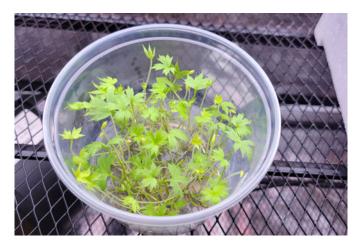
# **GROWERTALKS**

# Features

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# **Tissue Culture: The Next Frontier**

Bill Calkins



Producing from tissue culture (TC) is certainly cutting -edge and highly scientific, although it's not brand new to the world of plant propagation. In fact, it's more than a century old, going all the way back to the father of plant tissue culture—Gottlieb Haberlandt in 1902 when he theorized that artificial embryos could be extracted from vegetative plant cells. In the 1920s, scientists cultured small root tips from peas and maize, and in the 1950s, virus-free plants were recovered from infected plants via meristem culture. For decades, these plant cell

technologies have been developed and refined.

Pictured: Two mature leaves and an active shoot on all inputs is the ideal spec. Plantlets with very small or no roots require additional crop time.

Moving to current day and focused on floriculture, more and more crops are being offered from TC and greenhouse professionals are gravitating to this type of young plant input for many advantageous reasons. Some of these include a desire for disease-free plants, the reduced need for greenhouses of mother plants to produce on a commercial scale, allowing for the storage of viable plant genetics for many years, increased production speed and many more.

But with any "new" technology, there's bound to be a steep learning curve. TC plantlets are not like unrooted cuttings, plugs or liners—they're quite different and require new skills in the early stages of propagation. Once acclimated, most growers do find that subsequent processes are much the same as plugs or liners. Thankfully, there's significant research underway and new strategies for TC production being shared. Now is the time to begin learning ways to produce TC-raised crops efficiently and effectively, and refining techniques that will allow your operation to benefit from this new frontier.

The remainder of this article will draw heavily on a presentation given by Dr. Nathan Jahnke, Culture Research Manager at Ball Horticultural Company, and Deanna Felton, Propagation & Production Manager at Sunbelt Greenhouses in Georgia. Nathan and Deanna presented on the subject of TC at AmericanHort's Cultivate'23 event, bringing research findings and practical application together to help demystify TC and offer guidance for those of you interested in achieving maximum success with the young plant form. (To help with the content, please refer to "The

Language of Tissue Culture" sidebar.)

#### **Components of success**

Like all aspects of plant propagation, successful production begins even before young plants land on your loading dock. Choosing a reliable supplier or source is critical. This is the case with TC and the lab producing plantlets must be carefully considered. Both Nathan and Deanna felt having a close relationship with source labs, and providing clear expectations and feedback is paramount. (More on that later in this article.)

Crop selection is also a key piece of the puzzle, as some crops work well for your entry into TC, while others are more challenging and should probably be reserved for future years when you've gained experience. Crops Deanna described as "low difficulty" include ferns, Rex begonia, heuchera, gerbera and hosta. The "moderate difficulty" group includes crops like alocasia, calathea, dieffenbachia, echinacea and monstera.

For more experienced TC producers, aglaonema, Ficus elastica, Ficus lyriata and single-stem philodendron might be potential crops to consider. Of course, these lists aren't exhaustive by any means, so talk to your peers who've tried TC production and your trusted brokers to determine what makes the most sense for your operation.

Again, not unique to TC, but still vitally important to success, is preparation, inspection and grading once plantlets arrive. Deanna explained that before shipments arrive, you'll need to print out the packing list from the lab and schedule labor accordingly. Staging trays, tags and soil will help your team hit the ground running without costly delays.

You'll also want to make sure labor is scheduled based on arrival time because as soon as TC plantlets are unpacked, the proverbial clock is ticking. Nathan said this can be quite a challenge depending on the lab. There are many points where TC doesn't arrive on time—lack of communication on the export, delays with customs, secured flights and truck delivery. This is why the relationship is so important and where a trusted broker and distributor can make a significant difference. They have strong relationships, formal communication and possibly even pre-secured logistics.

Prepping your growing area is a best practice Nathan shared, and this includes cleaning and disinfecting all tools, surfaces and people. Dialing in your humidity levels based on the TC crops coming in is also critical, as plantlets need immediate hydration and acclimation.

TC plantlets must be inspected before propagation can begin and this is a process that needs to be in place every time a shipment arrives. Check all plants for signs of contamination. Unless there's breakdown or severe contamination covering the crop, this isn't usually an issue. A good rule of thumb would be to segregate those plantlets in different trays just in case. More importantly, examine all plantlets for breakdown, focusing on roots and growing points. Inspection should also include verifying counts (bags, flasks, plantlets) to ensure consistency. If you notice variability, talk to your supplier.

Because TC material can be variable in terms of size, having a grading protocol will help you and your team understand what you're dealing with and how to manage the crop in propagation. This is where good training comes into play. Some growers have found success hanging up photos of plantlets (showing a range of quality specs) near the sticking line to remind the team of what should and should not be planted. Sometimes, a plantlet is not of high enough quality to stick, but you'll only know that if attention is paid to grading. Grade plantlets at the time of stick and once they're rooted. If you're a young plant supplier, you'll also want to grade them prior to shipping to ensure uniform trays going out the door.

Before sticking, your team needs to ask some questions. Nathan shared some thoughts on the question, "What are you willing to stick?" Do your plantlets have roots or no roots? Are they out of spec in terms of size? Ideally, he said,

you want two mature leaves and an active shoot on all inputs.

In terms of roots, plantlets with very small or no roots will require an additional two to three weeks or more in crop time. Large roots could result in crops finishing one to two weeks ahead and requiring PGR treatments. At this point in the realm of TC, it's likely everything stuck on the same day will not be shippable on the same day due to the amount variability within flasks. This can greatly improve as the lab-grower relationship develops over time and with precise feedback.

#### **Acclimating TC plants**

Once you've worked through the steps leading up to sticking, your attention should turn to acclimating your TC plants. Remember, they're moving from a very protected environment in a flask to a greenhouse—a brand new and much harsher environment. Your number one goal is to limit dehydration. There's a lot of plant science behind exactly why TC plantlets are prone to dehydration and Nathan and his team have been working hard to create some best practices to avoid it.

Some common and traditional tactics can be employed to reduce dehydration, such as tenting propagation benches with Reemay or plastic and shading greenhouses. You might also put trays into a cooler or mist chamber to help with hydration. These structural methods are recommended, but might not be enough. Like mentioned, the greenhouse environment can be harsh—exhaust fans running, supplemental heat leading to dry conditions or natural environments and seasons can all impact your goal of limiting dehydration. Some additional solutions Nathan mentioned range from booms running over Reemay tents adding moisture, installing humidity reservoirs under benches and installing dry foggers to add moisture to the air.



Lighting is also an important factor in TC acclimation. Nathan and Deanna discussed the "myth" that TC needs low light conditions after sticking and agreed that's not necessarily true, and like many greenhouse processes, it really depends on various factors. The type of lighting used is a key factor and research shows that's due to radiant heat coming off of the fixture. HPS lighting creates a lot of heat (causing dehydration), while LED fixtures don't create nearly as much.

Pictured: Inspect tissue culture plantlets every time a shipment arrives. Train your team and have a process in place for this.

Lastly, successful acclimation requires water management—both in your substrate and in the air. Nathan explained substrate moisture should be in the level two to three range—not bone dry, but not wet enough to easily squeeze water from the tray cells. Too-dry substrate can lead to wilting within an hour, which you want to avoid. As important (or maybe more) is the air moisture in your greenhouse. Minimizing constant leaf wetness is

critical and irrigation with the smallest droplet size possible is a best practice to follow. To help stop young plants from dehydrating, your team needs to minimize leaf movement from fans and outside factors. Watch the plants closely, and if you see movement, find the cause and make changes.

### Providing feedback & final tips

As mentioned above, having a close relationship with TC labs and your young plant distributors is key to fostering a feedback loop that leads to the best inputs possible for your specific production scenario. Deanna has a lot of experience with TC crops and offered a word of advice: Feedback is a crucial component to future success. In her experience, keeping all parties abreast of positive or negative observations throughout the entire production process

is necessary. Report back on quality when you receive plantlets, status at the time of stick, if timing and growth is on track, and how crops look when it's time to ship.

Before wrapping up their Cultivate'23 presentation, Nathan and Deanna mentioned a few things to explore as you move into the world of TC propagation.

Consider utilizing a spreader or surfactant in propagation. Products such as Capsil can help water spread across a surface and reduce the threat of dehydration. But be careful and use low rates (e.g., 2 oz. per 100 gal.), as they can potentially melt tender, young plants.

Trial rooting hormones before using. Research has shown a benefit on slow-rooting crops like aglaonema, but not on others such as echinacea. Defoliation in ficus has been seen when using rooting hormones.

## The Language of Tissue Culture

**Acclimation/Hardening:** The process of adjusting plants to a different environment by controlling light, temperature, humidity, air movement and nutrition.

**Agar/Media:** Liquid, or more usually a semi-solid substance, in which plantlets are grown. It may contain nutrients, carbohydrates, plant hormones and other substances to steer plant growth.

**Deflasking:** The removal and sticking process of tissue culture plantlets.

**Ex Agar:** Without agar; plants can be shipped in containers that do not contain agar.

Flask/Vessel/Bag: Containers used to house and produce plantlets. These may also be used to ship plantlets.

In Vitro: The process of growing a plant in a container outside or apart from its living, original plant.

**Micropropagation:** The process or method of propagation for tissue culture with small plant parts, usually shoot tips in a lab setting.

**Plantlet:** A plant grown from tissue culture that has a growing tip, petioles and stem; it may also (but not always) have leaf blades, callus and roots.

Stage 3 Tissue Culture: The most commonly available input; another name for plantlets that usually have roots.

**Subculture:** The process of cutting a clump of shoots into smaller clumps or individual shoots during tissue culture production.

Vitrified: The water-soaked appearance of leaves is often seen when plants have been covered in agar.

**Water Roots:** Roots that have grown in agar. These roots may not take up water well, but can continue to grow and develop root hairs after deflasking. **GT** 

View Dr. Nathan Jahnke and Deanna Felton's full presentation as a multi-part video series from Tech On Demand. They dive far deeper into all of these topics and much more!