

## Abstracts of the 1971 Annual Meeting of the North Central Division of The American Phytopathological Society

*A controlled high relative humidity system for the study of corn leaf segments inoculated with Phyllosticta zeae.* A. BOOTSMA, T. J. GILLESPIE, & J. C. SUTTON (Univ. Guelph, Ontario, Can.). Saturated air streams at two different temperatures provided a means of maintaining high relative humidity (RH) inside a 17- $\times$ 8- $\times$ 4.5-inch chamber in the dark at 20 C. Chamber air was sampled through a dew point hygrometer that monitored dew point depression continuously. The system was capable of controlling RH to within  $\pm 0.6\%$ . Corn-leaf segments were inoculated with *Phyllosticta zeae* conidia and incubated at 94, 96, and 98% RH. Germination maxima were 0, 29, and 60%, respectively, after 20-hr incubation. In the presence of free moisture, germination was 95%. Lesions developed only on corn leaf segments maintained under free-moisture conditions. This humidity controlling system is adaptable for studying other host-pathogen combinations on intact leaves.

*Mechanical injury of epidermal cells in local lesion virus infection.* I. P. BUTZONITCH & W. J. HOOKER (Mich. State Univ., E. Lansing). After mechanical inoculation of upper leaf surfaces of *Tetragonia expansa* with tobacco necrosis virus, silicone rubber negative replicas were made of epidermal cell surfaces at intervals for recording evidence of injury. Positive replicas were then prepared with transparent nail polish. Approximately 1.2% of the epidermal cells appeared injured or had collapsed 45 min after inoculation. Of such cells, ca. 20% recovered within 21 hr after inoculation. Surfaces were again smooth and cells appeared turgid. The remaining 80% of injured cells collapsed further and appeared dead. Lesions did not become apparent until 24-28 hr after inoculation. They were first evident as several flaccid cells. Adjacent cells developed similar symptoms, and within 4-5 hr, the lesion had become macroscopically evident. In ca. one-half the lesions observed, a cell which showed initial flaccidity at the 45-min observation and later recovered was present in the lesion center. The other lesions were not preceded by cell flaccidity, and seemed to have developed from cells which appeared uninjured during the period of observation. Epidermal cells similarly treated without virus evidenced either injury followed by recovery or collapse. Local lesions did not form.

*Pathogenicity of Penicillium species on corn ears.* R. W. CALDWELL & J. TUIE (Purdue Univ., Lafayette, Ind.). The pathogenicity of 12 species of *Penicillium* on a double-cross corn hybrid, Indiana 253, was evaluated in 1969 and 1970 field studies in Indiana. At full silk or at the onset of denting, ears were injected at the butt and tip, or the silks were sprayed with a spore suspension. Pathogenicity was evaluated by estimating the per cent of the ear visually rotted and also by determining the kernel infection by plating out surface-disinfected kernels on corn steep agar containing 100 ppm Tergitol NPX. Injection at full silk was the most effective inoculation procedure, and resulted in 5-15% ear rot by *P. oxalicum*. None of the other species tested caused visible ear rot. Over 50% of the kernels from ears injected with each of seven *P. oxalicum* and seven *P. funiculosum* isolates were infected. At least one of the seven *P. cyclospium*, six *P. citrinum*, seven *P. expansum*, five *P. variabile*, and seven *P. viridicatum* isolates tested gave 10-50% seed infection. Kernel infection by seven *P. brevis-compactum*, two *P. frequentans*, three *P. palitans*, two *P. purpurogenum*, and two *P. urticae* isolates was less than 10%. Delaying harvest 2 months increased seed infection as follows: *P. brevis-compactum*, 10%; *P. citrinum*, 15%; *P. cyclospium*, 10%; and *P. viridicatum*, 33%.

*The effect of harvest technique on recovery of wheat from experimental plots with and without stem rust.* L. CALPOUZOS, M. E. MADSON, & J. R. WELSH (Univ. Minn., St. Paul, Colo. State Univ., Ft. Collins). Loss of yield due to normal harvesting methods was determined from plots of wheat rusted with *Puccinia graminis tritici* and non-

rusted plants in Minnesota and Colorado. In plots containing up to eight 5-m rows, all wheat heads were collected by hand from 1-m lengths of three interior rows. The remaining wheat (minus border areas) was harvested with a 2-row cutting rig and a Vogel threshing machine. The hand-picked heads were threshed by hand, and every grain was collected. Five wheat cultivars were tested and replicated 5 times, each cultivar with three treatments: early epidemic; late epidemic; and disease-free. Mean yield was consistently lower, and error mean square of yield was consistently higher for machine harvest compared to hand harvest. For the five cultivars, the average yield loss due to machine harvest was 13% in disease-free plots, 19% in late epidemic plots, and 34% in early epidemic plots, showing that the percentage loss from machine harvest operation increased with earlier epidemics.

*Some characteristics of the host-selective toxin from Helminthosporium maydis race T. J. C. COMSTOCK* (Mich. State Univ., E. Lansing). The fungus was grown in still culture on modified Fries' solution plus 0.1% yeast extract. Toxicity of culture filtrates to corn seedlings with Texas male sterility (TMS) reached a maximum in 12-15 days at 22 C. Culture filtrates (pH 3.5) were concentrated and extracted with methanol, followed by butanol. After removal of butanol, the aqueous solution was adjusted to 4% of the original volume of filtrate. This preparation caused 50% inhibition of root growth of normal (N) and TMS cytoplasm corn seedlings at 1:100 and 1:2,000 dilutions, respectively. Toxin purified further by gel filtration (Sephadex G10) caused 50% inhibition of TMS and N corn at 16 and 1,360  $\mu\text{g}/\text{ml}$ , respectively. Thirteen corn lines with N cytoplasm were tested and found to be approximately equal in tolerance; 10 TMS lines were much more sensitive. Tolerance of several nonhost plants (tomato, cucumber, radish, sorghum, barley, and *Lolium* sp.) was comparable to that of N corn. Crude and partially purified toxin was heat-labile. No reduction in toxicity was evident after storage at  $-10\text{C}$  for 30 days. Activity was reduced 50% in 24 hr at 25 C and pH 7.0, and  $< 20\%$  at pH 3.5. Toxin had an approximate  $R_F$  of 0.85 on paper chromatograms developed with propanol:acetic acid:water (200:3:100).

*A selective medium for isolation of pectolytic soft rot bacteria from soil.* DIANE A. CUPPES & A. KELMAN (Univ. Wis., Madison). In a comparison of various selective media for isolation of soft rot *Erwinia* spp. from soil, best recovery of *E. carotovora* was obtained with a modification of Beraha's pectate medium (CVP) containing: crystal violet, 1.5 mg;  $\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$ , 0.6 g;  $\text{NaNO}_3$ , 2 g; Na polypectate, 30 g; and agar, 3 g/liter. Soft-rot *Erwinia* isolates formed distinctive colonies in deep, circular depressions in the pectate-agar substrate. Percentage recovery of *E. carotovora* [(number of colonies on CVP medium  $\div$  number of colonies on optimal growth medium)  $\times 100$ ] was 77% from a cabbage field soil to which a suspension of *E. carotovora* was added 1 hr prior to isolation. On the CVP medium, ca. 90% of the background population of soil bacteria could be eliminated. In isolations with this medium from cabbage, potato, and carrot field soils, the pectolytic Gram-negative bacteria commonly found were not species of *Erwinia*, but fluorescent pseudomonads capable of rotting potato tuber slices. When 0.1 ml of a 50% aqueous solution of  $\text{MnSO}_4 \cdot \text{H}_2\text{O}$  was added to the CVP medium by a surface-plating procedure, the percentage recovery of *E. carotovora* from field soil to which it was added remained about the same, but the numbers of other Gram-negative bacteria, including the pectolytic pseudomonads, were markedly reduced.

*A model system for studying systemic fungicides.* L. V. EDGINGTON & J. SCHOOLEY (Univ. Guelph, Ontario, Can.). A model system for evaluating system activity of fungicides was developed. We applied the test fungicide in a 50- $\mu\text{liter}$

aliquot as a band across a cucumber leaf, and studied subsequent movement of fungicide by its therapy of powdery mildew-diseased cucumber and bioassay of plant tissue. Mildew was rated 6-7 days after inoculation and fungicide application to study basipetal vs. acropetal movement, uptake via adaxial vs. abaxial leaf surface, and uptake in veinal vs. interveinal leaf areas. Concomitant studies in which leaf sections were frozen and bioassayed on potato-dextrose agar against *Penicillium* were used to corroborate the mildew therapy tests. We have used this method to determine that Thiabendazole [2-(4-thiazolyl) benzimidazole] moved only acropetally, would penetrate either adaxial or abaxial surfaces equally well, and was taken up similarly in both veinal and interveinal areas.

*Failure to recover turnip mosaic virus from infected mature cabbage and cauliflower.* I. R. EVANS (Univ. Guelph, Ontario, Can.). Three isolates of turnip mosaic virus (TuMV) were obtained from widely separated localities in Southern Ontario and maintained in Tendergreen mustard. The three isolates were readily aphid- and mechanically transmissible to Wisconsin Golden Acre cabbage and Stokes Perfected Snowball cauliflower seedlings at the first true leaf stage. Each isolate induced a severe mosaic and chlorotic ring-type symptom in subsequently formed leaves of cabbage and cauliflower seedlings, respectively. Negatively stained leaf dip preparations of each isolate from these leaves, examined with an electron microscope, revealed numerous filamentous particles characteristic of this virus. The three isolates could be transferred from these seedlings mechanically, or by aphids, to mustard or rutabaga. As growth of the seedlings continued, virus symptoms became less pronounced in both cabbage and cauliflower. At maturity (4 months after infection), none of the isolates could be recovered from cabbage or cauliflower tissue which appeared symptomless and indistinguishable from noninfected controls. Virus particles were not detected in leaf dip preparations made from both inner and outer leaves of these infected cabbage and cauliflower plants.

*Etiology, impact, and distribution of Swiss needle cast of Douglas fir in Michigan.* K. F. FORD & H. L. MORTON (Univ. Mich., Ann Arbor). *Phaeocryptopus gäumannii* produced perithecia on current year's needles of *Pseudotsuga menziesii* during the first week of August 1969. Periodic perithecial examination and daily attempts to collect ascospores showed that the spores were formed in early spring and were released intermittently from 19 May 1970 to 21 August 1970. Spore production was greatest during June and July. A correlation ( $R^2 = .39$ ) was found between total ascospores produced in a 24-hr period and total rainfall over 48 hr, including the previous day. A 21-year-old plantation, composed of 184 trees, was examined for disease impact. Trees were grouped into six classes based on percentage defoliation and infection. Eighty-eight per cent of the trees were infected; mortality amounted to 4% over the 1967-1970 period; 8% of the trees showed no infection. Mean height and diameter at breast height ranged from 25 ft 3.6 inches, respectively, for class I (healthy) trees, to 9.0 ft 1.4 inches, respectively, for class V (defoliation greater than one half) trees. Variation in growth loss may indicate inter- or intraspecific resistance. Infected plantations were found in the south central, southeastern, and northern Lower Peninsula of Michigan.

*The succession of organisms in preservative treated wood.* H. E. FUNKE, J. T. WORTENDYKE, & H. L. MORTON (Univ. Mich., Ann Arbor). To determine if there was a succession of microorganisms in wood treated with 98.5% creosote (C), 15% pentachlorophenol (P) in No. 2 fuel oil, or 4.5% tanalith (T) in water, stakes were placed in the soil at Ann Arbor (AA), Pellston (Pel), and Iron River (IR). After various exposure periods, five randomly selected stakes/treatment were removed from each plot. Isolations were made from two sides of each stake by aseptically removing  $\frac{1}{16}$ -inch-thick facings from the underground portion and

dissecting eight chips from the exposed surfaces at a point 2 inches below the soil. Four chips were placed on a preservative medium, and four on PDA. No microorganisms were isolated from the initial controls. After 8 months' exposure, all stakes remained sterile at Pel, while bacteria were found in the C and T treatments at AA and, in all treatments, at IR. No fungi were found. After 1 year, bacteria were isolated from all treatments at all field locations except at Pel, where bacteria had not colonized the P and T treatments. Fungi were found at all locations in C and T treatments only. These data show the importance of geographic location and preservative on microorganism succession, and support other studies where bacteria precluded fungal colonization of woody substrates.

*Seasonal fluctuations in susceptibility of apple to the collar rot disease.* J. E. GATES & D. F. MILLIKAN (Univ. Missouri, Columbia). The susceptibility of several commercially important apple trees to the collar rot disease was studied over a 3-year period. All cultivars were highly susceptible in mid-April. Other periods of susceptibility include late June-early July and late August-early September. These three periods occur during blossoming, flower bud differentiation, and the onset of dormancy. The highly susceptible cultivar, Grimes Golden, may show an additional period of susceptibility in mid-March. Biochemical investigations of an alkaline extract of the inner bark tissue showed the presence of a substance which stimulated the in vitro growth of *Phytophthora cactorum* and appeared to be quantitatively associated with susceptibility. The precise identity of these substances has not been determined, but their UV absorption is similar to that of nucleic acids. These observations concerning the timing of host susceptibility are useful in determining the timing of applications of chemicals to control the pathogen.

*Inhibition of soybean hypocotyl elongation by Rhizoctonia solani.* C. R. GRAU & C. A. MARTINSON (Iowa State Univ., Ames). *Rhizoctonia solani* inhibits soybean hypocotyl elongation when used at moderate inoculum levels. This phenomenon is exhibited when seedlings are grown in Vermiculite or natural soil infested with *R. solani*. Hawk-eye soybeans were used because of their rapid emergence. Seedlings were grown in plastic vegetable crispers in a growth chamber at five temperatures ranging from 20 to 30°C in total darkness. The degree of inhibition was a function of temperature and inoculum density when compared to healthy controls; optimum temperature for inhibition of hypocotyl elongation was 25°C. The amount of inhibition varied with different *R. solani* isolates. The seedcoats were colonized within 24 hr after planting, and the degree of inhibition was correlated with the length of time the seedcoat remained attached to the cotyledons. Hypocotyl lesions sometimes formed were not necessarily associated with the inhibition phenomenon.

*DIALNOSE: a computer-assisted disease diagnosis program for use with a telephone.* A. L. JONES (Mich. State Univ., E. Lansing). A FORTRAN II program developed by Larry Morse for identifying plant specimens at Michigan State University was modified to assist in plant disease diagnosis. Information on disease symptoms used in the identification program are coded into a matrix by crop, with each element describing how specific disease/symptom couplet combinations relate. A predetermined disease and symptom description list and a coding sheet assist the user in executing the program from touch-tone telephone or teletypewriter. The operator can readily interact with the computer to adjust symptom information to obtain a probable diagnosis. Failure to obtain a complete answer initially results in a listing of useful symptoms for separating the remaining diseases. Following each diagnosis, a list of signs and symptoms is provided for the identified disease in decreasing order of importance. The program is written in FORTRAN IV for use with a time-sharing computer system equipped with an audio response unit. Though de-

signed primarily for use by Cooperative Extension workers from their offices, it may be used in plant disease clinics or in courses on disease diagnosis.

*Reaction of Phaseolus lunatus to Pseudomonas syringae.* D. J. HAGEDORN, R. E. RAND & S. M. SAAD (Univ. Wis., Madison). Thirty-nine lines of lima beans (*Phaseolus lunatus*) from commercial sources and 344 Plant Introductions (P.I.'s) were tested in the field and greenhouse for their reaction to *Pseudomonas syringae*. A technique for supplementing naturally occurring inoculum was used, wherein lima bean seeds were dusted with dried, finely ground, infected leaves, and planted every third row in field plots. An atomizer at 15 psi was used to inoculate young, first trifoliate leaves of greenhouse-grown plants. Inoculum was grown on nutrient-agar-glycerol medium for 24 hr. The cells were washed with sterile distilled water, and inoculum level was adjusted to  $1.5 \times 10^8$  cells/ml. In the 1969 field plot, 42 P.I.'s appeared to have tolerance to *P. syringae*, but when 16 promising lines were tested in the greenhouse, they were susceptible. The 1970 field plot contained 143 of the best P.I.'s from 1969 and 23 new accessions. Fourteen P.I.'s were significantly more tolerant than the control, Early Thorogreen, the most commonly grown cultivar in Wisconsin. When 12 of the most tolerant lines were tested in the greenhouse, they were susceptible. The only lima bean which was tolerant in both field trials was P.I. 183412, now being evaluated for disease reaction in the greenhouse.

*Systemic activity of benomyl fungicide against Cladosporium leaf mold and Verticillium wilt of greenhouse tomatoes.* J. C. LOCKE & R. J. GREEN, JR. (Cornell Univ., Ithaca, N.Y., Purdue Univ., Lafayette, Ind.). Benomyl fungicide showed systemic activity in tomato against *Cladosporium* and *Verticillium* with a combination of preventive and curative fungicidal properties. This study utilized three methods (tissue plating, tissue incorporation, and vascular fluid incorporation) in assaying for benzimidazole carbamic acid, methyl ester (BCM), the breakdown product of benomyl, in plant tissue. Vascular fluid incorporation was the most quantitative method. *Penicillium oxalicum* was completely inhibited in vitro at 0.5 ppm benomyl, and *P. expansum* and *P. funiculosum*, at 0.1 ppm. Benomyl at 25 ppm either as a soil incorporation or a soil drench was effective in protecting tomato from infection by *V. albo-atrum*. A 25-ppm benomyl soil incorporation also protected against *C. fulvum* infection. At this level of treatment, no detrimental effect was noted on foliage or root system development. Although BCM accumulated in leaves at concentrations of at least 25 ppm, the compound was not detected in mature fruit.

*Elm mosaic virus in Iowa.* D. E. MAYHEW & A. H. EPSTEIN (Iowa State Univ., Ames). Three declining Moline elm (*Ulmus americana*) trees observed in western Iowa showed symptoms of dieback of upper branches, sparse foliage, smaller leaves with chlorotic ring patterns, and enations growing interveinally on the undersides. The disease was established in American elm seedlings in the greenhouse by bud-grafting from diseased trees and by mechanical inoculation using several different buffers. All symptoms except the enations were reproduced under experimental conditions. The herbaceous hosts *Nicotiana*, *Cucurbita*, *Vigna*, and *Chenopodium* species (some previously unreported) developed necrotic and chlorotic lesions and ringspots when inoculated. The causal agent was identified as elm mosaic virus (EMV), based on host range similarities and positive microprecipitin and immunodiffusion tests when reacted with EMV antiserum provided by J. P. Fulton, University of Arkansas. This is the first report of EMV in Iowa.

*The distribution of maize dwarf mosaic, bromegrass mosaic, and clover yellow mosaic virus in roots of corn, wheat, oats, and peas.* H. E. MOLINE (Iowa State Univ.,

Ames.). Electron-microscopic examination of root tips of peas, wheat, oats, and corn showed that clover yellow mosaic (CYMV), bromegrass mosaic (BMV), and maize dwarf mosaic virus (MDMV) occurred in meristems of infected roots. The viruses were applied by rubbing, spraying, pin pricking, or injecting roots of plants grown on germination papers. Concentrations of the three viruses in roots and shoots were measured by infectivity assays. CYMV infected pea roots after transmission by all methods, and was readily translocated to shoots from inoculated roots. The concentration of CYMV recovered from pea roots was as high as that recovered from shoots. BMV infected wheat and oat roots after transmission by rubbing, but infection of corn roots was erratic. BMV was translocated to shoots from inoculated wheat and oat roots but not from corn roots. BMV concentrations were similar in roots and shoots of wheat and oats, but only low quantities of virus could be recovered from corn roots. MDMV rarely infected corn after inoculation of roots, but it was translocated from shoots to roots. However, MDMV was recovered only in low quantities from those roots.

*Systemicity of Thiabendazole and Terrazole in tomato seedlings.* G. J. MULLER & M. B. LINN (Univ. Ill., Urbana.). Thiabendazole[2-(4-thiazolyl) benzimidazole] (TBZ) and Terrazole [5-ethoxy-3-(trichloromethyl)1,2,4-thiadiazole] were found to be systemic in Bonny Best tomato seedlings. These compounds are known to be systemic in other crops. The roots of twenty 2-week-old seedlings growing in vermiculite were exposed to 25, 50, or 100 mg/liter active ingredient of the wettable powder formulation of TBZ or the emulsifiable concentrate of Terrazole. Three tissue samples (1 g each) were taken from (i) entire plants, except roots; (ii) stems only; or (iii) leaves and petioles. Each was bioassayed separately using *Penicillium atrovenetum* to detect the presence of TBZ and *Rhizoctonia solani* to identify Terrazole. Bioassays showed the presence of the fungitoxicants in all tissues from treated plants. Thin-layer chromatography showed that the toxicants had the same  $R_F$  values as the original compounds. Assays of seedlings transferred from treated to nontreated medium detected TBZ up to 7 days after removal and Terrazole for only 1 day. After 48-hr exposure to the fungitoxicants, seedlings showed symptoms of toxicity.

*Association of maize dwarf mosaic virus with the small seed disease of grain sorghum.* C. L. NIBLETT & L. K. EDMUNDS (Kansas State Univ., ARS, USDA, Manhattan, Kans.). Field plots of RS 671 hybrid sorghum were subjected to naturally occurring aphid populations and below-normal temperatures at several stages in development of inflorescences. Sorghum greenbug (*Shizaphis graminum*, biotype C), a vector of maize dwarf mosaic virus (MDMV), was present from anthesis to milk stage in different plantings. When minimum temperature of  $< 16$  C for 4 or more consecutive days coincided with soft dough stage, pigmented, necrotic lesions developed on rachises and rays of panicles. Premature shrinkage of seed ensued on affected panicles. A virus isolated from these panicles was partially purified and identified as MDMV, strain A. Similar small seed was reproduced in the greenhouse when RS 610 sorghum plants were inoculated at anthesis with this virus, then subjected to  $< 16$  C between milk and soft dough stage. Results closely follow observations made among irrigated grain sorghum fields in southwestern Kansas in recent years, where both sorghum greenbug and corn-leaf aphid (*Rhopalosiphum maidis*) have occurred after panicle exertion, indicating that MDMV-A is the primary cause of the small seed disease.

*Uptake and translocation of benomyl in creeping bentgrass.* J. F. NICHOLSON, W. A. MEYER, & J. B. SINCLAIR (Univ. Ill., Urbana). Root uptake and translocation of benomyl and its breakdown product, MBC (methyl-2-benzimidazole carbamate) were studied in Toronto creeping bentgrass (*Agrostis palustris*) stolons having either one

or two root systems. The roots of stolons with a single root system were treated with 5, 12.5, or 25 mg/liter benomyl for 24, 48, or 96 hr. Bioassay and thin-layer chromatography showed presence of fungistatic compounds throughout all tissues of stolons with a single root system. This showed translocation of benomyl upward in the transpiration stream to the growing point. When benomyl at 100, 500, or 1,000 mg/liter was sprayed on leaves, only upward translocation was noted. Using a double-cup technique, a second and separate root system was established five nodes from the first. When this system was exposed to either 25, 100, 500, or 1,000 mg/liter benomyl for 4 days, the fungistats were found to be translocated against the transpiration stream and back into the crown of the first root system. This indicated lateral movement of the fungistats.

*Relationship of cell length and colony type in Pseudomonas syringae.* J. D. OTTA & O. M. BAIN (South Dakota State Univ., Brookings). Rough and smooth cultures of *Pseudomonas syringae* cannot be differentiated serologically on the basis of their heat-stable antigens. Apparently, some factor other than a change in the heat-stable antigen is responsible for the appearance of rough colony types in this species. One obvious difference between rough and smooth cultures is the occurrence of extremely long cells in rough cultures. Therefore, we measured the cell lengths of 18 rough and 55 smooth cultures grown for 24 hr on King's Medium B agar to determine if there was a relationship between cell length and colony type. The average cell lengths of 55 smooth cultures ranged between 1.1 and 3.8  $\mu$ , whereas the averages of 18 rough cultures ranged between 5.4 and 29.9  $\mu$ . Individual cell lengths in rough cultures varied from 1.0 to 113  $\mu$ , whereas most cells of smooth cultures varied from 0.5 to 10  $\mu$  with a few rare cells longer than 10  $\mu$ . Increased cell length may be responsible for the appearance of rough colony types in previously smooth cultures of *P. syringae*. The controlling factor governing the appearance of long cells is unknown at present.

*An inducer of soybean phytoalexin.* J. D. PAXTON (Univ. Ill., Urbana). A phytoalexin inducer, which chemical tests indicate is a glycoprotein, is produced by *Phytophthora megasperma* var. *sojae*. Equal amounts of this inducer stimulate equal amounts of phytoalexin production in soybean cultivars which are resistant (Harosoy 63 and L2A) and susceptible (Harosoy) to *P. megasperma* var. *sojae*. The susceptible plant, however, does not initiate formation of the inducer by this fungus, whereas plants of the resistant cultivar can do so. Filter-sterilized sap from the susceptible plant also will not stimulate inducer production by *P. megasperma* var. *sojae*. This sap does not contain inhibitors of inducer formation, since mixed sterile sap of resistant and susceptible cultivars is a good substrate for inducer production. The inducer is catalytic in stimulating phytoalexin production, as concentrations of the inducer, one-half, one-fourth, or one-eighth the original concentration induce equal quantities of phytoalexin production from soybean cotyledons. Further studies are underway to identify the inducer and the plant substance (trigger) necessary for its production.

*Inhibition of bacterial blight of soybean by Bdellovibrio bacteriovorus.* R. H. SCHERFF (ARS, USDA, Univ. Missouri, Columbia). *Bdellovibrio bacteriovorus*, a small, comma-shaped bacterium, parasitic on some Gram-negative bacteria, was isolated from the rhizosphere of soybean roots. Three of the *B. bacteriovorus* isolates (Bd-10, Bd-17, and Bd-19) were each mixed with *Pseudomonas glycinea* at ratios of 1:1, 9:1, and 99:1, and rubbed onto the Carbendum-dusted trifoliates of 21-day-old Clark 63 soybean plants. The initial cell concentrations were  $10^8$  cells/ml. When Bd-17 was inoculated onto soybean trifoliates with *P. glycinea* at ratios of 9:1 and 99:1, bacterial blight symptom development was completely inhibited. Bd-10 caused no reduction, and Bd-19 was intermediate in in-

hibiting symptom development, as compared to plants inoculated with *P. glycinea* alone. There was little effect on disease development by any of the *B. bacteriovorus* isolates at the 1:1 ratio. The observed differences among *B. bacteriovorus* isolates in affecting bacterial blight development cannot be explained by differences in their efficiencies to kill cells of *P. glycinea* in vitro, because all three isolates were equally effective in producing plaques on lawns of *P. glycinea*.

*Variability in growth and sporulation among five isolates of Cephalosporium gregatum.* R. W. SCHNEIDER, J. B. SINCLAIR, & B. L. KIRKPATRICK (Univ. Ill., Urbana). Differences in cultural characteristics, growth rates, and sporulation of *Cephalosporium gregatum*, incitant of brown stem rot of soybean, were compared among five isolates from: Iowa (I), North Carolina (N), Illinois (L), Mexico (M), and the American Type Culture Collection (A). Isolates were grown on soybean-seed (SSA) and potato-dextrose agar (PDA) at 15, 20, 25, and 30 C for 20 days. Colonies were measured every 4 days. Colonies of M were reddish brown at all temperatures, and produced three or four concentric rings. Production of dark-brown-to-black conidia in colony centers of isolates N, I, M, and A on PDA at all temperatures after 12 and 16 days is reported for the first time. Only A produced these conidia on SSA at 25 C after 16 days. Optimum temperatures for growth on SSA were 20 C for N and L; and 25 C for I, M, and A. Optimum temperatures for sporulation were 15 C for N; 20 C for I, L, and A; and 25 C for M. L produced a greater number of conidia than did all other isolates at all temperatures. There were significant differences in growth rates among isolates within each temperature.

*Brome mosaic virus: Fixation enhancement in thin sections of young leaf cells of Zea mays.* J. D. SMOLIK & W. S. GARDNER (S. Dak. State Univ., Brookings). We have had difficulty identifying brome mosaic virus (BMV) in dense cytoplasm of root and shoot growing points. Now, we have resolved massive amounts of BMV, both loose and crystallized, in cytoplasm of very young corn leaves. Tissue was fixed in Langenberg's solution (5% glutaraldehyde in 0.05 M potassium phosphate buffer pH 6.9 containing 0.12% thioglycolic acid). Samples were rinsed 4 times in buffer, dehydrated in acetone, embedded in Araldite-Epon, sectioned, and stained with uranyl acetate and lead citrate. Osmic acid was accidentally omitted from some of the samples, but later ones were postfixed in buffered 1% OsO<sub>4</sub>. BMV was sometimes seen in samples fixed in OsO<sub>4</sub>, but other cellular constituents often obscured the virions. The full extent of intracellular BMV was revealed in samples not treated with OsO<sub>4</sub>. In these, the only commonly distinguishable cellular components were cell walls, cytoplasmic virions, and chromatin in the nuclei. Many of the virions appeared to have hollow centers. Viral- and nuclear-nucleoprotein apparently did not require OsO<sub>4</sub> for adequate fixation.

*Partial purification of pea seed-borne mosaic virus.* W. R. STEVENSON & D. J. HAGEDORN (Univ. Wis., Madison). The pea seed-borne mosaic virus was partially purified using the following procedure: Dark Skin Perfection pea was propagated 8 days at 24 C following inoculation. Infected tissue, chloroform, and 0.5 M borate buffer pH 7.5 in a 1:1:1 ratio was blended with 15% Al<sub>2</sub>O<sub>3</sub> and 0.02 M 2-mercaptoethanol, and incubated for 2 hr at 4 C. The emulsion was centrifuged at 2,200 g for 15 min, and the aqueous phase collected. Virus was precipitated from the aqueous solution by adding 0.02 M NaCl and 4% polyethylene glycol (mol wt 6,000), incubated for 1 hr at 4 C, and centrifuged at 9,000 g for 15 min. Pellets were resuspended in 0.05 M borate buffer and clarified by low-speed centrifugation. This step was repeated, and pooled supernatants were subjected to at least two cycles of differential ultracentrifugation. Pellets were resuspended in 0.05 M borate buffer and the final solution was colorless and opalescent.

Typical nucleo-protein absorption curves for a rod-shaped virus were obtained with 260:280 ratios of 1.15:1.25. Yields ranged from 50-80 OD units/ml from 120 g starting material. Inoculum with an OD of  $1 \times 10^{-5}$  was infectious for pea. Upon sucrose density-gradient centrifugation, five distinct and reproducible bands were detected.

*Direct comparisons of physical properties of maize dwarf mosaic and sugarcane mosaic virus strains.* M. TOSIC & R. E. FORD (Univ. Belgrade, Yugoslavia, Iowa State Univ., Ames). Physical properties of maize dwarf mosaic virus (MDMV), strains A and B, and of sugarcane mosaic virus (SCMV), strains A, B, D, H, Calif. Jg, and I were compared using uniform host, test, and environmental conditions. The host for virus replication was Golden Bantam sweet corn. Viruses were extracted from 14-day infected leaves, diluted 1:10 in 0.01 M K phosphate buffer at pH 7.0, treated, and assayed on sweet corn. Thermal inactivation of MDMV and SCMV strains were not significantly different. No infection occurred at 58 C; most isolates and strains were infective at 56 C; and five MDMV-A isolates were infective at 54 and three at 56 C. Dilution end points were most variable; MDMV-A and -B = 20,000; SCMV-B = 10,000; and SCMV-A, -D, -H, and Calif. Jg = 5,000. Longevity in vitro of MDMV and SCMV was similar, 12-14 hr in undiluted sap and 24-48 hr in buffer. Phosphate buffer specifically enhanced infectivity. MDMV-A seemed more stable in undiluted plant sap than MDMV-B or SCMV strains. This difference disappeared when buffer was used. Significant differences among physical properties of MDMV and SCMV strains were not apparent; therefore, valid criteria for strain differentiation must be resolved by host range, serological, and chemical comparisons.

*Monroe bean as a new local lesion host for bean common mosaic virus.* G. E. TRUJILLO & A. W. SAETTLER (Mich. State Univ., E. Lansing, ARS, USDA). Primary leaves of Navy (pea) bean (*Phaseolus vulgaris* 'Monroe') seedlings were dusted with Carborundum and rub-inoculated with sap expressed from plants infected with the type and New York 15 strains of bean common mosaic virus. Under greenhouse conditions of 22-27 C and natural light, necrotic local lesions ca. 1 mm in diam were consistently obtained. Lesions were initially observed and could be counted at 4-5 days after inoculation as red-brown spots on the upper leaf surface. Except for slight vein necrosis adjacent to a few lesions, no evidence of systemic virus movement was found. Single local lesions macerated in a small amount of buffer and inoculated to Carborundum-dusted primary leaves induced typical systemic common mosaic virus symptoms on the susceptible bean cultivar Rainy River. Number of local lesions produced was in-

versely related to dilution of inoculum, and was relatively uniform for opposite half-leaves. The local lesions formed on Monroe differ from those reported previously on other bean cultivars.

*Comparisons of Fusarium spp. and populations in cultivated and noncultivated soils.* CAROL E. WINDELS & T. KOMMEDAHL (Univ. Minn., St. Paul). Paired samples of cultivated (corn) and noncultivated (grasses, composites, and legumes) soil were collected throughout Minnesota for bioassay of *Fusarium* spp., sieved to < 250- $\mu$  particle size, and uniformly distributed on agar plates with an Andersen sampler (0.010 mg/plate). Usually, more colonies of *Fusarium* spp. developed from cultivated than from noncultivated soils. The highest ratios of occurrence of *Fusarium* in cultivated and noncultivated soil, respectively, were: *F. oxysporum*, 3:2; *F. roseum*, 5:1; and *F. epispheeria*, 6:1; but *F. solani* was up to 1.5 times more prevalent in noncultivated soil. *Fusarium moniliforme* occurred infrequently and subequally in both soils. Of *F. roseum* cultivars, Equiseti colonies were more numerous than both Graminearum and Avenaceum colonies combined. Thus, cultivation may differentially alter populations of *Fusarium* spp. in soil.

*The inheritance of downy mildew resistance in sunflower.* D. E. ZIMMER & M. L. KINMAN (ARS, USDA, Fargo, N.D., College Station, Texas). Field and greenhouse evaluations of over 200 introductions, lines, and cultivars were made in search of resistance to the downy mildew fungus, *Plasmopara halstedii*. HA 61 and two other rust (*Puccinia helianthi*)-resistant selections from the cross 953-88-3  $\times$  Armsvirsky 3497 were highly resistant to downy mildew. The Canadian line, 953-88-3, is known to carry the  $R_2R_2$  gene pair for rust resistance. All other entries, including the Romanian line, AD 66, and the rust-resistant Canadian lines, CM 90RR and S-37-388RR (reported to be downy mildew-resistant in their respective countries) were susceptible. The number of downy mildew-resistant and susceptible plants in the  $F_2$  and test cross  $F_1$  generations of crosses between HA 61 and lines susceptible to downy mildew and rust satisfactorily fit 3:1 and 1:1 ratios, respectively ( $P > .8$  and  $.9$ ). When tested to *P. helianthi*, race 1, the downy mildew-resistant  $F_2$  and test cross  $F_1$  plants segregated in ratios of 3 resistant:1 susceptible and 1 resistant:1 susceptible, respectively ( $P > .3$  and  $.2$ ). We conclude that the downy mildew resistance of HA 61 is conditioned by a dominant gene pair that we have designated  $Pl_2Pl_2$ . This gene pair is inherited independently of the  $R_2R_2$  gene pair that supposedly conditions the rust resistance of HA 61.

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