1.15 Å Resolution Structure of the Proteasome Assembly Chaperone Nas2 PDZ Domain

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Supplementary figures

Fig. S1. The PDZ domain of Nas2 binds Rpt5.

(A) Topology of Nas2 and truncations used in this study. Predicted region of PDZ domain is indicated. Both truncations bind to the C-domain of Rpt5 ($Rpt5^{C}$) as determined by GST-pulldown assays using assays described previously (data not shown; Lee *et al.*, 2011). (B) Quantitative analysis of Nas2-Rpt5 Cdomain interaction using Bio-Layer Interferometry on the BLItz (ForteBio). The C-domain of Rpt5 (Lee *et al.*, 2011) was captured on Ni-NTA biosensor (ForteBio), and binding with increasing concentrations of analyte was measured using the BLItz. Upper panel shows base-line corrected data for the binding of Rpt5 C-domain with Nas2. Table shows quantitative analyses for Nas2 and the truncated versions Nas2ND and Nas2LND.



Figure S2. Top: Asymmetric unit of Nas2 LND colored by secondary structure: sheet (magenta) and helix (cyan). The disordered region is indicated by the dashed line. N-terminal residues resulting from cloning that could be modeled are colored blue. A molecule related by a 2-fold crystallographic axis (y, x –z) is colored green and its respective N-terminal residues resulting from cloning are colored red. View is normal to the crystallographic 2-fold axis. The distance between C α atoms of Gly 124 and Gln 129 is 14.4 Å in the current asymmetric unit. However, a comparable distance is found between C α atoms of Gly 124 from a molecule related by a crystallographic 2-fold axis and Gln 129 in the asymmetric unit is comparable 14.9 Å. Therefore it is not entirely clear if the N-terminal residues are connected to Gln 129 as modeled in the current asymmetric unit or as indicated by the red arrow. However, the orientation of Gly124 and Gln129 in the asymmetric unit appears to be sensible. **Bottom:** Same as the top figure but viewed along the crystallographic 2-fold axis.



Fig. S3. Structural detail of Nas2 PDZ domain.

(A) Binding region and Fo-Fc electron density map (green mesh) contoured at 3σ for the disordered PEG 400 fragment. Pro 185 and Ser 200 were modeled in alternate conformations as shown. (B) Fo-Fc electron density map (green mesh) contoured at 3σ for the modeled sulfate ion coordinated between Arg 119 and Arg 120.

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0001	Nas2LND	GPLTRRASV	QYTIPFA	FISEVVPGS	SPSD <mark>KA</mark> DII	KVD D KL I	SIGNVHA	ANHSKLQNI	OMV V MK N EDR	PLPVLLLREC	GOILKTSLTPS	RNWNGRGLLO	GCRIQEL
0002	3rleA		PGGGEG	HVLRVQENS	P GHR AGL I	EPF D F I V	SINSRLNI	KDNDTLI	K dl lk an ve k	PVKXLIYSSI	LELR E TSV TPS I	ILWGGQGLLC	G VSIRFC
0003	3id4A		EP	VLENVQPNS	SAASKAGL	DAGD RIV	KV DGQP L	rQwvt f	7MLVRDNPGK	SLALEIERQC	SPLSLTLIPE:	SKFV	GIEPK-
0004	3i1eB		G V	YVLSVKED	PAAG-IL	AGDLIT	EIDGQS-	FKSSQEF	D YIHSKV G D	rv k ikykhgi	KNEEASIKLT	AIDKGIC	GILEHH-
0005	3qdvA		Q G I	VVNEVSPD	PAANAGI	OVNDLII	SVDNKP	SALETI	MAQVAEIPGS	VIP V VVM RD I	KOLTLOVTIO	SYP	
	-												
			:	:	:		:		:	:	:	:	100
0001	Nas2LND	LLLLLLLL	LLLLLLE	EEEEELLLI	HHHHLLLI	LLLEEE	EELLLL	LLLLLLHHH	HHHHHLLLL	LEEEEEELI	JEEEEEEELLLI	LLLLLLL	EEEEEL
0002	3rleA		LLLEEE	EEEEELLLI	HHHHLLLI	LLLEEEE	EELEELLI	LLLLHH	HHHHHLLLL	LEEEEEELI	LEEEEEELLI	LLLLLLL	EEEEEE
0003													
3id4	A		-LLLEEE	ELLLHHHH	ILLLLLL	SEEEELL	LLLI	сниннини	HLLLLLEEE	SEEELLEEER	SEEELLEEE	ELLEEEI	
0004	3i1eB		LE	EEEEELLLI	HHHL-LLI	LLLEEL	EELLEE-	LLLHHHHH	HHHHHLLLL	EEEELEELI	EELLEEELEI	LLLELI	EEELL-
0005	3advA			REFERING	ннннттт	LLLEEE	EELLEE-	Т.ННННІ	нннннтллл	REFERENCE	REFERENCE	ST.T	
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Fig. S4. Dali analysis of Nas2LND.

Upper panel shows results from a Dali search to identify the closest structural homologs. Lower panel shows secondary structure assignments by DSSP (H/h: helix, E/e: strand, L/l: coil). The most frequent amino acid type is colored in each column.