocado isolates than by the type culture. The avocado isolates have a temperature response similar to the type culture and to other *P. citricola* isolates from lilac, hops, and rhododendron, but grow more slowly at 30 C than do some of the other isolates. The

avocado isolates, as well as other *P. citricola* cultures, were low in virulence to avocado roots, but caused cankers on stems or branches when wound-inoculated, and produced a rot on unwounded avocado fruit. This is the first report of *P. citricola* on avocado.

Two previously undescribed genes for rust resistance in flax. D. E. ZIMMER & V. COMSTOCK (ARS, USDA, N.D. State Univ., Univ. Minn.). None of the six described genes which condition resistance in flax, *Linum usitatissimum*, to all North American races of flax rust, *Melampsora lini*, condition resistance against all South American races. The introduction of South American races into North America could be disastrous to the flax industry. Progenies of individual plant selections from accessions C.I. 1888, C.I. 1911, and C.I. 2008 of the Flax World Collection were resistant to selected races of rust which collectively are virulent on all known rust-resistance conditioning genes. These selections were also rust-resistant in field trials in Argentina, where monogenic lines with all known genes were susceptible. Segregation ratios obtained in the F₂ and testcross generations involving Bison, C.I. 389, as the testcross parent revealed that resistance of each selection was monogenic and dominant. The segregation ratios in the F₂ generation of crosses made to learn the allelic relationship between the new gene(s) and the five known rust-conditioning loci revealed that the resistance of C.I. 1911 and C.I. 1888 are conditioned by gene(s) at the P locus, and the resistance of C.I. 2008 is conditioned by a gene at the M locus. These genes have been designated P⁴ and M⁶.

Pepper virus strain identification in southern Florida. T. A. ZITTER (Univ. Fla., Belle Glade). A combination of differential pepper cultivars and immunodiffusion tests was used to determine the identity and distribution of tobacco etch virus (TEV) and potato virus Y (PVY) strains infecting peppers grown in southern Florida during the 1971-72 season. Pepper cultivars used in greenhouse tests included Early Calwonder, Yolo Y, Avelar, Agronomico 8, Florida breeding line 23-1-7, and Tabasco. A standard TEV antiserum reacted with all TEV isolates, whereas two separate antisera were needed for the detection of common and severe PVY isolates. Common isolates of TEV infected only Early Calwonder, Yolo Y, and Tabasco, and were found in Palm Beach, Hendry, Collier, and Lee Counties, whereas common PVY isolates infected only Early Calwonder and were found in Palm Beach, Hendry, Collier, and Dade Counties. Severe strains of both TEV and PVY infected all of the differential pepper cultivars in varying degrees. Avelar expressed the

greatest amount of tolerance. These severe strains were recovered from the East Coast and remote areas of Palm Beach County and from several locations in Collier County. No other pepper viruses or additional strains of TEV or PVY were recovered by these methods.

Effect of soil amendments on the generation time of Xanthomonas campestris in cabbage guttation fluid and on lesion development in the host. B. G. ZOLLER & T. KOSUGE (Univ. Calif., Davis). Guttation fluids were collected at dawn over a period of 6 months from cabbage plants that were grown in the field in soil amended with various sources of inorganic nitrogen and sulfur. The fluids were filter-sterilized and used as culture media for *Xanthomonas campestris*. Viable cell counts under standardized conditions were used to measure growth rates. Generation time of bacteria decreased in fluids from plants that were grown in soils amended with ammonium phosphate or ammonium sulfate as compared to bacteria in fluids from plants in unamended soils. The effect was observed in fluids that were collected up to 3 months after amendment. In contrast, death of *X. campestris* cells occurred in fluids from plants growing in soil treated with sulfur. Death rates were highest in fluids that were collected 2-4 months after sulfur amendment. Death or increased generation time of bacteria also was found in fluids from plants grown in soils amended with potassium sulfate, calcium sulfate, or ammonium sulfate 4-5 months before collection. Generation times in guttation fluids were inversely correlated with the rates of development of vein-blackening on the same plants artificially inoculated with *X. campestris* during the same time intervals after soil amendments.

External Fusarium moniliforme var. subglutinans associated with right-angle bending and twisting of sweet sorghum stalks. N. ZUMMO (ARS, USDA, Meridian, Miss.). Conspicuous stalk bending and distortion have been observed in sweet sorghum for several years in the southeast. A fungus tentatively identified as *Fusarium moniliforme* var. *subglutinans* was consistently isolated from the exterior of distorted portions of sorghum stalks 1.8 to 3 m aboveground. Many, but not all, affected plants showed Pokkah boeng and/or knife-cut symptoms. In some instances, spindle leaves did not unroll properly, giving the plant a ladderlike appearance. The same fungus was isolated from the compacted leaf tips, where the mycelium apparently acted as a binder to prevent natural unrolling. The fungus was not isolated from internal portions of distorted areas of stalks even though severe reddening was evident inside the stalks. Some varieties (Mer. 64-7) were more affected than others.