Supplemental Data. Gerber et al. (2009). The Plastidial 2-*C*-Methyl-D-Erythritol 4-Phosphate Pathway Provides the Isoprenyl Moiety for Protein Geranylgeranylation in Tobacco BY-2 Cells.



Figure S1. MALDI-TOF/TOF MS/MS spectra.

(A) Spectum representing the fragmentation of the C- terminal modified peptide DHDGGSRTKSAGPSAAPASKRGQKC + (1) miss cleavage + methyl + S-geranylgeranyl identified by MALDI- TOF-MS in Figure 6. The peptide with the corresponding parent ion mass $[M+H]^+ = 2756.620 \text{ m/z}$ (indicated by an arrow in Figure 6) was isolated and fragmented by LIFT CID for partial sequencing.

(B) Spectrum representing the fragmentation of the C- terminal modified peptide DHDGGSRTKSAGPSAAPASKRGQKCVIL + (1) miss cleavage identified by MALDI- TOF-MS (figure 6). The peptide with the corresponding parent ion mass $[M+H]^+ = 2852.680 \text{ m/z}$ (indicated by an arrow) was isolated and fragmented by LIFT CID for partial sequencing.



Figure S2. Addition of Ca^{2+} or the calmodulin inhibitor calmidazolium does not affect the localization of GFP-CaM61 or GFP-BD-CVIL.

(A) Localization of GFP-CaM61 in BY-2 cells treated with 1 mM calcium, showing GFP fluorescence associated with ER and perinuclear membranes, plasma membrane, and cytoplasmic strands.

(B) Localization of GFP-BD-CVIL in BY-2 cells treated with 1 mM calcium, showing GFP fluorescence associated exclusively with the plasma membrane.

(C) Localization of GFP-CaM61 in BY-2 cells treated with 5 μ M calmidazolium, showing GFP fluorescence associated with ER and perinuclear membranes, plasma membrane, and cytoplasmic strands.

(**D**) Localization of GFP-BD-CVIL in BY-2 cells treated with 5 μ M calmidozolium, showing GFP fluorescence associated with the plasma membrane. White bars correspond to 10 μ m.



Figure S3. Mevinolin treatment of BY-2 cell cultures arrests cell growth.

- (A) Control BY-2 cell culture.
- (B) BY-2 cell culture treated with 5 μ M mevinolin

Table S1. Chi-squared analysis of data in Figure 3*										
	Observed	Expected	O-E	$(O-E)^2$	$(O-E)^{2}/E$	X^2				
	(O)	(E)								
MV5: PM	161	161	0	0	0	0				
MV5:						P > 0.05				
Delocalized	3	3	0	0	0	Consistent				
Fos40: PM	81	233	-152	23,104	99.16	4,719.96				
Fos40:						0.001 > P				
Delocalized	157	5	152	23,104	4,620.80	Not				
						Consistent				
Fos40MV5:	10	215	-205	42,025	195.47	10,701.72				
PM						0.001 > P				
Fos40MV5:						Not				
Delocalzied	209	4	205	42,025	10,506.25	Consistent				
OC40: PM	47	330	-283	80,089	242.69	13,590.86				
OC40:						0.001 > P				
Delocalized	289	6	283	80,089	13,348.17	Not				
						Consistent				
OC40MV5:	0	409	-409	167,281	409	21,319.13				
PM						0.001 > P				
OC40MV5:						Not				
Delocalized	417	8	409	167,281	20,910.13	Consistent				
*All values were compared by Chi-squared analysis against the null hypothesis (i.e., no										
effect compared to the control). Values with a $P > 0.05$ were considered to be consistent										
with the null hypothesis. The only treatment found to be consistent with the null										

hypothesis was the MV5 treatment.

Table S2. Chi-squared analysis of data in Figure 4*									
	Observed	Expected	O-E	$(O-E)^{2}$	(0-	X^2			
	(O)	(E)			$E)^2/E$				
Fos40Gol20: PM	60	60	0	0	0	0			
Fos40Gol20:						P > 0.05			
Delocalized	242	242	0	0	0	Consistent			
Fos40Fol20: PM	164	47	117	13,689	291.26	363.31			
Fos40Fol20:						0.001 > P			
Delocalized	73	190	-117	13,689	72.05	Not			
						Consistent			
Fos40GGol20: PM	91	18	73	5,329	296.06	369.06			
Fos40GGol20:						0.001 > P			
Delocalized	0	73	-73	5,329	73	Not			
						Consistent			
Fos40MVA3: PM	145	29	116	13,456	464	578.03			
Fos40MVA3:						0.001 > P			
Delocalized	2	118	-116	13,456	114.03	Not			
						Consistent			
Fos40MV5Gol20:	32	16	16	256	16	17.08			
PM						0.001 > P			
Fos40MV5Gol20:						Not			
Delocalized	220	236	-16	256	1.08	Consistent			
Fos40MV5Fol20:	47	19	28	784	41.26	44.19			
PM						0.001 > P			
Fos40MV5Fol20:						Not			
Delocalized	240	268	-28	784	2.93	Consistent			
Fos40MV5GGol20:	152	9	143	20,449	2,272.11	2,415.11			
PM						0.001 > P			
Fos40MV5GGol20:						Not			
Delocalized	0	143	-143	20,449	143	Consistent			
Fos40MV5MVA3:	107	7	100	10,000	1,428.57	1,528.57			
PM						0.001 > P			
Fos40MV5MVA3:						Not			
Delocalized	0	100	-100	10,000	100	Consistent			

*All values were compared by Chi-squared analysis against the null hypothesis (i.e., no complementation compared to the Fos40 or Fos40MV5 controls). Values with a P > 0.05 were considered to be consistent with the null hypothesis. The only treatment found to be consistent with the null hypothesis was the Fos40Gol20 treatment.