

Collection of xylem sap

A, Root pressure method

a, Climbing plants (squash, cucumber etc.) or pot-grown plants (tomato etc.)

- 1, Water well and drain excess water.
- 2, Cut stem 10 – 30 cm above soil.
- 3, Remove all leaves from stump.
- 4, Rinse well the cut surface of stump with DW.
- 5, Lead the stem into the bottle or tilt the stump onto the bottle which is kept cool with ice block etc. (Fig. 1)
- 6, Collect drops from stem for several hours – 1 day.

*Contents of sap may change depending on the timing or condition of collection (day or night, time after cut of stem).

Fig. 1



- (Ref) • A. Oda et al, Possible involvement of leaf gibberellins in the clock-controlled expression of XSP30, a gene encoding a xylem sap lectin, in cucumber roots. *Plant Physiol.* 133: 1779-1790 (2003)
- S. Satoh Organic substances in xylem sap delivered to aboveground organs by the roots. *J. Plant Research* 119: 179-187 (2006)

b, Deciduous woody plants (sugar maple, birch, grape etc.)

- 1, Cut the branch (grape vine) or make small hole in the trunk and insert tube into the hole in early spring just before bud burst (timing is critical).
- 2, Collect drops into cooled bottle.

*Available only in early spring with specific plant species. The sap usually contains glucose and fructose (sugar maple sap contains sucrose).

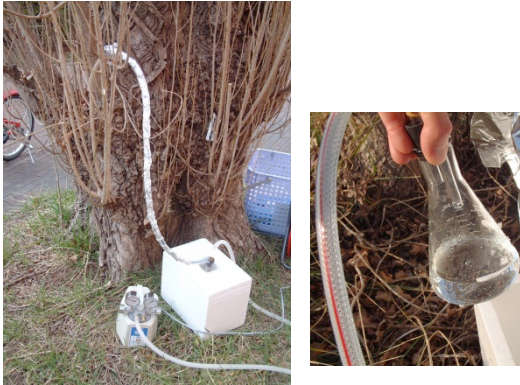
c, Field grown grass plants (rice, broccoli, tomato etc.)

- 1, Cut stem 5 – 10 cm above soil.
- 2, Remove all leaves from stump.
- 3, Rinse well the cut surface of stump with DW.
- 4, Attach sanitary cotton ball onto the cut surface of stump, wrap the cotton ball and top of stump with plastic film, and band it with a rubber band.
- 5, Collect the cotton ball after several hours – 1 day.
- 6, Put the cotton ball into centrifuge tube in which smaller tube or pipet tip is placed under the cotton ball to keep space from the bottom, and centrifuge at 2,000 rpm for 10 min and collect the sap from the bottom of tube.

B, Suction method (woody plants)

- 1, Cut side or main branch.
- 2, Rinse the cut branch surface with DW and connect it via a plastic or silicon tube to a sample collection container cooled with ice (Fig. 2). The container was maintained at - 0.08 MPa with a vacuum pump (Compact air pump NUP-2; AS ONE Co.) for several hours – 1 day.

Fig. 2



(Ref) J. Furukawa et al, Seasonal fluctuation of organic and inorganic components in xylem sap of *Populus nigra*. Plant Root 5: 56-62 (2011)

C, Pressure chamber method (pot-grown grass plants)

- 1, Put the plant with pot in a plastic container with a small hole on the top of container. Cut the stem above the height of top of container. Rinse the cut surface with DW and lead it through hole of container. Attach the rubber or silicon tube around stem to form a gasket.
- 2, Pump air into the container with the exhaust of a vacuum pump (e.g. Compact air pump 0.08 Mpa) or oxygen cylinder (e.g. 150 kPa).
- 3, Collect the sap from the top of stump with pipet etc.

(Ref) M. Erik et al, Chemical hydrophobicity and uptake by plant roots. Environ. Sci. Technol., 43:324-329 (2009)