Abstract

We used static light scattering to measure the 2nd virial coefficient of the eye lens protein, bovine γB-crystallin, which exhibits liquid-liquid phase separation related to cataract disease. The measured molecular weight was (2.3 × 10^5 ± 1.2 x 10^4) grams/mole (mean ± std. dev.), within two standard deviations of the sequence value, 20992.7 grams/mole. At pH 4.4, where no phase separation occurs, we measured the repulsive, dimensionless 2nd virial coefficient, B_T 8.1 ± 1.0, exceeding the values of 4 and 5.4 that would correspond to hard spheres and hard sphere dimers, respectively. At pH 5.4, our measurements suggested B_B, consistent with net attractive interactions and phase separation at this pH, though higher concentration measurements are needed to quantify B_B at this pH. These measurements suggest bovine γB-crystallin has a crossover from attractive to repulsive interactions near pH 5.

Materials & Methods

- Nuclear and cortical material from calf lenses.
- Homogenated and centrifuged cytoplasm.
- Size, ion exclusion chromatography to purify γB.
- Concentrations measured by UV spectrometry.

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References and Further Reading


Conclusions

- Measured molecular weight (22659 ± 1230) grams/mole.
- Measured a dimensionless second virial coefficient at pH 6.5, B_T (8.1 ± 1.0).
- At pH 5.4, negative second virial coefficient, crossover pH between pH 5.4 and 4.4.
- Crossover pH may have well depth 1.93kT and the fraction of surface with charge property A near 83%.
- Models used were simple, experiments run into roadblocks with filters and time. Hopefully further research will bypass these blocks.