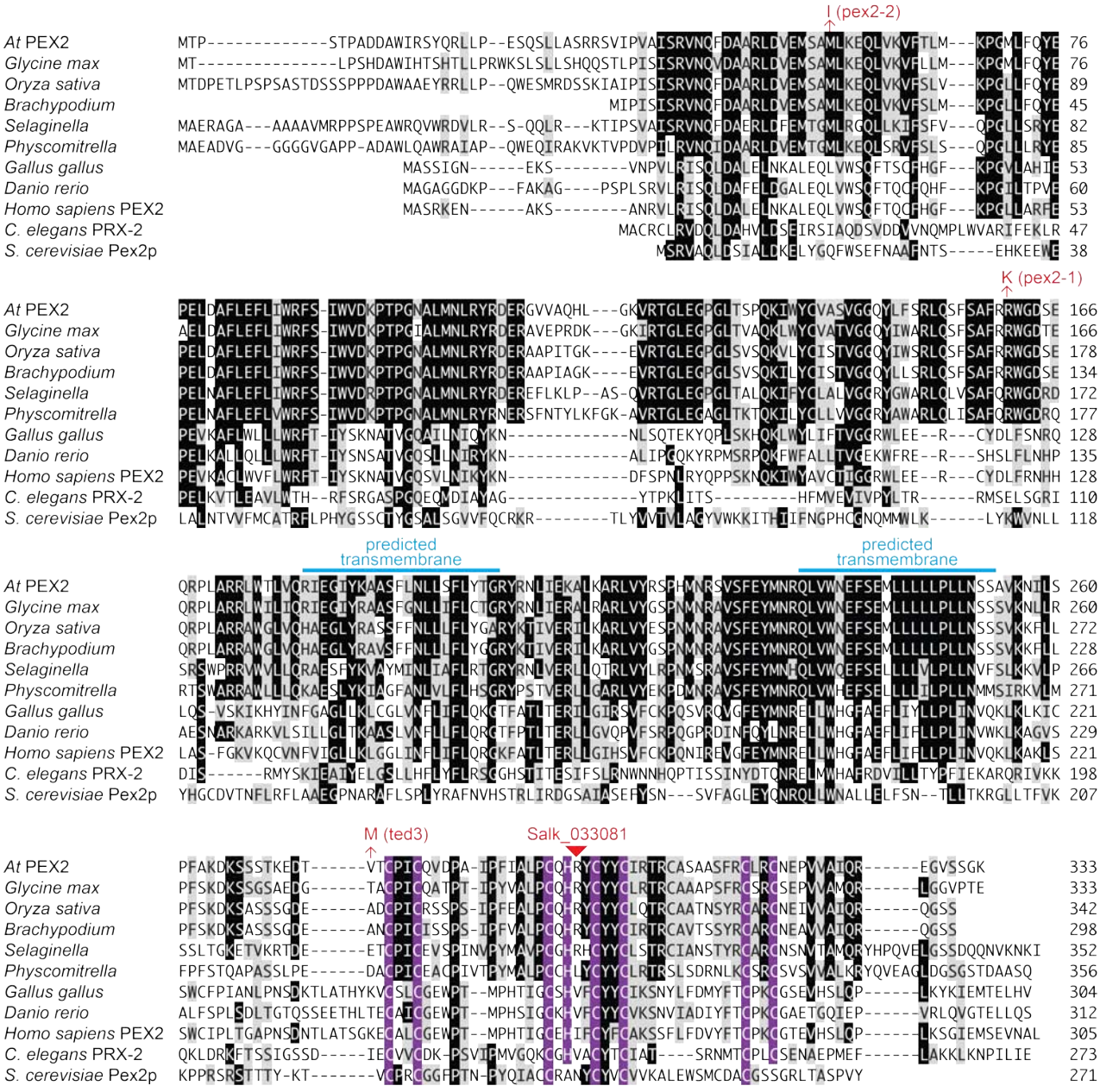
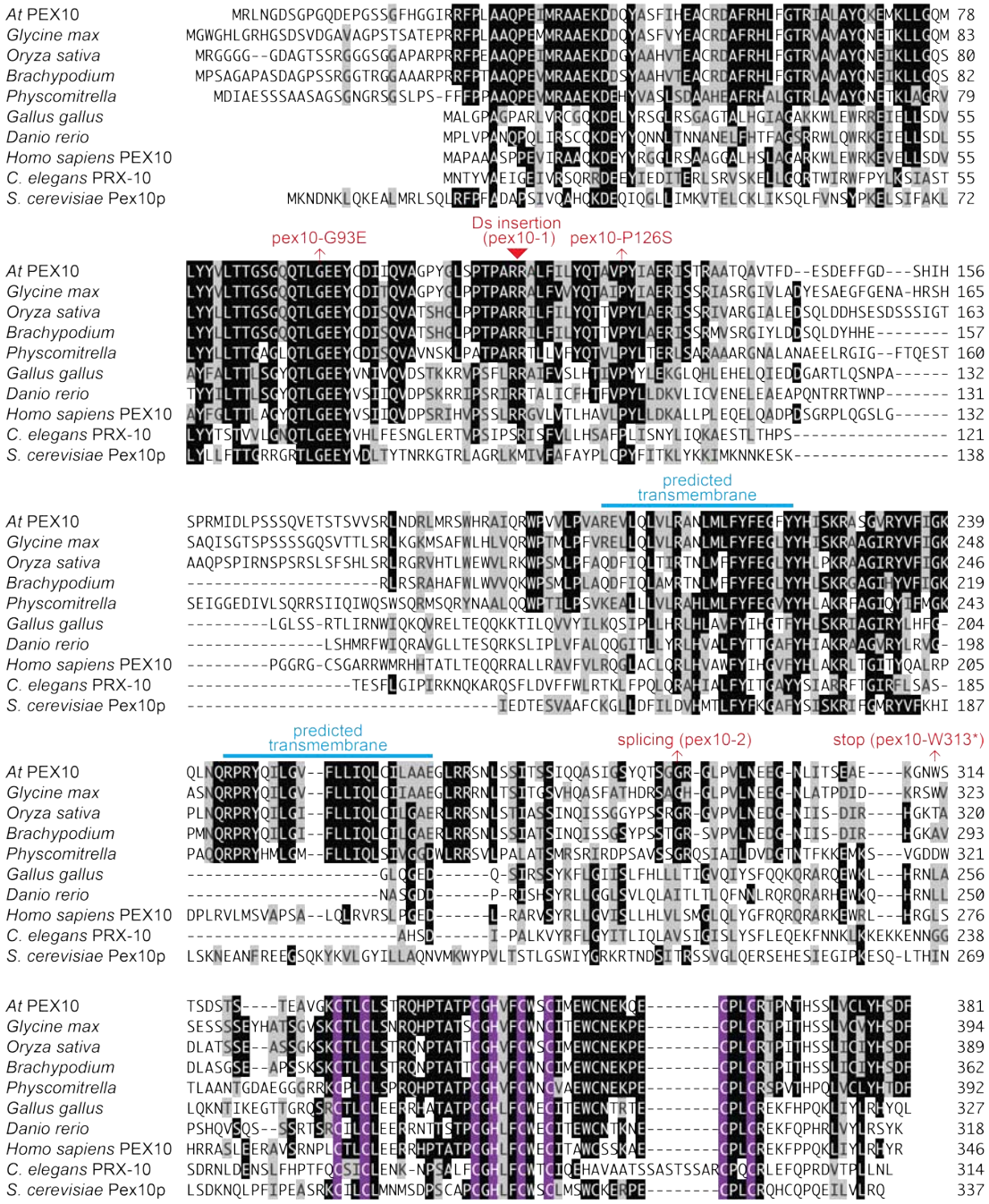
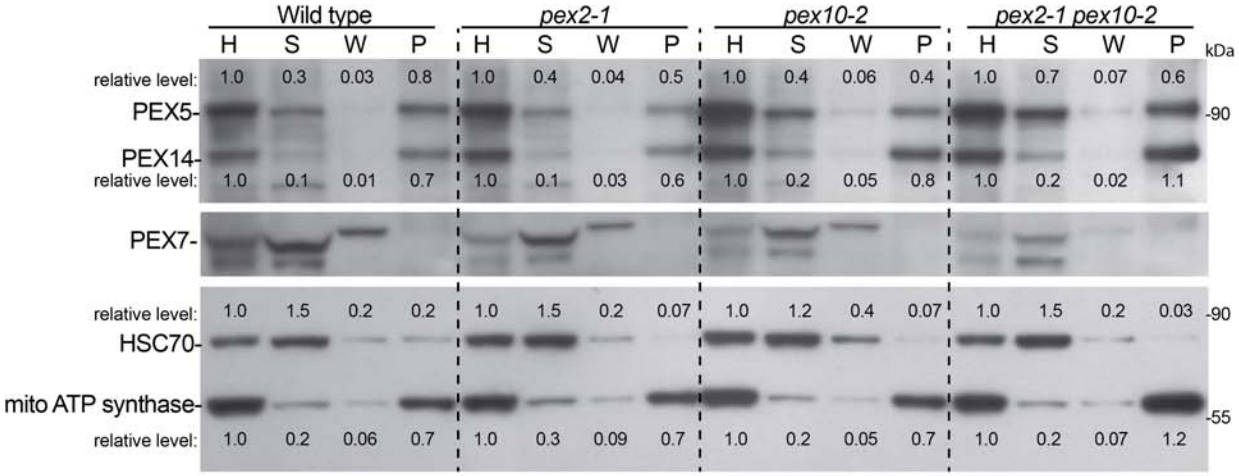


Supplemental Figure S1. Alignment of PEX2 proteins from various organisms.
Arabidopsis (*At*) PEX2 (At1g79810) was aligned with homologs from *Glycine max* (XP_003530138.1), *Oryza sativa* (Os05g0275700), *Brachypodium distachyon* (XP_003573500.1), *Selaginella moellendorffii* (XP_002976539.1), *Physcomitrella patens* (XP_001781834.1), *Gallus gallus* (NP_001008454.1), *Danio rerio* (XP_684073.2), *Homo sapiens* (NP_000309.1), *Caenorhabditis elegans* (CAA92640.2), and *Saccharomyces cerevisiae* (NP_012325.1). The alignment was generated using Lasergene MegAlign (DNASTAR, Madison, WI) with the Clustal W default settings and the Gonnet series protein weight matrix. Residues identical in at least six sequences are shaded in black or purple; chemically similar residues are shaded in gray. Transmembrane domains predicted using the ARAMEMNON database (Schwacke et al., 2003) are indicated in blue, metal-coordinating residues of the RING-finger domain are highlighted in purple, and positions of *Arabidopsis pex2* mutants are indicated in red.



Supplemental Figure S2. Alignment of PEX10 proteins from various organisms.
Arabidopsis (*At*) PEX10 (At2g26350) was aligned with homologs from *Glycine max* (XP_003544551.1), *Oryza sativa* (Os07g0608800), *Brachypodium distachyon* (XP_003559973.1), *Physcomitrella patens* (XP_001777562.1), *Gallus gallus* (NP_001185583.1), *Danio rerio* (NP_001005994.1), *Homo sapiens* (NP_722540.1), *Caenorhabditis elegans* (NP_001021200.2), and *Saccharomyces cerevisiae* (NP_010551.1). The alignment was generated using Lasergene MegAlign (DNASTAR, Madison, WI) with the Clustal W default settings and the Gonnet series protein weight matrix. Residues identical in at least five sequences are shaded in black or purple; chemically similar residues are shaded in gray. Transmembrane domains predicted using the ARAMEMENON plant membrane protein database (Schwacke et al., 2003) are indicated in blue, metal-coordinating residues of the RING-finger domain are highlighted in purple, and positions of *Arabidopsis pex10* mutants are indicated in red.



Supplemental Figure S3. Membrane association of peroxisome matrix protein receptors in *pex2-1*, *pex10-2,* and *pex2-1 pex10-2*.

Light-grown 8-d-old seedlings were homogenized and the resultant extracts were centrifuged to separate soluble proteins from proteins associated with organellar membranes. Homogenate (H), soluble (S), wash (W) and pellet (P) fractions were processed for immunoblotting and serially probed with the indicated antibodies. HSC70 and mitochondrial membrane complex V (mito ATP synthase) were used as cytosolic and organellar controls, respectively. Numbers above or below the bands indicate the relative level of the indicated protein in the S, W, or P fraction relative to the amount in the H fraction. This experiment was repeated three times with similar results.

Supplemental **Table 1.** *Markers used in recombination mapping of pex2-1 and pex10-2*

Marker	Nearest gene	Restriction enzyme	Fragment size (bp) Col-0	Fragment size (bp) L*er*	Primer sequences
LCS1127	*At1g32130*	*Taq*I	206	138, 68	CAAAATCCTGAAGTGGCTCTGTAG ATTTCGATTTCGTTTTTGTG
T14L22	*At1g51913*	*Hinf*I	~700	~450, ~250	GAAAAATTAAAGCCCAACC CACCCGCAAACACAACAACC
SNP10592	*At1g69250*	*Rsa*I	115, 29	144	AGCGACGGCAACCTAAAAAAGTAAAG AGGTTCAAGCACATTGATGATACTCTGTA
SEC238	*At2g18950*	*Dde*I	145, 69	214	GAGTTCTGCGTTGTGATTCGAGTAAAGTTG GAAAACCTGTAAAACGCATCTAACGAGTC
SEC241	*At2g20770*	*Pvu*II	177, 27	204	CAACTCAAACTTATTCACCATCAAACAACAA TATCTCTTCCTTGACATGGTCGACCCAGCT
SEC244	*At2g25095*	*Ava*II	197, 28	225	TAAAAATAGTAAACTTGTCCCGATAAA GTCGATAGATTCACGGACGTATTAGAAAGG
SEC245	*At2g28360*	*Rsa*I	116, 28	144	CTGCAGCTTCGTTTCTAGGATATTTGGTCAT CTATAAGAATTGAAGAAAGGCGATGAAGTA
SEC246	*At2g32340*	*Rsa*I	114, 27	141	TTTACCTGTTCCGCCAATCACTCCTC GAACCTGCGTCGCTATCTGCTTCCTCCGTA

Supplemental **Table 2.** *Primers used for amplification and sequencing of PEX2 and PEX10*

Gene/Accession #	Primer names	Primer sequences
PEX2/At1g798100	PEX2-12 and	CATGGATGAATCTCTGTGGGTGTGA
	PEX2-13	GAAAGAAAAAGAAGAGTAGCGGAAGTGAGAA
	PEX2-14 and	TCGCTATGGTTGATTCTTTTCTCCTT
	PEX2-15	GCTGCTTGCCTTGGCTTTTTACACTTC
PEX10/At2g26350	PEX10-3 and	GACCACCTAACATCTCCAACTCTTTT
	PEX10-4	ATGGCCACTCGACTGATCCCGCTCTAC
	PEX10-5 and	AAGGAGTGGCCGTTGGGTGCTGAC
	PEX10-6	TTCTTGGGGTTTTCCTTCTAATC
	PEX10-7 and	CCTATAGAAGCCTGCTGAATGGAG
	PEX10-8	TCAGGGCAACAAACTTTAGGAGAGGAAT
	PEX10-9 and	TGGAACTGCGGTCTGGTACAATATGAACAAAG
	PEX10-10	CTTTGAATTAAAAAAAAATATCAAAGGC

Supplemental **Table 3.** *PCR-based markers for determining mutant genotypes*

Mutant	Primer names	Primer sequences	Restriction enzyme	Product size (bp)	
				Wt	Mutant
pfl36/pex2-1	PEX2-18	TGCGTTGCCTCCGTTGGTGGTCAG	*Dpn*II	65, 18	93
	PEX2-DpnII[1]	CATACAGACCTGCTCAGAATCACCCGAT			
pex2-2	PEX2-20	GCTTATTATATGTGGTCTTTGATGGCAGTC	*Nde*I	134	102
	PEX2-NdeI[1]	GAGTGAAAACCTTAACGAGCTGTTCCTTCCA			
ted3	TED3-1	ATGCTTTTGTTGCTTCTCCCGCTGCTC	*Tsp*451	341, 99	440
	TED3-2	TCTAAGACACCGGAACGATGCTGCTG			
pfl81/pex10-2	PEX10-11	CGTTGAAGTTGAATCGGAGGTAGAC	*Pst*I	92, 28	120
	PEX10-PstI[1]	AATATAGTTTTGGTATTGTTCCTGCA			
pex10-P126S[2]	PEX10-8	TCAGGGCAACAAACTTTAGGAGAGGAAT	-	229	229
	PEX10-TaqI	ACGAAACCTAATTCTCTCTGCGATATATC			

[1]dCAPS oligonucleotide (Michaels and Amasino, 1998; Neff et al., 1998); the underlined nucleotide differs from wild-type sequence to create a restriction site in either the mutant or wild-type PCR amplicon.

[2]*pex10-P126S* was amplified with the listed primers and sequenced with primer PEX10-8 to confirm genotype.

Supplemental literature cited:

Michaels SD, Amasino RM (1998) A robust method for detecting single-nucleotide changes as polymorphic markers by PCR. Plant J **14:** 381-385

Neff MM, Neff JD, Chory J, Pepper AE (1998) dCAPS, a simple technique for the genetic analysis of single nucleotide polymorphisms: experimental applications in *Arabidopsis thaliana* genetics. Plant J **14:** 387-392

Schwacke R, Schneider A, van der Graaff E, Fischer K, Catoni E, Desimone M, Frommer WB, Flugge UI, Kunze R (2003) *ARAMEMNON*, a novel database for Arabidopsis integral membrane proteins. Plant Physiol **131:** 16-26