

Figure S1:

А

A, Multiple sequence alignment of Cdc48/p97, Drg1, and Rix7/NVL over the D1 and D2 AAA-ATPase domains. The alignment was generated with ClustalW [76] and displayed with Jalview [77]. Secondary structure elements (α -helices and β -strands) are according to the crystal structure of p97 (pdb 3CF2) [20]. In the D2 domain, the positions of α -helix 8 and of the last α -helix are according to secondary structure prediction (PSIPRED) [78]. Numbering of α -helices and β -strands as well as nomenclature of sequence elements is according to [17]. Elements of the ATPase core are coloured in red (α -helices) and green (β -strands), the lid domain and its α -helices are depicted in light blue. The additional α -helix of classical clade AAA-ATPases and the post helix7 insertion are indicated in dark blue. Specific ATPase elements are labelled. The classical Walker A (K>A, defective nucleotide binding) and Walker B mutations (E>Q, impaired ATP hydrolysis) are also indicated [11]. Unique features of the classical clade are highlighted: (i) replacement of the generally conserved sensor-II arginine at the base of α -helix7 by an alanine; (ii) short insertion within the arginine finger region leading to the occurrence of two conserved arginines that are usually separated by proline and glycine. **B**, Representation of the secondary-structure distribution within each ATPase domain with the α/β domain depicted in green (β -strands) and red (α -helices) and the α -helical elements of the α -helical lid domain depicted in green (β -strands) and red (α -helice) and the post helix7 insertion are shown in dark blue.



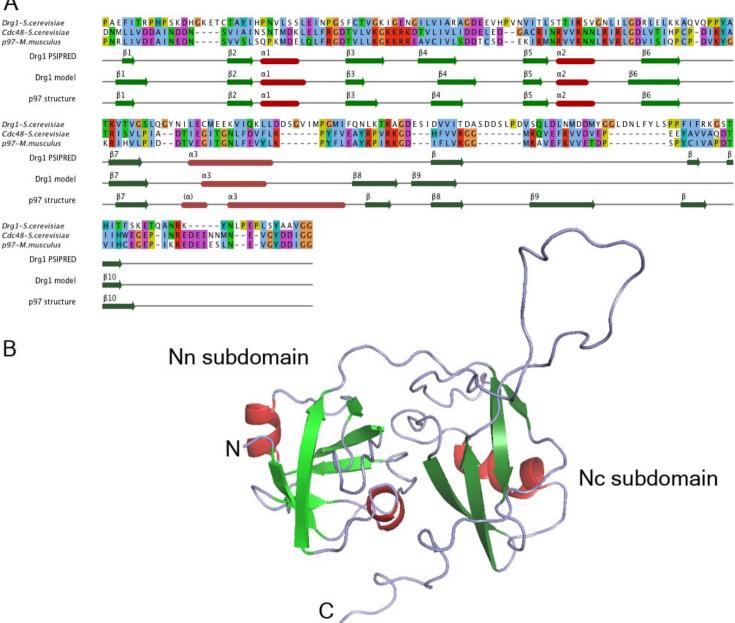


Figure S2:

A, The N-terminal domain of Drg1 is composed of two sub-domains: a double- ψ β barrel Nn-domain and a four-stranded β -barrel Nc-domain. A, Multiple sequence alignment of Drg1 (*S. cerevisiae*, amino acids 31-249), Cdc48 (*S. cerevisiae*, amino acids 33-218) and p97 (*M. musculus*, amino acids 23-208). The alignment was generated with ClustalW [76] and displayed with Jalview [77]. Secondary structure elements (α -helices in red and β -strands in green) are derived from secondary structure prediction (PSIPRED) [78], a Drg1 structure model, or crystal structures of p97 (pdb 3CF2, 1R7R, and 1E32) [20, 22, 23]. Numbering of α -helices and β -strands is according to [22]. Note that the alignment was refined in order to properly position the predicted β -strand 10 of Drg1. **B**, Structure model of the N-terminal domain of Drg1. The structure model was calculated, based on HHpred alignments [79], by the MODELLER software from a p97 reference structure (pdb 3CF2) [20]. The PyMOL program was used to display the structure [80]. As in A, the structural elements of the N-terminal are distinguished from the ones of the Nn-domain by the use of darker colours.

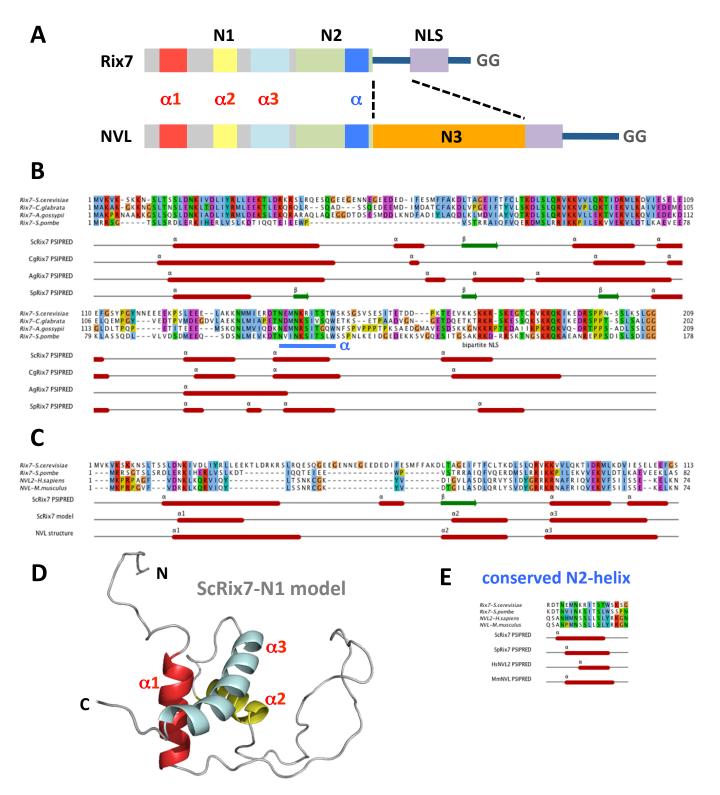


Figure S3:

Structural analysis of the N-terminal domain of Rix7. **A**, Schematic representation of the N-terminal domains of Rix7 and NVL. The distinct N-terminal regions are denoted by N1, N2 and N3 and they are followed by a bipartite nuclear localization signal (NLS). Relevant predicted α -helices are indicated. The two glycines (GG) indicate the start of the AAA-domain D1. **B**, Multiple sequence alignment of the N-terminal domains of fungal Rix7 proteins (*S. cerevisiae*, *C. glabrata*, *A. gossypii* and *S. pombe*). The alignment was generated with ClustalW [76] and displayed with Jalview [77]. Secondary structure elements (α -helices in red and β -strands in green) are derived from secondary structure prediction (PSIPRED) [78]. The conserved α -helix at the end of the N2 region is underlined in blue. **C**, Multiple sequence alignment of the N1 regions of fungal Rix7 (*S. cerevisiae* and *S. pombe*) and mammalian NVL (*M. musculus*). Secondary structure elements are derived from secondary structure prediction (PSIPRED) [78], a Rix7 (*S. cerevisiae* amino acids 1-113). The structure of mouse NVL (pdb 2RRE) [29]. **D**, Structure model of the N1 region of Rix7 (*S. cerevisiae* amino acids 1-113). The structure model was calculated, based on HHpred alignments [79], by the MODELLER software from a mouse NVL reference structure (pdb 2RRE) [29]. The PyMOL program was used to display the structure [80]. The colour of the α -helices is as in A. **E**, Multiple sequence alignment of the conserved α -helix at the end of the N2 region of fungal Rix7 (*S. cerevisiae* and *S. pombe*) and mammalian NVL (*H. sapiens* and *M. musculus*). The position of this α -helix (red) is derived from secondary structure prediction (PSIPRED) [78].

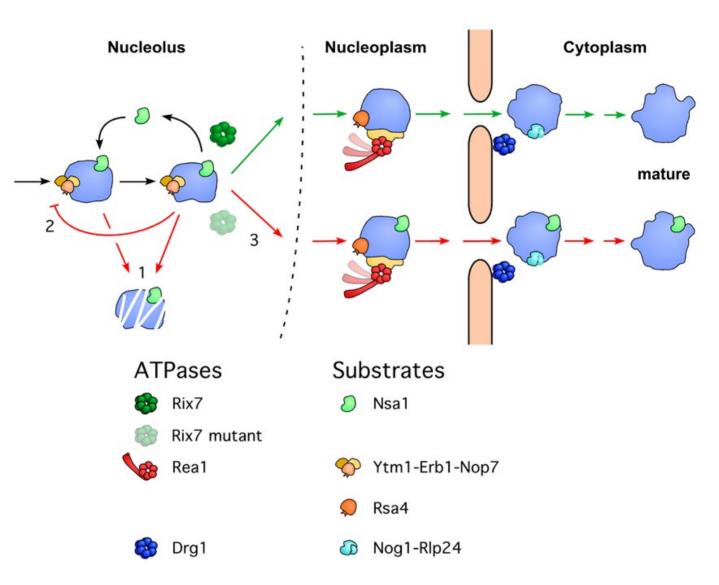


Figure S4:

Effects of mutational inactivation of Rix7 on the fate of Nsa1-containing pre-60S particles. In wild-type cells (RIX7, upper green pathway), Rix7 releases Nsa1 from late nucleolar pre-60S particles. Upon mutational inactivation of Rix7 (rix7, lower red pathway), the majority of pre-60S particles do not further evolve and are degraded (1), while a fraction of pre-60S particles gain an earlier composition and are retained in the nucleolus (2). Nsa1 remains associated with pre-60S particles that have escaped from disassembly or nucleolar retention, and accumulates over time on 'aberrant' cytoplasmic 60S subunits (3). [15, 19]. For simplicity, the Rea1-mediated release of the trimeric Ytm1-Erb1-Nop7 sub-complex has been omitted.

D1_C.glabrata VAVLKKLAKLIQKN-EPMMLVGKAGTGKTFLINELSKVMKTH D1_S.cerevisiae VSSLRQLGRKIQNS-TPIMLIGKAGSGKTFLINELSKVMCH D1_K.factis KLALQKLAEGIQRN-KPVMLVGKAGSGKTFLINELSKKMNCD D1_C.globosum VENLENLSSMLQQS-GPVLLHGLSGAGKTSLVHEISRELGKQ	DS I VK I HLGEQ TDAKLLIGTYTSGDK PGT SS VVK I HLGEQ TDAKLLLGTYTSGEK PGS KEMVTLHLNEQ TDAKMLLGLYTTDSK PGS	FEWRA GVLA TA V KEGR WVLIEDIDKA PTDVLSILLSILE FEWRS GVLTTA V KEGR WVLIEDINKA PTEVLSVLITLLE FOWR FGVLTTA V KEGR WVLIEDLDRA PTEVM STLLPLIE	
DI_S.pombe QENLNRITPYLVQK - RPILLAGPEGIGKKFLITQIAAKLG DI_M.musculus CKNLQLLAVSVASQ - NAVLLEGPIGS GKTSLVEHLAAVTGRTK DI_H.sapiens CKSLQTLAMAVASQ - NAVLLEGPIGC GKTSLVEYLAAVTGRTK	PQLLKVQLGDQTDSKMLLGMYRCTDVPGE PQLLKVQLGDQTDSKMLLGMYRCTDVPGE	FVWQPGTLTQAATKGYWILLEDIDYAPLDVVSVLIPLLE FVWQPGTLTQAATMGHWILLEDIDYAPLDVVSVLIPLLE	- HGELLIPGHODC KVAP TFQLFATRRLLSCGGSWYRPMN - NGELLIPGRODC
DZ_C.glabrata SLRLMEQIAMSIQNNEPLLEV GETGTGKTTVVQQVAKLHKT- DZ_S.cerevisiae SLRLMEQISVCIQNTEPVLLVGETGTGKTTVVQQLAKMLAKK- DZ_K.factis SLRLMEQIGVAVKMCEPTLLVGETGTGKTTVVQLAKMLAKK- DZ_C.globosum AKRLLEQISVAVKMCEPTLLVGETGTGKTTVQLAKLGK- DZ_S.pombe SLGLEQLAAGVQTNEPLLLVGETGTGKTTTIQLAGLLGQK- DZ_M.musculus SVLLEQLAVCVSQGEPVLLVGETGTGKTSTIQVLAHITGH- DZ_M.sapiens SVLLEQLAVCVSKGEPVLLVGETGTGKTSTIQVLAHITGH-	LTVINVSQTETCDLLGCKKPVNS-108-SFV LTVINVSQTESCDLLGCKKPVNS-107-SFV LVAVNLSQQSEAGDLLGGFKPVSS-116-GFA VAVNLSQQTESSDMLGCKKPINA-103-GFM LRVVNMNQQSTGSDMLGCKKPIDA-90-ALL	FNEVECS LVK TI A CEWLL DEVNLA TADTLE SISDLITE FNEVECS LVK AV MCEWLL DEVNLASADTLENIADLISES FAFVECKIVKAV MCOWVLL DE INLASEDTLENIADLISES FSFVECALVKAV SCHWVLL DE INLASLETLE FICULSSY A FVECALVKAV SCHWLL DE INLASLETLE EN COLLESC	DS RS ILL SEX GDA EPIK AH - PDFRIFACMN A TDVGK D - L DS RS ILL SEX GEA EKIKAH - PDFRIFACMN PATDVGK D - L PS LLL SET GEI ERIQAH - PNFRVFGAMN PATDVGK D - L ES - GILL SER GDI EPIV PH - KNFRIFGCMN PSTDVGK E - L SG - SLVL DR GDT EPIV RH - PDFRIFACMN PATDVGK K - L
D3_C.globosum ERNLENLVRATSTRRYPILIQCPTSACKTSMIEYLANTCNK D3_S.pombe QKNLENIARACSTRMFPILIQCPTSSCKTSMIEYVAKKTCHK D3_M.musculusKLNLRDIARVVSACTYPVLIQCETSVCKTSLIRWLAARSCNH D3_H.sapiens KLNLRDIVRVVSACTYPVLIQCETSVCKTSLIQWLAAATCNH	F V R I N NH E H TO L QE YL G T W V TO D T G K F V R I N NH E H TO L QE YL G T W V TO D	L S FK E GV L V E AL NK G YWI VL DE L NL A P TD VL E AL N R LL DD N L S FK E GV L V E AL HK G YWI VL DE L NL A P TD VL E AL N R LL DD N K F QE GV L VQ AM R G SWI VL DE L NL A P TD VL E AL N R LL DD N S S FR E GV L VE AL N K G YWI VL DE L NL A P TD VL E AL N R LL DD N L V FN E GV LI DAM RK G YWI VL DE L NL A P TD VL E AL N R LL DD N L V FK E GV LI DAM RK G YWI I L DE L NL A P TD VL E AL N R LL DD N	IR E L F I P E TQE VV H P H - P D F L F A TQN P P G I YGG R K I L S - IR E L L I P E TQE VV H P H - P D F M L F A TQN P A G I YGG R K V L S - IR E L L I P E TQE I V R P A - E N F C L F A TQN P P G L YGG R K V L S - IR E L L I P E TQV L V K P H - P E F M L F A TQN P P G L YGG R K V L S - IR E L L Y T E TQE VV R A H - P R F M L F A TQN P P G L YGG R K V L S - IR E L L Y T E TQE VV R A H - P R F M L F A TQN P P G L YGG R K V L S -
D4_C.glabrata LRKLVVLVSSAMKNKEPILLVGETGCGKTTVFQLLSKVLRNN- D4_S.cerevisiae MRRLSVLVSSCLKNKEPVLLVGETGCGKTTICQLLACFMGRE- D4_K.Martis MKRLAVLVNSCLKNNEPVLLVGETGCGKTTICQLSNYYKKE- D4_C.globosum MRRLYVLVKRAVKNNEPLLLVGETGCGKTTVCQLLAEFEKQE- D4_S.pombe MIRLFCLVWRCLLAKEPVLLVGDTGCGKTTVCQLLAEFEKQE- D4_M.musculus MRRLAVLVGRALEFGEPVLLVGDTGCGKTTVCQMFSALANQK- D4_M.musculus MRRLAVLVGRALEFGEPVLLVGDTGCGKTTICQVFAALANQK-	LYSVNCHLNMEISDFLUGLKPVKIIKL	FEWHOGPLVLAM EDSFFLLDEISLADDSVLEKLNSVLEVE	KC-LVLAEKUSPESKDNEVELLIAU-KHPKILAIMNPGGDFGKME-LS-
DS. C. alabarum V.MNAMPVI.PALOCT - V.PILLECNPCVCVTTLVTALAPACCPP	L T R I N L S E Q T D L V D L F G A D A P G E R S G E - L T R I N L S E Q T D L V D L F G T D A P G E R T G E - L T R I N L S D Q T D L MD L F G T D V P V E G A E A G N - L V R I N L S D Q T D L MD L F G S D V P V E G G E G G Q - L V R I N L S E Q T D I T D L F G A D L P V E G G R G G E	FLWHDAPFLRAMCKGEWYLLDEMNLASQSVLEGLNACLDHR FYWRDAPFLRAMQKGEWYLLDEMNLASQSVLEGLNACLDHR FYWRDAPFLEAMQKGEWYLLDEMNLASQSVLEGLNACLDHR FAWRDAPFLAAMRNGHWYLLDELNLASQSVLEGLNACLDHR FAWRDAPLLAALKACFDHR	С - Е А Y I P E L D I S F S C - H P N F L V F A A Q N P Q Y Q G C G K G L C - E A Y I P E L D K V F T R - H P N F L V F A A Q N P H Q Y Q G C K G L G - E V Y I A E L D Q V F K R - H P D F K L F A A Q N P H Q G G K K G L N - E A Y I P E L D K V F K R - H P N F R V F A A Q N P H Q G G K K G L D K V F K R - H P N F R V F A A Q N P C A Q K K G L - C - E I V Y P E L D K V F K R - H P N F R V F A A Q N P F R Q G G K K G L
D6_C.glabrata NFDAYESLIRCIKYNWPAIIVGPSKSGKTELINFIADVVGQK D6_S.cerevisiae NVAYESVLKAINNNWPLVLVGPSNSGKTELINFIADVVGQK D6_K.lactis NFNYYESVLCVKNWPLULVGPSNSGKTELIAFVAALAGKS- D6_C.globosum RLKEIESVNSIEQDLPCILVGPSGSGKSTLLAHVAALAGKS- D6_S.pombe QLPVLESVITCINKKWPCILVGDIATGKTCILRLAALAGKS- D6_Musculus SFQSLEPIMKCVQMNWWILVGDIATGKTCILRLAALAGK D6_M.sapiens SFQPLESIMKCVQMSWMVILVGPASVGKTSLVQLLAHLTGHT	T I E F S M S - D - V D S MD I L G G Y E QL D L 9 1 V K L V V F P L N A - D - V D A MD L I G G F E QA D P 8 7 P R I K E M A V N S - D - T D T MD L I G E Y E Q I D I - 9 1 G S L K I M A M N S - A - M D T T E L L G G F E Q V D I - 109 - G S	FEW FDGMLVKAVEE GHWLIID NANLCSPSVLDRINSLLET- FEWLDGVIVRAVET GAWLVLDNANLCSASVLDRLNSLLERP FEWFDGYLLKAVEE GHWFVLDNANLCSPAVLDRLNSLLEH- FEWDSMLVOALKSCOWLLMDNVNFCNPSVLDRLNALLEP-	D - G S L M I N E C S L T D G K P R V K P H R D F R L F L T M D F K F G L S - N - G I L S I N E H S G P G G E P R I I T P H P D F R I F L T V D P R Y G E L S - K - G V L I V N E K T T E D G H P K T I K P H P N F R L F L T V N P V Y G E L S - G G V L T I N E R G M V D G T C T V T P N P N F R L F L S M D I H G E I S -
Zndary structure 0 B1 01	β2 α2	Walker B β α2 β3 α3	Pre-Sensor-I β β β4
D1_C.glabrata INMIG <mark>K</mark> SAWNIV <mark>S</mark> LEEPSD KDLEEILSKKFSLLTRLIP- D1_S.cerevisiae YNLNMIGMRIWNVIELEEPSE EDLTHILAQKFPILTNLIP- D1_K.lactis S DLIGMRLWEKILVDEPSE DDLKLILQKRFSLLTALIP- D1_C.globosum NLIGLRWHLLHYKIPR DDLKEVITGKYPLLHKYTP- D1_S.pombe KILGQRLWQILDLTYQPD ECVEVVSTLPVLSIICP-		SIYMNTKFISLNK GAHTRVY SV EDLIKL CERLDILFKNN DIYRN PQFVSLNK GVQ PRIISV EDLIK LSQRITELFEAN ACTS GSTRISI GTAIDED V STRDIKK SPIDILFAA	CINKPDQLIQSSYDSIFSEAADCFACAICEFKALEPIIQAICESLD QVVK DQLISSIYDDIFAEAVDCFVCSISEYCAVQPLVQKICEELE CKTCDCFIDTDRMEIEAVDCFVCSMHEDCAVLIVAALAKEMH
D1_H.sapiens S-HATLLDKYWTKIHLDNLDKRELNEVLQSRYPSLLAVVD		– – DL F SQR S F L A T S K – – I Y R R L C L R D F Y K F I K R V A F L Y H K F A <mark>P</mark> Q E V S E A E R E N R R V V L E <mark>G R E L <mark>S</mark> L R D L L N WC N R V A H <mark>G</mark> F D <mark>P</mark> T A <mark>P</mark> E E V S E A R R E N K R <mark>P</mark> T L E <mark>G R E L S L R D L L N WC N R I</mark> A H S F D S S</mark>	MIPSOHVVISQELQDAVFKEAIDMFGAFIPSRDGFDLVVRNVAIELN SSTALLHIFQEANDCFTAMLSEQTKKLRMAEVIGSRLN SLSASLNIFQEALDCFTAMLSEHTSKLKMAEVIGSKLN
D1_M.musculus S - HATMLDK YWTK IHLHN NK KDLNEVLQSKYP SLLAATD D1_H.sapiens S - HATLDK YWTK IHLN NK RELNEVLQS RYP SLLAVD D2_C.glabrata PSGI RSR FTE IYVHS PDRDISDLLAI DKYIGKYSV D2_S.cerevisiae PMGI RSR FTE IYVHS PDRDISDLLS I DKYIGKYSV D2_K.factis PPGI RSR FTE IYVHS PDRDISDLLS I DKYIGKYSV D2_C.globosum PLGL RSR FTE IYVHS PDRDISDLLS I DKYIGKYSV D2_S.pombe EPSF RSR FTE IYVHS PDRDK DLLS I I QKYIGSLCI D2_M.musculus PPGI RNR FTE LYVE ELE - SKEDLQILI VDYLKGLNV D2_M.sapiens PPGI RNR FTE LYVE ELE - SKEDLQVLI VDYLKGLSV		– – DL F SQR S F L A T S K – – I Y R R L C L R D F Y K F I K R V A F L Y H K F A <mark>P</mark> Q E V S E A E R E N R R V V L E <mark>G R E L <mark>S</mark> L R D L L N WC N R V A H <mark>G</mark> F D <mark>P</mark> T A <mark>P</mark> E E V S E A R R E N K R <mark>P</mark> T L E <mark>G R E L S L R D L L N WC N R I</mark> A H S F D S S</mark>	MIPSOHVVISQELQDAVFKEAIDMFGAFIPSRDGFDLVVRNVAIELN SSTALLHIFQEANDCFTAMLSEQTKKLRMAEVIGSRLN SLSASLNIFQEALDCFTAMLSEHTSKLKMAEVIGSKLN
D1_H.sapiens S-HATLLDKYWTKIHLDNLDKRELNEVLQSRYPSLLAVVD		- DLF SQRS FLATSK I YRRLCL KOFYK FIK KVAFLYHK F A QE VS EAR FEN RKVIEG RELSL ROLL NWCNR VAHG FOR T REEVS EARRENKR PTLE GRELSL ROLL NWCNR I AHS FDS S 	MIESOHVVISQELQDAV FKE A I DME GAF I ESRDGFDLVVRNVA I ELN
D1_H.sapiens S - HATLLDK YWTKIHLDNLDK RELNEVLQS RYPSLLAVVD- D2_C.glabrata		- DLFSQRSFLATSK I YRRLCL KOFYK FIKRVAFLYHKF ARQEVSEARRENKR PTLEGRELSL ROLLNWCNRIAHSFDS KLSENNVIVDGSNQRPHFSIRTLTRTLLYVCDIVHVY 	MIESOHVVISQELQDAVFKE AIDMEGAFIESRDGFDLVVRNVAIELN
D1_M.sapiens S - HATLLDK YWTKIHLDNLDK - RELNEVLQSRYPSLLAVVD- D2_C.glabrata		- DLF SQRS FLATSK I YRRLCL KOFYK FIK KVAFLYHKF A QE VS EAR RENRK PTLE GREL SL ROLL NWC NRIAHS FD FT FE E VS EARRENKR PTLE GREL SL ROLL NWC NRIAHS FD S 	MIESOHVVISQELQDAVEKEAIDMEGAFIESKDGFDLVVRNVAIELN
D1_M.sapiens S - HATLLDK YWTKIHLDNLDK - RELNEVLQSRYPSLLAVVD- D2_C.glabrata		- DLF SQRS FLAT SK I YR RLCL KOFYK FIK NAFLYHK F P C E V SE AR REN KR M T LE G RE L SL ROLL NWC NR I AHS FD S 	MI B SOHVVI SQELQDAV KK E ALDME GAF L B SRDGF DLVVRNVA LELN

Figure S5:

Multiple sequence alignment of the Rea1 ATPase domains was done with ClustalW [76] and displayed with Jalview [77]. Secondary structure elements are depicted below (see also Figure S6). Elements of the ATPase core are coloured in red (α-helices) and green (β-strands), the lid domain is depicted in light blue and clade-specific insertions are indicated in dark blue. Specific ATPase elements are labelled.

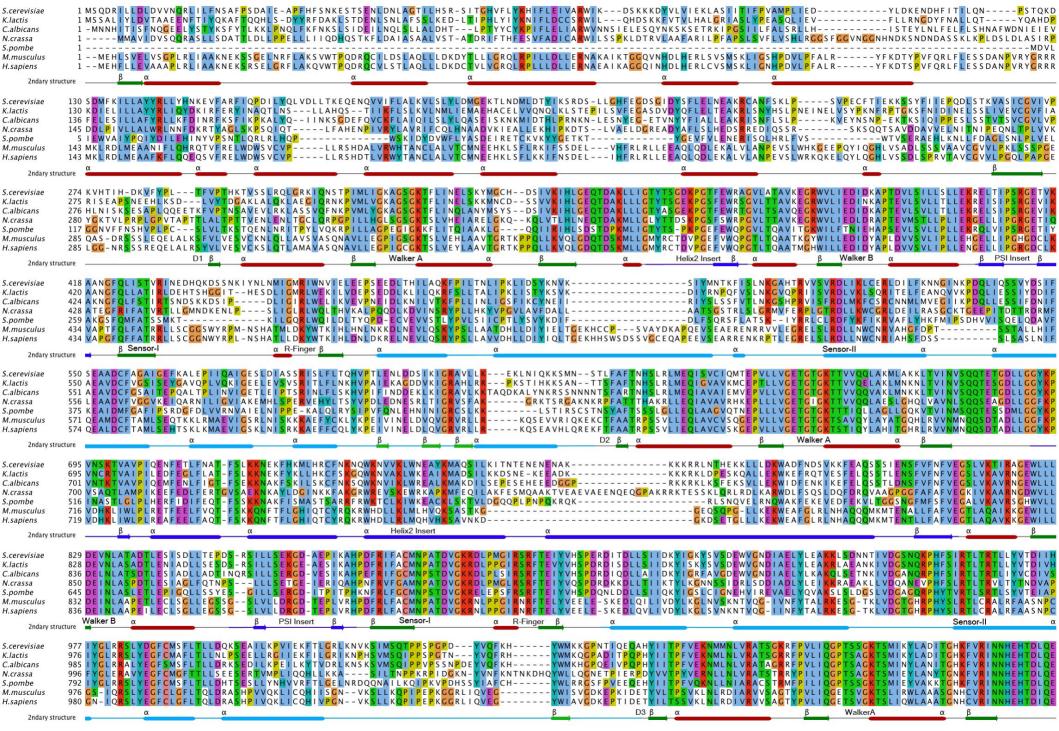


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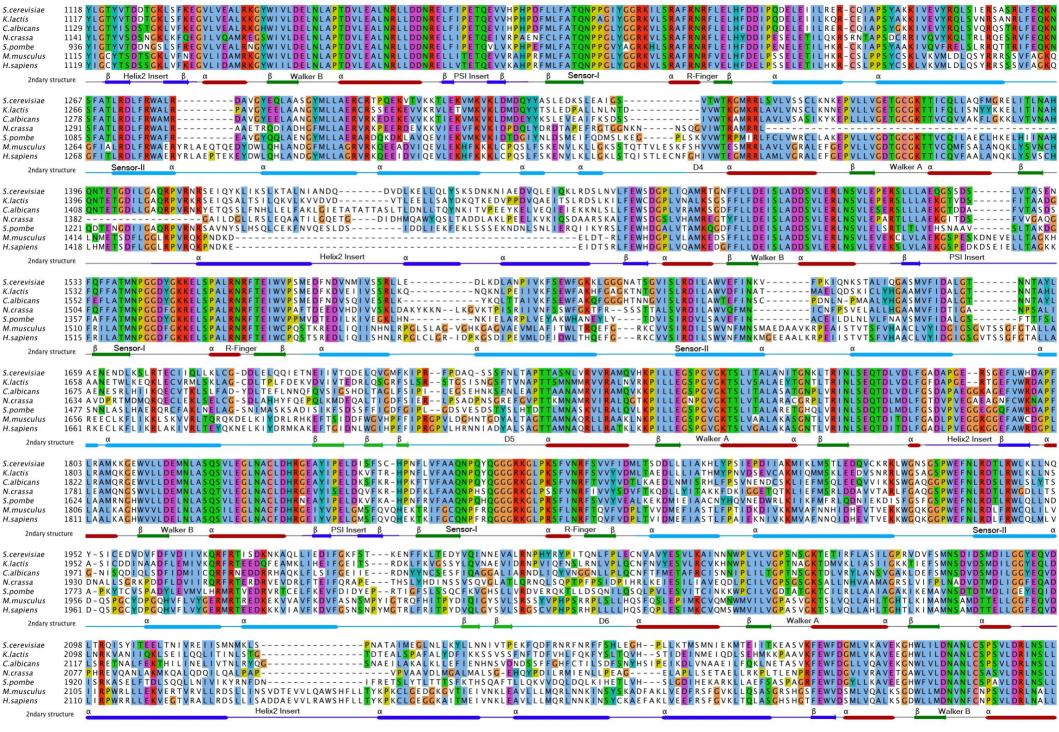


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S.cerevisiae K.lactis C.albicans N.crassa S.pombe M.musculus H.sapiens 2ndary stru	2201 E R P N G SL S I N E H S 2052 E H - K G V L I V N E K 2255 E P - G G V L T I N E R C 2260 E P - G G V L T I S E R 8	QEDGQPRVLKPHPNFR LTDGKPRVVKPHRDFR LEDGQPRVLKPHANFR GPGGEPRILTPHPDFR TEDGHPKTIKPHPNFR MVDGSTCTVTPNPNFR MIDGSTPTITPNPNFR βlinsert β β	LFLTMDPKYGELSRAM LFLTMDPKFGELSRAM LFLTVDPKYGELSRAM LFLTVDPRYGELSRAM LFLTVNPYYGELSRAM LFLSMDPIHGEISRAM LFLSMDPYHGDISRAM Sensor-I α	RNRS VEIYLEG RNRGVEIFLLK RNRGLEIYISGEEDC RNRGLEIYISGEGDA	S I QDNLDLKVLLHS I S T PDNLDLKVLLHS I	L G L V G D S V C N I L L A L G L V G N S V C D I L L A	LHTEIQSLVRQSPASS LHTETRSTVVQSPTSS	S V S T L S H T A I L I V Q Y S V S T L I Q T A I L I V Q Y	L Q R <mark>G</mark> L S L H K A F F E L Q R <mark>G</mark> L S L D R A F S E	ACWE VY VC S QHS ACWE VY VC S QHS	A A N Q K L V P A N R K L V
S.pombe M.musculus	2285	L P VRASTODRKILGIE LEQRATEFDRQILG A PWEAKNAG - HQTIAP EALTEIDKKQMSLLEP RETWGHSTLATGLWPD	L G E N I D F VS I D D G I K K C S S Q VG T E S E VE A R L R F S P C T T D E S L Q I K VDGS L Q R F H T A T R I L N A P I S S A VD T L A S N I S Y S V P S A L F A T E D S R L S V S V P S A L F A T E D S H L S T	SLQLDATSMSKIPLS IEELDISDKRVVPTS GA-ENADVLAPLAFE IKYVFETMGKIEIDC VRSEGOILAYCLNRM	5 A F I S P R L S K L Q S L V/ 5 S F N L D K E S A I K S F S I D V L S L G D T R L L E A F M(N Y M Y I A H A I I L A L F S M S L K T S S W T R S O P L T	A L HDMS L L G E V S P Y L I DDA F <mark>S L G N I S</mark> QA S T N G L I K S S P R Q L K L L R K V L L T I L OD L E N I MOT C S P D	L M S T I N M N A I Y L L N A S A L W S F V Q L K N F P L I 	– – DWN RN V RN S QL F D L K DWA S V V F S S S E F A – – – – – – – – – – – – – – – – – – –	E – – V D F C D A V T L M Q T S K S Y T E K L C D T – – – <mark>P</mark> A A S Q R L A Q L S L S I K A D A E L L L A T N O D W M L R V K W L	TNFITESGVFSE LKYIEQTSLVDI LSYISAEDAGAL LKNLVQKIYCAD CHLAKNIPOGLE	VEAIYSP SSSVYFP RQAVASL YFNHMDL SVRVHLE
K.lactis C.albicans N.crassa S.pombe M.musculus	2417 LNR I VONTL SNG 2437 VYMI ANN I VOKKI 2355 YG SAPNKMLMP - 2218 KASR FMDI Y EYP 2554 ASATAL RKFYSTS 2559 ASAASLRNFYSHS	KEFCYSQSLIPTLNSYI DFIESQNFHFLANVYI LHPLLNPPM /QLREVVGLIQTINDFQ ;LSAGVSNVLKILQTNI	L P T V S K Y S D K I K S P E S A T V K QN S P L S L T S E S V A S I L Q N H P T V T T S E S I I P L E Q D R - E G L A A W I S V I L T S H L E L P E T Y A S T D F V I P L D P RWNMQ A T D E F V I P L D P RWNMQ A	VYFFSLIWYVLEAKC TMIFEVLKEIVELNL AGCYELYLDIQSAQS GLLFVSAHEILDLTE LDIIRN <mark>S</mark> LDFD <mark>P</mark> QSE	SKL DF AQKKA VNGKI TVS KIESNAMDKKV SAMEAQLGKVNVSKP EVNRLAVSTSN QQAKQLFALLESVAN	N E L S Y L E L <mark>S</mark> T A V A F H E L S F I E K S A A <mark>S T</mark> L S Q L N R L Q R <mark>S</mark> W I A D K – S <mark>T</mark> Y L L K S A S A V Y K T F I Y L D R E K R I F T	G R N V K N V P H I E V Y K L I G R Q I K S P P K L E L F R F V A S L S R D S T V N A A R F I N V S <mark>S</mark> F K G S T P S L W N L I E A N L V S V G S K K L R S <mark>S</mark>	L K S L I E F A L H S I K N – V K S L Q <mark>G</mark> F I I S C F L N S L A S M L K L F K A Y M S E T L N Q F S K F L I E I A S A N V L R M S F E F H K D <mark>P</mark> E N Y	– I <mark>P</mark> L F S V – E KMYN F K T D L F T – G V I Y C I N E <mark>S</mark> R AW – K E R R A S N I VYK L S Y D V I R H S <mark>P P H E I V A N L A</mark> A	SLFQLYILWIGA PLHELLLILIGN ILRRLMLFWTRT HFLKLVVLWKNI FFELIDGLILLW	Y SC S KDR II K S G HE K F E A L I V P V V W T N V P V V Q S P Q G V
K.lactis C.albicans N.crassa S.pombe M.musculus	2564 DETKLRVYC 2586 NESKLKVYC 2494 NAKFELAR FOAH 2364DCDISKFYCYT 2704 VPDAHINKILCS 2709 VSDASANEILGSL	2 E L F T KWA S – M S C S I L – 2 E L I V KW <mark>S</mark> E R Y L <mark>S G</mark> H D R . T Q G S N L L R K A L S V L Q E . KM L G EWM F T L T E K T K – / <mark>Q W</mark> R D R F W T V A D <mark>T</mark> V T V D	P = Ε I T P F L N I N E D L DQF QQA I A - DDR KMLA T I L E YLE L L E S F L P K DS L AS G L A L L A L HWHW VS K A P G L A L L A L HWHW VS K	DE I AQPAVL KTGNS - S I FNSQFELNKGVA - RDFVVGFKLSTGLS - EKFSELQNLSTGLH - HLIYQIPQLLVNQEN	MDMIWEEVRI INEIWEYCR MEALWNQLR MQAIWDKWH NKYYKELQTV <mark>SE</mark> HIH	R	Q F E S L <mark>S G</mark> D F D QM V K K (R L F K I I E N F D T V S K L (E M E R L A D R F D S L RW K / T V D K L L T Q Y V N A N I <mark>P</mark> K L Q A F L G R P F P F K D K I	QF F D S Y P L I S QL S E T QF P E S D I V T S F G Y S A DA S – I P T L R S V QE A S I S ME T T A C E V V G T S L V V D C F L QL QA L N R A	F N A L F M D I L N - - N I A D L Y K S I I S G D Y MA R A Y T V V R S - - - L S L L N K V L V E N - - L A I G E Q L <mark>P V F G</mark> E C	INTAESENILEKL QRDELSSILEKL GRGDASALVREL EVGDIYSYLKIL GWQEDINRLQV	. K E <mark>G I S T L</mark> E Q G I D Q L E S E I S T L G K G V N E L / A S E W N L K
S.pombe M.musculus H.sapiens	2498 KSSKQVILPENL 2854 KSLLQAWGLILRA 2859 KSLLQAWGLILRA	NILEDVNLDELKNLVS	T Q C S E L K S R G L S R G F L IA Q C L E L K A K G L S L G F L	E KAH – DASS PSQPDL	A S L D L L H I F I K Y T T S S L VH L T R R V Q L W P	S	V C F H S R I S <mark>G</mark> N L – – – – – A D F I A QA C L R R S S K H I	D – – QMDEEISHHIAF	CLKHT <mark>P</mark> IA <mark>P</mark> QKLW	/NLWSLLHIEMLS	ILLHGISF AQEVSCL
K.lactis C.albicans N.crassa S.pombe M.musculus H.sapiens	2758 F DV LWT E KNAKLT 2758 F S L LWKKENG K F V 2783 F D LWKKENG K F V 2697 S P LQ VDCLLAQE 2565 D S TKAVAP VLTYF 3001 WS E LF TS TF G S S V 3009 WS E LF NSMFMS F V	/ SHTKAVF S	RKSNNLKSFSGNQIKQ IKSNDFYEFSGGQIEQ TKAVQFNNTSGKSLEQ LKYDATSSASMHELHS AFTSIDENANLESANI LPNIQQEVPRSRWDS LPGMQQREAPKSVLDS α	T L R D G R V L T E S I I K S T S F D L K S V S K S V I E F L E T <mark>E L P</mark> I M G K I L A K S F E H A R L A L L Q H F M D F T L K G P G S L C R A V F S K	5	I L K DWVL K I I A V H L L L I <mark>S</mark> WF <mark>G - I I L K A L L L L K L ME E I MAA H G I L L Q R Y A V I I S I F L WD V <mark>S G L P L L S</mark> S S H V</mark>	K V S S	L D L D E L N E M <mark>G</mark> N V Q V A E L A D L I T K L S L <mark>P</mark> Y E E L A E S S L L W T N MA V <mark>P</mark> S V A E	Y S L S I K E L E A E VA L S H P E L VL D K V S L A L L E Q G T N F A I D N F V S I L E A C K A F L F R R T D S R L Q G L V L	T S E C	AHQRQEY
C.albicans N.crassa S.pombe M.musculus H.sapiens	2867		ETRVTFAEYIFPALDL. WFKPVFAKYLGASLFL DSHSFVAVKLLVDSVNM. HLVVIFERHFYKALRA FISYTYTERFIHSLRF ALPKELLCLLLTSLHY VLPKELLCQLLTSLHH	I D S S R S L C A V V N A S E L S L C L A A A E K G L Q P R S L K D S W L S S N Q Q K M L K F I G E G E G K Q N L P E A A	2 N L G K A W V L F A V G L L C E L G K A W V S F S L G L I A Y T S V A W V Q F S L A S I K N Q G M A Y I Y F A S G M L A R R G R L W V S L G L L Q I C	Q L Y V P S S P Y D P A I R L L Y V P D S P Y D P A I K K L F I P D K I F D P H H R L V Y V P D K P F D P A L L Q T W L P R A R F D P A V K	DYVIYDNYVAHKSLT EYVLYKVFEDRKVFG AEIEQEEHREIYESL PLLTVESLRHYLESL KAYKLRYAQDELRQL	ESIIHNWKCAGSVLF GNLIS <mark>S</mark> WE <mark>S</mark> ARRVFS QQQIAALEAFELAFT YKE <mark>S</mark> QILEIAESLNS QCELKTRNLLSHLQT	G	VDSYTQKLVPEV KEMYLESTLKHL HAG-LRCKLLME VNSVMRRLVSTE HPHIKLLPQRIS	/ S S D D - I D L E E E VA <mark>G L GE</mark> - I S N T <mark>P</mark> N I D D
K.lactis C.albicans N.crassa S.pombe M.musculus	2968 A P QS P R VY R T G MS 2971 E P K K P R VF R P NH 2994 A P NN P K VY R D S K 2936 P P A V Q S I Y R P G D 2805 D S S F S T VY R S L N B 3301 S L S K K QAF R P H L F 3309 H L L K K QAF R P QL F	- I DS L F DEWSAFML S - I DG L F DEWNSIMES E L RG L QGE F NNVL NAL S I V PL Y SEL E F F MKSV S Y E T L V OE I H H Y V T SI	MSSRQIKELVSSYKCN VDVTPVISLLESSTEY IDMCPVESLLKSVFTS VQNDVAASHLRSISTD VLNQYIFELAMRLSKE AKVTAVQDLLMRLQA AKAPAVQDLLTRLLQA		S D S R F KMF E QNT S Q F S VNMMEMF QNNT S Q F A S E E L S L VE RNL VA L Y V V E E A K S F V T KWK A Y L L K E E A S WOO S HH O F	L S RL S S T Y E V Y S D I I MRL K KNYL L Y S D L V G R F I G R F NA Y Q D L I E R I R E A Y P Q F V D V R K RL V E E Y A L Y P D T	NDIFAGYVYAMKFGFI DDILCGYIYGMKLGFI TMPLVSFLRCLQLGL YELILSFISFMIYGII VAPLLASILOLOHGM	NV I S L GAENESQELD EMITID – KNQG KYL S L G QGA V P – A K S S D E E L L MF EA K R R L D E R S R L VA S E V HA S L H S G V	ISPLFTVDCLAVT YPQTWSIDVCDFT TDFKSMVKATPFL QILSTLILTLVDP LCAENLGALAMAV	R E K S I L S H F S R L S L K S V E R V F P S V S G T V W D A E A K S S F A R S L S F D D V (L A F P S V G P S F P T	N K F L K N Q / Q D F A K T V - S L <mark>P</mark> L R S L / S N L I E Q I T Y H A H A D A

	K.lactis C.albicans N.crassa S.pombe M.musculus	3109 N 3130 S 3072 E 2945 K 3451 L 3459 L	MEDTS I E KVLMY VGSHDVEEILIH I DSSAAE FVMAY FLSFVQAIVAVE VLDLNDSIRFEI CSVNAEALRSLGKLMVKRAARKE CSVKSEEVLRGLGKLILKRSGGKEI α		C Y A H K D S E S G F A H R S S E N E Q Q Q K Q L P E N A S R L C S E K Q - H S S D T L L Y L R S H V L C K G E L D	M I D S L N E I L Q P L Y 2QQTQS I L I QS L Q T L Y L R QA VH E S F Y A F F 7HS L A N S F V L L A N E F Y 2QRALLLF R H L C Q E I I	Y RWS L R RMKS E QE A E Q Y RWS L R R I KE E KE N L Q HE EWS KK L E A D R KA E E Q I H N A K I K Q K E L E E I E E N EWDE QE R I A QE KA E Q	K N S L F K F N D N S D D QGS L Y K Y N D P D E D K N S L F R F K G S L E D E E A K N R L Y R Q R E F N F D E S S L Y R Y R S R S G R T A L	- I EAEFKAMFPDYEDS - I EGDFKQLFPDYED FDQEEFDQLFPDYDD KNDYLKVFINYDD SEEEEELELRKLFP	VSLDSPVEESSA (LDIDGTTNANTL DEEQDADAPKKK VEPEVEPEVVIE HEKDFADILMEP	T – – – – S L I D T Y Y E I A K KKNNAS FES I Y ES I A S KQI <mark>P R G</mark> NRDLS I L L A E R K – – R F L Q L Q F A F W S T L E E K K K V S D G R E E A
	S.cerevisiae K.lactis C.albicans N.crassa S.pombe M.musculus H.sapiens	3213 T 3216 I 3249 D 3182 A 3059 L 3600 A 3609 G	Y I S VF DK – DHDANFSSELKSGAII Y I E I F ED – ASNVSLSQVMRKGSE Y I KEYLL – QEKDSVI DI VHQGSEL HEKIFLSPQEPQQSI RILLTQVARI YNEI YSEKMNVI PLEQLMNTGSYL P DPTLLC – QSSMQAVMLI HQQLCLI TNPALLS – QNSMQAVMLI HQQLCLI α	TTILSEDLKNTRIEELKSG SVILLRSCSDFKLYNLNES ITHLNQYENNLYKISNNAS RIARAERNAAAGEPEMDAL AKKIKVKNPDMIASSGFDI SFARSLWYQQAVPPHEAKH NFARSLWYQQTLPPHEAKH	SLSAVINTLDAETQS MVLAAIKTLSEKTNS MITSLVNELANTHTK LLPATILAFEDQIKA VSVVLMMGVKSTNER YLSLFLSCYQTGASL YLSLFLSCYQTGASL	F K N T E V F G F F K D S S N T E F F N QA V S F Q Y W T P D	QLL QS QLLACTLS SNTL RLL QS QLLACTLS HNTL		DFYHDFSIPEFOKAG FYRCTSRIETKAQH NFYQDSNPTEVKRAVT NFYDSILVEVRKALT NFYSDPNPSKAIEVRC DFYQHPNVAEARQCOF DFYQHPNVPEARQCOF A	VI QG LWN S VI VI QG LWN N VI MTT VYN L SK LVTAL KKRFQEL DLIKIVES RAI VLQGFSE - AVS VLQGFSE - AVS	K L L K QWP E HAT L K E L Y TL L K EWP E HAT L K D L F K L L DOWP E HAT L K N I V QT I E E I G H QT L A D V I S L I K NWP E N F V L R G L K Q L Q DWP E H P L Q L L H L L Q DWP E H P A L E Q L L α
Schward 396 B	K.lactis C.albicans N.crassa S.pombe M.musculus H.sapiens	3326 R 3357 F 3295 I 3168 D 3747 V 3756 V	VC K E F I – S Y P V S T PMA R L L Q K I E Q A T NE F L – S Y P I G L P L G R Y L Q K L E Q A C D K V L – D M S I D D P L A A I I S P ME K I A I D A I L – N L S P F S P I A E Y L S K L E R Y	I Y T F V V EWQ K Y A S S K V S I Y T F I A EWQ K Y A S S Q V S L Y A H V Y EWH E G G WA T K A H K V F H L L S EWE K L A S R E Y S	LQAHVDSITNLIVSW LEDQFVVLTNLIVSW ATVLYEKLRDTICDW LANEMDLIKKKIIDW	/ R R L E L H T W K A L F QME / R K L E L S T W K S L F E F E / R R L E L S <mark>S</mark> W S R L L D A E / R K F E L S N W N N L L K L E	DMN <mark>ST</mark> KTIGNWWFYLF EKSVEKNIGKWWFYLL AKKCHDDAKSWWFIAY EYKLSERVYPRLYSIL	ETIIVGTENANIDVSE EIIIIPMLGDNEKEPA GAVILQPCSWLQEGKD OFIILKPFFENSKFTK	RSGSLLSALN VTLLSALN LYAYAVELLGALE ONLCESASIIV	YFF <mark>SKSTLGE</mark> FAN YFMSKVTCGEFTI YFNTASLGQFTS YFITDLTVGEFOL	RLRLVKAFCKHVSVMH RLKLLKAFKNHAFEIN RLNLLRQLRNQLELLM CLKCLLSFSOHAASLR
Spentify 3434	K.lactis C.albicans N.crassa S.pombe M.musculus H.sapiens	3468 G 3497 - 3442 V 3311 - 3893 Q 3902 Q	KTSIISQALQNIISFYEQFTS KTHPIVDALCNFVTFYEQFQP DEPQLSVIRDAVQNFITYHARYER ICHGIDAMLLNIYHYFEQFLSS VE-GKDSLCSVLWNLYHFYKQFLD	V V N D K I K T <mark>G K</mark> K L L E KE I S E T V L S T I E D S K K R L E KD I N E K V K E A I A A G R T P L D R K M K D K V S E A I H T Q K Q S L E N S I K E P V K A K I M E L R S P I E K E L K E	VILLA <mark>SWKDVNID</mark> AL VILLA <mark>SWKDVNID</mark> AL VLLLSKWRDKNIEAL RILLM <mark>SWKDTN</mark> VYAL FVKISKWNDVSFW <mark>S</mark> I	K QS S R R S HN S L F K I V K QS A R R S HN N L Y K I V R A S A R K S HQK L F K L V K E S A K K S HA E L F K V L K QS V E K TH R T L F K F M	V R K Y R T L L S E P L T S M I E V R K Y R A I L S T P V Q P I I E V R K F R G V L S Q P V S G I T E I R Y R E V L R Q P V S S Y L S M K K F E A V L S E P C Q S C L V	S G I D S A L P V QG I S E S T NGM T S S S S QG L P E E E H K S L V A Q K H DWD S E S D K E E Q P D C L P K P T E	TKQFERVTISHFRSIC SSSALINLPQIQII IVDKAAVQADQEA LLDTENNSAMWVAKK EATMETSPIQGLNNAL	V S <mark>G A </mark> K S I V T N I P A <mark>G</mark> N E R Q I V Q S I P A L V L Q L I P S L E S Y N L S P S Y I E K M D T R E T L L A Q P V E W K	GW
Klactis 371 34	S.pombe M.musculus H.sapiens	3434 - 4041 E 4050 E	GELL R R L P R L S K R M R K M C L V F M K E	ANVERPGSTFTNMVSNLTT SPLPHLVESLDOFTGGVIS	DARELM – – KLIPETI SVSELOSLKVDPSAE	KEKOHSEAKHILLOK	CHLLLTETFKILKAFGL CHRALSDLFKHLANIGL	QYRVKAGTEENLSNLR SYRKGLAWARSKSPQE SYRKGLAWARSKNPQE	LLHLHPLDLRSALSI	STREADS RLLT	EISSLWDGCOKYFYRS
3799 VRLFEKWLPSLLDYAAQTLSVISKYSATSEQ QKILLDAKSTLSSFFVHFNSS-RIFDSFLESYSRFELFINELLKKLENAKETON-AFVFDIIIEWIKANKGCP-IKKEQKR Klactis 3801 FRHICKWLPQLIDYIEVVEDSTAHEVPRSTVSYLYSKQKLLDFLAQSEMF-RIYDDITENILENENAFIKEFISAISAMNGTNSFVVDLLESWLQQALSHDVIFDT Calicians 3849 ISVKMUPKLDYGITTLESITKFGENDTNVSYLYSKQKLLDFLAQSEMF-RIYDDITENILENENAFIKEFISAISAMNGTNSFVVDLLESWLQQALSHDVIFDT Ncrassa 3849 ISVKMUPKLDYGITTLESITKFGENDTNVSYLYSKQKLLDFLAQSEMF-RIYDDITENILENENAFIKEFISAISAMNGTNSFVVDLLESWLQQALSHDVIFDT Ncrassa 3849 ISVKMUPKLDYGITTLESITKFGENDTNVSYLYSKQKLLDFLAQSEMF-RIYDDITENILENENAFIKEFISAISAMNGTNSFVVDLLESWLQQALSHDVIFDT Spombe 3645 ISVKMUPKLDYGITTLESITKFGENDTN-VSYFYLLKNKLELDSSINDENNV-VVQSIFFISSIEPCUSSSIRDQUKGINDLEFVCKKKRTEIPELSWCFKH.VSLQSLKSISRTQVDL M.musculus 4340 DLLSSDLSYFSVPTSQLPSACRMRRQDQLWQSAKKVDKKRQSCENLFHKWKDFEVCSSGLNCLSQVSAHLQCLASLFILPGIEVGUDSMALVESLEYVRGEVSKANDFTWKTHLTSDS-QEGNQMV M.musculus 4340 DLLSSDLSYFSVPTSQLPSACRMRRQDHWQSGTRLTEMEKTIKTWKADVDKRQSCENLFHKWKDFEVCSSALSCLSVSVHLQLESLEVVSKALGESKSIRDQQVSKKLQHTMKTHLTSDS-QEGNQML VALAS SGLPSSCRMRRQDHWQSGTRLTEMEKTIKTWKADVDKRQQSCENLFHKWKDFEVCSSALSCLSVSVHLQLESLEVVSKALGESKSIRDQQVSKKLGPLSALSVSKKNETTNFTNKTHLTSDS-QEGNQML VALAS VRTSQLPSACAMRRQDQLWQSGTRLTEMEKTIKTKTWKADVDKRQSCENKRQENKUSSLSVSKNIETALSUSVSKNIETALSLSVSKKNETETALSL VALAS SGLPSSCRMRRQDHWQSGTRLTEMEKTIKTKTWKADVDKRQSCENKRQENKUSSSUSVSKNIETALSVSVSKNIETALSUSVSKNIETALSVSVKKNETAKRYNNV	K.lactis C.albicans N.crassa S.pombe M.musculus H.sapiens	3734 - 3771 S. 3716 - 3577 - 4191 L. 4200 L.		GMAMTENLIFSLIASROPL GLSATDNLIHSLIVDRVPI SIGFLEGILYVMFKORMYL GVGLFNSMLSLOLGERAQL	SIFVHTYSKIESLRL KKLADNIDTFNSWYN SRTAEAERALAQAIT VEFTNELLALKNVYS TTITEOWILLRNUS	DLESVSLDC EFYTVANLNNLSATL FTKEVSGRGGEVAIC EVGVNGSPLESFNNS	H V L KH F D L S Y N F G T - _ A P V S V K D S I K L N I D N - Q S Q K S L T N - S S F N E V S S L G Y - D V A F P P OD S V O W T F R				
S.cerevisia S.cer	K.lactis C.albicans N.crassa S.pombe M.musculus H.sapiens	3801 - 3849 - 3782 - 3645 - 4340 D 4350 D	LLSSDLSY P S P V P TSOL P SACRMR	– – F R H I C KWL <mark>P</mark> QL I D Y I L E – – I S I V KMVL P K L L D Y G I T – – H G R V L RWV VQ I L K F A V Y – – D H D F E N R A QA V S M L C Q I R O D O LWO O S T A G L T E M L K T	Y V E D S T A H E V <mark>P</mark> R S T – T L E <mark>S</mark> I T K F G F N D T N – M V D V H G K L G D I S N D Q Y A I V I Q K H S S I S <mark>P</mark> T A I K T M K A G V D K I R Q O S	– – – – V S YL Y S S KQKL – V S V F Y L L KNKL N E L 2 V R S K L Q HWL D T F A T L S F Q S I G H E L S R F A D L C E T L F H TWE D F E V C S	L D F L A Q S E M F R I Y S L S I N D E N M V V V Q S A T K Q E S L P R L P T G F S S L S N K L F P S S I P - L Y A S S G L N C L S O V S A H L O G L	DTTENILENFNAFLKE SITIGNVEKFSKFIGE SALEQLHSDAEKELDG ADKVSSIRDQKGIND ASLFILPGIEVEOTDS	FISAISAMNGTNSF LMTTLTKWKLEHKQFA LRESLSSLLERPDMA LIEYCRKKRTELPELS RMALVESLDYLRGEVS	F VYDLLESWLQQ F VADTILNWIIN F VIRQVQLWTSA YCFKHLVSLQSL KAIDDFTAWKAR	2A L S – – – – H G D V I F D T I INSNT I NT I E S S T S L T F Q T V A I – – – – A N E L Q – . K S I S R – – – – T Q V D L T . L F V A H K – – O E G NOM V E
	S.cerevisiae K.lactis C.albicans N.crassa S.pombe M.musculus H.sapiens	3910 - 3911 D 3968 V 3899 E 3762 N 4488 E 4498 E	PSLEELDCKLRRISTAILISVQKLY ETIENVEKEFRTLSSSILVAVQKIN TTIDEFAKSALDLSSRILVALQKF DEFLNLMNFVLNLFDSLLSSIETA SFVEDFSEQVETAIRAILCTIONLA	V E NN	TTP I SEE 	D D KWL I E L N R R I V S N D D D WL I L S Q H K L I N Y D A GWF L A H S E G L E K A E T S S F I E M S S C F S K V P G H L T K L L E D D F W A S	NI KI ANCNKI FALI DDL YMKNLHLLTI <mark>S</mark> NKLKKC AINTLAMPRI TSVDGI VLRAFNLKFQ <mark>S</mark> MKLSSL SVSTLHVQKTI SSVSEL	VTTIKSTSFTAESSP- MEIISLVEHNQQSSK- LRLLKTSDLSQPAVRD KEKLRSSSVDKMSCQ- LERLKSCSEDGNTKH	NVAAAVSFTMPF LISALTSFTLPI AETALLRAILPN LLMLFLP KVFSQSCCLLVRLIPI	I NHYFSMLNDIL VNHYYQLCLKIF ILEQYAA <mark>T</mark> CQQNL YCEQFINLAESVL LC G F <mark>S</mark> DLVLFFL	NTTSENYFKFSKSTYI DKVRLNYDLSKATFI AQYADLHRSTCRLGYT DYFINVHNSNLDSLSK TVSLATHRSTAKLLSV

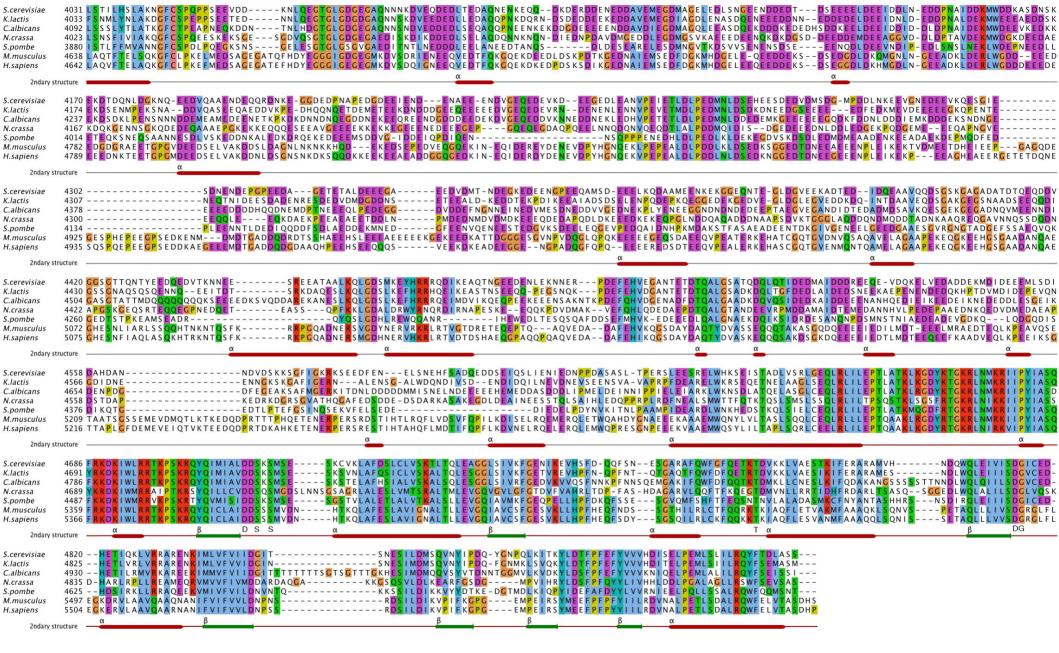


Figure S6:

Multiple sequence alignment of Rea1 full-length was done with ClustalW [76] and displayed with Jalview [77]. Secondary structure prediction of S. cerevisiae Rea1, done with PSIPRED [78], is depicted below and ATPase specific elements are labelled (α -helices are shown in red, β -strands in green, the AAA-ATPase specific lid domain is indicated in light blue and the clade-specific helix2 insert and pre-sensor-I insert (PSI) are indicated in dark blue. A red line marks the MIDAS domain.