

Magn. Reson. Discuss., author comment AC3
<https://doi.org/10.5194/mr-2021-21-AC3>, 2021
© Author(s) 2021. This work is distributed under
the Creative Commons Attribution 4.0 License.

Reply on RC3

Ulric B. le Paige et al.

Author comment on "Characterization of nucleosome sediments for protein interaction studies by solid-state NMR spectroscopy" by Ulric B. le Paige et al., Magn. Reson. Discuss., <https://doi.org/10.5194/mr-2021-21-AC3>, 2021

Dear prof. Nordenskiöld,

Many thanks for your comments and I apologize for the late reply.

Indeed, we used low Mg^{2+} (2 mM) with the intent to avoid close packing, which would be induced by high Mg^{2+} as shown by your lab, and here report our assessment on how this approach worked out. We did not systematically evaluate the Mg^{2+} -dependence of sedimentation efficiency or spectra quality. Judging from results from your lab it seems that both high and low Mg^{2+} conditions result in high quality spectra. In the discussion, we discuss explicitly the factors that likely contributed to the lack of pronounced ordering, including the choice for low Mg^{2+} : "First and foremost, the Mg^{2+} concentration used in our study is way below the minimum required to precipitate isolated nucleosomes (Berezhnoy et al. 2016; Wang et al. 2021), ..."

As for the in-rotor concentration estimates, we did not use an external standard, but did explicitly estimate the material loss due to cap clearing.

We added a sentences to make explicit that most of the PHD2 protein remained in the supernatant after sedimentation.

Thanks for pointing out the additional factors in mediated nucleosome contacts, we have rephrased this in the introduction.

We added the requested additional information on the SAXS data analysis in the M&M and a additional Table. As for the estimation of the length scale of ordering in the sediment, this should be taken as rough estimate based on the peak width. We rephrased the sentence to make this more clear. We also the the discussion on the SAXS data more explicit in terms of the comparison to literature data.

We further corrected the textual issues and added the info on the peak intensity error bars.

Best regards,

On behalf of the co-authors,

Hugo van Ingen