Electronic Supplementary Material (ESI) for Molecular BioSystems


# Supplementary Information

## Insights into the role of the beta-2 microglobulin D-strand in amyloid propensity revealed by mass spectrometry

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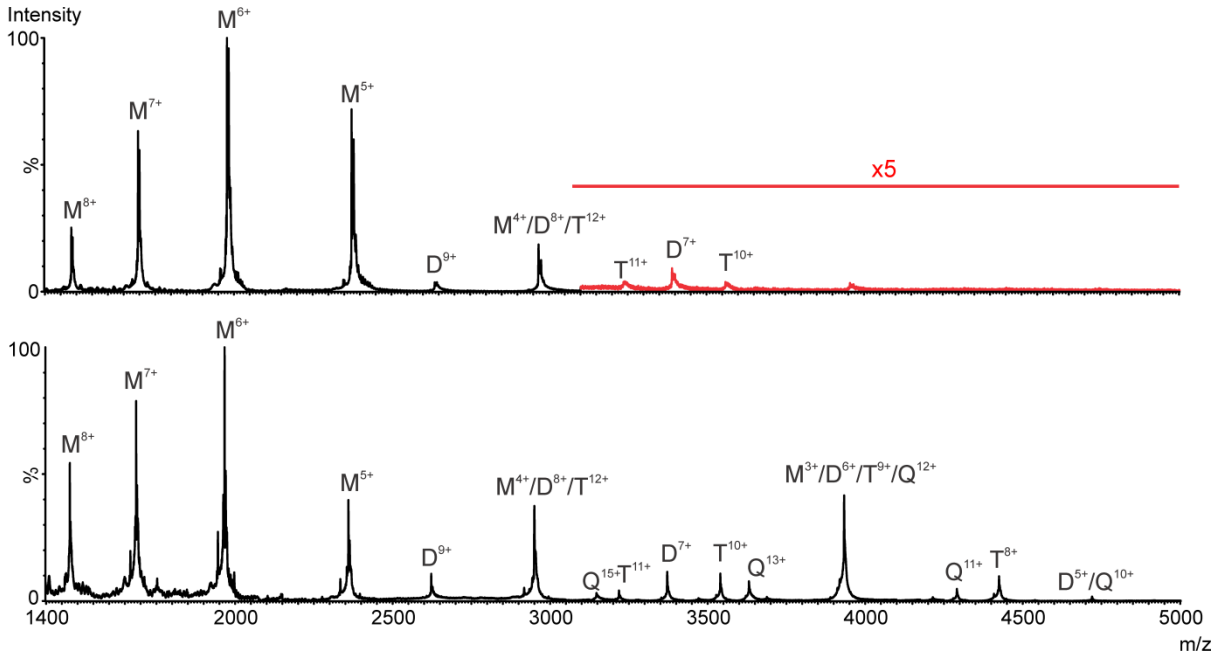


Figure S1. ESI-MS spectra acquired immediately after dilution of WT $\beta_2$m (upper) and H51A (lower) into the fibril-forming buffer (100 mM ammonium formate, pH 2.5). The WT mass spectrum was amplified (5x) from *m/z* 3000 to 6000 to highlight the higher order oligomers present.

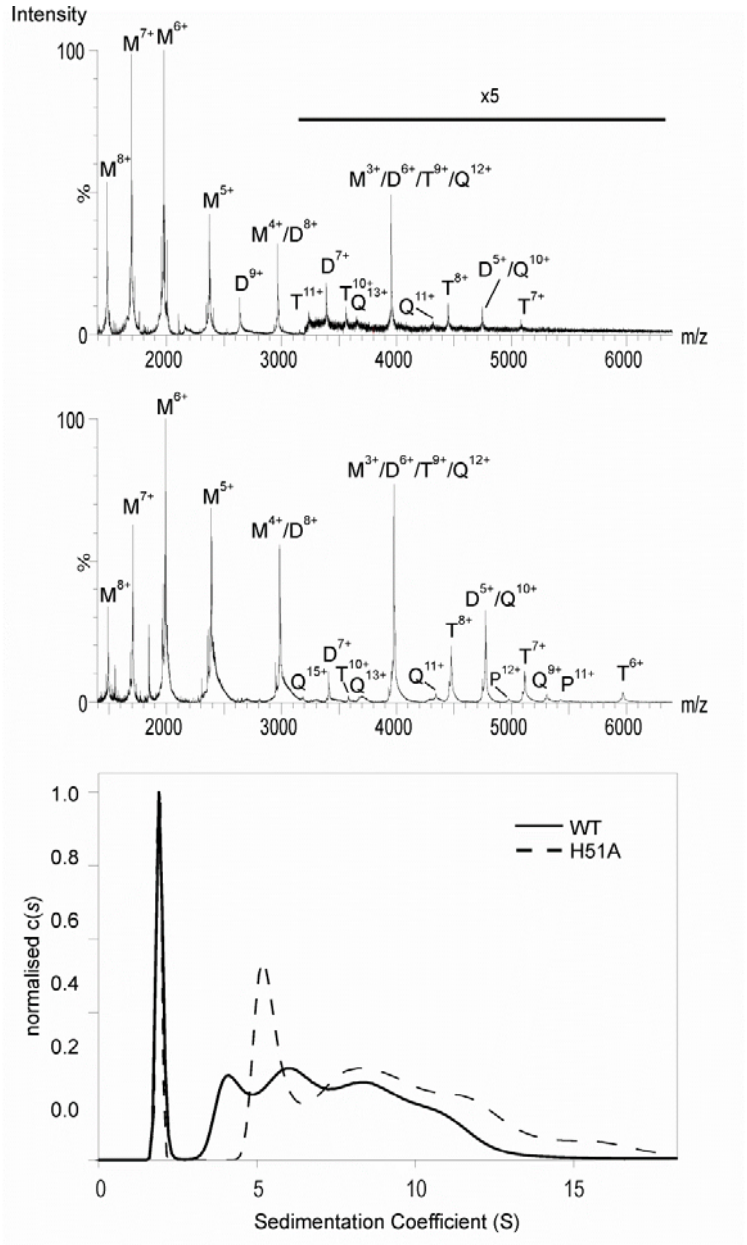


Figure S2. ESI-MS spectra acquired from WT $\beta_2$m (upper) and H51A (centre) at 0.4 mg mL$^{-1}$ after dialysis into the fibril-forming buffer (100 mM ammonium formate, pH 2.5) overnight at 4 $^{o}$C. The WT mass spectrum was amplified (5x) from *m/z* 3200 – 6400 to highlight the presence of the trimeric and tetrameric oligomers. Sedimentation velocity analytical ultracentrifugation analyses (lower) of the same samples from WT $\beta_2$m (solid line) and H51A (dashed line) were performed at 40,000 rpm at 25 $^{o}$C. The resulting broad peaks indicate interconverting species and therefore it was not possible to estimate masses for these data; hence all mass measurements were performed using ESI-(IMS)-MS.

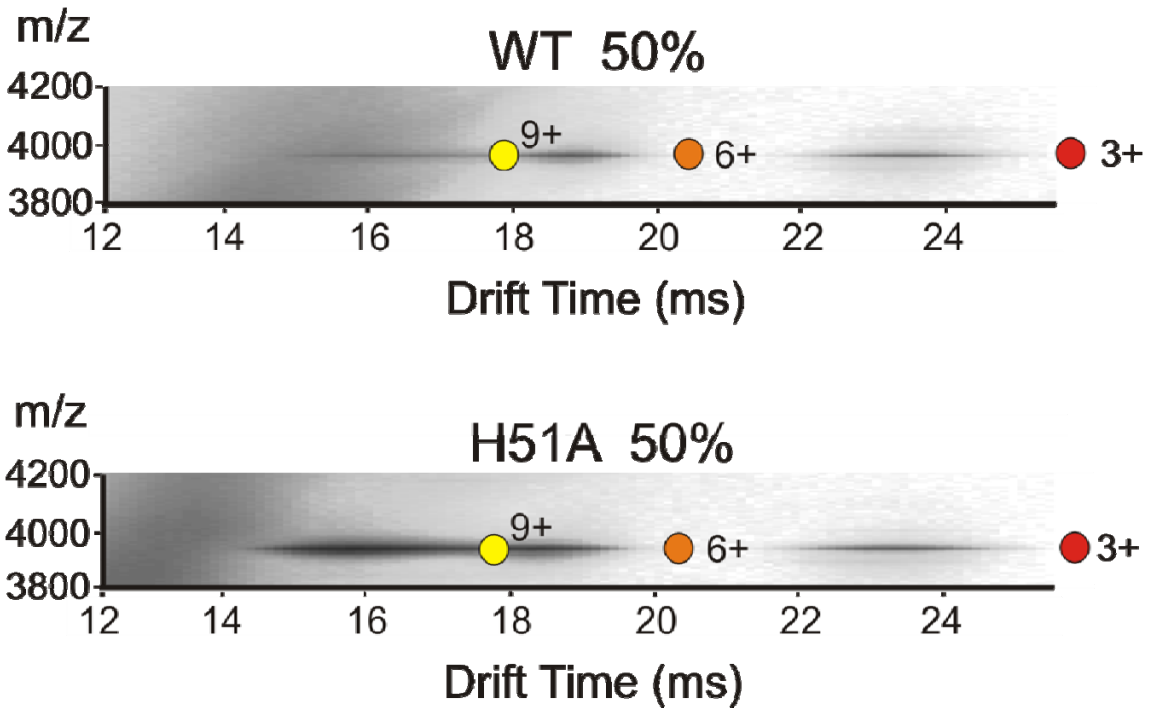


Figure S3. ESI-IMS-MS Driftscope plot of WT $\beta_2$m (upper) and H51A (lower) at 50 % of the respective lag-phases of the two proteins. The separated isobaric monomer 3+, dimer 6+, trimer 9+ ions are highlighted in red, orange and yellow, respectively.

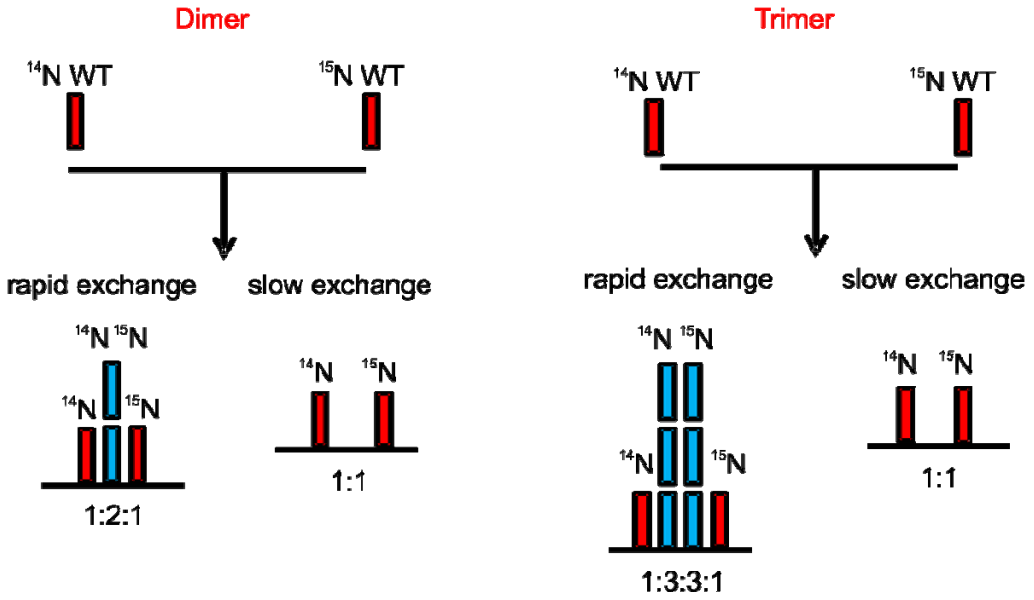


Figure S4. Schematic of subunit exchange showing the expected $^{14}N/^{15}N$ ratios for fully exchanged dimers and trimers, in addition to those expected for dimers and trimers that do not undergo exchange. Homogeneous $^{14}N$- and $^{15}N$-only dimers and trimers are shown in red, heterogeneous mixed $^{14}N/^{15}N$- dimers and trimers are shown in blue.

It has been reported previously[1] that mixed oligomers do not form during the ionisation process, and hence the oligomers observed are representative of the solution components

1. D. P. Smith, S. E. Radford and A. E. Ashcroft, Elongated oligomers in beta(2)-microglobulin amyloid assembly revealed by ion mobility spectrometry-mass spectrometry, *Proc. Natl. Acad. Sci. U. S. A.*, 2010, **107**, 6794-6798).