## Characterization and quantification of AAV capsid loading states by multi-wavelength AUC with UltraScan

Amy Henrickson<sup>1</sup>, Xiaozhe Ding<sup>2</sup>, Austin G. Seal<sup>3</sup>, Zhe Qu<sup>2</sup>, Lauren Tomlinson<sup>4</sup>, John Forsey<sup>4</sup>, Viviana Gradinaru<sup>2</sup>, Kazuhiro Oka<sup>3, 5</sup>, Borries Demeler<sup>1,6</sup>

<sup>1</sup>Department of Chemistry and Biochemistry, University of Lethbridge, Lethbridge, AB, T1K 3M4, Canada

 $^2$  Division of Biology and Biological Engineering, California Institute of Technology, Pasadena, CA, USA

<sup>3</sup> Gene Vector Core, Advanced Technology Cores, Baylor College of Medicine Houston, Texas, USA

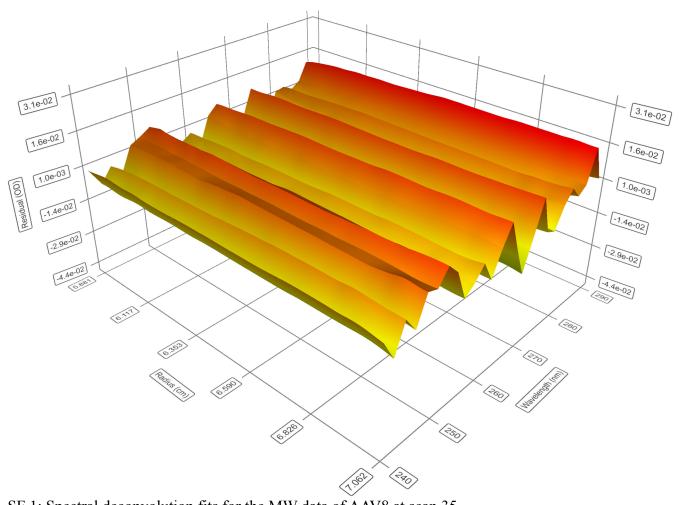
<sup>4</sup> Pharmaron Biologics Ltd, Speke, Liverpool, UK

<sup>5</sup> Department of Molecular & Cellular Biology, Baylor College of Medicine, Texas, USA

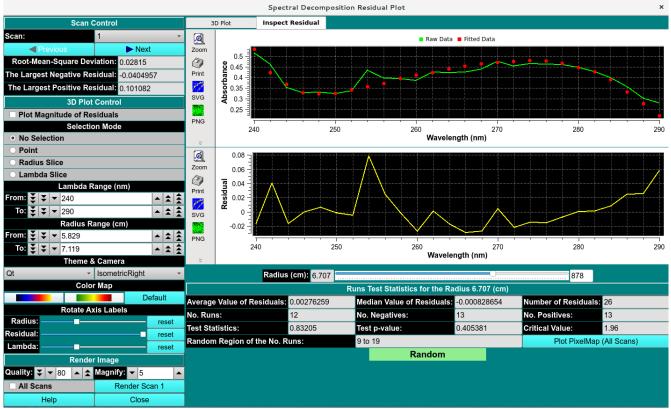
<sup>6</sup> Department of Chemistry and Biochemistry, University of Montana, Missoula, MT, USA

Corresponding Author: Borries Demeler (<u>demeler@gmail.com</u>)

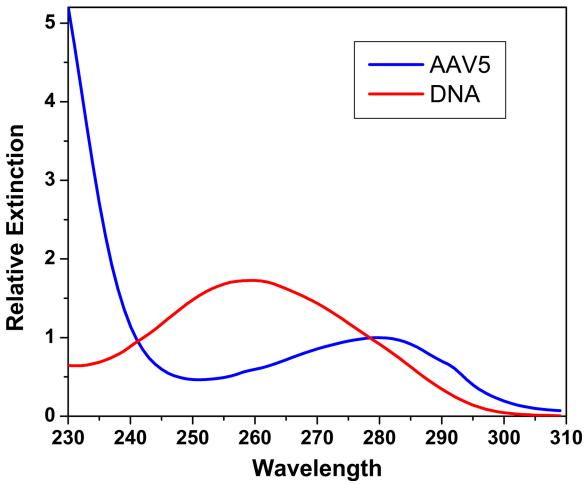
## SUPPLEMENTAL INFORMATION



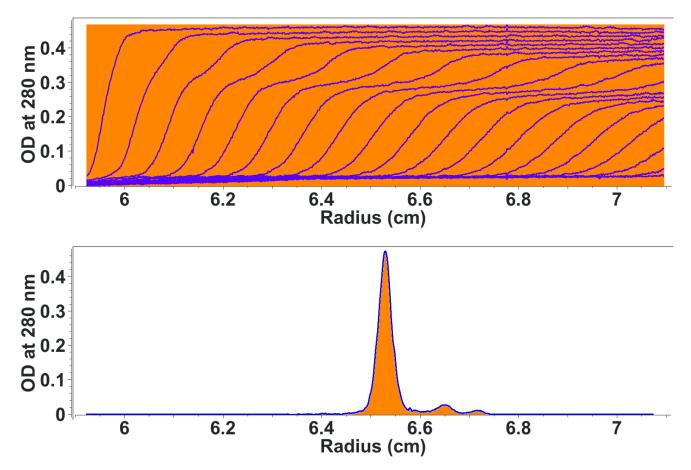
SF 1: Spectral deconvolution fits for the MW data of AAV8 at scan 35



SF 2: Screen shot of the spectral decomposition residual plot in UltraScan demonstrating the fit and residuals at individual radius positions for all wavelengths measured for AAV8.



SF 3: Extinction spectrum for AAV5 capsid protein (blue) and DNA (red) between 230-310 nm. Absorbance spectra are normalized for to have the same area under each curve.



SF 4: Sample quantity requirements – SV vs. ABDE. Required sample amounts are proportional to the orange shaded regions in an SV experiment (top) and an ABDE experiment (bottom). Since ADE experiments collect the entire signal only in the peak area, sample amounts are significantly reduced. In both experiments, the dynamic range of the detector must be observed to insure a linear response. In the case shown here, the amount of sample required for an ABDE experiment is 23.7 fold lower than in the SV experiment when integrating the orange area for both plots. This ratio increases if fewer peaks are observed, or decreases when additional peaks are observed.