

**Supplementary Figure S1** Secondary structure diagrams for P3 RNA domains of RNases MRP/P from various eukaryotes. The ACR triad corresponding to A37, C38, A39, which are an integral part of the Pop7 protein fold in *S. cerevisiae* RNase MRP, is shown in red. In *S. pombe*, the ACR triad appears to be replaced by a UCA triad (shown in green). It is not clear whether *S. kluyveri* RNase P possesses an unusually positioned ACA triad (red) or an alternative GCA triad (underlined in orange). The diagrams are based on [Tranguch & Engelke 1993, Ziehler et al. 2001, Li et al. 2002, Piccinelli et al. 2005, Lopez et al. 2009].

1 SS elements 2 Pop6_Sc SS 3 PNA inter	EO EEE	н0 ннннннннннннннн	E1 EEEEEEE	н1 ннннннннннн	E2 EEEEEEEE	н2 ннннннннннннннн	E3 EEEEEEEEEE EE	E4 SEEEEEEEE
<pre>A Dimerization Dop6_sc Pop6_sc Pop6_zr Pop6_cg Pop6_Kl Pop6_Kl Pop6_Xl Pop6_Hs(Rpp2!</pre>	4 GVYYNEISRD-LI 3 VIEYCNKETG-LG 4 RVOYEGINVE-II 5 KLKYNEIEVEGII 5)	DISSTOCLEDIKETVIP-SLAN LPNYSECLESIEDHVVP-DMLA LSNFSOCSEYIKNQILP-TMLG DSTYVECSTYIKEKIIPETLES	NG-NNSTSIQYHGIS GS-MMDSKISYRKVS EP-NTEFNYITTKVS AGWNNASETSVGAIISYS 30 VQE 40 VK	NDNIKKSVNKLDKQINMADRS: NDSIRRVVEKLQAESTP NDKIKDKYSSLEKNVKDNN NSNIKQCVDQLAAITAD GSKMRNLLGFAIGKME( GSK <mark>IRNLM</mark> AF <mark>A</mark> TASMA(	* LGLQQVVC <mark>IFSYG</mark> P ITC <mark>V</mark> FSYGP FVIIASYGD TFV <mark>I</mark> VAYGV GESVRQMLFVGSG <mark>R</mark> QPATRAIVFSGC <mark>G</mark> R	** ** ** HICKMLSILEIFKKGYIKNNK HICKMLSILEIYKKVIYKEDKT HICKMVTIVEILKKNFAKDTK HVQKALSILEIFKTTENGK ALGKTISCVEIMKRRLQ AT <mark>TKTVTCAEILKR</mark> RLQ	* * * * -KIYOWNKLTSPDIKREGRNELQEERL /KLHOWNKLNWETNKVETHNBLIHTRK -LHOWNRLSPEHIKPEKNBLLEVRT -ALHOFVKLDSTEIKAGRNELLDRKV -QLHOVTRIC-BRHTEETW-96 RNG -GLHOVTRLR-YRSVREVW-106 KNT	*** KVPILVTLVSD 144 RVPILVTITED 138 KVPIMVTALS 135 ULPILTLFTK 140 CPTICVLLSKD 124 VPGLAILLSKD 142
11 ALBA Sso10b S 12 ALBA Sso10b	SS	1	EEE MSSGTPTPSN <mark>VV</mark> L <mark>I</mark> <mark>G</mark> K	HHHHHHHHHHHH <mark>KPV</mark> MN <mark>YVL</mark> AALT-LLN	EEEEEEE HI -QGVSE <mark>IVIKA</mark> R <mark>C</mark> R	hhhhhhhhhhhhhh A <mark>ISKAVD</mark> TVEI <mark>VR</mark> NRF <mark>L</mark> PD	EEEEEEEEEEEEEEEEEEEEEEEEEEEEEEEEEEEEEE	SEEEEEEEEE SRV <mark>STIP</mark> IAIR <mark>K</mark> K 97
13 SS elements 14 Pop7_Sc SS 15 RNA inter 16 Dimerization	E0 EEE * ** * *	НО E1 НННННЕЕЕЕ ННННН **** ***** **	н1 нннннннннннннннн * *	E2 IHH EEEEEEE HHHHHH *** * ** * ** *	H2 HHHHHHHHH EI * * ** ***	E3 EEEEEEEEEE ** * *** * *	E4 EEEEEEEEE ** * * *	SEE
17 Pop7_Sc 18 Pop7_Zr 19 Pop7_Cg 20 Pop7_Kl 21 Pop7_Xl 22 Pop7_Hs (Rpp2(	14 RVTKHPSLKTI 4 LIKKHPTVKS 5 LVRKLPTVKT 10 LRRKLPTVKT 13 HTRR-PAP 0)22 LRKRLPS	LTHKQIHTTIFVKSTTYVSALK ISHKQVHSTLVLKSKTPVVSALK ISHKKIKSTMVVKSGTPFKSATK SHDKILTTIYVKSSPYISVVK -RPPRSPNDIVVNTKSD9RAQLA -RLPRRPNDIYVNTKSD9RAQLA	RINKFLDSVHK- RINKFLANVNR- RIDKLLN RTNRFLSNLHR- RCRQLVSSGDFREVRV- RCQKLLDGGARG	-QG-SSYAVLGMGKAVEKTL -TG-STYVIVLGMGKAVEKAL -QC-QRYVTVLGMGHSVTKTL -RK-SEYITLLGMGKAVFKTL -HGLGLAIGRAVNLAL -QNA <mark>G</mark> SE <mark>IYIHGLG</mark> LAIN <b>H</b> AI	ALGCHFODQK-NKK SLACHFODQL-QKR ALATTYOEKLHK SVAAHFNTMG-DYK 2LQLSFPGTLL NLALQLOAGS-FGS	IEVYTKTIEVLDEVITBOQAI VEILTKSVDWDELSNDD VQVMTKTIEVLDEVVEDT VDILTKTIEVLDEVFADD-G ISPSTSSVQLTDDLEPBG LQVAANTSTVELVDELEPBT	DIDMESDVEDDDK <mark>ETQLKKRAVSCVEL</mark> DDDPPDVDTELRKRTISCVEI ESIDDSDSDIETKLKKRLLSCVEV NDEDGNEVSTEDR <sup>®</sup> TELTKRTLSCVEV 	RIAV 140 RIAP 122 RIAR 121 RISP 135 RVAR 123 RVAR 123 RVAR 136
23 ALBA Sso10b S 24 ALBA Sso10b	SS 1 MSS	EEE HHHHHHH SGTPTPSNV <mark>V</mark> L <mark>I</mark> -GK <mark>K</mark> PVMNYVL	HHHHHHH AA <mark>L</mark> TLLN	EEEEEEE HHHHHHH -QG-VSE <mark>IVIKAR<mark>GRAISK</mark>AV</mark>	HHHHHHHHH TVEI <mark>VR</mark> NRF <mark>L</mark> PDK	EEEEEEEEEEEEE <mark>I</mark> E <mark>I</mark> KE- <mark>I</mark> R <mark>V</mark> G <mark>S</mark> QVVTSQDG	EEEEEEEEE Rosrv <mark>Stid</mark> i	EEEE A <mark>I</mark> R <mark>K</mark> K 97

**Supplementary Figure S2** A structure-guided sequence alignment for Pop6 (lines 1-10) and Pop7 (lines 13-22) versus Alba protein Sso10b (lines 11-12 and 23-24). The alignments are shown for *S. cerevisiae* (Sc, lines 5,17), *Zygosaccharomyces rouxii* (Zr, lines 6, 18), *Candida glabrata* (Cg, lines 7, 19), *Kluyveromyces lactis* (Kl, lines 8, 20), *Xenopus laevis* (Xl, lines 9, 21), and human (Hs, lines 10, 22) proteins. Lines 1,13- secondary structure elements according to the nomenclature from Figure 3; lines 2, 14 - secondary structures of Pop6, Pop7, respectively; lines 11,23- secondary structures of Alba Sso10b (PDB 1h0x). The residues conserved in Pop6 (lines 5-10), Pop7 (lines 17-22) and Alba proteins (line 24) are highlighted as follows: nonpolar aliphatic (GAPVLIM, yellow), aromatic (FYW, dark blue), polar uncharged (STCNQ, green), positively charged (KHR, light blue), negatively charged (DE, brown). The absolutely conserved glycine is highlighted in red. The shown conservation pattern for Alba is based on the alignment of Alba proteins with known structures (PDB ID 1h0x, 1y9x, 1nfj, 1nh9, 2h9u, 2z7c, 2bky, the alignment is not shown). Residues involved in interactions with the P3 domain RNA are marked by red asterisks in lines 3, 15; residues involved in the formation of the Pop6/Pop7 heterodimer are marked by black asterisks in lines 4, 16.



**Supplementary Figure S3** Conserved residues mapped to the surface of the Pop6/Pop7 heterodimer. The conservation is shown according to the alignment in Supplementary Figure S2. Non-conserved residues are represented in light blue (Pop6) and light yellow (Pop7); residues conserved in yeast (*S. cerevisiae, Z. rouxii, C. glabrata, and K. lactis*) are shown in blue (Pop6) and yellow (Pop7); residues conserved in all organisms in Supplementary Figure S2 (*S. cerevisiae, Z. rouxii, C. glabrata, K. lactis, X. laevis, and H. sapience*) are shown in dark blue (Pop6) and red (Pop7). The P3 domain RNA is shown in green.



**Supplementary Figure S4** The electrostatic potential of the surface of proteins **(A)** Alba 1hox, **(B)** Pop7, **(C)** Pop6. Positively charged areas are shown in blue, neutral- in white, negatively charged- in red. All proteins are shown in the same orientation.

## P3 domain upper strand loop

			65 66 69 69 68 71 71	
RNase	MRP	5′-	AG-U-AAUAU	-3′
RNase	Ρ	5′-		-3′

P3 domain lower strand loop

RNase	MRP	5′-	۱         ۱         ۱         ۱         1	3′
RNase	P	5′-	UUUACAGAA-G -3	3′

**Supplementary Figure S5** Sequence alignments for the upper and the lower strands of the loop region of the P3 domain RNA from *S. cerevisiae* RNase MRP and RNase P.



Supplementary Figure S6 A sample of a density modified SAD electron density map contoured at the  $1.5\sigma$  level.