For the student, the advantages are obvious; they earn some money, get some practical experience, get a real-life first hand look at their field of study. The experience enhances their employability after graduation. They may be more likely to remember what is taught in the classroom following practical work experience. By the same token, some previous teaching may take on new life and meaning.

For the cooperating employers in industry, the program can be what they make it. It enables them to look at the quality of one or several OH students presently enrolled at the University of Florida, and the program could conceivably be used as a continuous screening device for selecting future permanent employees. Some find that working with students can be stimulating and that students may have something of value to contribute that pay off eventually, in dollars and cents. Naturally, the cooperator has a closer tie with the OH Department and may feel more free to call on the faculty if the need arises. Since not every firm in the state can be a cooperator, there may be a source of pride involved, not to mention the satisfaction of helping guide an aspiring young person on a career that we ourselves have chosen.

So far the success rate of the OH 400 program is phenomenal and the OH Department is seeking cooperators to expand the program in every area of ornamental horticulture. If you are interested in giving one or more students an opportunity for full time practical work experience, please write or call the OH 400 advisor: Dr. Willard T. Witte, Ornamental Horticulture Department, University of Florida, Gainesville, FL 32611. (phone 904/392-1831) He is prepared to answer any questions a prospective cooperator may have.

Literature Cited

**A PARTIAL TISSUE CULTURE METHOD FOR PATHOGEN-FREE PROPAGATION OF SELECTED FERNS FROM SPORES**

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Abstract. Recently collected spores of several fern types were disinfested and successfully germinated on an agar-containing plant tissue culture (PTC) medium. Spores of maidenhair fern germinated within 5-7 days and prothallia were evident within 10-14 days. Approximately 3 weeks after initial culture of the spores, prothallia clumps were transferred intact to fresh PTC media. Clumps increased rapidly in size and required division and subsequent transfer to new media every 3-6 weeks. For several of the ferns tested, only the gametophytic stage was produced during culture in vitro on the agar-containing PTC medium. Gametophytic tissue comminuted for 5 sec in a blender containing 1/2 strength Murashige and Skoog (M&S) salts solution produced the sporophytic stage when transferred to soil. This method could be modified and developed for commercial production. It has the potential of reducing many of the manual transfers now required in current plant tissue culture propagation of ferns and appears to be effective in eliminating known-pathogens from propagative material.

Until recent years, ferns have been propagated by traditional methods (4) with new plants arising from spores, rhizome sections or crowns, divisions of mature plants, or from runners of mature plants that normally grow across the surface of commercial stock beds. However, the studies of Murashige (5) and his co-workers have contributed to the rapid multiplication in vitro of many ornamental plants by the method known as plant tissue culture (PTC).

The PTC method for the rapid propagation of ornamental plants (other than orchids) was initiated commercially in California with Boston-type ferns. Today many commercial PTC laboratories are producing Boston-type ferns and others by the method developed (6). Ferns can be rapidly produced *in vitro* while eliminating the vast number of stock plants previously required in the production cycle. A disadvantage in this production method is the amount of manual labor required to make the transfers of sporophytic tissue in what are described by Murashige (5) as Stage II and Stage III. Included also is the labor and time involved in transplanting Stage III plants to soil.

The method to be described in this paper employs a partial PTC method that eliminates much of the manual labor required by the PTC method currently used by commercial operations.

**Materials and Methods**

All fern tissue cultures were started from spores collected from spore-producing stock plants growing at this research center or from plants growing in commercial nurseries. Whenever possible, recently matured spores were collected just prior to use. Prior to placement on the PTC medium, spores were disinfested employing a method similar to that described by Smith and Yee (5) for *Nephrolepis hirsutula* (Forst.) Presl. (hairy sword fern). The method employed here was as follows: Spores were placed in a previously autoclaved centrifuge tube which contained sterile deionized water (SDW) with Tween 20 (5000:1) added and the suspension was mixed on a Vortex (Scientific Products, McGaw Park, IL) test tube shaker. Tubes were centrifuged for 5 min and the supernatant discarded. A solution of 1 part commercial laundry bleach (sodium hypochlorite, 5.25% by wt) and 49 parts SDW with Tween 20 (5000:1) was then added to each tube, the fern spore pellet resuspended on the Vortex shaker, and the tubes centrifuged for an additional 5 min. The supernatant was discarded, the spore pellet resuspended in SDW and the tubes again centrifuged. The preceding resuspension-washing-centrifugation process was repeated 3 additional times. The resultant spore pellets were resuspended in 0.03 fl oz (1 ml) of SDW after the last centrifugation and the spore suspension streaked onto the PTC media surface.

The PTC medium employed for both spore germination and culture of the ferns tested consisted of: Packaged

Murashige and Skoog salts mix [GIBCO, Grand Island, NY]; sucrose, 1.0 oz/0.26 gal (30 g/liter); thiamine-HCl, 0.4 ppm (0.4 mg/liter) and Difco Bacto Agar, 0.8% (8 g/liter). An initial pH of 5.7-5.8 was achieved with the addition of either IN HCl or IN KOH and the medium then autoclaved for 15 min. The medium was placed in 0.92 x 6.0 inch (23 x 150 mm) test tubes and the cultures incubated under 16 hr daily illumination of 100 ft-c produced by Gro Lux fluorescent tubes in a culture room maintained at 80°F (27°C).

Attempts were made to germinate spores and grow cultures of the following fern species: Adiantum tenerum Swartz (maidenhair fern), A. tenerum Swartz 'Pink Lady' (pink lady maidenhair fern), Cyrtomium falcatum Presl. (holly fern), Davallia bullata Wall. (rabbit's foot fern), Polystichum adiantiforme (Foerst.) J. Sm. (leaf fern), Platycerium bifurcatum C. Chr. (staghorn fern), and Pteris ensiformis Burn. 'Victoriae' (Victoria table fern). Rapid gametophytic growth was the desired end product in the in vitro part of this system and ferns producing sporophytic growth were eliminated from further evaluations. All transfers of plant material were performed employing aseptic techniques within a laminar-flow transfer chamber.

Once gametophytic growth was established in vitro, experiments were carried out to determine if the gametophytic stage could be divided using a sterile Waring blender containing a 1/2 strength M&S salts solution and whether the resultant gametophytic tissue suspension (GTS) could be applied onto a soil surface and induced to continue growth with the eventual development of the sporophytic stage. New 4 inch (12 cm) plastic pots and steam pasteurized soil mixes were employed. A minimum of 5 replicate pots were used in individual treatments. In detailed studies with maidenhair fern, 0.03-0.5 oz (1-10 g) of gametophytic tissue was added to 6.8 fl oz (200 ml) of sterile 1/2 strength M&S salts solution to produce the GTS. The gametophytic tissue of Adiantum tenerum and Polystichum adiantiforme was comminuted in a Waring blender for 1, 3 or 5 sec. The quantity of GTS applied to the surface of each pot was 1.7 fl oz (50 ml). Pots were covered with a plastic bag after infestation and watered by sub-irrigation. The plastic bags were removed after approx 30 days and the pots placed under intermittent mist (15 sec every 30 min) from 6 a.m. to 8 p.m. to initiate development of sporophytes. Several soil types, similar to those tested by Henley and Poole (1) were evaluated as growing media. Developing fern cultures were fertilized overhead, when needed, initially with 1/2 strength M&S salts, although later fertilization was achieved with 20-20-20 (0.25 lb/100 gal).

Results and Discussion

All fern spores tested, with the exception of those from leaf fern and rabbit's foot ferns, were successfully disinfested, germinated and grown by the PTC procedures mentioned. Growth on the agar-containing PTC medium resulted in only gametophytic growth with all remaining ferns tested except holly fern which produced sporophytes. Detailed studies with maidenhair fern produced the system described in Fig. 1, a system which allowed production of in vitro part of this system and ferns producing sporophytic growth were eliminated from further evaluations. All transfers of plant material were performed employing aseptic techniques within a laminar-flow transfer chamber.

Once gametophytic growth was established in vitro, experiments were carried out to determine if the gametophytic stage could be divided using a sterile Waring blender containing a 1/2 strength M&S salts solution and whether the resultant gametophytic tissue suspension (GTS) could be applied onto a soil surface and induced to continue growth with the eventual development of the sporophytic stage. New 4 inch (12 cm) plastic pots and steam pasteurized soil mixes were employed. A minimum of 5 replicate pots were used in individual treatments. In detailed studies with maidenhair fern, 0.03-0.5 oz (1-10 g) of gametophytic tissue was added to 6.8 fl oz (200 ml) of sterile 1/2 strength M&S salts solution to produce the GTS. The gametophytic tissue of Adiantum tenerum and Polystichum adiantiforme was comminuted in a Waring blender for 1, 3 or 5 sec. The quantity of GTS applied to the surface of each pot was 1.7 fl oz (50 ml). Pots were covered with a plastic bag after infestation and watered by sub-irrigation. The plastic bags were removed after approx 30 days and the pots placed under intermittent mist (15 sec every 30 min) from 6 a.m. to 8 p.m. to initiate development of sporophytes. Several soil types, similar to those tested by Henley and Poole (1) were evaluated as growing media. Developing fern cultures were fertilized overhead, when needed, initially with 1/2 strength M&S salts, although later fertilization was achieved with 20-20-20 (0.25 lb/100 gal).

Results of the study on different soil media where German and native Florida peat were employed alone and in various combinations with coarse builder's sand indicate maximum growth was achieved with a steam pasteurized mix containing 2 parts Florida peat + 1 part coarse builder's sand amended with 7 lb dolomitic limestone/yd². Initial results of studies presently in progress with the described method indicate maidenhair fern once established in PTC has the capability of increasing its wt in gametophytic tissue 10 or more fold within a month. In our studies 0.03 oz (1 g) of gametophytic tissue in 6.8 fl oz (200 ml) of 1/2 strength M&S salts comminuted in a blender for 5 sec produced more sporophytes than could be counted when 1.7 fl oz of the GTS was poured over the surface of the soil in a 4 inch pot. The lowest concn of GTS that still will produce a salable product is as yet undetermined. Gametophytic tissue of maidenhair increased through 8 transfers in PTC has produced healthy, stable plants. However, the number of transfers that will continually result in a stable, non-mutating system is still under investigation.

The method as described is non-complicated and employs a simple PTC medium. Once the GTS is produced there should not be a major problem to adapt it through mechanization to a commercial system which would keep it gently agitated and which could dispense it into soil-containing cavity trays, flats, pots, etc. The soil mix employed in the propagative unit should be sterilized, be lightly packed, be moist when the GTS is applied and should be watered by sub-irrigation, possibly by capillary mats. A polyethylene-enclosed bench or similar structure would be commercially acceptable as the area for placement of units to which GTS is applied. Relative humidity within the enclosed structure should be kept at 100% by periodic misting or more preferably by a device emitting a light fog-type mist. Extreme care must be taken to avoid introduction of algae to the growing area through contaminated water, pots, soil, etc. as

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Fig. 1. Cycle for production of fern plants from spores by the partial tissue culture method.
algae will compete with the developing gametophytes (2) and can eventually overgrow and kill the fern tissue.

The system also has the added advantage of developing fern plants from gametophytic tissue derived from disinfested spores, an ideal method for production of ferns free of known pathogens.

**Literature Cited**


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**CITRUS BLACKFLY CONTROL BY FOLIAR TREATMENTS OF DOORYARD CITRUS**

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Additional index words. Aleurocanthus woglumi, insecticides, mist blowers.

**Abstract.** The citrus blackfly, *Aleurocanthus woglumi* Ashby, was found in Fort Lauderdale, Broward County, Florida on January 28, 1976. Acephate and chlorpyrifos applied with a mist blower provided 91-100% and 82-98% control of second and third instar larvae and 82-97% and 69-94% control of pupae, respectively. At least three foliar spray applications of malathion were required to obtain 98% larval control, but mist blower applications of malathion did not provide greater than 89% control of citrus blackfly larvae. Naled provided no more than 77% control while a mist blower treatment of Pyrelin® and an ultra-low-volume application of malathion provided no control of this pest.

The citrus blackfly, *Aleurocanthus woglumi* Ashby, is potentially one of the most economically important pests of citrus in Florida. It is native to South Asia and has spread to Africa, Central and South America, Mexico, the Azores Islands in the Atlantic, and to Texas and Florida in the United States (5, 8). In the Western Hemisphere, it was first reported in the West Indies in 1913 and spread throughout the Caribbean until it reached Key West, Florida in 1931, from whence it was eradicated by 1937 (3). The second report in the United States was in 1955 in South Texas (6) where it is now well established.

More recently it has been found in Fort Lauderdale, Broward County, Florida on January 28, 1976, and now infests portions of Broward, Dade, and Palm Beach Counties. Research was initiated at the Agricultural Research Center in Fort Lauderdale to develop a program of chemical control to either eradicate or manage this pest. Insecticides have been evaluated for citrus blackfly control in Mexico (4), India (2), and the Sultanate of Oman (7); however, evaluations have not been made under Florida conditions, nor with recently developed insecticides. Efforts are being directed to develop control strategies in various environmental situations, e.g., homeowner yards, wholesale and retail nurseries, commercial citrus groves, and non-crop areas. Since most of the present citrus blackfly infestation is confined to homeowner yards, considerable effort is being directed toward finding insecticides which have low mammalian toxicity and which are relatively safe to the environment, while still providing satisfactory control of the citrus blackfly.

**Materials and Methods**

Mature, 8-16 ft-tall citrus trees (either orange, *Citrus sinensis* L., or grapefruit, *C. paradisi* Macf.) in homeowner yards in Fort Lauderdale were selected for these tests. In the test area almost all citrus trees were either moderately or heavily infested with the citrus blackfly, i.e., at least 50% of the tree’s leaves were infested with larvae and pupae. Eight newly developed but infested leaves from near branch terminals were removed from each test tree, placed in plastic bags, and taken to the laboratory where the percentage of larvae and pupae alive initially in the population was determined under a dissecting microscope. Mortality readings were made by probing each 2nd and 3rd instar larva and pupa on each leaf. If body fluids were present, it was considered to have been alive. Only trees with 75-100% living insects were used.

In Test 1, malathion was evaluated by 3 treatment methods at the rates given in Table 1: (a) plants were sprayed to the point of drip off with a high pressure (200 p.s.i.) truck-mounted sprayer; (b) plants were treated with a portable electrical mist blower at a rate of ca one-fifth gal of solution per tree; and (c) plants were treated by ultra-low-volume (ULV) at a flow rate of 2 gal maximum per hr. The ULV treatment was applied with a truck-mounted unit from the

<table>
<thead>
<tr>
<th>Method of Application</th>
<th>Rate lb AI/100 gal</th>
<th>% control following initial treatment</th>
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<tbody>
<tr>
<td></td>
<td>1 wk</td>
<td>2 wk</td>
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<tr>
<td>Foliar spray</td>
<td>1.25</td>
<td>73a</td>
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<tr>
<td>Mist blower</td>
<td>1.25</td>
<td>45b</td>
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<tr>
<td>Mist blower</td>
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<td>33b</td>
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<tr>
<td>ULV</td>
<td>0</td>
<td>0c</td>
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<tr>
<td>Untreated check</td>
<td>0</td>
<td>0c</td>
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*Adjusted by Abbott's Formula to untreated checks. Means in a column not followed by the same letter are significantly different (P=0.05) (Duncan's multiple range test).

*Applied at 2 gal maximum flow rate/hr.*