## Supporting Information

## Molecular Dynamics Simulations Reveal Differentiated Context –Dependent Conformational Dynamics of Two Proteins of the Same Family

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		β1	α1	$\alpha 2$	β2	
PYR1	TT		lllll	TT eee	TT	TT
	_				CL1	76
PYR1	.QLDPGS <mark>C</mark>	SSLHAQRIH	<b>AP</b> PEL <mark>VW</mark> SIVRI	RFDKPQTYF	HFIKS <mark>C</mark> SVEQNF	EMRV <mark>G</mark>
PYL1	YQLGNGRC	SSLLAQRIH	<b>AP</b> PETVWSVVRI	RFDRPQIYF	H <mark>fiksc</mark> nvsedf	EMRV <mark>G</mark>
PYL2	FEPDPTTC	T <mark>S</mark> LITQRIH	<b>AP</b> ASVV <mark>W</mark> PLIRI	RFDNPERYF	H <mark>FVK</mark> R <mark>C</mark> RLISGD	GDV <mark>G</mark>
PYL3	FPRSPNTC	TSLIAHRVD	<b>AP</b> AHAI <mark>W</mark> RFV <mark>R</mark> I	D <b>F</b> AN <mark>P</mark> NKYF	HFIKSCTIRVNGNG.I	IKEIKV <mark>g</mark>
PYL4	HEVGPNQ <mark>C</mark>	CSAVIQEIS	APISTVWSVVRJ	R <mark>F</mark> DN <mark>P</mark> QAYF	HFLKSCSVIGGDGD.	NV <mark>G</mark>
PYL5	HDVGPDQ <mark>C</mark>	CSSVVQMIH	<b>AP</b> PES <mark>VW</mark> ALVRI	R <mark>f</mark> dn <mark>p</mark> kvyf	N <mark>FIRQC</mark> RIVQGDGL.	HV <mark>G</mark>
PYL6	HVVGPSQ <mark>C</mark>	F <mark>S</mark> VVVQD <mark>V</mark> E	<b>AP</b> VSTV <mark>W</mark> SILSI	R <mark>F</mark> EH <mark>P</mark> QAYF	H <mark>FVK</mark> S <mark>C</mark> HVVIGDGR.	EV <mark>G</mark>
PYL7	HHCRENQC	T <mark>S</mark> VLVKYIQ	<b>AP</b> VHLV <mark>W</mark> SLVRI	R <mark>F</mark> DQ <mark>P</mark> QKYF	PFISRCTVNGD	PEI <mark>G</mark>
PYL8	HELVDNQC	SSTLVKHIN	<b>AP</b> VHIV <mark>W</mark> SLVRI	R <mark>F</mark> DQ <mark>P</mark> QKYF	PFISRCVVKGN	MEI <mark>G</mark>
PYL9	HLCRENQC	TSALVKHIK	APLHLVWSLVR	R <mark>F</mark> DQ <mark>P</mark> QKY}	PFVSRCTVIGD	PEI <mark>G</mark>
PYL10	HELVESQC	SSTLVKHIK	<b>AP</b> LHLV <mark>W</mark> SIVRI	R <mark>f</mark> de <mark>p</mark> qkył	(PFISR <mark>C</mark> VVQGKK	LEV <mark>G</mark>
PYL11	TSQKYHT <mark>C</mark>	GSTLVQTID	APLSLVWSILRI	R <mark>F</mark> DN <mark>P</mark> QAYF	QFVKTCNLSSGDG	GE <mark>G</mark>
PYL12	TSQEQHV <mark>C</mark>	GSTVVQTIN	APLPLVWSILRI	RFDNPKTFF	HEVKTCKLRSGDG.	GE <mark>G</mark>
PYL13	ESSKQKRC	RSSVVETIE	APLPLVWSILR:	SFDKPQAY	REVKSCTMRSGGGGG	KGGEGK <mark>G</mark>

	β3		β4	β5		β6
PYR1		TT —	<b>&gt;</b>	$\longrightarrow$	TT	→T
		Gate		L	atch	132
PYR1	CT <mark>RDV</mark> IVI	I <mark>SGLPA</mark> NT <mark>S</mark> I	ERLDILDDE	RR <mark>V</mark> TGFSII <mark>GGE</mark>	IRLT <b>NY</b> K <mark>S</mark> VTT	VHRFEKE
PYL1	CT <mark>RDV</mark> NVI	I <mark>SGLPA</mark> NT <mark>S</mark> F	RERLDLLDDD	RR <mark>V</mark> TGFSIT <mark>GG</mark> E <mark>I</mark>	IRLR <mark>NY</mark> K <mark>S</mark> VTT	VHRFEKEE
PYL2	SV <mark>RE</mark> VTVI	I <mark>SGLPA</mark> ST <mark>S</mark> I	ERLEFVDDD	HR <mark>V</mark> LSFRVV <mark>GG</mark> E <mark>I</mark>	IRLK <mark>NY</mark> K <mark>S</mark> VTS	VNEFLNQD
PYL3	TI <mark>RE</mark> VSVV	/ <mark>SGLPA</mark> ST <mark>S</mark> V	<b>EILEVLDEE</b>	KRILSFRVL <mark>GG</mark> EI	IRLN <mark>NY</mark> R <mark>S</mark> VTS	VNEFVVLEKD
PYL4	SL <mark>RQ</mark> VHVV	/ <mark>SGLPA</mark> AS <mark>S</mark> I	ERLDILDDE	RH <mark>V</mark> ISFS <mark>VV<mark>GG</mark>D<mark>I</mark></mark>	<mark>irl</mark> s <mark>ny</mark> r <mark>s</mark> vtt	LHPSPIS
PYL5	DL <mark>RE</mark> VMVV	/ <mark>SGLPA</mark> VS <mark>S</mark> I	ERLEILDEE	RH <mark>V</mark> ISFS <mark>V</mark> V <mark>GG</mark> D <mark>I</mark>	IRLK <mark>NY</mark> R <mark>S</mark> VTT	LHASDDE
PYL6	SV <mark>RE</mark> VRVV	/ <mark>SGLPA</mark> AF <mark>S</mark> I	ERLEIMDDD.	RH <mark>V</mark> ISFS <mark>V</mark> V <mark>GG</mark> D <mark>I</mark>	IRLM <mark>NY</mark> K <mark>S</mark> VTT	VHESEEDS
PYL7	CL <mark>REV</mark> NVI	K <mark>SGLPA</mark> TT <mark>S</mark> I	ERLEQLDDE	EHILGINII <mark>GG</mark> DI	IRLK <mark>NY</mark> S <mark>S</mark> ILT	VHPEMID
PYL8	TV <mark>REV</mark> DVI	K <mark>SGLPA</mark> TR <mark>S</mark> I	ERLELLDDN	EHILSIRIV <mark>GG</mark> DI	IRLK <mark>NY</mark> S <mark>S</mark> IIS	LHPETIE
PYL9	SL <mark>REV</mark> NVE	K <mark>SGLPA</mark> TT <mark>S</mark> I	ERLELLDE	EHILGIKII <mark>GG</mark> D <mark>I</mark>	IRLK <mark>NY</mark> S <mark>S</mark> ILT	VHPEIIE
PYL10	SV <mark>RE</mark> VDLE	K <mark>SGLPA</mark> TK <mark>S</mark> I	EV <mark>l</mark> eil <b>d</b> dn	EHILGIRIV <mark>GG</mark> DI	IRLK <mark>NY</mark> S <mark>S</mark> TIS	LHSETID
PYL11	SV <mark>RE</mark> VTVV	/ <mark>SGLPA</mark> EF <mark>S</mark> F	ERLDELDDE	SHVMMISII <mark>GG</mark> DI	<mark>irl</mark> v <mark>ny</mark> r <mark>s</mark> ktm	AFVAA.DT
PYL12	SV <mark>RE</mark> VTVV	/ <mark>SDLPA</mark> SF <mark>S</mark> I	ERLDELDDE	SH <mark>V</mark> MVISII <b>GG</b> DI	irl <mark>vny</mark> q <mark>s</mark> ktt	VFVAA.E
PYL13	SVRDVTLV	/ <mark>SGFPA</mark> DF <mark>S</mark> I	ERLEELDDE	SHVMVVSIIGGN	IRL V <mark>NY</mark> K <mark>S</mark> KTK	VVASPEDM

	β7	α	3
PYR1	T>	22222222222222	200000000000
		Recoil	187
PYR1	NRIWTVVL <mark>ES</mark> YV <mark>VD</mark> M	PEGNSEDDTRMFADTVVKL	VLQKLATVAEAMARNSGDGSG
PYL1	EEERIWTVVLESYVVDV	PEGNSEEDTRLFADTVIRL	NLQK <mark>L</mark> ASITEAMNRNNNNNNSSQ
PYL2	.SGKVYTVVL <mark>ES</mark> YT <mark>VD</mark> I	PEGNTEEDTKMFVDTVVKL	NLQKLGVAATSAPMHDDE
PYL3	KKKRVYSVVL <mark>ES</mark> YI <b>VD</b> I	PQGNTEEDTRMFVDTVVKS	NLQNLAVISTASPT
PYL4	GTVVVESYVVDV	<b>PPGNTKEETCDF</b> VDVIVRC	NLQS <mark>L</mark> AKIAENTAAESKKKMSL.
PYL5	GTVVVESYIVDV	PPGNTEEETLSFVDTIVRC	NLQSLARSTNRQ
PYL6	.DGKKRTRVVESYVVDV	PAGNDKEETCSFADTIVRC	NLQS <mark>L</mark> AKLAENTSKFS
PYL7	GRSGTMVMESFVVDV	PQGNTKDDTCYFVESLIKC	NLKSLACVSERLAAQDITNSIAT
PYL8	GRIGTLVIESFVVDV	<b>PEGNTKDETCYF</b> VEALIKC	ILKSLADISERLAVQDTTESRV.
PYL9	GRAGIMVIESFVVDV	PQGNTKDETCYFVEALIRC	NLKSLADVSERLASQDITQ
PYL10	GKTGTLAIESFVVDV	PEGNTKEETCFFVEALIQC	NS <mark>L</mark> ADVTERLQAESMEKKI
PYL11	EEKTVVV <mark>ES</mark> YV <mark>VD</mark> V	PEGNSEEETTSFADTIVGF	NLKSLAKLSERVAHLKL
PYL12	EEKTVVV <mark>ES</mark> YV <mark>VD</mark> V	PEGNTEEETTLFADTIVGC	NLRSLAKLSEKMMELT
PYL13	AKKTVVV <mark>ES</mark> YV <b>VD</b> V	PEGTSEEDTIFFVDNIIRY	NLTSLAKLTKKMMK

Figure S1. Aligned sequences of PYL members. Annotations of secondary structures are based on the closed conformation of PYR1 in 3K3K. Highly conserved positions are colored in red. Regions of interest are highlighted in yellow.



Figure S2. (A) Inter-subunit steric clashes in a homodimer structure modeled by replacing PYR1-open with PYR1-closed in the asymmetric homodimer. (B) The structure of closed form PYR1 homodimers bound with two ABA analogs, the recoil helix has been partially disrupted around Thr156.



Figure S3. RMSD distributions from the MD simulations. (A) RMSDs from the PYR1-closed reference structure (the closed subunit in PDB 3K3K); (B) RMSDs from the PYL10-o reference structure (PDB 3R6P); (C) RMSDs from the PYL10-r reference structure (the PYL-10 subunit in PDB 3RT0); (D) RMSDs from the PYR1-closed reference structure in the PYR1-HAB1 complex (the PYR1 subunit in PDB 3QN1); (E) RMSDs from the PYR1-open reference structure (the open subunit in PDB 3K3K). The legends indicate the simulated systems.



Figure S4. Comparisons of cross-correlations computed from different 100 ns simulations of the same system. (A) apo-PYR1-open; (B) apo-PYR1-closed; (C) ABA-PYR1-closed; (D) apo-PYR1-closed in apo-PYR1-HAB1; (E) apo-PYL10-o; (F) ABA-PYL10-o; (G) apo-PYL10-r; (H) apo-PYL10-r in apo-PYL10-HAB1.



Figure S5. Community structure of apo-PYL10-r.