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Spectographic Analysis of Water Agar, Potato-dextrose Agar, and V-8 Juice Agar. PATRICK M. MILLER. Formulae for many biological media specify use of agar that has been purified of contaminating metallic ions by some manner, as by washing with pyridine. Yet there is little readily available information on what these ions might be or in what concentration they are present in water agar. In addition, comparison of the metallic ion contents of V-8[1] juice agar and potato-dextrose agar (PDA) might aid in determining the reason why growth of several fungi on V-8 juice agar was superior to that on PDA.[2,3] Hence, analyses of Difco bacto-agar (water agar), Difco potato-dextrose agar (PDA), and V-8 juice agar[2] were made with an ARL 1.5-m spectograph. The following results are expressed as ppm of each metal that would be present 1) in 2 per cent water agar; 2) in 3.9 per cent PDA; and 3) in V-8 juice agar containing 2 per cent water agar, 3 g of calcium carbonate, and 200 ml of V-8 juice per liter. Amounts (in ppm) found in water agar, PDA, and V-8 juice agar, respectively, are as follows: potassium—0, 312, 400; calcium—120, 120, 800; magnesium—24, 39, 40; phosphorus—8, 19, 36; sodium—20, 39, 102; manganese—0.0, 0.1, 0.2; iron—2.0, 3.9, 2.8; aluminum— 3.8, 5.1, 2.6; copper—0.08, 0.31, 0.40; and boron—2.6, 3.5, 2.8. Copper is present in PDA and V-8 juice agar in amounts adequate to be toxic to some fungi, particularly some of the Phycomycetes. The absence of potassium and manganese in water agar might explain the failure of some fungi to develop on this media. Calcium and sodium are more plentiful in V-8 juice agar than in PDA, but there are adequate amounts of calcium in PDA and the significance of sodium in the nutrition of fungi is doubtful. Hence, the superior nutritional properties of V-8 juice agar must reside in other than its metallic ion content.—Department of Plant Pathology and Botany, The Connecticut Agricultural Experiment Station, New Haven 4, Connecticut.

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[2] Diener, U. L. 1952. A method for inducing abundant sporulation of Stemphylium solani in pure culture. (Abs.) Phytopathology 42: 7.
[3] Miller, P. M. 1955. V-8 juice agar as a general-purpose medium for fungi and bacteria. Phytopathology 45: 461-462.

Storing Inoculum of Pseudomonas glycinea in Host Tissue by Freezing.[1] F. I. FROSHEISER.[2] Cultures of *Pseudomonas glycinea* Coerper isolated from infected soybean leaves and maintained on agar media were sometimes unsuitable for use in producing bacterial blight when inoculated onto soybean plants. Apparently the bacteria lose their virulence when kept on agar media. A method of maintaining virulent inoculum of the alfalfa wilt bacterium, *Corynebacterium insidiosum* (McCall.) H. L. Jens., by keeping naturally infected host tissue in a freezer has been described.[3] It seemed possible that the same procedure might be effective with *Pseudomonas glycinea.*

Soybean leaves naturally infected with *P. glycinea* were collected in the field in August, put in plastic bags, and placed immediately in a freezer maintained at approximately $-18°C$. In June of the following year, leaves were removed from the freezer, placed in tap water for 30 minutes, then comminuted in a Waring Blendor for 3 minutes. The suspended material was filtered through several layers of cheesecloth to remove particles that might plug a sprayer nozzle, diluted with more water, and sprayed onto 6-week-old soybean plants in the field. A heavy infection of blight resulted.

Inoculum was similarly prepared from infected leaves that had been kept in the freezer for 20 and 31 months. For the preparation of the inoculum, 10 leaflets of average size, on which bacterial lesions covered approximately 2 per cent of the leaf area, were used for each liter of water. Leaves of soybean plants in the 1-trifoliate-leaf stage were inoculated in the greenhouse. Abundant infection was obtained with the inoculum of both ages.

This method of storing inoculum of *P. glycinea* requires very little time and effort, and the bacteria retain their virulence for more than 2 years. It also is very possible that such inoculum would comprise more virulent biotypes than would pure cultures maintained on agar media.—Department of Plant Pathology and Botany, Institute of Agriculture, University of Minnesota, St. Paul 1, Minnesota.

[1] Paper No. 3529, Scientific Journal Series, Minnesota Agricultural Experiment Station.
[2] Formerly Research Assistant, Department of Plant Pathology and Botany, University of Minnesota.
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ing the leaves before inoculation was greater than when the plants were incubated in the dry air of the greenhouse.

Heat treatments applied before or after inoculation with viruses on hosts other than bean have not as yet resulted in definite increases in numbers of lesions or in size of lesions. Heavy infection but no increase in local lesion numbers due to heat treatment has been observed for tobacco necrosis virus on sunflower; for tobacco necrosis virus on cowpea (*Vigna sinensis* (Torner) Savi); for spotted wilt virus on squash (*Cucurbita maxima* Dcne.); and for peach yellow bud mosaic virus on squash, sunflower, and sugar beet (*Beta vulgaris* L.).

Sunflower rust (*Puccinia helianthi* Schw. on *Helianthus annuus* L.) was the only infection tested on a host other than bean where heating before inoculation clearly increased infection. For all paired comparisons in which the test leaves were treated at between 10 and 20 seconds at 50°, the treated leaves had more pustules than the controls in 14 out of 17 pairs, and the gross number of pustules counted was 1902 for the heated and 907 for the control.—Department of Plant Pathology, University of California, Berkeley.

Stripe Rust Resistant Mutants Obtained from Irradiation of Gabo Wheat.[1] C. F. KONZAK, N. E. BORLAUG, ARISTEO ACOSTA, AND JOHN GIBLER.[2] Gabo, a spring wheat variety of Australian origin, is widely adapted on the Pacific Coastal Plain of Mexico. This variety is also adapted in Central Mexico at elevations up to 6000 ft, but cannot be safely cultivated there because of its extreme susceptibility to stripe rust (caused by *Puccinia glumarum* (Schm.) Eriks. & E. Henn.). Since the climate of some of the higher valleys of Central Mexico favors the development of very severe epiphytotics of stripe rust nearly every summer, the field conditions in that region are ideal for the recognition and testing of any plants suspected as mutants for resistance to this disease.

Experiments were undertaken to attempt to produce rust-resistant mutants by irradiation of seeds.[3] Dry seeds of Gabo were irradiated with doses of 15,000r of X rays or about 2.3×10^{12} Nth/cm^2 thermal neutrons at the Brookhaven National Laboratory in 1953. First generation plants were grown in plots near Ciudad Obregon, Sonora, Mexico, and second-generation progenies from these plants were grown at the experimental station at Chapingo during the summer of 1954. Notes were not taken on stripe rust reaction during 1954 because infection was light in that year, but numerous agronomically typical and atypical Gabo plants were selected for further testing. The X_3 and N_3 progenies from the selected plants were grown at Chapingo during the summer of 1955, and in this year stripe rust was especially severe. One N_3 line bred true for a high degree of stripe rust resistance; otherwise it was apparently typical of the Gabo variety. Twelve additional progenies from the X-ray and neutron treatments showed segregation for various degrees of resistance. These resistant types were classified as 1, 2, and 3 on a scale of 0 to 5, in which 0 represented highly resistant reactions and 5 completely susceptible reactions; the reaction of the parental variety was 4++. No variants resistant to stripe rust were found among a large number of nonirradiated Gabo lines tested under the same conditions. The fact that a true-breeding third-generation line was obtained suggests that the mutation is recessive in inheritance and similar to the natural types. Since the

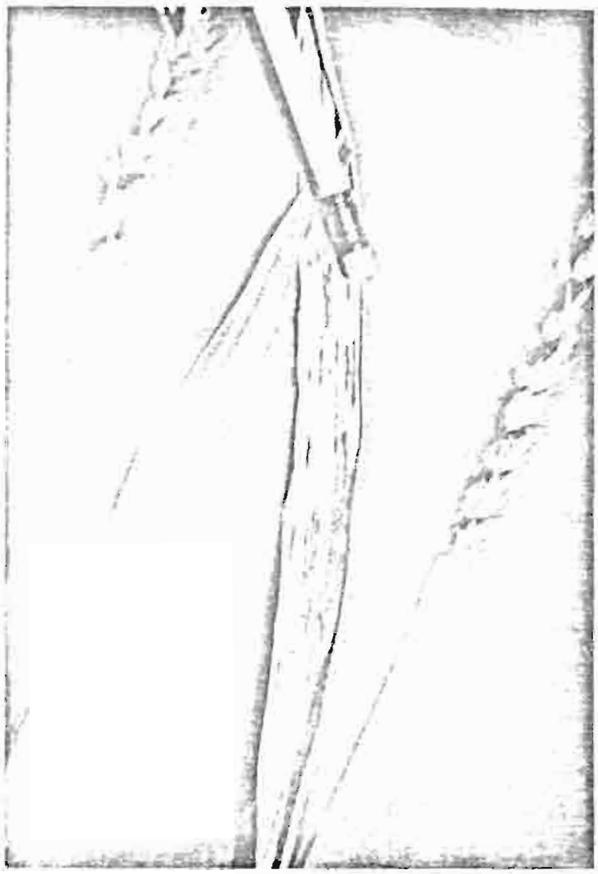

FIG. 1. Leaves showing stripe rust reaction. Left: mutant resistant to stripe rust, rust reaction type 1. Right: Gabo parent, rust reaction type 4++.

[1] Preliminary report on a cooperative project between Brookhaven National Laboratory and Rockefeller Foundation in Mexico through the Office of Special Studies of the Mexican Ministry of Agriculture. A portion of the work was carried out at the Brookhaven National Laboratory under the auspices of the U. S. Atomic Energy Commission.

[2] Respectively, Associate Geneticist, Brookhaven National Laboratory, Upton, L. I., N. Y.; Plant Pathologist, Rockefeller Foundation Mexican Program; Agronomist, Office of Special Studies del Secretario de Agriculture y Ganaderia; and Plant Pathologist, Rockefeller Foundation, now with the Colombian Agricultural program in Bogota, Colombia.

[3] Konzak, C. F. 1954. Stem rust resistance in oats induced by nuclear radiation. Agron. Jour. 46: 538-540.