

Abstracts of the Fifty-Seventh Annual Meeting of the Pacific Division  
of The American Phytopathological Society

*Isolation and culture of mycoplasma-like organisms (spiroplasma) from stubborn-affected citrus.* R. M. ALLEN. (Univ. Ariz., Tucson). Twenty-seven isolates of mycoplasma-like organisms (MLO) were isolated from stubborn-affected citrus tissues or filtered tissue extracts. Sources of MLO were leaves, bark, seed, or fruit mesocarp from 10 cultivars of citrus including orange, grapefruit, and tangelo. Time required for evidence of successful culture in cell-free liquid media, ranged from 10-60 days. Sub-cultures of isolates were successful at  $10^{-10}$  dilution but most reliable transfers were made at 4- to 5-day intervals using a  $10^{-1}$  dilution. A medium including 1% bovine serum fraction instead of 20% horse serum (v/v) has supported growth through 20 serial passages. Viability of MLO in most liquid media was lost after 12 days unless subcultured. MLO usually have small spherical bodies (0.1- to 0.3- $\mu$ m diam) with one or more helical filaments (0.2  $\times$  1.0 - 15.0  $\mu$ m) attached. Filaments of one slow-growing isolate, possibly a different strain, attained a length of 45.0  $\mu$ m.

*Ultrastructure of the hypersensitive reaction induced by potato virus X.* A. V. ALLISON & T. A. SHALLA. (Univ. Calif., Davis). Expanding necrotic lesions induced by PVX in leaves of *Gomphrena globosa* L. were studied by fluorescence- and electron microscopy. Amorphous inclusion bodies, typical of those induced by this virus in systemic host plants, were observed in cells adjacent to the necrotic region. These contained smooth and beaded sheets often formed into scrolls with associated virus particles. Small virus aggregates were found in the cytoplasm, but massive virus accumulations were rarely encountered. Membrane-bound bundles of virus particles were frequently observed outside the protoplast, between the plasmalemma and the cell wall, and traversing the plasmodesmata. These sacks of virus particles were embedded in a matrix identified as callose by histochemical staining and fluorescence microscopy. Wall modifications included depositions of an electron-dense material resulting in thickening of the wall several cells distant from the necrotic region. These deposits are associated with boundary formations and deposition of callose around the plasmodesmata.

*Conformation of pinwheel inclusions induced by tobacco etch virus.* J. H. ANDREW & T. A. SHALLA. (Univ. Calif., Davis). Root tips and leaf cells of tobacco plants (*Nicotiana tabacum*, var. 'Havana 425') infected by TEV were studied using thin-sectioning, freeze-etch, and particle extraction methods for electron microscopy. Pinwheels that abutted to plasmodesmata in the apical meristem and elongation regions of roots were examined in serial sections. In cross section, pinwheel arm length was maximum at the cell wall and decreased with height, indicating that the arms were triangular. To eliminate the possibility that this appearance was an artifact of sectioning angle, models of pinwheels with either rectangular or triangular arms were constructed and cut in various planes. Evidence obtained by comparing these models confirmed that the triangular shape was legitimate. Examination of inclusions in shadowed or negatively stained dip preparations, and in freeze-etched cells verified that pinwheel arms were triangular. Therefore we conclude that, at least in the early stages of formation, the pinwheel inclusion as a whole is conical, rather than cylindrical, as it is usually described.

*Influence of milling of air dry soil upon apparent inoculum density and propagule size of Verticillium albo-atrum.* L. J. ASHWORTH, JR., D. M. HARPER, & H. L. ANDRIES. (Univ. Calif., Berkeley). Field soil was dried to ca. 7% moisture in 48 hours at 22-25 C, passed through a 2-mm

sieve, then ground with either a rotating flail-type mill (FTM) or mortar and pestle (MP). Triplicate 15-g samples were wet-sieved and separated into fractions having particles ranging from 37-120  $\mu$ , 18-37  $\mu$ , and 14-18  $\mu$ . Soil residue fractions were cultured as reported earlier. Soil passed through the FTM and MP had, respectively 2.2 and 12.3 propagules/g soil. Regardless of milling, 50-70% of the propagules were in the 37- to 120- $\mu$  fraction of soil. About 30% and 5% of propagules were in the 18- to 37- $\mu$  fractions of the FTM- and MP-ground soil, respectively. No propagules were in the 14- to 18- $\mu$  fraction of FTM soil but ca. 40% of all propagules of MP soil were in this size range. These data indicate that milling procedures can affect apparent inoculum densities of soil and also can affect propagule size by breaking microsclerotia into smaller viable units.

*Control of Fusarium stem rot of carnations in automated irrigation culture.* R. BAKER & H. R. KINNAMAN. (Colo. State Univ., Fort Collins). High incidence of *Fusarium* stem rot of carnations (incited by *Fusarium roseum* f. sp. *cereale* 'Avenaceum') occurred in gravel culture when plants were irrigated by automated systems dispensing nutrients. The frequent inundations of lower branches with solutions apparently predisposed tissue to infection by the pathogen. Benomyl was applied initially in aqueous suspensions in single drench treatments to plants in both soil and gravel at 0.15 g/meter<sup>2</sup>. In either medium, the fungitoxicant (detected by bioassay) reached a maximum in tissue 4 wks after application; however, only in plants growing in soil did the concn exceed the ED<sub>50</sub> level for *F. roseum*, in this case being three times that found in those in gravel. This was at least partially explained when benomyl applied to gravel was recovered in high concentrations in leachates. Nevertheless, in subsequent trials, loss due to *Fusarium* stem rot in carnations in gravel culture treated with 3  $\mu$ g/ml of benomyl applied with each irrigation through automated systems averaged 34%; loss in nontreated controls was 71%.

*Epidemiology of Rhizoctonia solani damping-off of radish.* D. M. BENSON. (Colo. State Univ., Fort Collins). Inoculum density vs. disease incidence curves for pre-emergence damping-off of radish (caused by *Rhizoctonia solani*) transformed to the semilogarithmic, log-log, or log-probit transformation suggested synergism. At 22, 26, and 30 C, log-log slopes (1.31-1.48) were parallel with position dependent on the influence of temperature. At 15 and 20 C, slopes (2.57-2.19) were significantly different. Position of the inoculum density vs. disease incidence curves as interpolated by the ID<sub>50</sub> value (i.e., the inoculum density required for 50% disease incidence) was 0.76 propagules/g (p/g) soil at 26 C, 1.6 p/g at 30 C, 2.8 p/g at 22 C, 5.2 p/g at 20 C, and 7.0 p/g at 15 C. Fungal growth rate was directly correlated with temperature. Germination and emergence of radish seedlings at 20 to 30 C was similar. Susceptibility period of radish to pre-emergence damping-off was similar (42-53 hrs) at 20 to 30 C but was longer (100 hrs) at 15 C. The interaction of fungal growth rate with host maturation rate, explained the change in slope of the inoculum density vs. disease incidence curve at 15 and 20 C.

*Genetics of hypersensitive-fleck resistance to Trichometasphaeria turcica in Sorghum bicolor.* R. R. BERGQUIST & O. R. MASIAS. (Univ. Hawaii, Honolulu). *Sorghum bicolor* I.S. accessions 2663, 2680, 2687, 2760, and 8777 were resistant to *Trichometasphaeria turcica* (*Helminthosporium turcicum*) in multiple-location field tests and greenhouse studies in Hawaii. The reaction was characterized by formation of minute necrotic reddish-purple or yellowish-tan flecks within 2-3 days after inoculation.

Flecks on resistant plants remained static without sporulation. *T. turcica* isolates from Johnson grass, sorghum, and corn produced only flecks on F<sub>1</sub> hybrids of resistant sorghum lines crossed to susceptible cytoplasmic sterile A-Line S9326-OL, suggesting a dominant gene(s) conditioning resistance. The above sources of resistance were heterogeneous and heterozygous for *T. turcica* reaction, suggesting random mating in the resistant populations. The F<sub>2</sub>, F<sub>3</sub>, and backcross populations demonstrated one or two genes conditioning resistance. Different backcross-two plants carrying one dominant gene from each of the Indian sorghum selections had similar hypersensitive-fleck reactions.

*A selective medium for the isolation of Pythium aphanidermatum from field soil.* T. J. BURR. (Univ. Ariz., Tucson). Oospores were the sole survival structure of *Pythium aphanidermatum* in naturally infested field soil. Population counts determined on a species specific isolation medium from naturally infested soils of various types ranged from 10-250 oospores of *P. aphanidermatum*/g soil. The medium contained Difco cornmeal agar (1.7%), pimarinic (Myprozone, potency 92.2%, American Cyanamid) 100 µg/ml, streptomycin sulfate 200 µg/ml, rose bengal 250 µg/ml, and benomyl 5 µg/ml. Dilution plates were incubated at 35 C, the optimum temperature for *P. aphanidermatum*. Oospores, zoospores, sporangia, and mycelial fragments of *P. aphanidermatum* were capable of greater than 90% germination and/or growth on the selective medium and ca. 97% of a known oospore population was recovered from artificially infested field soil. Besides being species specific, which eliminates the need for isolation and identification of all colonies on dilution plates, it is possible to identify the originating colony propagule and subsequently isolate pure, single-oospore isolates from this medium. The medium also allowed demonstration of a linear relationship between oospores/g soil and soil dilution which eliminates preparation of numerous soil dilutions when dealing with soils containing populations of *P. aphanidermatum* of unknown density.

*Potted ornamental plants: another source for introduction of Pseudomonas aeruginosa into hospitals.* J. J. CHO, S. K. GREEN, M. N. SCHROTH, & S. D. KOMINOS. (Univ. Calif., Berkeley; and Dept. of Pathol., Mercy Hosp., Pittsburgh, Pa.). The plant pathogen *Pseudomonas aeruginosa* is second only to the Klebsiella-Enterobacter group of bacteria in causing hospital-acquired infections. Its introduction into hospitals has been attributed to fresh vegetables. We now report potted ornamental plants as another source of introduction. The bacterium was isolated on Müller-Hinton medium, containing 0.03% cetrimide and acetamide broth, at 42 C, from leaves and soil samples of potted ornamental plants including chrysanthemum, African violet, azalea, and hydrangea. Eighty to 90% of the chrysanthemums from various nurseries harbored the bacterium on the leaves. Populations of *P. aeruginosa* on chrysanthemum leaves were as great as 50 cells per leaf. Every soil sample examined harbored *P. aeruginosa*. Two soil samples each contained approximately  $5.0 \times 10^5$  cells/g. All isolates that were tested rotted tomatoes and celery. Several isolates were identified by pyocine typing as common strains prevalent in hospitals.

*Infectivity of tobacco mosaic virus protein.* G. W. COCHRAN. (Utah State Univ., Logan). Tobacco mosaic virus protein prepared from whole virus by the acetic acid method of Fraenkel-Conrat induced atypical necrotic lesions on ultrasonically inoculated half leaves of 'Scotia' and 'Persian' beans. Inoculation of 'Turkish' tobacco with macerates of the atypical bean lesions induced typical tobacco mosaic virus symptoms in four experiments. Tobacco plants inoculated

directly with virus protein, or macerates from normal bean leaves, remained healthy. The protein's atypical infectivity for beans was unaffected by preincubation with pancreatic ribonuclease. Adding virus protein to phenol-prepared TMV RNA at the instant of inoculation gave a three-fold increase in typical lesions on beans. These results suggest that both virus protein and full-length viral RNA are needed for typical bean lesion development. Because fully typical virus was produced in tobacco (via bean) from only virus protein inoculum, one virus protein subunit probably carries a complete TMV genetic complement. This appears to be the first report of protein serving as a carrier of genetic information.

*Effects of soil moisture related to potato scab control.* J. R. DAVIS & N. K. NIELSEN. (Univ. Idaho, Branch Exp. Stn., Aberdeen). Lesions of potato scab (*Streptomyces scabies*) were significantly deeper when 'Russet Burbank' tubers were grown at 45% ASM-FC (45% of the available soil moisture at time of irrigation) compared with tubers grown at 60% ASM-FC or 75% ASM-FC. Russet Burbank tubers from plots maintained at 45% ASM-FC or 60% ASM-FC showed no difference of scab surface coverage, but scab coverage was significantly less with tubers grown at 75% ASM-FC. Aerial growth of *S. scabies* on potato peel media prepared from potatoes grown with several moisture treatments (45% ASM-FC, 60% ASM-FC, 75% ASM-FC, and 90% ASM-FC) and ability of tubers to respond with suberization was closely correlated with scab severity in the field. Moisture treatments showed an effect on plant nutrition as evidenced by petiole uptake data. There existed a highly significant correlation between Ca and scab severity and negative correlations with K, PO<sub>4</sub>, and Mn. Scab severity with given moisture treatments was also positively correlated with Ca accumulation in tuber peelings. Tubers grown at 45% ASM-FC were significantly more susceptible to potato scab and showed more Ca in the peel than tubers grown at higher moisture levels. Differences of lesion depth, pathogen growth on peel media, suberization response, and tuber chemistry suggest differences of host susceptibility with increased moisture.

*Aseptic germination of teliospores of Sphacelotheca reiliana.* I. S. FARAG, H. S. FENWICK, & W. R. SIMPSON. (Univ. Idaho, Moscow). Teliospores of *Sphacelotheca reiliana* germinate poorly on water agar, and bacterial and fungal contaminants occur when the teliospores are seeded on rich media. A method developed in our laboratory permits teliospore germination free of contaminants and enabled us to run a comprehensive study on spore germination. This method is: Place 1 g of teliospores in a 50-ml centrifuge tube, add 10 ml 5% Clorox; centrifuge at 3,000 rpm for 90 sec, decant. Add 10 ml sterile H<sub>2</sub>O, centrifuge 60 sec, decant. Add 10 ml 5% thymol, centrifuge 90 sec, decant. Add 10 ml sterile H<sub>2</sub>O, centrifuge 60 sec, decant, repeat. Add 10 ml sterile H<sub>2</sub>O, transfer 0.5 ml to corn meal agar (CMA) containing streptomycin (200 mg/l) and lactic acid (0.1 ml/l). Incubate at 25-28 C; examine at 12-hr intervals for 8 days. Germination was initiated at 36 hr with a maximum (71%) at 5.5 days.

*Determination of the time of infection of corn head smut.* I. S. FARAG, H. S. FENWICK, & W. R. SIMPSON. (Univ. Idaho, Moscow). Susceptible hybrid sweet corn seed was planted 3.8 cm deep in soil infested with teliospores of *Sphacelotheca reiliana* (Kuhn) Clinton at the rate of 33.3 g/0.5 kg of soil. At 24-hr intervals for 8 days, 180 germinating seeds and seedlings at the same stage of growth were extracted and treated as follows: (i) one-third were soaked 20 min in a 26.6 mg/ml solution of 75% WP pentachloronitrobenzene (PCNB), (ii) one-third were treated

as above but rinsed with tap water, and (iii) one-third were rinsed with tap water only. All were then transplanted into sterile soil and maintained until signs of the disease were visible. Treatments (i) and (ii) killed the spores in soil adhering to plants upon extraction. Infection was initiated 3 days after planting, with a maximum (66.7%) 6 days after planting. Coleoptile lengths were 1.3-2.5 cm at 3 days and the first true leaf was formed at 6 days. Exposure of plants to the inoculum beyond 6 days did not increase the level of infection.

*Prokaryotic-like structures associated with albino disease of Prunus avium.* E. R. FLORANCE & H. R. CAMERON. (Ore. State Univ., Corvallis). Three albino diseased sweet cherry (*Prunus avium*) trees were selected for a developmental cytopathological study. Controls were healthy-appearing trees selected in the same orchard and healthy virus-free trees maintained under screen at Corvallis. Tissue samples from diseased and healthy trees were collected monthly from April through September 1972. The distal 2-5 mm of leaf midvein and a portion of peduncle were fixed in glutaraldehyde and postfixed in osmium tetroxide for electron microscopy. Prokaryotic-like structures were observed in mature sieve tubes of samples taken from diseased trees during June and July. No structures were found prior to June or after July in samples from diseased trees, or in tissue from control trees. The structures ranged from 0.15 to 0.55  $\mu$  in diameter and up to 1.3  $\mu$  long. They were spherical to oblong, bound by a unit membrane, and contained a dense nucleoid area. The structures were present in both leaf and peduncle sieve tube elements. No virus particles were observed. The results suggest that albino may be caused by a prokaryotic structure rather than the previously reported virus.

*A screening technique useful in selecting for resistance in alfalfa to Phytophthora megasperma.* F. A. GRAY, R. B. HINE, M. H. SCHONHORST, & J. D. NAIK. (Univ. Ariz., Tucson). A screening technique, consisting of planting alfalfa seed in infested soil, was developed for selecting plants with resistance to damping-off and root rot caused by *Phytophthora megasperma*. Seedling disease increased in direct proportion to inoculum concentration. Pre- and post-emergence damping-off was influenced by inoculation technique. Approximately 10,000 seed of the cultivar 'Hayden' were planted in flats containing soil infested with three Arizona isolates of *P. megasperma*. After 2 weeks' incubation in growth chambers maintained on a 12-hr light cycle at 24 C and 12-hr dark cycle at 18 C, only 2% of the original stand remained. After an additional 6 weeks, plants having little or no root rot were selected for polycrossing. When seed from this polycross (PX) were compared with certified Hayden, the PX showed approximately 50% increase in seedling survival.

*Stalk blight of sugarbeet seed crops caused by Fusarium oxysporum f. sp. betae.* D. C. GROSS & L. D. LEACH. (Univ. Calif., Davis). In July, 1971, leaf wilt and seedstalk blight occurred on sugarbeets in the Willamette Valley, Oregon. *F. oxysporum* was isolated from discolored vascular tissue of blighted plants. Root dip inoculation with spores from single spore isolates caused wilt and death of sugarbeet seedlings. Oregon stalk blight isolates resemble Colorado isolates of *F. oxysporum* f. sp. *betae* in cultural characteristics and pathogenicity on sugarbeet seedlings. The Willamette Valley supplies seed for California beet sugar production. Cultivars grown for California were particularly susceptible to stalk blight. In severely affected fields, over 50% of the plants showed symptoms. Invasion of inflorescences occurred and 1 to 3% of the seeds from severely affected fields carried the pathogen.

*Light- and electron microscopy investigations of wheat smut fungi.* M. GROVE & W. M. HESS. (Brigham Young Univ., Provo, Utah). Caryopses of wheat were studied during infection by *Tilletia caries* and *T. controversa*. Initially the pathogens penetrated embryos of the host. After the embryo tissue was consumed by the fungi, endosperm tissue was penetrated and consumed. Initial penetration of endosperm tissue was in localized areas. During infection, the fungus mycelium of both fungi spread intercellularly, but was occasionally seen intracellularly. As the host tissue was consumed, teliospores were formed. At maturity, a host cell layer 4-6 cells thick remained which retained the mature teliospores. Histochemical studies indicated that carbohydrates and lipids of the host disappeared soon after invasion by the pathogens. Nuclei and cell walls remained intact after other cell components decomposed.

*Electrophoresis of long flexuous viruses using agarose-acrylamide gels.* K. M. MAKKOUK & D. J. GUMPF. (Univ. Calif., Riverside). The recent use of agarose together with acrylamide has facilitated the use of disk gels for the electrophoresis of rod shaped viruses. Using gels of 1% acrylamide and 0.5% agarose, and a buffer composed of 0.036 M Tris [tris(hydroxymethyl) aminomethane], 0.03 M sodium-dihydrogen phosphate, and 0.001 M disodium EDTA (ethylenediaminetetraacetic acid) adjusted to pH 8.3 with sodium hydroxide for both the gels and the reservoirs, long flexuous rods of potato virus Y (PVY) and tobacco etch virus TEV could be electrophoresed. Both PVY and TEV gave a single infectious band when electrophoresed for 6 hr at 5mA/gel (200 V). Some virus aggregation was apparent as a band on top of the gels. Migration was faster using a buffer system of 0.04 M Tris and 0.001 M Na<sub>2</sub>-EDTA adjusted to pH 8.3 with acetic acid. Both TEV and PVY migrated slower than TMV. In gels of 0.7% agarose alone or 2% acrylamide with 0.5% agarose, neither TEV nor PVY were able to penetrate the gels.

*Olive sickle leaf virus found in 'Dwarfing Manzanillo' from Israel.* W. O. MC CARTNEY. (State of Calif., Dept. of Food and Agric., Sacramento). Olive sickle leaf symptoms were found in olive, *Olea europaea* L. 'Dwarfing Manzanillo' scions imported in 1970 from Israel. Symptoms occurred both on the top stock and the 'Mission' understock 2 years after grafting. In contrast to the form of sickle leaf disease present on the olive cultivar Mission in California, symptoms found in the Dwarfing Manzanillo variety are more severe. The California form is evidenced by shortened sickle-shaped leaves often found scattered over the whole tree. The symptoms of the Dwarfing Manzanillo form of olive sickle leaf are: (i) leaf deformity and stunting of lateral branches due to blind shoot buds and shortened internodes; and, (ii) affected apex leaves are blotched, streaked, curved and puckered, and light-green with white markings. The general appearance of the affected plant is that of a bushy shrub. This form of olive sickle leaf virus may be a threat to the olive industry.

*Infection of wheat by soil-borne inoculum of Cercospora herpotrichoides.* MARY L. MC COY & R. L. POWELSON. (Ore. State Univ., Corvallis). The influence of three different soils on the amount of infection in wheat by soil-borne inoculum of *Cercospora herpotrichoides* was studied. Three different soils (sand, sandy loam, and silt loam) were infested with conidia of *C. herpotrichoides* at inoculum densities of 2,000, 10,000, 30,000, and 50,000 spores/g soil. Regression lines drawn from points on an arithmetic plot of the data suggest that infection of wheat by soil-borne inoculum of *C. herpotrichoides* is under the influence of the host rhizosphere. Regression lines drawn

using the multiple-infection transformation show that more spores of *C. herpotrichoides* are required to incite disease in wheat with the silt loam soil than in the sandy loam soil or sand. The decrease in disease incidence in the soils was correlated with higher rates of microbial immobilization of sucrose with a resultant shrinkage of the rhizosphere influence.

*Effect of light on uptake of Verticillium dahliae spores by mints.* H. A. MELOUK, C. E. HORNER, & V. Q. PERKINS. (USDA, ARS; Ore. State Univ., Corvallis). Inoculation of mint shoot tip cuttings with a spore suspension ( $10^6$  spores/ml) of *Verticillium dahliae* by dipping the cut ends in the inoculum for 45 min resulted in a rapid uptake of spores. Uptake of spores by peppermint (*Mentha piperita* 'Mitcham') and spearmint (*Mentha cardiaca* 'Scotch') cuttings placed under fluorescent light (6,456 lx=600 ft-c) at 23 C was double that in the dark at the same temperature. Spore uptake under light, as determined by the propagule counts using the dilution plate assay, was 20,000 and 50,000 spores/g fresh wt of stem tissue for peppermint and Scotch spearmint, respectively; whereas spore uptake in the dark was 10,000 and 24,000 spores/g of tissue for peppermint and Scotch spearmint, respectively. After inoculation, cuttings were placed in sand for rooting and sampling to determine fungus populations. In inoculated peppermint cuttings (both in dark and light), *V. dahliae* propagules increased rapidly and were highest 2 weeks after inoculation and then declined in the third week.

*The ultrastructure of a rickettsia-like organism from a peach tree affected with phony disease.* G. NYLAND, A. C. GOHEEN, S. K. LOWE, & H. C. KIRKPATRICK. (Univ. Calif., Davis). Electron microscopy revealed a rickettsia-like organism in vessels only of roots of a peach tree affected with phony disease. The organism was seen in each of over 1,000 ultrathin sections from rootlets taken from a peach tree that exhibited classic symptoms of phony disease at the Southeastern Fruit and Tree Nut Research Station in Byron, Georgia. No organisms were found in approximately 1,200 sections of rootlets from four nearby healthy-appearing trees. The most prominent feature of the cells of the organism is the circumferential folds of the outer wall which are oriented in a regular annular or spiral pattern. In thin sections the outer wall appears notched, wavy, or rippled. The cells are about  $2.3\mu \times 0.35\mu$ , slightly smaller than a similar organism associated with grape Pierce's disease. The organism has a double-layered outer wall, double-layered cytoplasmic membrane, and cell contents similar to described rickettsiae.

*Transfer of R factors carrying carbenicillin resistance to plant pathogenic pseudomonads.* N. J. PANOPOULOS, W. V. GUIMARAES, & M. N. SCHROTH. (Dept. of Plant Pathology, Univ. of Calif., Berkeley). Antibiotic resistance (R) factors were transferred from *Pseudomonas aeruginosa* to *Pseudomonas phaseolicola* and *P. lachrymans*. Genetically marked derivatives of *P. aeruginosa* which had acquired the plasmids R18 and R91 from clinical strains highly resistant to carbenicillin (CB) by conjugation were used as donors. The frequencies of transfer per donor cell for factor R18 were:  $2.4 \times 10^{-4}$  and  $2.1 \times 10^{-4}$  to strains HB-36 and R2QHB of *P. phaseolicola*, respectively, and  $2.0 \times 10^{-4}$  to *P. lachrymans* strain PL-11. Factor R91 was transferred to PL-11 with a frequency of  $7 \times 10^{-7}$ . No transfer of R91 to the *P. phaseolicola* isolates was obtained. Inheritance of the R factors did not alter the negative oxidase reaction of the recipients or the ability to induce hypersensitive reaction in tobacco. Neither plasmid was transferable from *P. aeruginosa* to *P. pisi* 10, *P. solanacearum* 10, *P. glycinea* NCPB 1271, *Xanthomonas vesicatoria* 1, or *Agrobacterium tumefaciens*.

*Variation among isolates of Prunus necrotic ringspot virus from hops.* E. G. PROBASCO & C. B. SKOTLAND. (Wash. State Univ., Irrig. Agric. Res. Ext. Center, Prosser). Fourteen *Prunus necrotic ringspot virus* isolates, each from a different hop cultivar, were maintained in and transferred weekly to healthy cucumber cotyledons. Incubation periods of the isolates ranged from 60-84 hrs and lesion color ranged from light green to dark yellow in cucumber cotyledons. The incubation periods of yellow-lesioned isolates (60-72 hrs) were shorter than those of green-lesioned isolates (72-84 hrs). Single lesion transfers to cucumber cotyledons from one of the yellow-lesioned isolates produced both yellow-lesioned and green-lesioned cultures. Purified preparations of yellow-lesioned isolates were consistently infective in cucumber cotyledons at dilutions 2-4 times greater than those of green-lesioned isolates. Symptoms of the isolates varied in a hop seedling clone. Yellow-lesioned isolates produced ringspots in inoculated leaves and systemic symptoms consisting of ringspots, linepattern, and mottling while the green-lesioned isolates frequently produced either ringspots in the inoculated leaves or systemic symptoms. The yellow-lesioned isolates generally produced more severe symptoms than the green-lesioned isolates.

*Control of the citrus nematode, Tylenchulus semipenetrans, with foliar Vydate sprays on Valencia oranges in southern California.* J. D. RADEWALD, D. ROSEDALE, F. SHIBUYA, & J. NELSON. (Univ. Calif., Riverside). Seven-year-old Valencia oranges on Troyer citrange root heavily infected with the citrus nematode were first sprayed with Vydate [S-methyl 1-(dimethylcarbamoyl)-N-[(methylcarbamoyl)oxy] thioformimidate] (D-1410) at 1816 g/378.5 liters water on 1 August 1969. Seventy-six liters or 363.2 g of Vydate per 0.404 hectare was applied to the young trees. The spray schedule was followed monthly for the first year and on a 2- to 3-month schedule thereafter to date. Soil and root samples were taken at each spray date. Two months after the initial spray, the nematode count from the sprayed trees was reduced in the roots and soil by 53% and 68%, respectively. After 8 months, the population from the sprayed trees decreased in the roots and soil by 95% and 94%. Two years after initiation of the experiment, populations from the Vydate-sprayed trees were reduced by 98% on the roots and 95% in the soil. No phytotoxicity was ever observed. In other experiments on citrus the chemical has been ineffective. The reasons for the failures are under investigation.

*Properties of a toxic compound produced by a mutant of Corynebacterium michiganense.* P. V. RAI. (Rocky Mtn. Coll., Billings, Montana). Mutation in bacteria generally causes a change in their virulence. The object of this study was to determine the mechanism of sub-virulence in a pink mutant of *Corynebacterium michiganense*. The properties of its extracellular toxin were studied and compared to that of the wild-type toxins. The toxin from the mutant required considerably more time to wilt tomato cuttings than the toxins from the wild-type isolate. Sephadex column chromatography of the toxin preparation from the wild-type isolate yielded three fractions. The second toxic fraction eluting from the column was the most potent toxin, having a molecular weight of 129,700 and the empirical formula of  $C_{473}H_{490}O_{449}N$ . The toxin from the mutant had a molecular weight of 21,480 and an empirical formula of  $C_{32}H_{68}O_{33}N$ . All toxins migrated towards the negative pole in the electrophoresis. Paper- and thin-layer chromatography of the hydrolysate of the toxin from the mutant showed seven sugars and six amino acids. The mutant was unable to produce the high molecular weight second toxic fraction associated with the wild-type isolate.

*Effect of temperature on curly top resistance in Phaseolus vulgaris.* M. J. SILBERNAGEL & A. M. JAFRI. (USDA, ARS; Western Region, Irrig. Agric. Res. Ext. Center, & Department of Plant Pathology, Wash. State Univ., Pullman). Greenhouse and growth chamber studies indicate that resistance to curly top virus in certain snap bean (*Phaseolus vulgaris*) varieties may break down at high temperatures but is not influenced by the stage of plant development at time of inoculation. Inoculated symptomless plants do not contain the virus. Resistant plants that become infected exhibit some tolerance even after initial mild symptom development, since symptoms do not become as severe as on a susceptible variety. In a susceptible variety, higher incubation period temperatures increased curly top incidence and symptom severity and reduced the number of days to symptom expression.

*Ultrastructural and histochemical investigations of Ipomoea batatas infected by Rhizopus stolonifer.* K. O. SMITH & W. M. HESS. (Brigham Young University, Provo, Utah). Four regions of infected sweet potatoes were examined; noninfected tissue, tissue degraded by the fungus, tissue at the "zone of transition", between the degraded tissue and the healthy tissue, and noninfected tissue 1 mm from the "zone of transition". Degradation of host tissue in advance of the fungus was evident at the ultrastructural level, and all cellular membranes of infected tissues were significantly affected. A pectin stain used with fixatives for electron microscopy was used to study the breakdown of the middle lamella. In some tissue, there was degradation of the middle lamella before degradation of the cell wall. In other tissue, there was simultaneous degradation of the middle lamella and other cell wall layers, and in still other tissue the other cell wall layers degraded before the middle lamella.

*Germination of Pythium aphanidermatum oospores in soil.* M. E. STANGHELLINI. (Univ. Ariz., Tucson). Mode of oospore germination of *Pythium aphanidermatum* in field soil is regulated by the presence or absence of an exogenous source of nutrients. In the presence of such nutrients, oospores germinated exclusively by germ tubes (direct germination). Zoospore production (indirect germination) from germinating oospores in soil occurred only in the absence of exogenous nutrients and was restricted to the surface water of saturated soils. Oospores germinated in nutrient-amended soils maintained at various moisture levels ranging from saturation ( $pF=0$ ) to the permanent wilting point (PWP,  $pF=4.2$ ). However, percentage oospore germination and germ tube growth rates were reduced at the lower soil moisture levels. Colonization of alfalfa seeds, sown in soil containing a natural population of 80 viable oospores of *P. aphanidermatum*/g soil, occurred at all soil moisture levels except the PWP. Directly germinating oospores rather than zoospores apparently function as the major root infective units in field soil and wet soil conditions favor the activity of *Pythium* by increasing nutrient availability for oospore germination.

*The cytology of dormant and pregermination oospores of Phytophthora capsici.* L. W. STEPHENSON, D. C. ERWIN, & J. V. LEARY. (Department of Plant Pathology, Univ. Calif., Riverside 92502). Oospores from crosses of  $A^1 \times A^2$  isolates of *Phytophthora capsici* were produced in clear V8 juice broth. When 32-day-old oospores, stained with iron hematoxylin or acetocarmine, were observed by bright-field microscopy, a nucleus in each of the two pellucid bodies (ca. 5-7 $\mu$  diam) could be clearly differentiated. Nuclei were observed in optical sections of whole mounts of oospores with a phase-contrast microscope (NA 1.4). In most instances the two nuclei in dormant oospores either closely approached

or made contact with one another. Delayed karyogamy was associated with dormancy. After fusion of the nuclei (e.g. pellucid bodies), in some oospores a rugose structure resembling a spore could be differentiated in living and stained whole mount preparations, in optical sections, and in paraffin embedded sections. Reduction division of the fusion nucleus occurred in the rugose sporelike structure. In some optical sections a germ tube was observed to emanate from the rugose structure.

*Sporophore production and heterothallism in Clitocybe tabescens.* H. TANG & R. D. RAABE. (Dept. Plant Pathology, Univ. Calif., Berkeley 94720). Sixty-five single-spore isolates from a single basidiocarp of *Clitocybe tabescens* produced in pure culture were tested for their ability to produce sporophores in pure culture. Four isolates produced abnormal, stunted sporophores which did not bear basidiospores. Of 140 random pairings made between the single-spore isolates, 16 produced normal sporophores. The single-spore isolates varied in cultural characteristics from the parent isolate. Also, the cultural characteristics of the paired isolates which produced sporophores were different from those of either of the single-spore isolates used in any pairing. Sporophores developed best on two-thirds strength potato-dextrose agar and developed equally well at 18, 21, 24, and 27 C. Light was found not to be necessary for sporophore initiation but was necessary for maturation of the sporophores. Sporophores were found to be negatively geotropic and positively phototropic with the latter being dominant.

*Factors of resistance to curly top virus in Lycopersicon esculentum.* P. E. THOMAS & M. W. MARTIN. (USDA, ARS, Western Region, Irrig. Agric. Res. and Ext. Center, Prosser, Washington). The tendency to escape curly top infection possessed by four tomato cultivars (C5, C193, C27, and CVF4) was segregated into at least four measurable factors. This was achieved by selectively precluding the expression of specific resistance factors under controlled conditions and then measuring the decrease in tendency to escape infection which resulted. Results indicate that C27 and C193 possess a factor not present in C5 or CVF4 which is operative under field but not greenhouse conditions. Vector nonpreference contributes to the resistance of C5 and CVF4 but not of C193 or C127. Field resistance and vector nonpreference account for about half the total resistance available in the four cultivars. The remaining resistance is uncharacterized. Evidence suggests it may be attributable to one minor factor, possessed by C5 and C193, and at least one major factor possessed by all four resistant cultivars.

*Increased tolerance to benomyl in greenhouse populations of Botrytis cinerea.* A. G. WATSON & C. E. KOONS. (Cal-Fla Plant Corp., Fremont, California). Poor control of *Botrytis cinerea* was observed in a greenhouse bed which had been sprayed with benomyl at rates of 124.4 and 248.8 g per 378.5 liters (4 and 8 oz per 100 gal) approximately every week for the past 2 years. A leaf disk assay procedure was used to determine whether there had been selection for increased tolerance to benomyl among the greenhouse populations of *B. cinerea*. Chrysanthemum (*Chrysanthemum morifolium*) plants, cultivar 'Iceberg', were grown in 15-cm diam pots. The soil was drenched three times with 250 ml of the benomyl solution at weekly intervals. The concentrations of benomyl used were 0, 5, 25, 50, 250, and 500  $\mu$ g/ml. One disk of each leaf pair was placed on a water agar plate seeded with conidia of *B. cinerea* from chrysanthemum; the other disk was placed on water agar seeded with the same concentration of wild *B. cinerea* isolated from sources other than chrysanthemum plants. Greenhouse isolates completely

colonized leaf disks from plants treated with all concentrations of benomyl. Wild populations colonized only the disks treated with 0 or 5  $\mu\text{g/ml}$  benomyl.

*A leaf disease of chrysanthemums incited by Pseudomonas viridiflava.* A. G. WATSON, N. J. PALLERONI, J. J. CHO, & C. E. KOONS. (Cal-Fla Plant Corp., Fremont, Calif.; Dept. of Bacteriol. and Immunol.; and Dept. of Plant Pathol., Univ. Calif., Berkeley). In late 1972 a bacterial leaf spot was observed on young chrysanthemums, cultivar 'Mountain Sun'. The succulent leaves characteristic of this variety became water soaked during periods of high humidity in the greenhouse. These water soaked areas often collapsed and blackened. Isolations were made on the Miller-Schroth selective medium developed for *Erwinia amylovora*. A rapidly growing bacterium was consistently isolated, which was readily differentiated from other bacteria after 24 hr. Bacteriological tests indicated that the pathogen was *Pseudomonas viridiflava*. Subsequently, this bacterium was found on a number of chrysanthemum varieties in California and Florida, where it caused marginal leaf spots. Cuttings from major propagating companies harbored the bacterium as an epiphyte. This bacterium has also been reported as a weak pathogen on several hosts, including chrysanthemum, in England.

*Lipid metabolism during germination of spores of Ustilago maydis.* D. J. WEBER & J. L. BUSHNELL. (Brigham Young Univ., Provo, Utah). Free fatty acids decreased in spores of *Ustilago maydis* that were germinated in 1% sucrose and 0.2% casamino acids until none could be detected after 24 hours. The total fatty acid composition varied in concentration during the germination process. The  $\text{C}_{18}$  fatty acid increased while  $\text{C}_{18:1}$  decreased and the  $\text{C}_{16}$  fatty acid

decreased while  $\text{C}_{16:1}$  increased. Acetate  $1\text{-}^{14}\text{C}$  was readily incorporated into diglycerides, phospholipids, and sterols, but not into the free fatty acids during the germination process. The spores were able to catabolize the lipid reserves (free fatty acids) and at the same time synthesize lipid components (diglycerides, phospholipids, and sterols) during the germination process.

*The effect of oil on the uptake and translocation of methyl-2-benzimidazolecarbamate and its hydrochloric acid salt in cotton.* A. I. ZAKI & D. C. ERWIN. (Department of Plant Pathology, Univ. Calif., Riverside 92502). The hydrochloric acid salt of  $^{14}\text{C}$ -methyl-2-benzimidazolecarbamate ( $^{14}\text{C}$ -MBC-HCl) applied to the second internode of cotton plants (cultivar SJ-1) with or without a paraffinic oil (Orchex N 795) (20%, v/v) penetrated the bark and translocated upward, presumably through the xylem, and gradually accumulated in the leaf blades. Less than 1% of the label moved downward to the lower internode and roots. Quantitatively, the portion of  $^{14}\text{C}$  detected in the leaves of MBC-HCl-oil treated plants increased with time (between 4 and 11 days after treatment) to a higher value than in plants treated with MBC-HCl alone, but the oil did not alter the general distribution pattern of MBC-HCl from the treated area to the rest of the plant. In other experiments, plants were treated with equal amounts of  $^{14}\text{C}$ -MBC-HCl or  $^{14}\text{C}$ -MBC each incorporated with the paraffinic oil or with a naphthenic oil (Orchex N 792) at different concentrations (0.0, 1, 5, 10, and 20%). Fourteen days after treatment when the leaves were analyzed for total radioactivity, the  $^{14}\text{C}$  in the leaves increased with increase in concentration of either oil between 1 and 10%. In the absence of oil, MBC-HCl was taken up and translocated more efficiently than MBC.