Review of: "Blocking a dead-end assembly pathway creates a point of regulation for the DEAD-box ATPase Has1 and prevents platform misassembly"

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Potential competing interests: The author(s) declared that no potential competing interests exist.

The manuscript of Xin Liu et al; nicely describes the role of Has1 and Rrp5 proteins in controlling proper assembly of 40S-plateform and processome. The authors show that Rrp5 interacts first with Rrp36 on the body, to avoid early assembly of Rrp5 into the platform that might leads to an end-end unproductive assembly pathway. It is also shown that he helicase activity of Has1 is required to release Rrp5 and subsequently re-localization to the platform very early in 40S subunit assembly. These functional interactions are driven by the first S1 domain of Rrp5. In addition, the data indicate that Rrp5 S1 domain and timely interactions leads to strong depletion of the A2, but not A0, cleavage products as well depletion (Rps27, 13 and 22) or enrichment (Rps1, 14 and 22) of specific components. Depletion of the S1 domain leads also to defects in start and stop codon selection, reading frame maintenance but not the decoding indicating that the A-site is properly formed. Interestingly, inactivation of Has1 does not affect assembly of early processsome components (UtpA-Utp8, or UtpB-13, -Rrp5 or RocK1) but the assembly of late binding factors Pno1 and Enp1. The studies and observations also indicate that Has1 and relocalisation of Rrp5 to the platform might stabilize the UtpB conformational switch and then the assembly of the late processome.

To recapitulate, this is an integrative work that utilize structural studies and combines biochemical, molecular and genetic approaches. My major criticism is the organization and/or the style of the manuscript. It is not always easy to read, at least for a not native English speaker. I also believe that the manuscript can be simplified and/or shortened (more concise) to make the points more straight forward. In other hand, figures can also be enlarged a little bit (in particular 5A and 5, left panels) and the color codes be checked (for instance Figure 4A).

Other points

page 3: “these two ribosomal proteins”

- which ones Rps1 and Rps27, or Rps1 and Rps22/uS8 or Rps27 and Rps22/uS8 ?

page 5: “For these experiments we expressed and purified recombinant Rrp5 and Rrp36..”

- it is MBP-Rrp36 instead of Rrp36

page 6: “(Figure S1A), as well as within the 5’-end of 5.8S rRNA (data not shown) ….”

- data can be provided as S-data

page 7 “Previous work had shown that Rrp5 bound the DEAD-box ATPase Has1(Khoshnevis et al., 2016).” :

- correct ?

page 8 “Previous work had shown that these mutants could provide for Has1’s roles in 60S assembly, ….“

- this is correct?

Page 9 “Notably, Rrp36 was accumulated in ribosome bound fractions (Figure 5A).”

- Figure shows Free and “processome” fractions

page 10 First paragraph:

- Dim1 it is also down regulated in both Has1 mutants. Authors should comment this in the text

page 14 “we note that by Western analysis of gradient centrifugation experiments Rcl1 and Bms1 are depleted from early pre-ribosomes”

- depletion of Bms1 is less evident in figure 5C compared to 5B. Indicate if changes are significant or not.