

Measurement of Potassium Content in *Arabidopsis*

Mitsuru Abo¹, Yuriko Osakabe², Kazuko Y-Shinozaki³ and Etsuro Yoshimura^{3*}

¹Agricultural Chemistry, Meiji University, Kawasaki, Japan; ²Gene Discovery Research Group, RIKEN Center for Sustainable Resource Science, Tsukuba, Japan; ³Graduate School of Agricultural and Life Sciences, The University of Tokyo, Tokyo, Japan

*For correspondence: ayoshim@mail.ecc.u-tokyo.ac.jp

[Abstract] Potassium is an essential element in plant growth and has an important role in regulating cell water potential and turgor in osmotic regulation. Potassium content in plants is high compared to trace elements, however, it is difficult to measure a relatively small change of potassium content in the large total. Here, we describe a procedure for measuring potassium in *Arabidopsis* that is easy to handle in preparative scale and avoids contamination.

Materials and Reagents

1. *Arabidopsis* seedlings
2. Milli-Q water
3. Nitric acid (analytical grade for heavy metals) (Wako Pure Chemical Industries, catalog number: 140-04016)
4. Potassium standard solution (1,000 ppm) (Wako Pure Chemical Industries, catalog number: 165-17471)
5. Contaminon L (Wako Pure Chemical Industries, catalog number: 035-09311)
6. Murashige and Skoog salt (Wako Pure Chemical Industries, catalog number: 392-00591)
7. 2-(N-morpholino)ethanesulfonic acid (MES, Wako Pure Chemical Industries, catalog number: 345-01625)
8. Gamborg's Vitamin Solution (Sigma-aldrich, catalog number: G1019)
9. Difco Bacto agar (Becton, Dickinson and Company, catalog number: 214010)
10. Germination media (GM)-agar (see Recipes)

Equipment

1. Plates
2. Centrifugation tubes
3. A paper or aluminum foil
4. Atomic absorption spectrometer (AAS) or Inductively-coupled plasma optical emission

- spectrometer (ICP-OES)
5. Stainless-steel vessel (HU-25, o.d. = 55 mm x 104 mm) (SAN-AI science)
 6. Polytetrafluoroethylene (PTFE) vessel (o.d. = 28 mm x 40 mm) (SAN-AI science)
 7. Tetrafluoroethylene-perfluoroalkyl vinyl ether copolymer (PFA) cup (o.d. = 22 mm x 32 mm, 7 ml) (Sanplatec, catalog number: 0225R)
 8. Volumetric flask (100 ml)
 9. Ceramic scissors and tweezers (AS ONE Corporation, catalog number: 8-203-23, 7-166-11)

Procedure

1. *Arabidopsis* seedlings are germinated for 10 days on GM agar and then cultivated hydroponically with milliQ H₂O in plates for 5 days.
2. Plants are dissected into shoots and roots by cutting the bottom of the shoots with ceramic scissors and tweezers.
3. Samples are dried in disposable centrifugation tubes (50 ml) at 70 °C for 12 h. A paper or aluminum foil is placed over the samples during the drying process.
4. Dried samples (between 15 and 20 plants, corresponding to between 5 and 10 mg as dry weight) are placed into PFA cups with ceramic tweezers and weighed.
5. The PFA cups are placed in PTFE vessels, and nitric acid (1 ml) is added to the PTFE vessels outside the PFA cups. To avoid contamination of impurities in nitric acid, samples are decomposed by nitric acid vapor (not nitric acid liquid).
6. The PTFE vessels are set in stainless-steel vessels (HU-25) and heated at 150 °C for 10 h.
7. The decomposed samples in the PFA cups are diluted with 0.1 M nitric acid and poured into a volumetric flask (100 ml). The sample solution is filled up with 0.1 M nitric acid and subjected to analysis.
8. Potassium content in plants is measured with an atomic absorption spectrometer in emission mode. Standard solutions at concentrations of 0, 1, 2, 4, 8 ppm are prepared from a 1,000 ppm reference solution diluted with 0.1 M nitric acid (It also can be measured by ICP-OES).

Notes

1. To avoid potassium contamination from hands, use rubber gloves during sample treatments.
2. Take care when handling dried samples, which may be attached to vessel walls or

- scattered due to static electricity.
3. Dried samples are very hygroscopic. It is necessary to weigh them quickly for an accurate content calculation.
 4. Contaminon L is used for heavy metal cleaning. Glass vessels are immersed in Contaminon L overnight and rinsed off with MilliQ water before use.

Recipes

1. GM (germination media)-agar
 - 1x Murashige and Skoog salt
 - 3% sucrose
 - 1x Gamborg's Vitamin solution
 - 0.05% 2-(N-morpholino) ethanesulfonic acid (MES)
 - 0.8% Difco Bacto agar
 - Adjusted to pH 5.7 with 1 M KOH

Acknowledgments

This protocol is based on the procedure described by Kojima and Iida (1986).

References

1. KOJIMA, I. and IIDA, C. (1986). [Phase Digestion of Botanical Samples Polytetrafluoroethylene Bomb](#). *Anal Sci* 2: 567.
2. Osakabe, Y., Arinaga, N., Umezawa, T., Katsura, S., Nagamachi, K., Tanaka, H., Ohiraki, H., Yamada, K., Seo, S. U., Abo, M., Yoshimura, E., Shinozaki, K. and Yamaguchi-Shinozaki, K. (2013). [Osmotic stress responses and plant growth controlled by potassium transporters in *Arabidopsis*](#). *Plant Cell* 25(2): 609-624.