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## Small angle x-ray Scattering demonstrates the morphological heterogeneity of extracellular vesicles

Extracellular vesicles (EVs) are functional lipid bi-layer enclosed nanoparticles secreted by cells and involved in intercellular communications, and disease progression. EVs have therapeutic potential as drug delivery vehicles, and diagnostic potential as a source of biomarkers. EVs are heterogenous in their morphology both in terms of EV and non-EV nanoparticles, making isolation and characterisation of distinct EV sub-populations difficult. Here we have conducted synchrotron-based small angle X-ray scattering (SAXS) to assess the morphological heterogeneity of EVs isolated by different methods.

EVs were isolated from murine N2a neuroblast cells with Size exclusion chromatography (SEC), Capto Core multimodal chromatography and density gradient ultracentrifugation. Isolated EVs were subjected to SAXS at the Australian synchrotron. The generated spectra were averaged and subtracted from background PBS for further analysis.  $R_g$  was calculated in RAW from the Guinier region and used to generate Indirect Fourier Transforms and  $P(r)$  to determine particle size and shape. Averaged spectra were also fit to a theoretical poly-disperse vesicle model generated using SASview.

This research demonstrates the morphological heterogeneity of EVs and the associated challenges of isolating distinct EV subpopulations. SAXS was successfully employed to detect the size of EVs but may not be able to provide high detail due to EV aggregation, and EV heterogeneity. Although spectra were comparable to vesicle models. This research evaluates SAXS for the study of EVs and suggests the use of higher purity isolation techniques to obtain sufficiently homogenous populations for SAXS analysis.

### Level of Expertise

Student

### Presenter Gender

Man

### Pronouns

He/Him

### Do you intend to attend UM2022

In person - Melbourne

### Students Only - if available would you be interested in student travel funding

Yes

### Students Only – Do you wish to take part in the Student Poster Slam

Yes

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Yes

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