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Fern Propagation Strategies at Casa Flora©

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INTRODUCTION

Casa Flora has traditionally specialized in production of tropical and hardy ferns over its 39-year history. That represents a long learning curve and a willingness to take risks. We have come a long way from those early days of producing tropical ferns from runner tips in beds of peat. We have gotten where we are today by introducing new taxa, insisting on quality, and pioneering new production methods. Fourteen years ago we bought a large tissue culture lab and two smaller ones in Florida producing tropical ferns. In 2 years we were producing 2 million plants. Two years later and with much difficulty, we started producing hardy ferns in the lab as well. Now we consistently ship over 106 fern taxa year round, many with their own protocols.

Ferns, going back 300 million years, are much more primitive than most of the plants commercially propagated, and so have unique opportunities and problems. Much of coal is made up of fern fronds, and many of the ferns today are represented in fossils. Unlike higher plants, ferns exhibit alternation of generations. Alternation of generation simply means that the cells that produce the gametes (egg and sperm) make up a whole independent plant instead of being contained in and wholly dependent on the parent. As you might suspect, being a gametophyte — being haploid — makes the plant look different than its diploid parent. In almost all ferns, this gametophyte starts out as a single-celled organism surrounded by a hard shell — a spore — that enables the cell to survive harsh environments until conditions are right to begin to grow. It then grows into a single cell layer, basically a heart shaped plant called a prothallus. When it is mature and environmental conditions are right, it releases sperm (pollen) that swims to and fertilizes an egg, which develops into a diploid (normal) plant — a sporophyte. When this plant matures and the conditions are right, it produces spores that start the cycle over.

Some ferns for one reason or another have developed other propagation methods as well. Some have branching stems that increase the number of growing points over time. Others have modified stems (stolons) that specialize in developing a growing point away from the parent. Still others form bulbils on the leaf veins or have buds on the leaf that grow when conditions are right. About the only vegetative propagation structure ferns don't have that higher plants do are well-developed axillary buds. Because of this you just can't take cuttings.

THESE DAYS CASA FLORA USES TWO PRIMARY METHODS OF PRODUCING FERNS — SPORE AND TISSUE CULTURE

Production from spore, in most cases, means you get sexual recombination of genes with the resultant phenotypic variation. Since this is the primary way most ferns reproduce, production is relatively straightforward and large numbers of plants can be propagated in a small space.

Spores are readily available from hobbyists if you want to produce small numbers of plants, but production on a commercial scale requires significant investment in stock maintenance. Most temperate ferns have spore that ripens once a year, therefore you must have sufficient stock plants to get enough spore for the following year. Spores ripen at different rates along leaf and care must be taken in harvesting the spore so you don't collect immature spore. The spore is mobile, so foreign spores can land on leaves and then be collected along with the desired spore — creating a mixture. Most spores looks alike, mixtures are only apparent when sporophytes get larger. I am sure any of you that have gotten spore-sown material have noticed a few odd ones.

Spore loses potency as it ages and must be stored correctly. Spore also seems to have distinct time windows in which it germinates and develops best. There are two windows of 4 or so weeks in the spring and fall when temperate spore seem to perform best. The weeks to transplantable sporophyte varies with taxon, time of year, and environmental conditions. Spore derived plants are variable just as seed derived plants are.

Ferns have developed many of the same strategies higher plants have for fertilization. There are those that share their genes with other individuals, those that only share with themselves, and those that don't share at all. The prothalli of a given population are specialized for the environment they usually develop in. In those species that germinate in areas where a favorable environment persists for months, trading of genes between individuals is common. As the environment gets less favorable, prothalli tend to become male and then female, fertilizing others by chance but fertilizing themselves just to make sure. Finally, there are species that are apogamous — no sexual recombination at all, the maternal mother cells never divide to form an egg, forming a plant instead — the progeny is identical to the parent — usually. This seems to be an adaptation to dry conditions where little free water is available for fertilization. While you might think that self-fertilizing prothalli would produce a homogeneous population, the high ploidy levels and what appears to be a propensity for faulty recombination of genes actually yields as much diversity as in those ferns that trade genes.

While some spore can remain viable for decades, in general fresher is better so we keep spore refrigerated in sealed containers so it doesn't desiccate. Green spore like *Osmunda* must be sown immediately. Many different substrates can be used for germination. An old Cub Scout project germinated spore on a brick. Black basalt dust is used in Germany. However, we use a good quality peat lite bark mix with an over layer of high quality peat. Pasteurization with boiling water or by microwave seems to be the best way to pretreat the substrate. Autoclaving will destroy the antibacterial and antifungal properties of the peat as well as foster the bloom of whatever fungal contaminates there are on the spore. We filter our spores with lens paper, upholstery cloth, or 200 mesh screens. This excludes most trash. Spores can be distributed by water or dilution with fine particles. Cover with translucent/transparent covers such as shrink wrap or zip lock bags to let light in, keep humidity in, and keep contaminants out. Germinate in a cool dry area with dim filtered light. Germination usually occurs in 2 to 6 weeks with sporophytes appearing in 3 to 6 months. Some can take 2 years. Free water is required for mating in non-apogamous taxa. If the prothalli density is too high the prothalli may stop developing at the male stage so only sperm will be produced, severely decreasing the yield

of the containers. Fungus gnats and snails can be a problem if they get into the containers. Fungus, bacterial, and various slime molds are always possible. Transplanting must be done at the correct developmental stage and care must be taken to wean the plants from their high humidity environment.

Casa Flora used to produce all of our Boston fern types from stolons. Many of these plants had sterile spore or were clonal in nature. Stolons (or runners) allowed production of true-to-type plants. Unlike spore production or tissue culture, the propagules per unit area were low resulting in single crown liners and quite a bit of our greenhouse committed to stock production — beds of peat moss, lines of pots — all to produce and convert enough runners to plants. We produced millions this way.

Stock plants were either grown in the substrate or above the substrate. Stolon tips were allowed to come in contact with the substrate, usually high-grade peat, where they stop elongating and develop leaves. Once a plant is initiated on the stolon, development is rapid. *Nephrolepis* taxa take 6 weeks to become large enough to transplant.

A few ferns can be propagated by bulbils, buds on the rachis, plants arising de novo on the leaf surface, or by buds present at the apex of the frond. Propagules are true to type, develop rapidly, and don't require special handling since they are survival structures. The numbers of propagules these plants produce tend to be small so a lot of stock space is required for commercial quantities. We only produce one selection ('Oriental Chain') this way. In this case, we have a small demand for this selection and the specimen plants we keep are sufficient. If demand were to increase, we would put it into tissue culture to ensure availability.

PRODUCTION BY DIVISION IN THE GREENHOUSE

This is only needed if there is a problem with production by spore. Low division rates and adverse response to growth regulators result in large space requirements for stock. To overcome this, we propagate by division in the tissue culture lab.

TISSUE CULTURE PROPAGATION

Tissue culture (TC) allows propagation of a large number of plants in a small area. Since you can start with a single specimen, selected characteristics can be captured. In some cases, TC allows dissemination of sterile hybrids, i.e., *Dryopteris x australis*, a cross between *D. celsa* and *D. ludoviciana*. When first tissue cultured, there were relatively few in the entire world. Within 2 years, 50,000 had been produced. Because of the small propagule size, multiple crown clumps are usually produced. Since propagation is done in a constant environment and growth rate controlled by hormones, year round production can be achieved.

On the other hand, TC requires special facilities and specialized knowledge. Because of the initiation costs and time spent building up stock, it is too expensive for small numbers of product. There is a lag time for buildup, conventional propagation will beat TC in the first 12 months — the 13th month TC will exceed the total output of the year of conventional propagation. There is no horticultural check for mixtures and mutations during propagation of ferns. With conventional propagation, you see the adult plant characteristics at the start of each propagation cycle. With TC, the production cycle is so long and has so many propagation cycles that large numbers of plants are produced without ever seeing an adult plant and so a single mixture or mutation can replace a good portion of the propagation material

before it is identified. Replacing a TC crop that is mixed or mutated is expensive in both discarded product and replacement time. The ability to initiate material into the laboratory varies with type and season. It is not always possible to initiate product when you need to.

Whether we are starting from spore or an organized structure, the plant material must first be disinfested of any organisms that will grow in the TC medium. We usually use chlorine-based aqueous solutions. Generally, if a taxon comes true from spore, it is easier to derive plants from these than from more complex structures although it usually takes a few more months before clonal product comes out of the lab. Interestingly, ferns do not have a highly organized apical region and so there are no periclinal chimeras. Nor do they have developed axillary buds so initiating from organized tissue is a hit or miss proposition.

There are two basic propagation strategies with ferns. You can either propagate gametophytic or sporophytic tissue. Both require buildup of stock and conversion of that stock to the greenhouse. Gametophytic tissue really means growing and multiplying prothalli. Since prothalli cells are totipotent, subdividing them increases their numbers. Once they are mature, they can be transplanted to the greenhouse and allowed to form sporophytes. While vast numbers can be produced this way, timing the crop in the greenhouse is impossible and therefore impractical for large-scale production. We have recently discovered that the haploid prothalli cultures can mutate over time and need to be restarted on a regular basis.

Production of sporophytes basically means suppressing leaf growth and forcing the meristematic structures to branch rapidly. This yields a large number of propagules in a very small volume and thus lending itself to automation. In our case, we convert whole plants to meristematic clusters, then manipulate those clusters in various ways and then convert them back into plants. Ensuring that tissue having no unique visual characteristics is what it is supposed to be can be challenging. We have chosen to replace the propagation material very frequently in order to flush out any mutations or mixtures. This requires a support program that consistently supplies known true-to-type material at regular intervals and a postproduction program that finishes product periodically to ensure that it is true to type.

Once the TC material is of sufficient size to survive in the greenhouse (Stage 3), it is divided and transferred to nurse trays. This is done in a high humidity room with frequent misting so material does not desiccate. From there it is moved to a humid nurse house to root before it is hardened off in a brighter, drier environment. When ready to transplant, they are brought to the transplanter. The transplanter plants 12 at a time into the 72-cell tray. The tray is then inspected for misses and uniformity and any corrections are made. The tray is initially watered in with a preventative mix of fungicide and larvacide and accumulated on carts. They are then placed on production benches. Our benches are a combination of watering mats and ebb and flood. They are all rolling benches. They are kept fairly dry to encourage rooting for the first few weeks and then fertilized with each watering. Since we water from the bottom and only enough to saturate the plug, the salt concentration in the fertilizer solution does not change nor do we get a buildup of fertilizer on the surface of the plug. Bottom watering also helps us prevent common diseases on the ferns by keeping the foliage dry.

One of the worst disease problems is black rot of the foliage caused by *Pythium*. While controllable with Captan, it is easier to prevent its occurrence by keeping

the foliage dry when watering and having good air movement at night. A high peat content in soil helps keep bacterial and fungal problems to a minimum.

Two pests we treat for regularly are fungus gnats and shore flies. While shore flies are more numerous and are the ones that hit your hand when you wave it over a flat, their larvae eat algae and live under the benches and other areas algae accumulates. Fungus gnat larvae do the actual plant damage but for us, the adults are not as numerous. We use a rotation of Adept and Enstar II as well as Nemashield — a beneficial nematode drench. We have seasonal problems with caterpillars on 72-cell liners. You can smell them before you see the damage. Treating with Dipel or 0.5X strength Orthene is satisfactory.

Benches are scouted daily for problems, to monitor treatments, and to correct imperfections in the trays. We find that it is much easier to make corrections to a flat when it is partially grown than just before shipping when small plants are hard to see and time pressures abound. Having done that, when we come on Thursday and Friday to pull flats for Monday shipment most of the corrective work has already been done. Regardless, we check the flats an additional time, water them, and then give them a preventative spray to make sure we don't have problems in the shipping box. Shipping actually started on Thursday with the pulling of flats and the making of boxes and shipping labels. Monday morning box filling starts.