WSDA NURSERY TEA PROJECT REPORT, 2022

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Camellia sinensis is an evergreen plant whose leaves and leaf buds are used to produce tea. Tea plants generally flourish in tropical climates (e.g., India), but there exist cultivars (C. *sinensis* and C. *assamica* x C. *sinensis* hybrids) that are well suited for production in Mediterranean climates (e.g., western Washington). The U.S. is the fourth largest importer of tea in the world after France, Malaysia, and Pakistan, and the estimated total wholesale value of the U.S. tea industry as of 2021 was $13.47 billion (Statista, 2022). Although tea is the second most widely consumed beverage in the world (Tea Association of the USA, Inc., 2022), there is essentially no research to support its production and marketing in the U.S. Located in a favorable growing climate, Washington State University Mount Vernon Northwestern Washington Research and Extension Center is well positioned to take a leadership role in researching tea as a new crop for the Washington nursery industry. This project will develop methods for propagating tea from cuttings, establish a tea planting, and develop a tea propagation and planting guide, thereby supporting a sustainable emergence of tea production in Washington. This project addresses two of the nursery RFP priorities: 1) Establishment of new cultivar trials conducted within the state and relevant to the climate and hardiness zones of the Washington nursery industry, and 2) Nursery best practices.

Objectives

1. Test and develop methods for propagating tea from cuttings.
2. Establish a tea planting at WSU Mount Vernon NWREC to provide the nursery industry with performance information as well as cuttings for development of their own propagation blocks.
3. Develop a tea propagation and planting guide for the nursery industry to support sustainable sales of tea nursery stock.

Research site, methods and results

The WSU Mount Vernon NWREC field site is located in the maritime Pacific Northwest, with a Mediterranean where the summer climate is mild and humid with 15.0 °C average daily temperature, 80% relative humidity, and 170 mm rainfall (20-year average; AgWeatherNet, 2020).

Objective 1. Test and develop methods for propagating tea from cuttings. In general, clonal propagation (cuttings) of tea is recommended because of the high level of heterozygosity in seeds (Zee et al., 2003). In Year 3 of this project we built on lessons learned in Years 1 and 2. First, propagation is most successful when cuttings are from recently matured shoots (bark slightly red). Second, shoots collected from late August through September may be ideal. And third, cuttings are sensitive to environmental factors light, temperature and relative humidity. We carried out 3 experiments to test propagation methods for tea under environmental conditions of northwest Washington: 1) assess time of year for tea shoot collection; 2) evaluate processing time following tea shoot collection to simulate shipping conditions; and 3) test two propagation media for tea shoot propagation.

We collected shoots of tea cultivar '1-2' from 21-year old plants from John Vendeland from his home tea garden planting in Burlington, WA. This cultivar is originally from Ilahee Hills Tea Farm in Salem, OR, which was discontinued at the end of 2020 and the breeding block is now part of Minto Island Growers in Salem, OR. Shoots collected from this cultivar were used for these three propagation experiments at WSU NWREC. The following methods were used for all three experiments except where differences are noted.

Shoot collection and cutting methods. Cuttings for all experiments were healthy shoots that had an initiated axillary bud but before initiation of flowering from the axillary bud (Fig. 1). The cut shoots were wrapped with moist paper towels, placed inside a plastic bag and brought to the WSU Mount Vernon NWREC greenhouse.

The shoot cuttings were washed to remove any potential contamination then processed for propagation. We used three-node cuttings, cut 1 cm below the node, and removed the leaf attached to the third (bottom) node keeping the first two leaves intact. Cuttings were dipped into rooting hormone (Dip'n Grow, Clackamas, OR) for 3-4 mins and placed in treepots (MT38BT, Stuewe and Sons, Tree Seedling Nursery Containers, Tangent, Oregon) filled with propagation media (peatmoss, vermiculite and perlite in 5:3:2 ratio) and watered well.

Treepots were placed in a clear plastic box, misted and closed to maintain relative humidity at 90–100%, and placed in a grow room with about 50% light, and 72-77 ^{o}C temperature (Fig. 2). In the propagation area, temperature and relative humidity data were recorded every 15 min using a data logger (Hobo Onset, Bourne, MA). The cuttings were monitored every week and watered every 2 weeks if the potting mix in the treepot was dry. The cuttings were misted if the relative humidity inside the propagation chamber fell below 70%.

Experimental design. The design for all three experiments was a completely randomized design with 6 replications. The number of plants per experimental unit was 6 for Expts. 1 and 2, 5 for Expt. 3, and 10 for alternative Expt. 1 (explained below).

Data collection. Data were collected for survival, leaf drop and shoot growth of the cuttings. Survival was measured as the total number of cuttings per experimental unit with either old, mature leaves intact or with new sprouts. Leaf drop was measured as the total number of cuttings per experimental unit that dropped both old, mature leaves. Shoot growth was measured as the total number of cuttings per experimental unit that showed new sprout growth.

Data were collected once a month for all three experiments: 1) for experiment 1, on the 5th of each month, from October 2021 to April 2022; 2) for experiment 2, on the 16th of each month, from November 2021 to June 2022; 3) for experiment 3, on the 5th of each month, from October 2021 to June 2022.

Experiment 1. To evaluate the time of year to collect tea shoot cuttings for successful propagation and based on successful propagation dates in our 2020 study, we collected tea cuttings at 2-week intervals, on 1 Sept., 14 Sept. and 1 Oct. 2021. The cuttings were processed and placed in boxes in the grow room (Fig. 2) as described above. On 15 Nov. the cuttings were moved from the grow room to a greenhouse at WSU NWREC for ease of care and maintenance and to accommodate the increased number of cuttings. The cuttings remained in the plastic boxes but the lids were removed, and the boxes were placed directly on the greenhouse bench with lights (Sun Blaze T5HO-48LED, Sunlight Supply, Vancouver, WA).

Additionally, based on preliminary results and feedback from John Vendeland (owner of the tea plants where we collected shoots), we collected shoots on 5 Nov. and 3 Dec. 2021 and 12 Jan. and 18 Feb. 2022 as alternative Experiment 1. Adding additional dates provided information regarding how successful tea propagation can be with different stages of stem/wood development. However, the new cuttings from November onward were placed in a fir mulch propagation media from Experiment 3 because of the higher survival success seen in the fir mulch compared to the standard potting media. The cuttings were processed and placed inside the mist chamber 1 with an automatic misting system (Drum-based misting system, MistAway Systems, Houston, TX) set up @ 30 s every hour (Fig. 3). The processed cuttings from 5 Nov. 2021, 3 Dec. 2021, and 12 Jan. 2022 were added to mist chamber 1. An additional mist chamber (mist chamber 2) was constructed on 18 Feb. with a different misting system (model #549, Mist Timer II, Drips Inc., Concho, AZ) set up @ 10 s every hour for 2-5 pm (Fig. 4) and cuttings from 5 Nov. and 3 Dec. 2021 were moved from mist chamber 1 to mist chamber 2 on 18 Feb. 2022 to provide a transition to greenhouse conditions. Cuttings from 18 Feb. 2022 were processed and placed in mist chamber 1 along with cuttings from 12 Jan. 2022 on 18 Feb 2022. In September 2022, rooted cuttings from our 2020-21 experiment will be transplanted into the field, and in May 2023, rooted cuttings from our 2021-2022 experiment will be transplanted into the field (see Obj. 2 below).

Results. While average temperature was similar for the grow room and the mist chambers (66–70 ^{o}F), the difference between minimum and maximum temperature in the grow room was 16.3 ^{o}F while the difference was 32.2 ^{o}F in mist chamber 1 and 37.6 ^{o}F in mist chamber 2 (Table 1). The grow room had the greatest average relative humidity (97%) followed by mist chamber 1 (90%), while the average relative humidity in mist chamber 2 was relatively low (61%) (Table 1). All cuttings collected on 1 and 15 Sept. and 1 Oct. did not survive (Fig. 5). Cuttings collected on 1 Sept. immediately started to decline while cuttings collected on 15 Sept. and 1 Oct. had a sharp decline after the first month. However, this may have been due to the move from the growroom to the greenhouse bench without a transition phase in a misting chamber. As of 8 July 2023, cuttings collected on 5 Nov. and 1 Dec. 2021 have approximately 97% survival and cuttings collected on 12 Jan. and 18 Feb. 2022 have approximately 80% survival (Fig. 6). For all cutting dates, new shoots emerged before the leaves that were initially present on the cuttings started to drop.

Changes to the experiment. We collected new shoots in September 2021 rather than August because our 2020 data showed that shoots collected in August had a low rooting rate. We did not request cuttings from NPGS in Hawaii in August 2021 because our propagation methods are not yet confirmed, and information is needed from Experiment 2 to provide shipping instructions to the curators (i.e., should ice packs be included with the shipped shoots). We did not transfer rooted cuttings to the hardening tunnel in May 2022 as this year has been unseasonably wet and cold and we were unable to complete our field preparations on time.

Experiment 2. The failure of tea shoots shipped from NPGS in Hawaii to root, indicated that we needed to explore shipping conditions if nurseries are going to be able to utilize this germplasm collection. A first step was to determine if shoots need to be kept cool during shipping, and so we tested 3 treatments: 1) processed cuttings immediately after collection (control): 2) stored cuttings in a cardboard box for 3 days at ambient temperature; 3) stored cuttings in a Styrofoam cooler for 3 days with three icepacks (Nordic Cold Chain Solutions, Hatfield, PA). Shoots were collected on 5 Nov. for all treatments as described above. Temperature and relative humidity were recorded inside the Styrofoam cooler for treatment 3 every 15 min. using a data logger (Hobo Onset, Bourne, MA). For treatments 2 and 3, the cardboard box and Styrofoam cooler were closed tightly and placed in our laboratory where temperature was maintained at 70-74 ◦F. For rooting, cuttings were processed as described above except that the media in the treepots was fresh ground fir mulch and perlite (see Expt. 3 below). This media was chosen because of the higher survival success seen in Experiment 3 with the fir mulch and perlite compared to the standard potting media. Cuttings from all three treatments were placed in boxes and set in the growroom as described above until 15 Nov. when the cuttings were transferred to the WSU Mount Vernon NWREC greenhouse bench and placed under lights (Sun Blaze T5HO-48LED, Sunlight Supply, Vancouver, WA) with the box lids removed. Transition through misting chambers was not carried out for this experiment.

Results. The temperature at ambient room temperature averaged 70.3°F and average relative humidity was 38% all three days, while in the box with icepacks, the average temperature was approximately 50 ^{o}F at the beginning of the experiment and increased 5 ^{o}F over the 3 days (Table 2 and Fig. 7). The relative humidity in the box with icepacks was about 55% throughout the experiment (Fig. 7). After shoots were propagated, survival after 7 months was similar for shoots that had been stored in a box for 3 days with and without cold packs (approximately 60%) (Fig. 8). In contrast, the shoots that were propagated immediately after collection had a 40% survival. For all three treatments, leaf drop started within 1 month of collection and new shoot growth started 2 months after collection. However, results were likely impacted by the lack of transition from the growroom to greenhouse conditions, and in our 2022-23 experiments cuttings will be rooted in the mist chamber 1, acclimated in mist chamber 2, and then placed on the greenhouse bench.

Experiment 3. We compared the potting medium that is standardly used at NWREC and was successful for us in our 2020-21 study, with a potting medium recommended for Camellia propagation (Arthur, 2019). The 2 treatments in this experiment were: 1) standard medium of peatmoss, vermiculite and perlite in 5:3:2 ratio; and 2) fresh ground fir mulch and perlite in 6:4 ratio. A factor to consider for tea propagation is the pH of the potting medium, which should be between 5.0 and 4.0 (Wilson and Clifford, 1992). We measured the pH of the media before adding the cuttings using the slurry method (Gavlak et al., 1994). Shoots were collected on 15 Sept. and processed and placed in boxes in the growroom as described above. The plastic boxes were transferred to the NWREC greenhouse bench on 15 Nov. and the lids were removed to acclimate the shoots to more light. Transition through misting chambers was not carried out for this experiment.

Results. The pH of the standard media was 4.66 and the pH of the fir mulch media was 4.12 on average (range 3.97 to 4.27). The pH for the fir mulch media was measured first using the standard slurry method and then a second time by soaking the media in deionized water as it was difficult to use the slurry method because the large size of particles. After 8 months, the overall survival of cuttings was approximately 8% for the standard media and 48% for the fir mulch media (Fig. 8). Leaf drop began for both treatments within the first month and shoot growth did not start for the fir mulch media treatment until about 4 months after the beginning of the experiment, while shoot growth started after about 6 months for the standard media treatment. However, results were likely impacted by the lack of transition from the growroom to greenhouse conditions, and in our 2022-23 experiments cuttings will be rooted in the mist chamber 1, acclimated in mist chamber 2, and then placed on the greenhouse bench.

Objective 2. Establish a tea planting at WSU Mount Vernon NWREC with domestically sourced planting stock. In June 2019, we procured 500 seedlings from a commercial tea farm in Alabama (Fairhope Tea Plantation) and established half in the greenhouse and half in the field in October 2019. In June 2020, we transplanted the remaining seedlings from the greenhouse into the field. In July 2020, November 2021, and April 2022, we assessed plant survival and plant height of tea plants in the field while overall plant health was measured in July 2020 and April 2022. Plant survival was measured as the total number of alive plants indicated by presence of healthy leaves or a sign of new growth. Plant height was measured from the base to the tip of the plant. Number of leaves were counted as the total number of leaves present in a plant. Number of branches were counted that were arising from the main stem. Overall health was recorded on a 5-point scale where 1 = very poor, 2 = poor, 3 = average, 4 = good, and 5 = very good.

Results. Plant survival decreased from 57% in July 2020 to 38% in April 2022 for October-planted tea plants, and declined from 96% to 69% for June-planted tea plants over this same period of time (Fig. 10). Increased plant survival from Nov 2021 to April 2022 was due to plants dropping leaves in the Fall and resprouting in the Spring. In July 2020, both October and June-planted tea plants had similar plant height (17.5 cm) while in April 2022, June-planted tea plants had greater height (23.8 cm) than October-planted tea plants (21 cm). In July 2020, June-planted tea plants had good health and October-planted tea plants had poor health, while in April 2022, both October and June-planted tea plants had very poor health.

Changes to the experiment. We had planned to transplant rooted cuttings to the field in May 2021 but did not do so as the seedlings that were transplanted in the two previous years were not thriving in the field. We continued to monitor plant growth in the field and on 4 Feb. 2022 we measured the field soil pH and found that it was 6.2 whereas 5.0–5.6 is ideal. In June 2022 we established a new site in a former blueberry field where the pH is 5.2, and we will move the tea plants on 20 July and will transplant the rooted cuttings in September, when fall rains start.

We had planned to request tea seedlings from NPGS in Hawaii in July 2021 but did not do so as our propagation methods had uncertain survival and it in unclear what conditions are needed for successful shipping.

Our goal was to establish a tea planting at WSU Mount Vernon NWREC with domestically sourced planting stock. We hoped that the heterozygosity of the tea seedlings from Alabama might result in a few plants with winter hardiness in our climate. We now aim to accomplish the field planting with plant material that we have propagated from John Vendeland. When plants become established in the field, likely after 4 years, cuttings will be available to the nursey industry to facilitate the availability of tea in Washington.

Objective 3. Incorporate new information we gain from studies into tea guide. Our studies on propagating, establishing and maintaining tea cuttings and seedlings will be recorded and used to develop a guide for growing tea in Washington, targeted to the nursery industry as well as their customers. This guide will support the expansion of sales of tea nursery stock in Washington. When complete, the guide will provide instructions on Propagation, Site Selection and Preparation, Planting, Irrigation and Nutrition, Pest and Disease Management, and Pruning and Training. We will post information on our website https://vegetables.wsu.edu along with PowerPoint presentations and posters to disseminate project outputs.

Currently, we have focused our writing on propagation based on our experimental results. However, our results indicate more research is needed to better understand optimal timing for collecting cuttings in Mount Vernon and optimal conditions for rooting the cuttings under our greenhouse conditions. Additionally, more research is needed regarding size of cutting (1-node vs 3-node) and potting media (peat moss vs bark mulch). Our 2022-23 experiments will provide greater clarity regarding these points. For field establishment and maintenance, we have provided information based on publications in other regions and experience in our region regarding pests of significance. Sections that will be added this year include When there is adequate experience in our region, we will update this information accordingly. Sections on Irrigation and Nutrition, Pest and Disease Management, and Pruning and Training. Additionally, in the upcoming year we will post our preliminary findings to our website along with resource information from other regions.

References

Arthur, B. 2019. Propagation for the home gardener. Soil3. https://blog.soil3.com/propagation-for-the-home-gardener

Gavlak, R.G., D.A. Horneck, and R.O. Miller. 1994. Soil, plant, and water reference methods for the Western Region. WERP 125: 37-42.

Statista. 2022. Tea market in the US. https://www.statista.com/study/15224/tea-market-in-the-us-statista-dossier/

Tea Association of the USA, Inc. 2022. Tea fact sheet – 2022. https://www.teausa.com/teausa/images/Tea_Fact_2021.pdf

Zee, F., D. Sato, L. Keith, P. Follett, and R.T. Hamasaki. 2003. Small-scale tea growing and processing in Hawaii. New Plants for Hawaii. University of Hawaii-Manoa Cooperative Extension Service NPH-9.

Zee, F. 2005. How to root tea cuttings. USDA/ARS, Tropical Plant Genetic Resources Management Unit, Pacific Basin Agricultural Research Center, Hilo, HI.

Table 1. Environmental conditions in propagation areas for Objective 1.

Location	**Min. Temp. °F**	**Max. Temp. °F**	**Avg. Temp. °F**
Grow room Inside Boxes (Sept-Oct)	64.5	80.8	70.3
Grow room Outside Boxes (Sept-Oct)	49.4	81.6	67.1
Mist Chamber 1 (Nov-June)	55.5	87.7	68.1
Mist Chamber 2 (Feb-June)	54.5	92.1	66.1
Greenhouse Bench (Jan-June)	55.3	104.8	69.5

Location	**Min Rel. Hum. %**	**Max Rel. Hum. %**	**Avg. RH %**
Grow room Inside Boxes (Sept-Oct)	47.2	100	97.6
Grow room Outside Boxes (Sept-Oct)	33.6	86.7	60.4
Mist Chamber 1 (Nov-June)	11.3	100	90.0
Mist Chamber 2 (Feb-June)	18.6	96.5	61.3
Greenhouse Bench (Jan-June)	16.0	100	66.2

Table 2. Temperature in the cooler box to simulate shipping conditions.

Avg Temp °F		Min °F	Max °F
Ambient	Cold Pack	Cold Pack	Cold Pack
70.3	48.8	47.8	68.8
70.3	50.3	49.2	51.8
70.3	53.7	51.8	57.0

Figure 1. Cuttings collected in Mount Vernon for propagation at WSU Mount Vernon NWREC.

Figure 2. Tea cuttings inside plastic boxes in the greenroom at WSU Mount Vernon NWREC.

Figure 3. First mist chamber in WSU Mount Vernon NWREC greenhouse.

Figure 4. Second mist chamber in WSU Mount Vernon NWREC greenhouse.

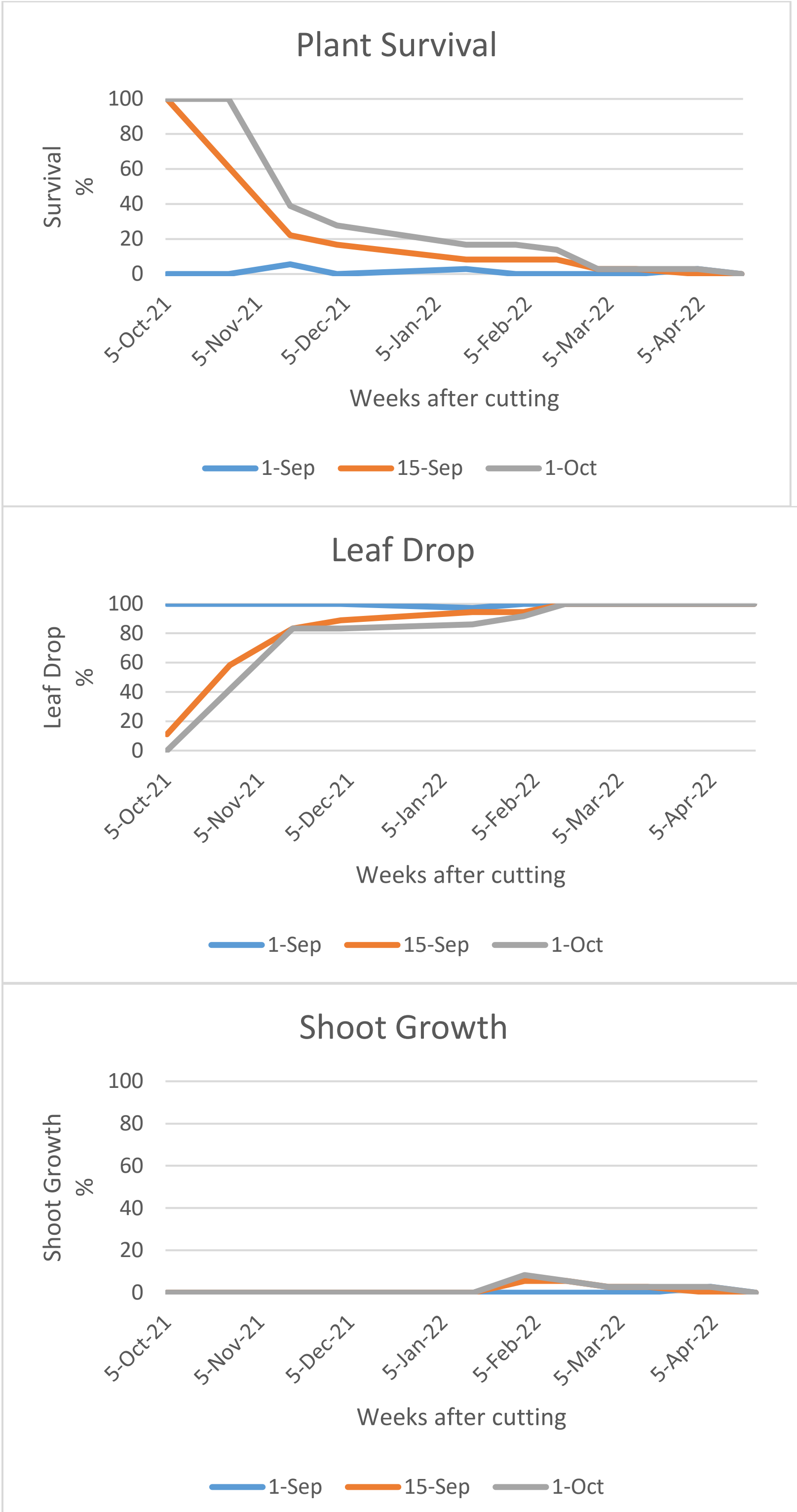

Figure 5. Experiment 1 plant survival, leaf drop and shoot growth measured every month from October 2021 until April 2022 for cuttings collected on 1 and 15 Sept. and 1 Oct. 2021 and placed in a growroom at WSU Mount Vernon NWREC.

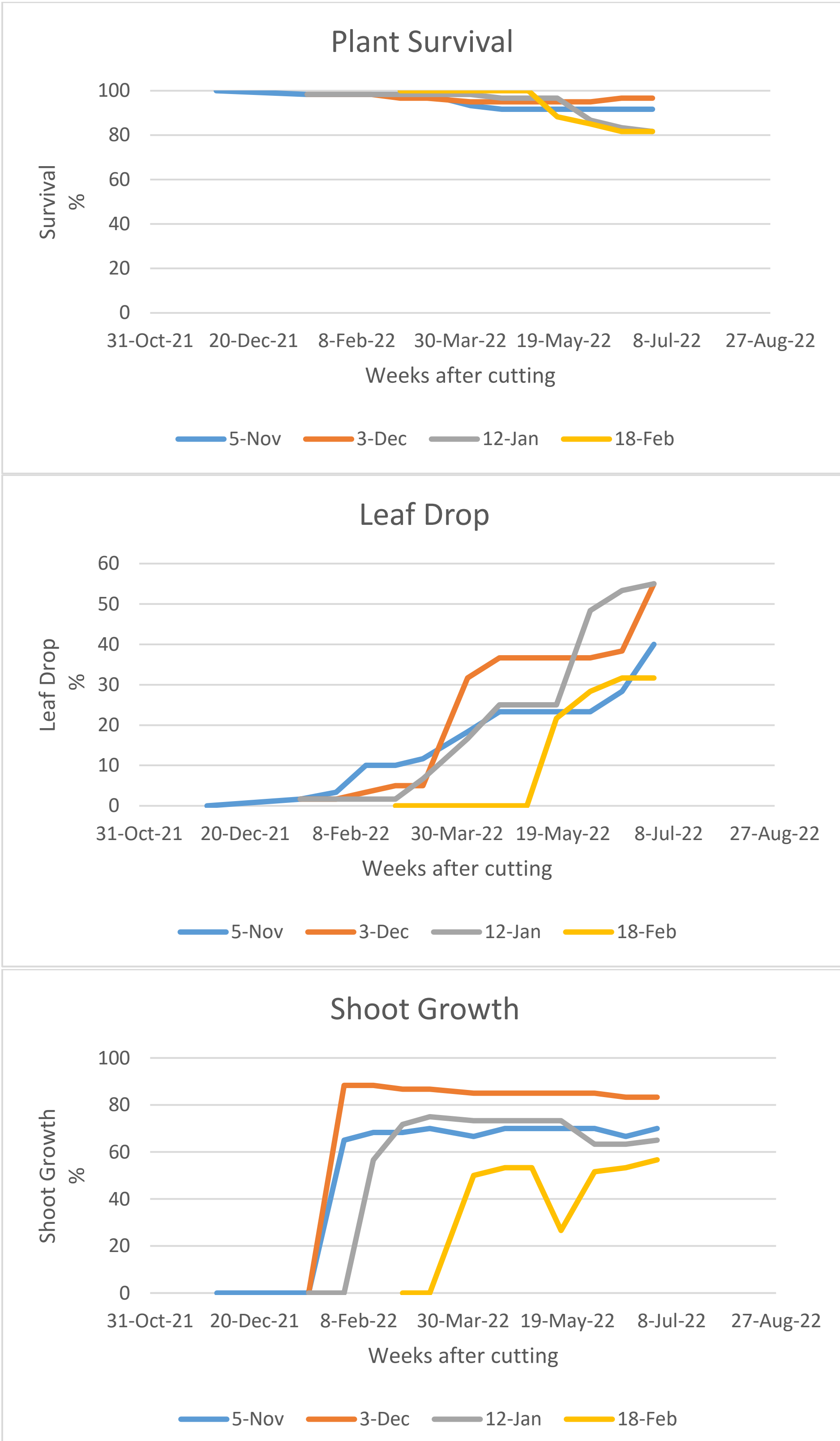

Figure 6. Experiment 1 plant survival, leaf drop and shoot growth measured every month from October 2021 until July 2022 for cuttings collected on 5 Nov. and 3. Dec. 2021 and 12 Jan. and 18 Feb. 2022 and placed in a misting chamber at WSU Mount Vernon NWREC.

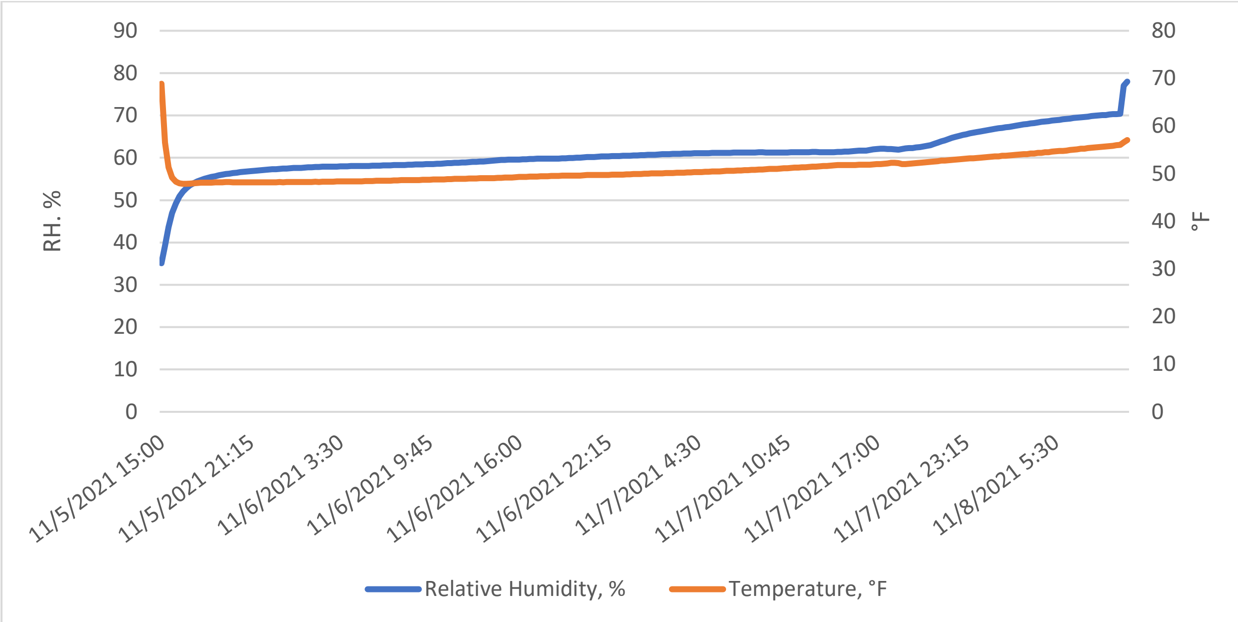

Figure 7. Experiment 2 relative humidity and temperature measured for 3 days inside the Styrofoam cooler where cuttings are stored with cold packs at WSU Mount Vernon NWREC.

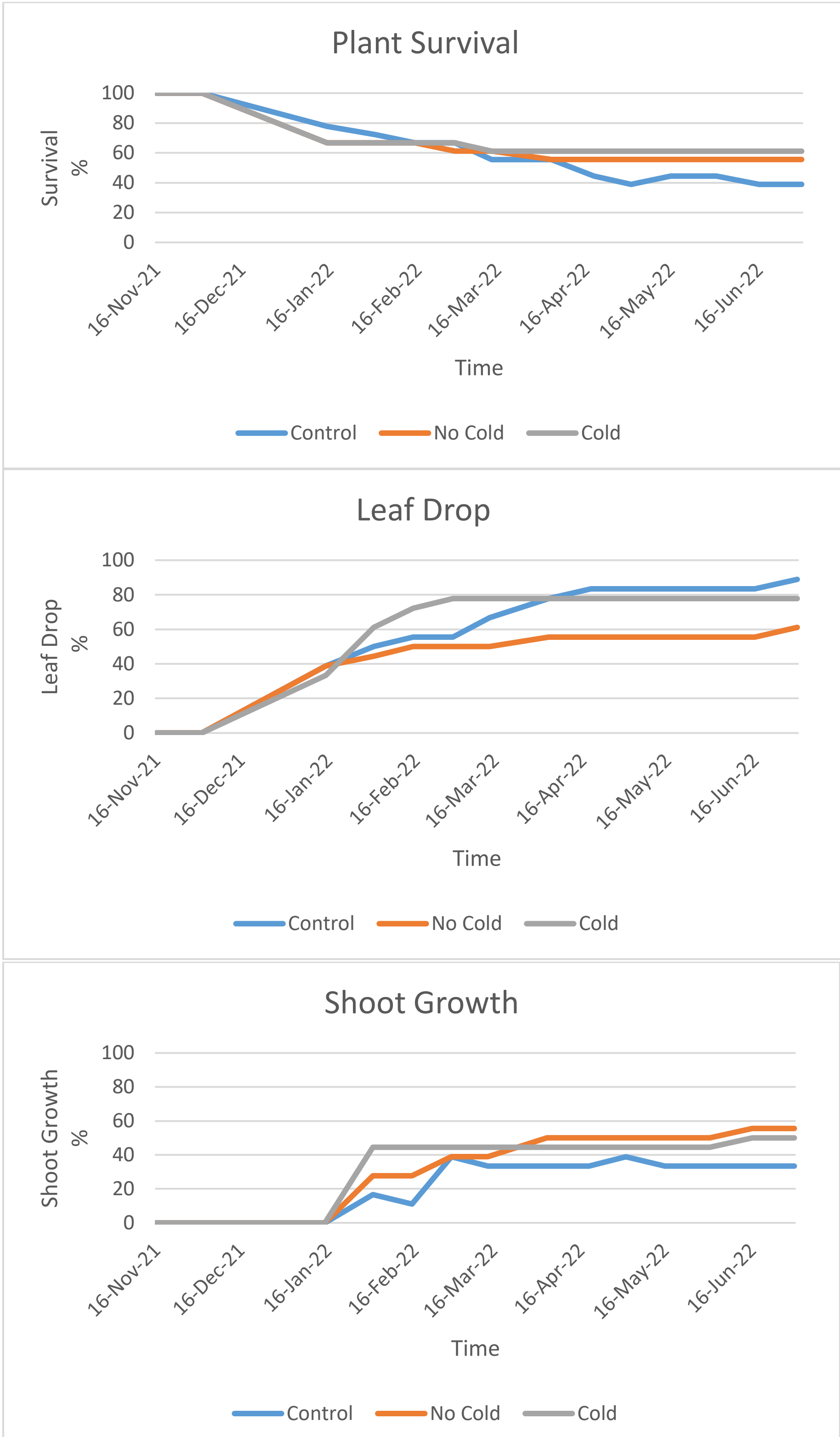

Figure 8. Experiment 2 plant survival, leaf drop and shoot growth measured every month from November 2021 until June 2022 for cuttings immediately processed after collection (control), first stored in a box with and without cold packs for 3 days prior to propagation at WSU Mount Vernon NWREC.

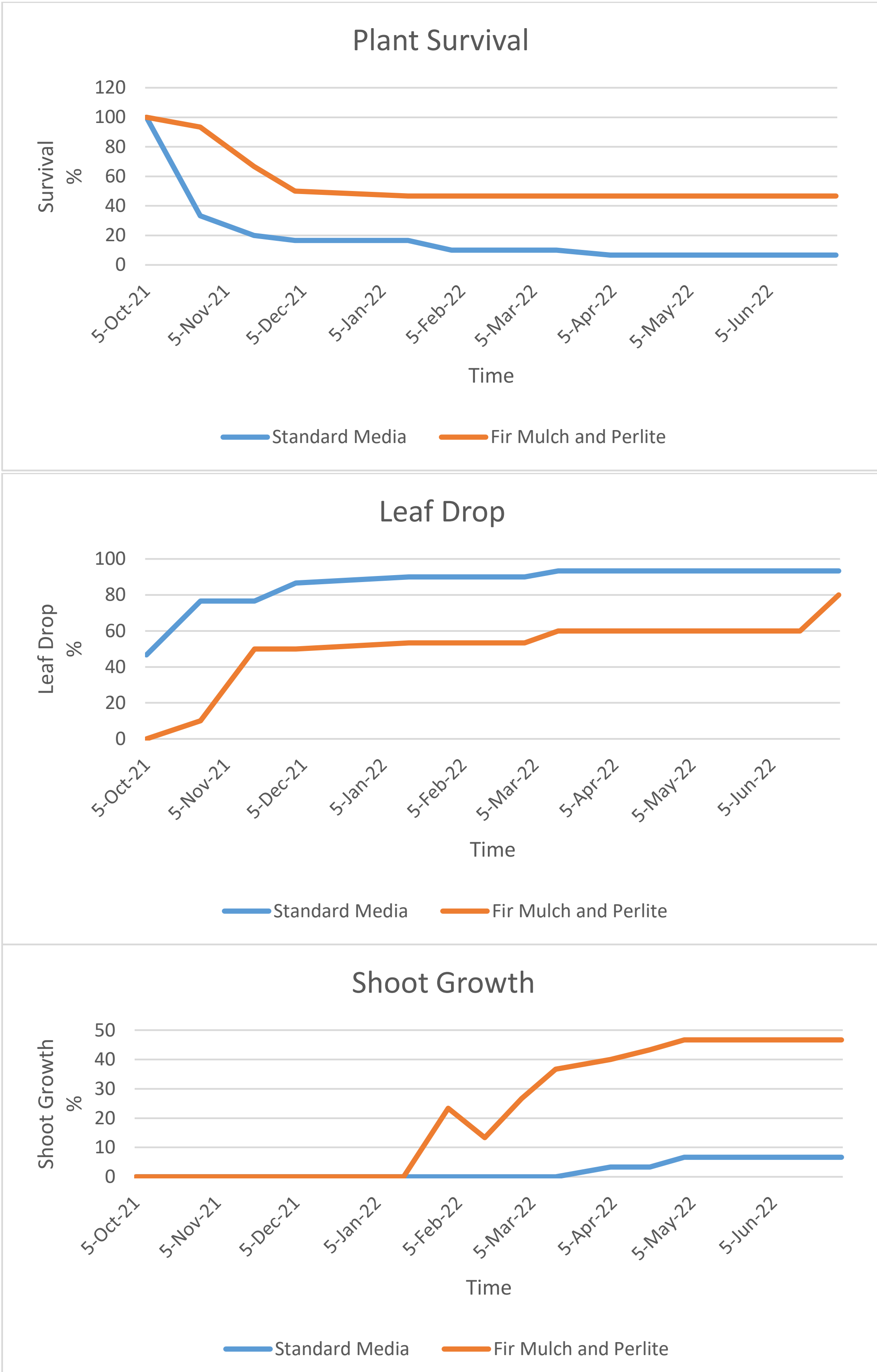

Figure 9. Experiment 3 plant survival, leaf drop and shoot growth measured every month from October 2021 until June 2022 for cuttings collected on 15 Sept. and propagated using a standard or firbark-based media at WSU Mount Vernon NWREC.

Figure 10. Plant survival and plant height measured in July 2020, Nov. 2021 and Apr. 2022, and plant health rating measured in July 2020 and Apr. 2022 for tea cuttings collected from Alabama and transplanted to the field in October 2019 and June 2020 at WSU Mount Vernon NWREC.