A Simple Method for Producing Cercospora arachidicola Conidial Inoculum

D. H. Smith

Assistant Plant Pathologist, University of Georgia, College of Agriculture Experiment Stations, Georgia Station, Experiment, 30212.


The author thanks Brenda Campbell for technical assistance.

ABSTRACT

Abundant quantities of Cercospora arachidicola conidia were produced on a peanut leaflet-oatmeal agar medium when cultures were continuously illuminated under cool-white fluorescent lights for 14 days at 28 C. Phytopathology 61:1414.

Additional key words: peanut leaf spot, sporulation.

The Cercosporaceae have the following requirements for in vitro growth and sporulation (2, 6, 7). Cercospora arachidicola Hori, one of the principal peanut leaf spot fungi, grows slowly and sporulates poorly in vitro (1, 3, 5, 8). Landers (4) developed a chemically defined medium for C. arachidicola which is useful for quantitative measurement of vegetative growth. Abdou (1) found that C. arachidicola sporulated on peanut leaflet extract, oatmeal, lima bean, and mycophil agar media.

The purpose of this note is to describe a technique for producing abundant quantities of conidia for use in pathogenicity studies.

Peanut oatmeal agar (POA) used in this experiment is prepared as follows: Arachis hypogaea L. ‘Argentina’ leaflets (50 g) + 500 ml of distilled H₂O are placed in a Waring Blender for 10-15 sec, and the resultant slurry is filtered through cheesecloth. Oatmeal (15 g in 500 ml distilled H₂O) is boiled for 15 min, then filtered through cheesecloth. Equal volumes of peanut leaflet and oatmeal filtrates are combined with 20 g agar/liter and autoclaved for 15 min at 121 C.

Suspensions of spores and mycelium of C. arachidicola are stored in sterile distilled H₂O at 5 to 6 C. To prepare inoculum for greenhouse pathogenicity studies, a plastic syringe (manufactured by Becton, Dickinson & Co., Rutherford, N.J.) with a 23-gauge needle is used to flood each POA plate with 2 ml inoculum. The narrow orifice of the needle results in a more uniform dispersion of the spores and mycelium than does a pipette with a larger orifice.

Plates of POA are incubated for 14 days at 28 C. A single layer of cultures is continuously illuminated with two 15-w cool-white fluorescent lights at a height of 31 cm above the level of the cultures.

To prepare the inoculum, 20 ml sterile distilled H₂O and two drops of Tween 20 (polyoxyethylene sorbitan monolaurate) are added to each 90 mm petri dish culture. Conidia are easily suspended with a sterile camel’s hair brush.

We have used this method to produce inoculum for numerous pathogenicity tests and to screen more than 800 peanut introductions for resistance to C. arachidicola. With 10-15 POA plates, it is possible to obtain enough conidia to inoculate all leaves of 400 3-week-old plants. The reproducibility and simplicity of this procedure should encourage additional research on the pathogenicity of C. arachidicola.

LITERATURE CITED