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LS-8-P-3489 Short- and long-distance signal transmission – following tagged proteins and their mRNA across graft junctions in Arabidopsis

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Grafting is an ancient technique in plant propagation, often used to join productive shoot systems (scions) onto disease-tolerant or otherwise vigorous root systems (rootstocks). In successful grafts, the long-distance transport systems rejoin early, supplying shoots with water and soil nutrients via the xylem, and supplying roots with the products of photosynthesis via the phloem. Grafts are also used to investigate whether endogenous compounds or the products of transgenes can be transported along certain pathways between rootstock and scion. The assumption is that for species with both internal and external phloem, bidirectional phloem transport is possible, but that for other species, this is either not possible or rare.

We investigated the capacity of the limited phloem system in *Arabidopsis thaliana* hypocotyls to carry macromolecules both shootward from roots and rootward from shoots. Rootstocks or scions expressing tagged proteins were grafted in the hypocotyl to non-expressing scions or rootstocks, respectively. We traced protein and mRNA from tissues expressing either green fluorescent protein (GFP), glucuronidase (GUS), or larger GFP-tagged proteins moving from transgenic scions to wild-type (WT) rootstocks, or vice versa. Free GFP (27 kDa mwt), expressed either throughout the tissue or only in phloem companion cells, moved rapidly via reconnected phloem from GFP-expressing scions to WT root tips, but moved only slowly from GFP-expressing rootstocks into the phloem of WT scions. Larger proteins such as GFP-sporamin (67 kDa) or GUS (68 kDa) also moved readily in the phloem from scion tissue into WT roots, and in WT scions on transgenic rootstocks, GUS protein was detected in phloem of many leaves. GFP associated with the endoplasmic reticulum rarely crossed the graft junction in either direction, and was detected within 2-4 cells of the junction. Similarly, transgene mRNA from scions was readily detected in WT rootstocks, but transgene mRNA from rootstocks was only detected in WT scion tissue very close to the graft junction. Clearly, non-native proteins and mRNA can move bidirectionally in phloem, although there is substantially greater movement towards the root tips. Tracer proteins were found only in phloem in the destination WT tissue and accumulated in companion cells. We speculate that similar to the introduced tracer proteins, many endogenous macromolecules may be able to move bidirectionally once they enter the phloem. Escape from the phloem may then require specific unloading signals, and the tight control of unloading may be key to regulation of development by abundant phloem-mobile proteins and RNA species.