

APPENDIX 2: FOLIAGE SAMPLING PROTOCOL

Please observe the following guidelines to ensure proper sample collection. Collect foliage from the measurement plots indicated in the document “foliage package list”. Foliage collections will be made from December 1 to February 15. Foliage samples will be collected from five dominant trees in each measurement plot unless specific trees are indicated in the foliage package list. To ensure that the same trees are sampled for foliage at each measurement period, sample trees must be clearly identified with paint or flagging and a list of foliage sample tree numbers for each plot must be maintained and provided to FPC.

Collect pine needles from the uppermost laterals which contain a full year's complement of foliage. Select needles from the first flush produced during the just completed growing season on a primary lateral branch in the upper 1/3 of the live crown. Remove intact fascicles (generally consisting of 2-3 individual needles), including the bundle sheath, from sampled branches. Collect whole, healthy (i.e., free of insect and disease damage) needles that are typical of the crown (i.e., include 4-, 5-, and 6-needle fascicles if representative).

Each sample is a composite of exactly 100 fascicles collected from the five sample trees in each measurement plot (20 fascicles from each tree).

Clean foliage samples of contaminants (e.g., soils, twigs, etc) after collection; however, do not wash samples. Place the composite sample of fascicles from each measurement plot in separate lunch-bag-size paper sacks clearly labeled with Sample number associated with the FPC study and plot number where the sample was collected. Please align all fascicles in the same direction (e.g., place bundle sheaths either all up or all down). Keep needles as straight as possible during collection and shipping. Large paper clips may be used to hold the bags closed; do not use staples or rubber bands. Maintain foliage samples as close to 4°C as possible until drying is begun (i.e., keep in an ice chest during collection and transportation). Dry foliage in a forced-air oven at 65-70°C (149-158°F) for a period of 24-48 hours. Oven temperatures must not exceed 70°C to prevent gaseous losses of nitrogen. Discuss alternative drying methods with FPC staff. Once thoroughly dried, store samples in a cool, dry place until sent to the lab. If drying facilities are not available, green and respiring tissues may be sent in sealed, plastic bags, labeled as described above. Green tissue (foliage that has not been dried) must be shipped to the lab overnight. Samples should be sent to Waters Agricultural Lab (address is below). **Include a copy of the completed Foliage Packing List and Foliage Packing Label inside the shipping box.** Send a copy of the completed Foliage Packing List along with the shipping company name and package tracking number to Tim Albaugh (tim_albaugh@vt.edu).

Send foliage samples to:

Waters Agricultural Lab

POB 382

257 Newton Highway

Camilla, GA 31730-0382

Contact Tim Albaugh (919-451-7328, tim_albaugh@vt.edu) with any questions or concerns.