P. F. KABLE, E. ZEHR & W. F. MAI: A simple apparatus for the study of soil moisture relations of plant parasitic nematodes under monoxenic conditions.

The apparatus herein described is reported because it is suitable for a wide range of studies on nematodes involving the interactions of environmental, host plant and soil microbial factors. The system is basically the modified Haines apparatus widely used by students of the moisture relations of nematodes (Haines, 1930; Jones, 1959) but it has the advantage of sterility. The apparatus is inexpensive compared with alternative equipment for maintaining sterility.

The items of equipment used in the apparatus are one 250 ml beaker, one 8 cm diameter Buchner type sintered glass funnel with ultra-fine porosity disc, one autoclavable transparent nylon film bag of approximately 15 X 30 cm, autoclavable adhesive tape, plastic tubing to fit stem of funnel, and glass tubing to fit plastic tubing.

The apparatus (Fig. 1) is prepared as follows. Approximately 100 ml of air dry soil, sand or other porous medium is placed in the 250 ml beaker. The volume of porous medium can be varied but it should be recognized that a small gradient in moisture tension will exist between the top and the bottom of the medium in the funnel. The importance of this gradient should be evaluated for each experiment, and will depend upon the depth and porosity of the medium and the tension used. An ultra-fine porosity sintered glass funnel of the same diameter as the beaker (approximately 8 cm) is loosely fixed stem upward to the top of the beaker by two small pieces of autoclavable adhesive tape. This unit is placed in an autoclavable nylon film bag with the stem of the funnel protruding. The bag is tightly sealed with autoclavable adhesive tape around the funnel stem. An access slit, approximately 2 cm long is made in the nylon bag at the junction of funnel and beaker. The slit is closed with autoclavable tape and the units sterilized by autoclaving. After cooling, plastic tubing is attached to the stem of the funnel, and distilled water is added to the funnel base and tube. The unit is then carefully inverted, so that the dry soil or medium is allowed to pour from the beaker into the funnel, and air is not permitted to enter the base of the funnel. By connecting the glass tube to the plastic tubing, and adding more water a simple water manometer is formed. The tube and water need not be sterile for many types of experiment since bacteria cannot pass through the ultra-fine sintered glass disc.

Tensions of up to 100 cm of water can be readily obtained in the test soil or medium with this arrangement, and higher tensions (up to approximately 600 cm water) may be reached by various arrangements of water and mercury manometers (Collis-George & Blake, 1959; Collis-George & Sands, 1959), pumps and manostats in the system (Jones, 1959).

The porous medium is saturated by reducing the tension to zero, and then the desired tension is applied. Seeds or seedlings of test plants, appropriately disinfested, are introduced with sterile instruments through the access slit, the beaker being lifted aside. Seeds or young seedlings are best introduced before saturation of the medium as they are embedded in friable air dry medium without the disturbance of structure which can occur in damp media. In many media germination of seeds of some plant species will not occur at low moisture tensions, probably because of poor aeration. A moisture tension appropriate to the medium and seed should therefore be applied during germination. After germination any tension desired may be applied.

Nematodes, free from contaminating micro-organisms, test micro-organisms, or mixture of micro-organisms, can be added at any position in the porous medium, at any time, by means of a sterile hypodermic needle. The access slit is resealed with tape after each operation. Constant moisture tension can be maintained for long time periods in such an apparatus. Constant lighting and temperature can be achieved by placing the apparatus in a growth chamber. Oxygen diffusion rates in the porous medium can be measured by platinum microelectrodes (Lemon & Erickson, 1955). Nutrition of plants growing in this apparatus could be manipulated by use of appropriate porous media and of nutrient solutions in the water manometer.

Contamination is always a possibility in this system, so each experiment should be well replicated. At the conclusion of each test, samples of the porous medium and plants from each funnel should be tested on several media of wide spectrum, to detect contaminating micro-organisms.

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Fig. 1. Modified Haines apparatus for control of soil moisture tension under monoxenic conditions.