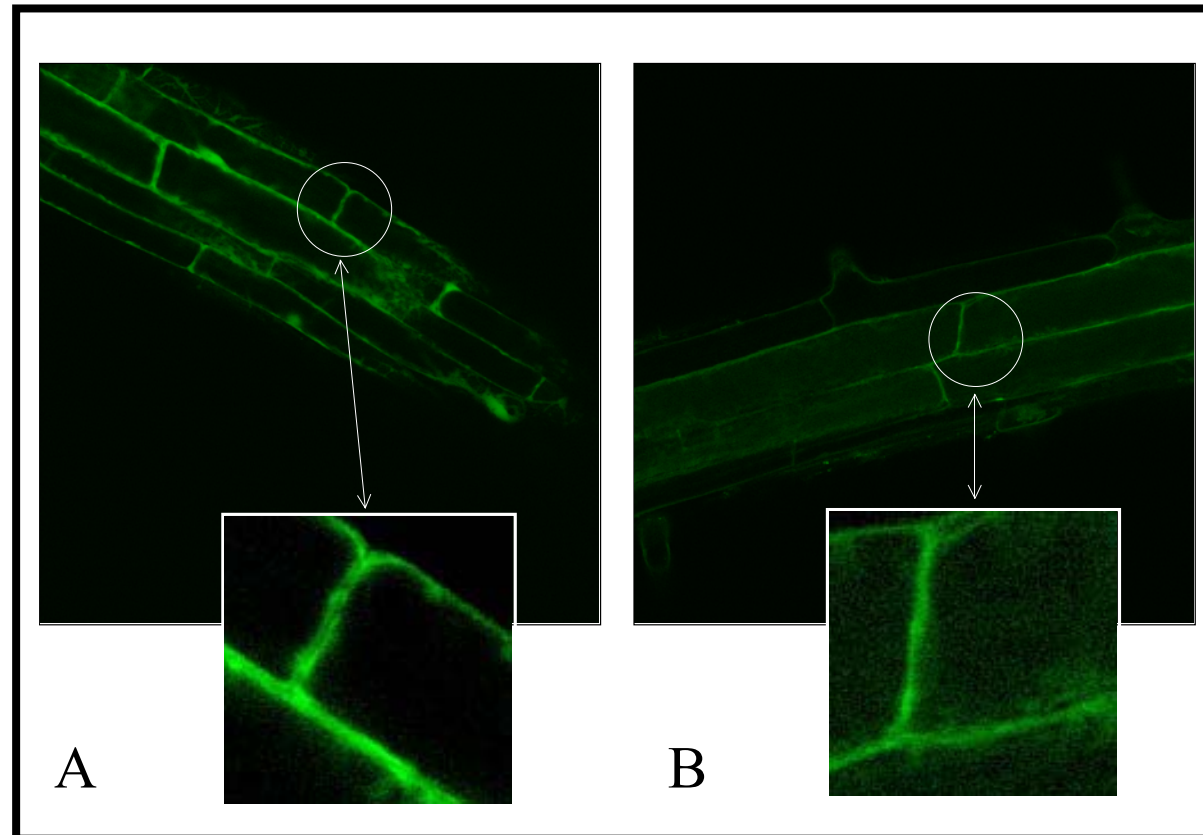
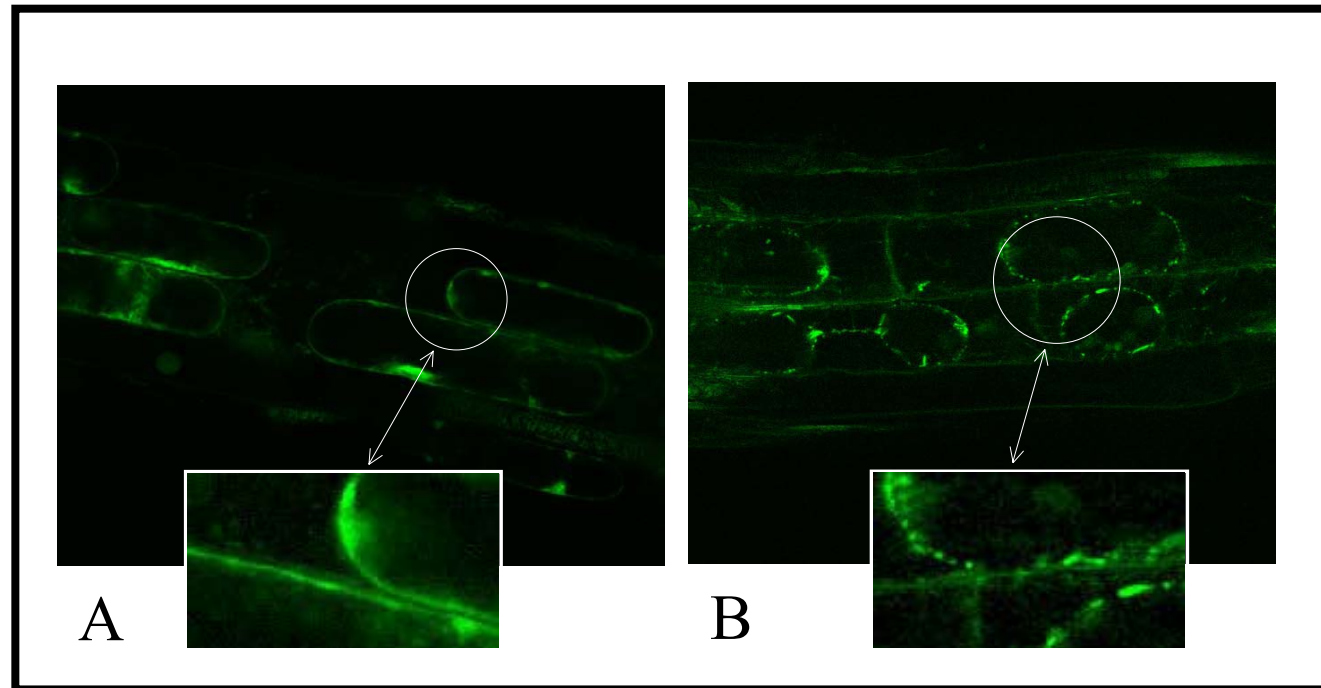
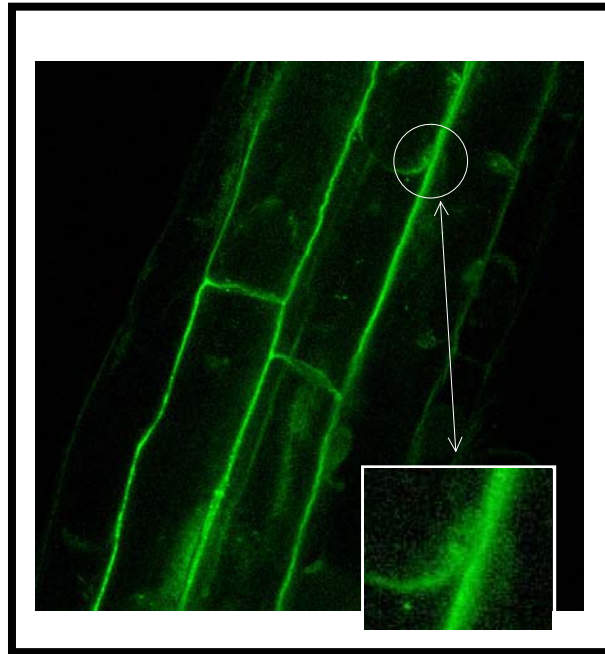


**Supplementary****Material****Figure S1**

Confocal optical sections of root segments from *Arabidopsis* expressing GFP:Aequorin in the cytoplasm (A: smGFP5:Aeq) and in the extracellular space (B: *chit*GFP5:AQ). In the cytoplasmic expression line (A) the cell wall (a non-fluorescent dark line) can be distinguished from the cytoplasm. In the apoplastic line (Fig. S1B), however, the situation is less simple. Although, only the cell wall seems to fluoresce we tried to get a clearer picture and separated cytoplasm and cell wall by plasmolysis (see Fig. S2). Excitation with Argon laser beam lines 458 nm, 476 nm, and 488 nm; emission at 500-540nm; Leica TCS SP confocal laser scanning system; HC PL APD objective (40x oil).

**Figure S2**

Confocal optical sections of plasmolysed root segments from *Arabidopsis* expressing GFP:Aequorin in the cytoplasm (**A**: smGFP5:Aeq) and in the extracellular space (**B**: chitGFP5:AQ) after 15 min treatment with 500mM mannitol. As in Fig. S1 the cell wall (a non-fluorescent dark line) can be distinguished from the cytoplasm in the cytoplasmic expression line (**A**). Plasmolysed cells of the extracellularly expression line (**B**), however, also show fluorescing walls. The bright agglutinated clumps of indicator sticking at the plasma membrane are probably internalization effects caused by the hyperosmotic treatment. Excitation and emission settings as in Fig. S1.



**Figure S3**

Confocal optical section of a plasmolysed root segments from *Arabidopsis* expressing a GFP:Aequorin fusion (*chitGFP5:AQ*) in the extracellular space after 15 h treatment with 100  $\mu$ M cycloheximide and 15 min with 500 mM mannitol. The cycloheximide treatment blocks trafficking and yields a better contrast showing indicator mainly in the cell wall. Excitation and emission settings as in Fig. S1.