

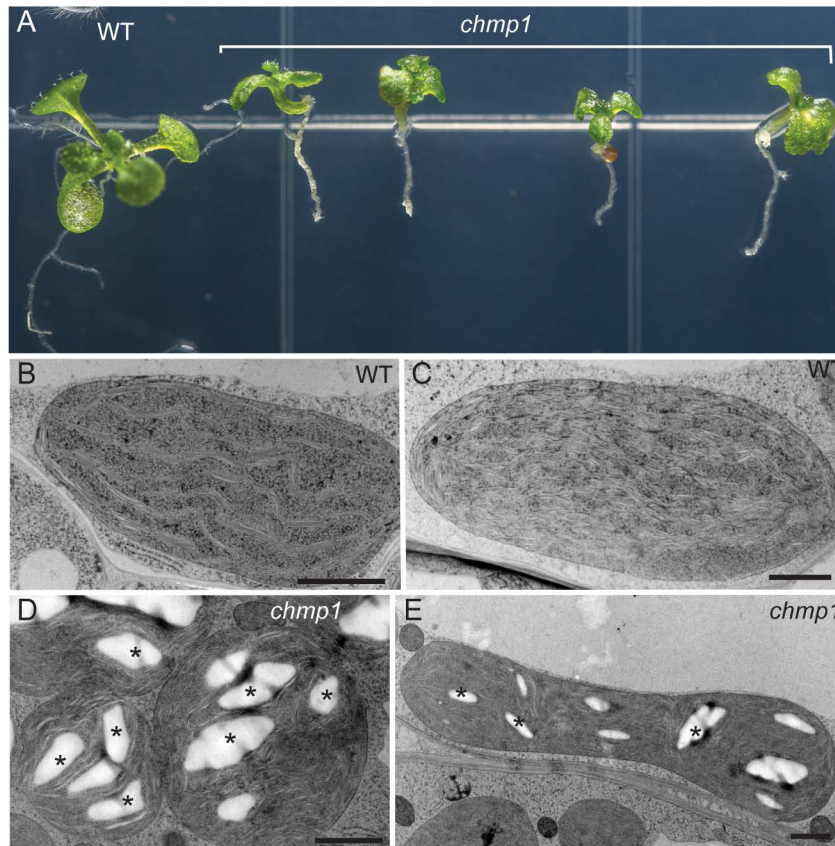
**Supplemental Figure 1.** Plastid Defects in *chmp1* Mutant Seedlings.

(A) to (F) Imaging of chlorophyll autofluorescence (magenta) at the hypocotyl of WT and *chmp1a chmp1b* mutant seedlings.

(G) and (H) Detail of a mutant plastid cluster and a connecting plastid bridge (arrowhead).

(I) to (L) Immunofluorescence detection of FTSZ2 rings (arrowheads) in WT and *chmp1a chmp1b* mutant seedlings.

Bars: 500  $\mu$ m in (A) and (D), 50  $\mu$ m in (B), (C), (E), (F), 2  $\mu$ m in (G) to (L).



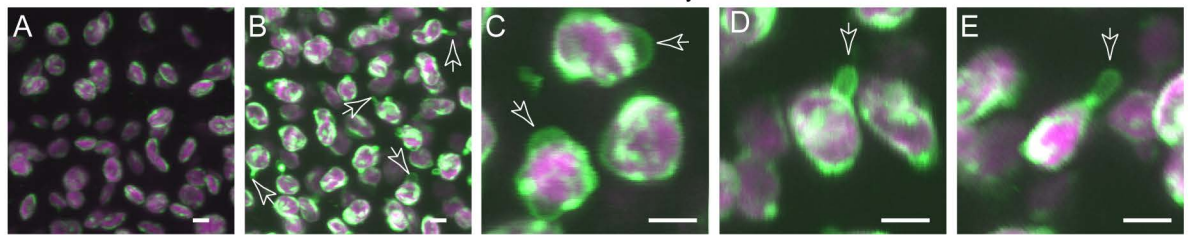
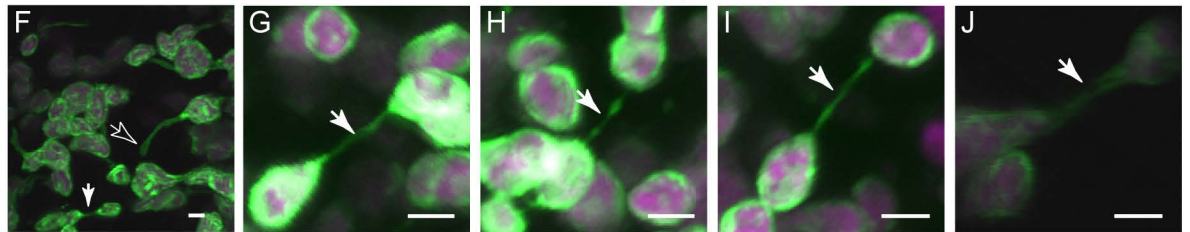
**Supplemental Figure 2.** Seedling Morphology and Chloroplast Ultrastructure.

**(A)** Twelve-day-old WT and *chmp1* seedlings.

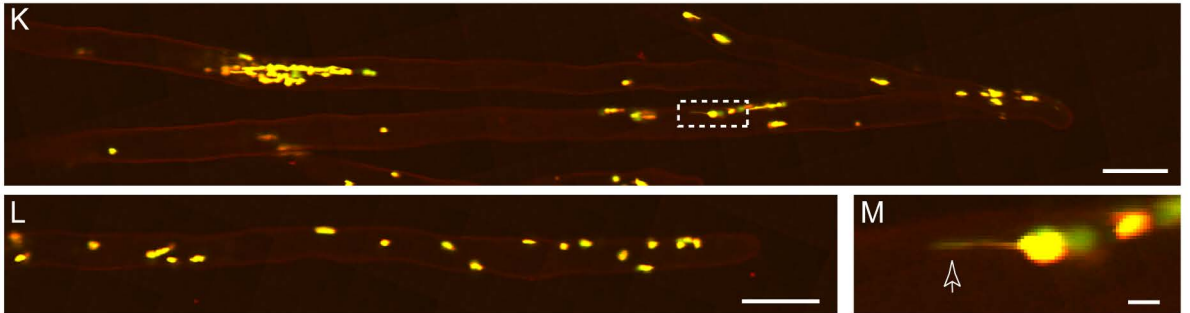
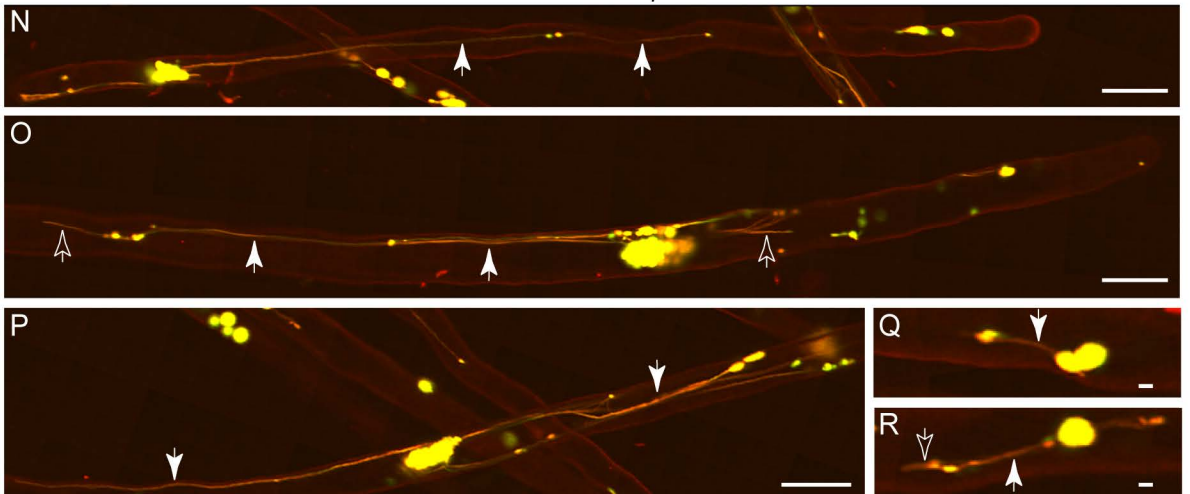
**(B) to (E)** High-pressure frozen/freeze substituted cotyledons from WT and *chmp1* seedlings. The *chmp1* chloroplasts accumulate large starch granules (asterisks) but show normal organization of thylakoid membranes.

Bars: 1 μm.

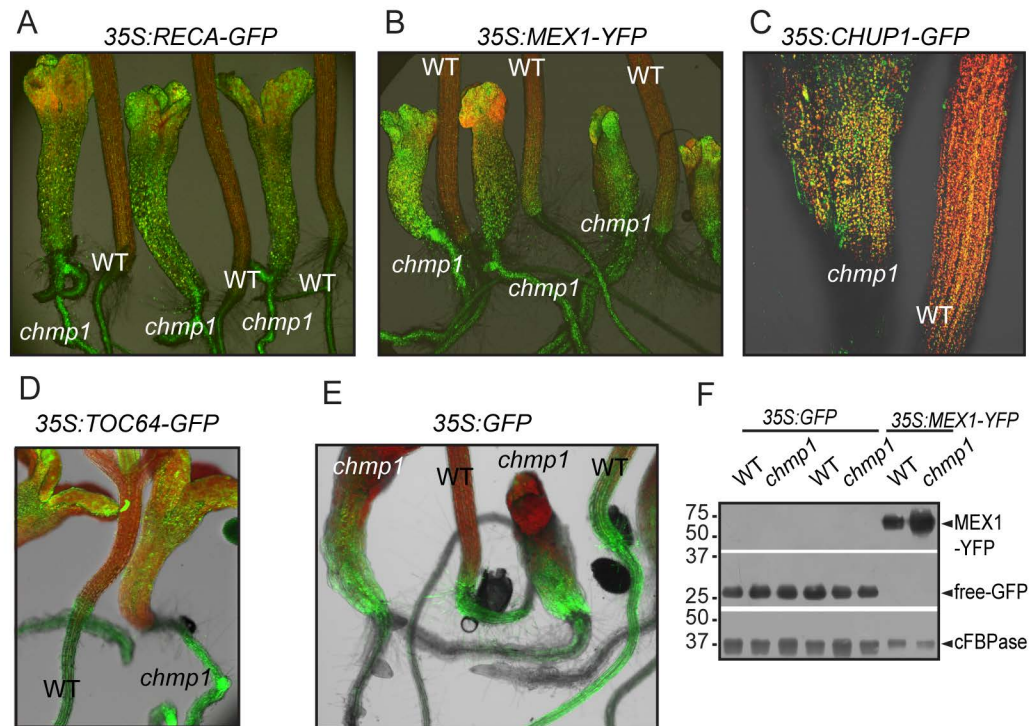
## MEX1-YFP in WT embryo cells

MEX1-YFP in *chmp1* embryo cells

## RECA-GFP in WT root hairs

RECA-GFP in *chmp1* root hairs

**Supplemental Figure 3.** Abnormal Plastid Morphology in *chmp1* Embryos and Root Cells. **(A) to (E)** Embryo cells expressing the inner envelope marker MEX1-YFP. Chlorophyll autofluorescence in magenta. **(K) to (R)** Root hairs stained with propidium iodide (red) and expressing the stroma marker RECA-GFP (green). Yellow denotes overlap of both signals. Open arrows indicate stromules and plastid extensions and closed arrows show interconnecting plastid bridges. Boxed region in **(K)** is shown at higher magnification in **(M)**. Bars = 2  $\mu$ m in **(A) to (J), (M), (Q), (R)**, 20  $\mu$ m in **(K), (L), (N) to (P)**.



**Supplemental Figure 4.** Protein Expression in WT and *chmp1* seedlings. **(A)** to **(E)** Confocal imaging of control and *chmp1* mutant seedlings expressing RECA-GFP, MEX1-YFP, CHUP1-GFP, TOC64-GFP, and cytosolic GFP under the control of *CaMV35S*. Chlorophyll autofluorescence is shown in red. Note that the signals from fluorescently-tagged plastid proteins but not from cytosolic GFP are stronger in *chmp1* than in WT seedlings.

**(F)** Immunoblot detection with anti-GFP antibodies indicates that MEX1-YFP but not cytosolic GFP accumulate at higher levels in the *chmp1* mutant compared to control seedlings. Cytoplasmic FBPase was used as loading control. Densitometric analysis of this immunoblot is shown in Supplemental Table 1.

Supplemental Table 1: Densitometric quantification of western blots shown in Figure 2 and Supplemental Figure 3.

	<b>WT</b>		<b><i>chmp1</i></b>		Fold increase
	Average	Std Dev	Average	Std Dev	
Mex1/FBPase ratio	1.190032	0.617486	3.642005	1.333038	3.060426
Tic110/cFBPase	0.217453	0.041303	1.067541	0.375686	4.909298
Toc75/cFBPase	0.201725	0.58522	1.302932	0.58522	6.458938
PsbA-D1/cFBPase	1.472533	0.239303	1.605705	0.57613	1.090437
FtsZ2/cFBPase	0.336573	0.105448	2.875513	1.731814	8.543514
PEX14/cFBPase	0.149051	0.016022	0.648455	0.137086	4.350569
VDAC/cFBPase	0.455402	0.042822	1.7901	0.172553	3.930815
GFP/cFBPase	0.666119	0.053307	0.961918	0.094021	1.444063

	<b>WT 6d</b>		<b>WT 12d</b>		<b><i>chmp1</i> 12d</b>	
	Average	Std Dev	Average	Std Dev	Average	Std Dev
Tic110/cFBPase	0.31	0.18	0.31	0.16	0.72	0.18
TIC110/PBA1	0.60	0.43	0.57	0.28	1.09	0.1
TIC110/H3	0.54	0.31	0.63	0.34	1.14	0.42
PDV2/ cFBPase	0.11	0.05	0.12	0.05	0.72	0.36
PDV2/PBA1	0.21	0.08	0.22	0.10	1.18	0.69
PDV2/H3	0.19	0.08	0.21	0.09	1.0	0.46
PSAF/ cFBPase	1.15	0.19	1.08	0.17	1.19	0.10
PSAF/PBA1	2.14	0.41	2.01	0.47	1.67	0.33
PSAF/H3	1.88	0.03	1.99	0.32	1.63	0.14

Supplemental Table 2: Densitometric quantification of western blot shown in Figure 4N

	<i>chmp1</i>		WT	
	N	no N	N	no N
Free GFP	9665.267	39082.63	6196.832	20976.07
GFP-ATG8	6990.196	17216.87	7056.368	6347.711
GFP/ATG8-GFP ratio	1.382689	2.270019	0.87819	3.304509
Fold increase of GFP/ATG8-GFP ratio upon starvation		<b>1.641742</b>		<b>3.762863</b>
cFBPase	17172.67	20295.24	30688.47	18005.72
Free GFP/cFBPase	0.562828	1.925704	0.201927	1.164967
GFP-ATG8/cFBPase	0.407053	0.848321	0.229935	0.352539
Total GFP + GFP-ATG8	<b>0.969882</b>	<b>2.774025</b>	<b>0.431862</b>	<b>1.517505</b>