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Supporting information for article:

Crystal structure of the spliceosomal DEAH-box ATPase Prp2

Andreas Schmitt, Florian Hamann, Piotr Neumann and Ralf Ficner

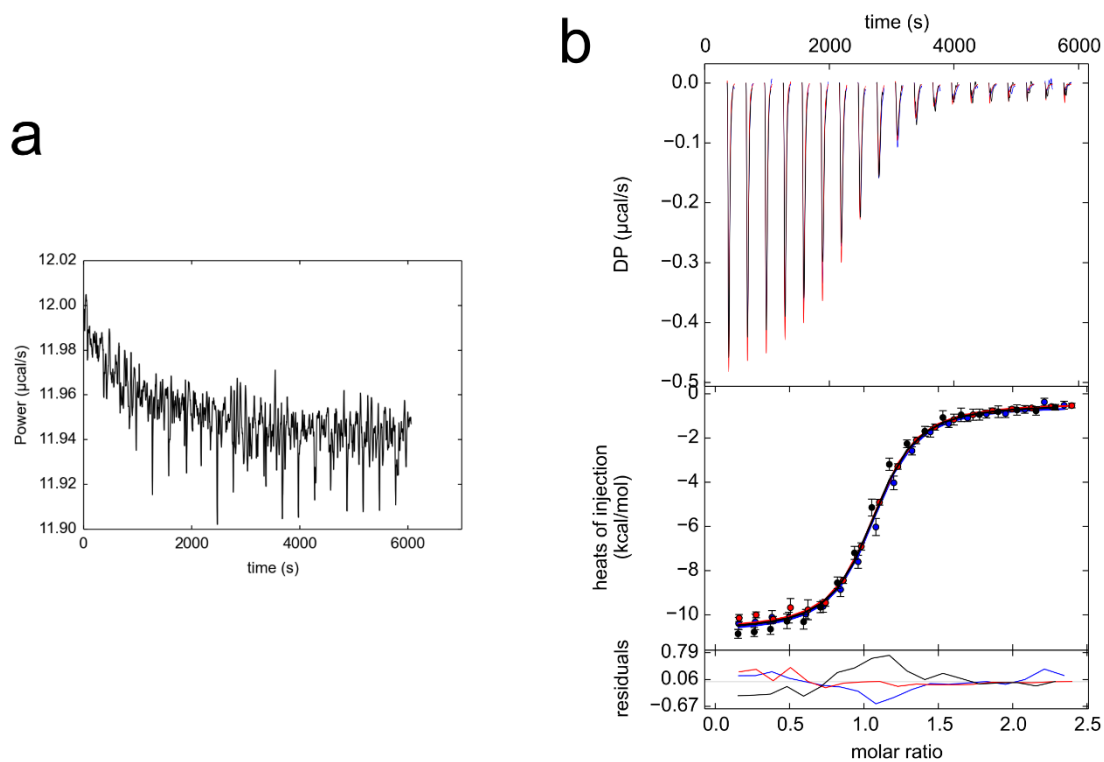


Figure S1 Isothermal titration calorimetry of Prp2 with AMPPCP and ADP. (a) The thermogram of the titration of AMPPCP to Prp2 did not result in thermic signals suited for integration, indicating there is no binding. (b) The binding of ADP was assessed and a dissociation constant (K_d) of 179 nM was determined ($n= 1.01$, $\Delta H= -10.63$ kcal/mol, $\Delta S= -5.06$ cal/mol*K). Individual triplicates are depicted in red, blue and black. In the upper panel the thermograms are displayed, the middle panel shows the individual fittings with the correspondent residuals in the lower panel.

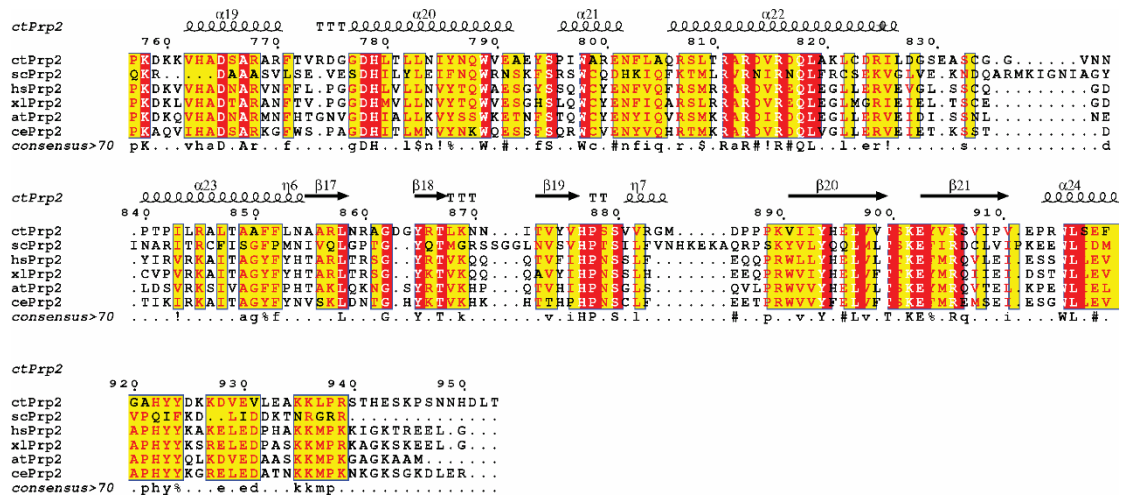


Figure S2 Sequence alignment of Prp2 from *Chaetomium thermophilum* (ct), *Saccharomyces cerevisiae* (sc), *Homo sapiens* (hs), *Xenopus laevis* (xl), *Arabidopsis thaliana* (at) and *Caenorhabditis elegans* (ce). Secondary structure elements from CF2 are displayed on top of the corresponding sequence.

ctPrp2

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1          10
ctPrp2  M S S K R Y A F V P . . . . . M D D E P A P . S . . . . .
scPrp2  M S S I T S E T G K . . . . . R R V . . . . .
hsPrp2  M A T P A G L E R . . . . .
ctPrp43 M A D I T K G T K R L . . . . . S G E A E G S . . . . .
scPrp43 M G S R R R F S . . . . . S E H P D P . V . . . . .
hsPrp43 M S K R H R L D L . . . . . G E D Y P S G . K . . . . .
ctPrp16 M G R Y D D . . . . . D R S . . . . . K K R R . . . . . K L
scPrp16 M C H S G R E . . . . . E R . . . . . I K D I . . . . . F K E L
hsPrp16 M G D T S E D . . . . . A S . . . . . I H R L . . . . . E G
ctPrp22 M R R R E Q Q Q L H L H S E A R S R A G F L P L T S S P C C C A D Q P Q L A D K M D . . . . . D L E . . . . . S L E L L S L
scPrp22 M S D I S K L I G A I . . . . . V G S D D P V I . I E F V L N I I N . K S G N L Q E F I R N I Q R L D A G I S Y E D S I K M Y N A F L G K Q E E K V R N K V K S S P L
hsPrp22 M A V A V A M A G A L . . . . . I G S E P G P . . . . . A E . . . . . E L A . . . . . K L E Y L S L
consensus>70 M . . . . .

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ctPrp2

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ctPrp2  . . . . . K R T . . . . .
scPrp2  . . . . . K R T . . . . .
hsPrp2  . . . . . K R T . . . . .
ctPrp43 . . . . . K R T . . . . .
scPrp43 . . . . . K R T . . . . .
hsPrp43 . . . . . K R T . . . . .
ctPrp16 . . . . . K R T . . . . .
scPrp16 . . . . . K R T . . . . .
hsPrp16 . . . . . K R T . . . . .
ctPrp22 . . . . . K R T . . . . .
scPrp22 . . . . . K R T . . . . .
hsPrp22 . . . . . K R T . . . . .
consensus>70 . . . . .

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ctPrp2

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ctPrp2  . . . . . Y E V T R Q N D N A . V R . . . . . I E P S S L G E E E D K E . . . . . A K . . . . .
scPrp2  K P A R A A E R E A R A L . . . . . L E K N R S Y R L L E D S E S S E . . . . . E T V . . . . . S R A G S S L Q K R R K R K H L R K K R E E E E E E A S E K G K R K T G G S K
hsPrp2  . . . . . Y E V T R Q N D N A . V R . . . . . I E P S S L G E E E D K E . . . . . A K . . . . .
ctPrp43 . . . . . Y E V T R Q N D N A . V R . . . . . I E P S S L G E E E D K E . . . . . A K . . . . .
scPrp43 . . . . . Y E V T R Q N D N A . V R . . . . . I E P S S L G E E E D K E . . . . . A K . . . . .
hsPrp43 . . . . . Y E V T R Q N D N A . V R . . . . . I E P S S L G E E E D K E . . . . . A K . . . . .
ctPrp16 . . . . . Y E V T R Q N D N A . V R . . . . . I E P S S L G E E E D K E . . . . . A K . . . . .
scPrp16 . . . . . Y E V T R Q N D N A . V R . . . . . I E P S S L G E E E D K E . . . . . A K . . . . .
hsPrp16 . . . . . Y E V T R Q N D N A . V R . . . . . I E P S S L G E E E D K E . . . . . A K . . . . .
ctPrp22 . . . . . Y E V T R Q N D N A . V R . . . . . I E P S S L G E E E D K E . . . . . A K . . . . .
scPrp22 . . . . . Y E V T R Q N D N A . V R . . . . . I E P S S L G E E E D K E . . . . . A K . . . . .
hsPrp22 . . . . . Y E V T R Q N D N A . V R . . . . . I E P S S L G E E E D K E . . . . . A K . . . . .
consensus>70 . . . . .

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ctPrp2

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          20          30
ctPrp2  . . . . . K . . . . . S . . . . . D R R R D R . . . . . D R R D R . . . . . R D . . . . . R . . . . .
scPrp2  . . . . . K . . . . . S . . . . . D K N S . . . . . A L Q L K R S . . . . . R Y . . . . . D P N K
hsPrp2  Q Q T E K P . . . . . E S . . . . . E . . . . . D E W E R T E R E R L Q D L E E R D A F A E R V R . . . . . Q . . . . . R D . . . . . K . . . . .
ctPrp43 . . . . . K R A G T . . . . . D G K D R D R . . . . . D R D R E D R S . . . . . K D . . . . . R . . . . .
scPrp43 . . . . . K R A G T . . . . . D F N P L R . . . . . S D D R F G . . . . . D I P A . . . . . R . . . . .
hsPrp43 . . . . . K R A G T . . . . . I . . . . . Q L D L D D . . . . . N . E D . . . . . E . . . . .
ctPrp16 . . . . . K R A G T . . . . . I . . . . . Q L D L D D . . . . . N . E D . . . . . E . . . . .
scPrp16 . . . . . K R A G T . . . . . I . . . . . Q L D L D D . . . . . N . E D . . . . . E . . . . .
hsPrp16 . . . . . K R A G T . . . . . I . . . . . Q L D L D D . . . . . N . E D . . . . . E . . . . .
ctPrp22 . . . . . K R A G T . . . . . I . . . . . Q L D L D D . . . . . N . E D . . . . . E . . . . .
scPrp22 . . . . . K R A G T . . . . . I . . . . . Q L D L D D . . . . . N . E D . . . . . E . . . . .
hsPrp22 . . . . . K R A G T . . . . . I . . . . . Q L D L D D . . . . . N . E D . . . . . E . . . . .
consensus>70 . . . . .

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ctPrp2

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          40          50          60          70
ctPrp2  . . . . . R D R K R D R S H S R S R S K D . . . . . H Y R D R S R S P S Y K . . . . . S R R R D G D D S P . . . . .
scPrp2  V F S N . . . . . T N Q G P E . . . . . K N N L K G E Q L G . . . . .
hsPrp2  . . . . . D R . . . . . T R N V L . . . . . E R S . . . . . D . . . . .
ctPrp43 . . . . . E T S . . . . . I P . . . . .
scPrp43 . . . . . E T S . . . . . I P . . . . .
hsPrp43 . . . . . E T S . . . . . I P . . . . .
ctPrp16 . . . . . S H S N N N . . . . . G S L Q . . . . . S R S T P R . . . . . G S R L E G A R T P . . . . .
scPrp16 . . . . . L D S P V Q K . . . . . K P A P T R L E K R I D K L K A Q L R Q Y S P T V K D P S F N S E Q Q T Q N G H A E T K D Y E P T R S E V V E D R E W Y D N D D D Y G N L V . . . . .
hsPrp16 . . . . . R D R S R H S . . . . . S R S E . . . . . R D G G S E R . . . . . S R R N E P E S P H R R P K D A A T P . . . . .
ctPrp22 . . . . . K . . . . . D R Y R . . . . . S R S R S P . . . . . A R F R S . . . . . R S L S R S R S K S P R . . . . . R G R R K S S R Y D . . . . .
scPrp22 . . . . . K . . . . . D R Y R . . . . . S R S R S P . . . . . A R F R S . . . . . R S L S R S R S K S P R . . . . . R G R R K S S R Y D . . . . .
hsPrp22 . . . . . K . . . . . D R Y R . . . . . S R S R S P . . . . . A R F R S . . . . . R S L S R S R S K S P R . . . . . R G R R K S S R Y D . . . . .
consensus>70 . . . . .

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ctPrp2

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          80          90          100
ctPrp2  . . . . . R R R S R S . . . . . P R D R S S N T H S A . . . . . T T T T . . . . . Y D E K I T S N N E L T . . . . . T F L N
scPrp2  . . . . . S Q K K S . . . . . K . . . . .
hsPrp2  . . . . . R R R S R S . . . . . P R D R S S N T H S A . . . . . T T T T . . . . . Y D E K I T S N N E L T . . . . . T F L N
ctPrp43 . . . . . H H R G D D . . . . . G . . . . . P E L S P G F D A D A E N A L . . . . .
scPrp43 . . . . . H H R G D D . . . . . G . . . . . P E E A K L . . . . .
hsPrp43 . . . . . H H R G D D . . . . . G . . . . . P T P S Y K Y N E W A D D R R H L G S T P R L S R G R . . . . .
ctPrp16 . . . . . P E P L S E . . . . . L . . . . . F R R P P P R D L D D A P Q L Y K I Y D G H V T G I K D F G A F V N L H G V K . . . . .
scPrp16 . . . . . P E P L S E . . . . . L . . . . . P S P V L H K V Y E G K V R N I T T F G C F V Q I F G T R . . . . .
hsPrp16 . . . . . P E P L S E . . . . . L . . . . . E R N L D R . . . . . W R D K H V D R P P P . . . . . E E P T I G D I Y N G K V T S I M Q F G C F V Q L E G L R
ctPrp22 . . . . . E R N L D R . . . . . W R D K H V D R P P P . . . . . E E P T I G D I Y N G K V T S I M Q F G C F V Q L E G L R
scPrp22 . . . . . E R N L D R . . . . . W R D K H V D R P P P . . . . . E E P T I G D I Y N G K V T S I M Q F G C F V Q L E G L R
hsPrp22 . . . . . E R N L D R . . . . . W R D K H V D R P P P . . . . . E E P T I G D I Y N G K V T S I M Q F G C F V Q L E G L R
consensus>70 . . . . .

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ctPrp2

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          110          120          130          140
ctPrp2  K . . . . . D D D R E A K L A E L R L K S R Q . . . . . Y L A K R E A . . . . . E K L A L L R K Q V A E E T A . . . . . E . . . . . G L L G D S . . . . .
scPrp2  . . . . . D . . . . . E . . . . . L . . . . .
hsPrp2  . . . . . D . . . . . E . . . . . L . . . . .
ctPrp43 . . . . . D . . . . . E . . . . . L . . . . .
scPrp43 . . . . . D . . . . . E . . . . . L . . . . .
hsPrp43 . . . . . D . . . . . E . . . . . L . . . . .
ctPrp16 . . . . . D R D . . . . . W Y G D D D L G G H T F G D E T H N E F G . . . . . D D S Y W A A . . . . . Q . . . . .
scPrp16 . . . . . D R D . . . . . W Y M M D E G Y D . . . . . E F H N P L A . . . . . Y S S E D Y V . . . . . R . . . . .
hsPrp16 . . . . . G . . . . . K I D G L V H I S A F G . . . . . Q R V N H P S D A V S M G Q N V K V I K I E G . . . . . N R I G L S M K D V D Q E T G . . . . . M D L A P Q M R I G S G A N M E A L G . . . . . G
ctPrp22 . . . . . M K N C D G L V H I S E M S D . . . . . Q R T L D P H D V R Q G Q H I F V E V I K I Q N G N K I S L S M K N I D Q H S G . . . . . E I R K R N T . . . . . E S V E D R G . . . . .
scPrp22 . . . . . K . . . . . R W E G L V H I S E L R R E G R V A N V A D V V S K G Q R V K V K V L S F T G . . . . . T K T S L S M K D V D Q E T G . . . . . E D L N P N R R R . . . . . N L V G E T N E E T S M
hsPrp22 . . . . . K . . . . . R W E G L V H I S E L R R E G R V A N V A D V V S K G Q R V K V K V L S F T G . . . . . T K T S L S M K D V D Q E T G . . . . . E D L N P N R R R . . . . . N L V G E T N E E T S M
consensus>70 . . . . .

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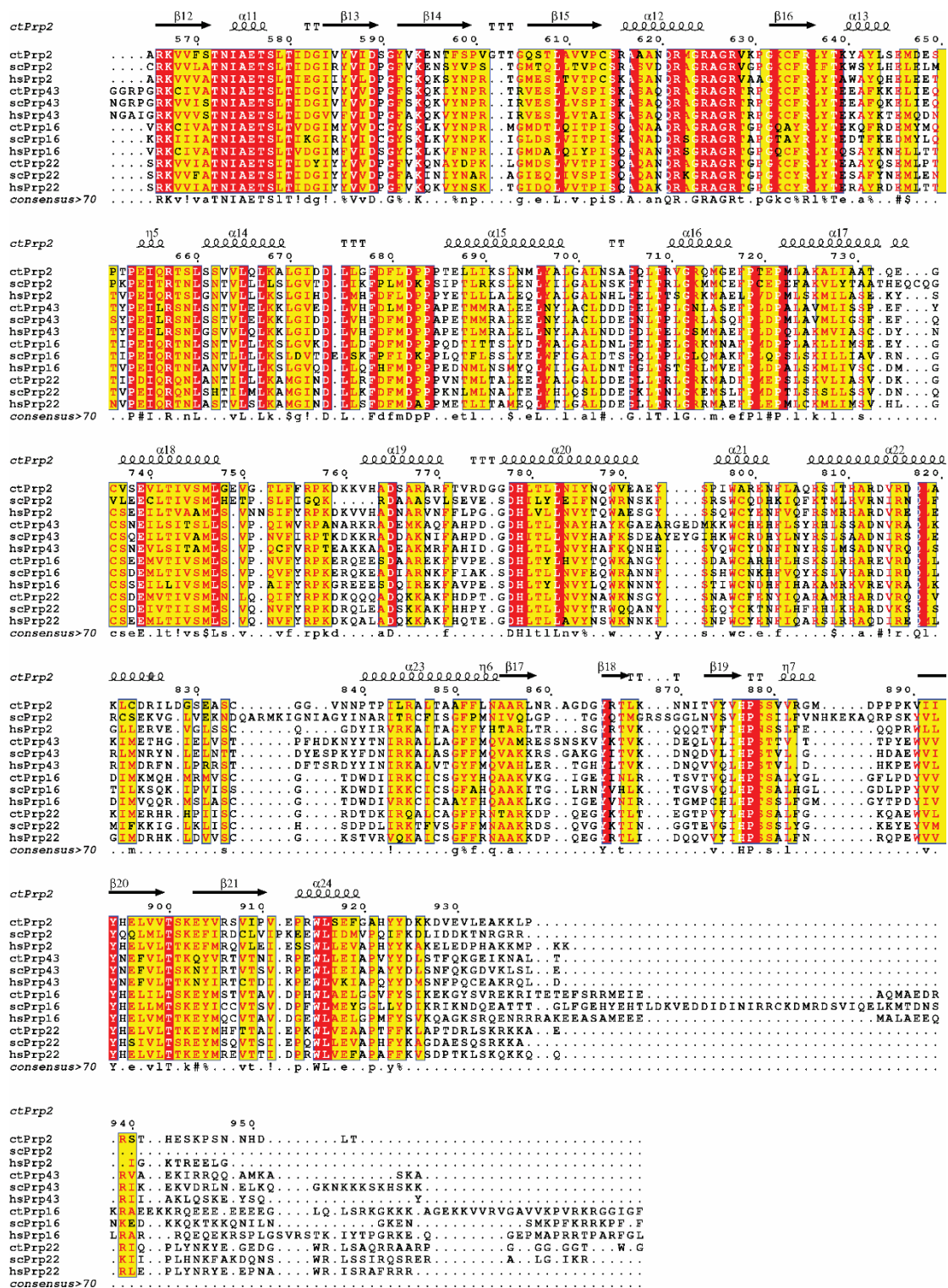



Figure S3 Sequence alignment of Prp2, Prp43, Prp16 and Prp22 from *Chaetomium thermophilum* (ct), *Saccharomyces cerevisiae* (sc) and *Homo sapiens* (hs). Secondary structure elements from CF2 are displayed above the corresponding sequence.

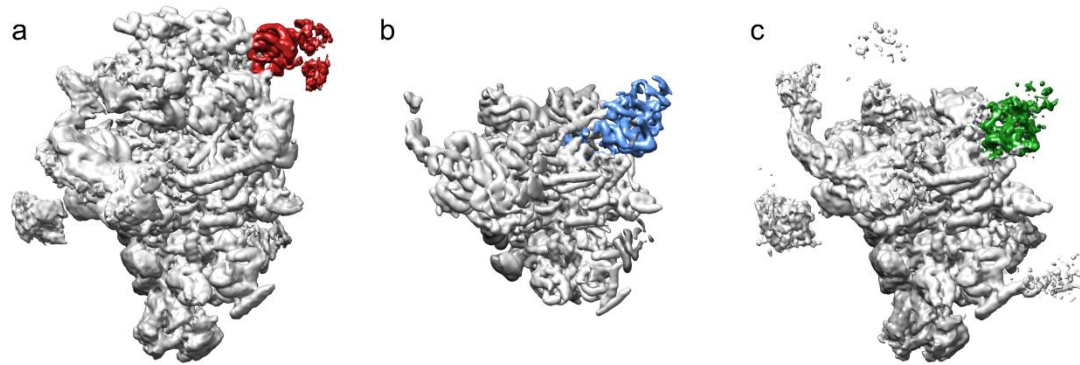


Figure S4 Position of DEAH-box helicases in the spliceosome. The cryo-EM maps of the *Saccharomyces cerevisiae* B^{act} complex (a, Yan *et al.*, 2016), *Saccharomyces cerevisiae* post-catalytic P complex (b, Liu *et al.*, 2017) and *Saccharomyces cerevisiae* spliceosome complex remodelled for exon ligation (c, Fica *et al.*, 2017) superimposed and are shown in grey and DEAH-box helicases of each complex are depicted in red (Prp2), blue (Prp22) and green (Prp16), respectively. All DEAH-box helicases are found in the periphery of their respective spliceosomal complex.



Figure S5 Secondary structure prediction of N-terminal extension of *Chaetomium thermophilum* Prp2. The secondary structure prediction was performed using the GeneSilico metaserver (Kurowski & Bujnicki, 2003). Used prediction server for the metasearch are stated the left followed by the corresponding prediction on the right. Residues predicted to be part of a helical secondary structure are highlighted by “H” whereas residues predicted to be part of beta strands are displayed as “E”.