Erratum

In the article "Seasonal changes in dehydration tolerance of xylem ray parenchyma cells of *Stylax obassia* twigs that survive freezing temperatures by deep supercooling" by S. Fujikawa et al. [Protoplasma 197: 34–44 (1997)] on p. 35 the section Material and methods was fragmentary and should be completed with the following:

Preparation of samples for electron microscopy

Fresh xylem was cut into small blocks (about $3 \times 3 \times 4 \text{ mm}^3$ in the radial, tangential and longitudinal directions, respectively). Each block was fixed on a freeze-fracture holder with starch paste. These samples were equilibrated and cooled to the desired temperature in the same manner as for DTA. Samples were kept overnight at each desired temperature and were cryofixed by abrupt immersion in melting freon 22 (-150°C). For osmotic manipulation, small blocks were immersed directly in solutions of 5.4 osmol (46.2% (w/w) sorbitol in distilled water, corresponding to freezing-induced dehydration at -10°C) or 10.8 osmol (62.0% (w/w) sorbitol, corresponding to freezing-induced dehydration at -20°C) at room temperature (22°C) for 2 h and then cryofixed. For reference, fresh samples were cryofixed at room temperature to reveal structures without the effects of cooling.

Assessment of freezing injury

Freezing injury was assessed by measuring the leakage of electrolytes from damaged cells. To measure the leakage of electrolytes, fresh xylem was cut into small blocks ($1 \times 1 \times 4 \text{ mm}^3$). From 8 to 12 blocks were placed in the bottom of a test tube. The test tubes were equilibrated at -5°C and cooled at a rate of $0.1^\circ\text{C}/\text{min}$ in a programmable freezer to desired temperatures. Frozen samples in test tubes were equilibrated at desired temperature overnight and thawed

at 0°C for 4 h. 2 ml of distilled water were added to each test tube which was then shaken overnight at 4°C . The electroconductivity of each solution was measured at 22°C with a conductivity meter (CD-35MII; M&S Instruments Inc., Tokyo). The material was then boiled for 10 min and shaken overnight, and then electroconductivity was determined. The electroconductivity before boiling was calculated as a percentage of that after boiling to give the relative electroconductivity (Fujikawa and Miura 1986).

Electron microscopy

Cryofixed samples were processed for observation with a cryo-scanning electron microscope (Cryo-SEM, JSM 840A, equipped with CRU-40; JEOL Co. Ltd., Tokyo) by the previously described method (Fujikawa 1991a). In brief, a cryofixed sample was transferred to a cold stage in the specimen-preparation chamber of Cryo-SEM, equilibrated at -105°C , fractured, etched, and evaporated with platinum-carbon and then reinforced by evaporation of carbon. After observation and photographic recording at an accelerating voltage of 5 kV on the cold stage of the Cryo-SEM at -164°C , the evaporated film (freeze-replica) was extracted from the sample to make a freeze-replica. Freeze-replicas were also produced independently with a freeze-etching apparatus (JFD 7000; JEOL Co. Ltd.). These replicas were examined with a transmission electron microscope (1200EX; JEOL Co. Ltd.).