

MOLECULAR AND GENETIC ANALYSIS OF MOR1

- a microtubule-associated protein in the model plant *Arabidopsis thaliana*

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The microtubule cytoskeleton is important for plant development, not only for the role it plays during cell division, but because of the role microtubules play in orienting cellulose deposition in the cell wall. To understand how microtubules function in plants, we have investigated MOR1 (microtubule organization1), a 217 kDa microtubule-associated protein (MAP) that is the *Arabidopsis* member of the pan-eukaryotic Dis1/TOGp family of MAPs.

How do MOR1 and its homologues bind to microtubules, and is there a specific microtubule-binding site, perhaps in the C-terminus as suggested by *in vitro* microtubule-binding assays [Twell et al (2002) Nat Cell Biol 4:711]? We constructed six fusions of MOR1 fragments and yellow fluorescent protein (YFP) and investigated their microtubule-binding properties *in vivo*. Transient expression of these proteins in leek epidermis failed to reveal any microtubule labelling. However, tubulin immunofluorescence microscopy showed that some N-terminal but no C-terminal constructs caused microtubule disruption,

suggesting that these fragments interact with microtubules. No microtubule labelling was observed in *Arabidopsis* seedlings stably transformed with the *YFP-MOR1* constructs nor was microtubule disruption observed. However, plants expressing constructs which caused disruption in the transient assay were less fluorescent. Reduced expression of these microtubule-disrupting *YFP-MOR1* transgenes was demonstrated by quantitative realtime PCR. These results, and our analysis of MOR1 gene sequences, indicate that microtubule-binding in MOR1 is not limited to the C-terminal domain.

