THE RHEOLOGICAL PROPERTIES ON AN INSULIN ANALOGUE USING RHEO-SMALL ANGLE NEUTRON SCATTERING

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Research work Sponsored by

UMD, College Park

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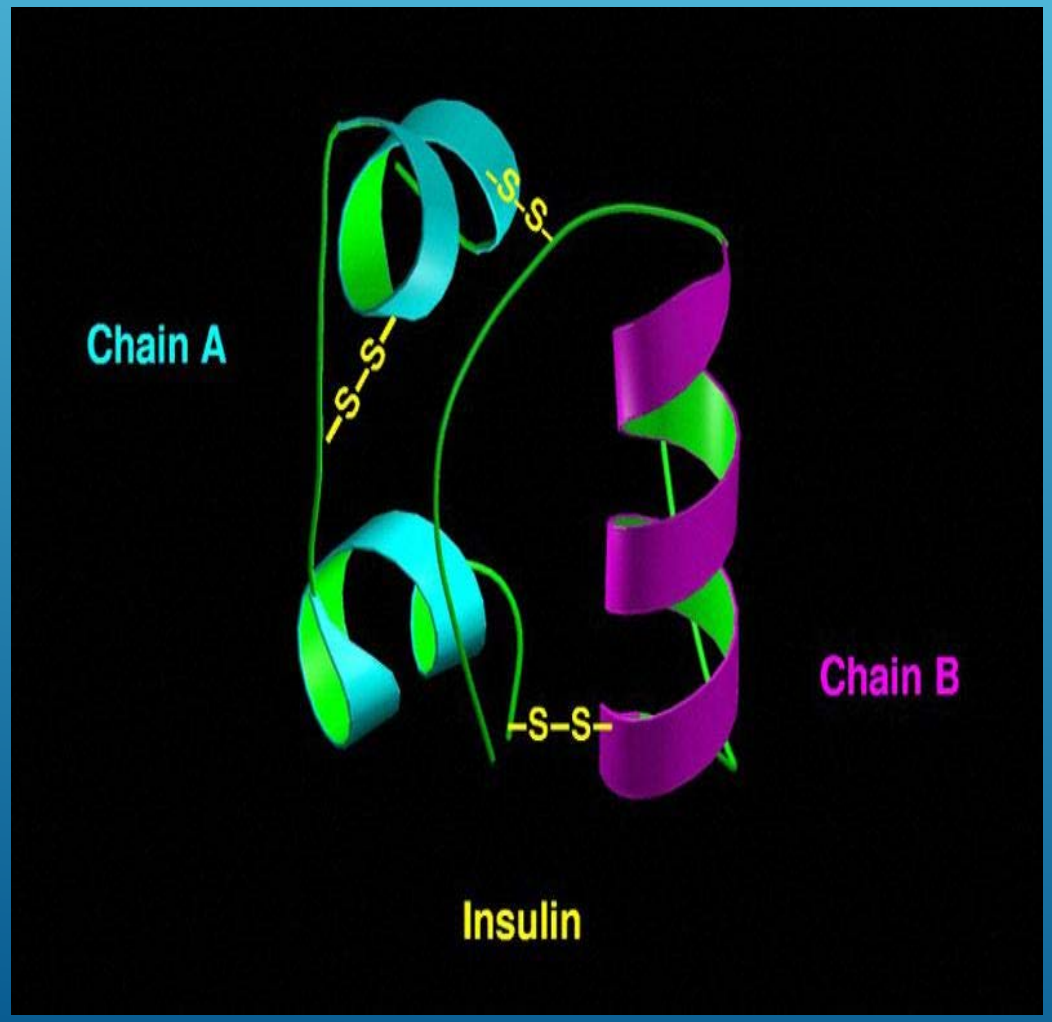
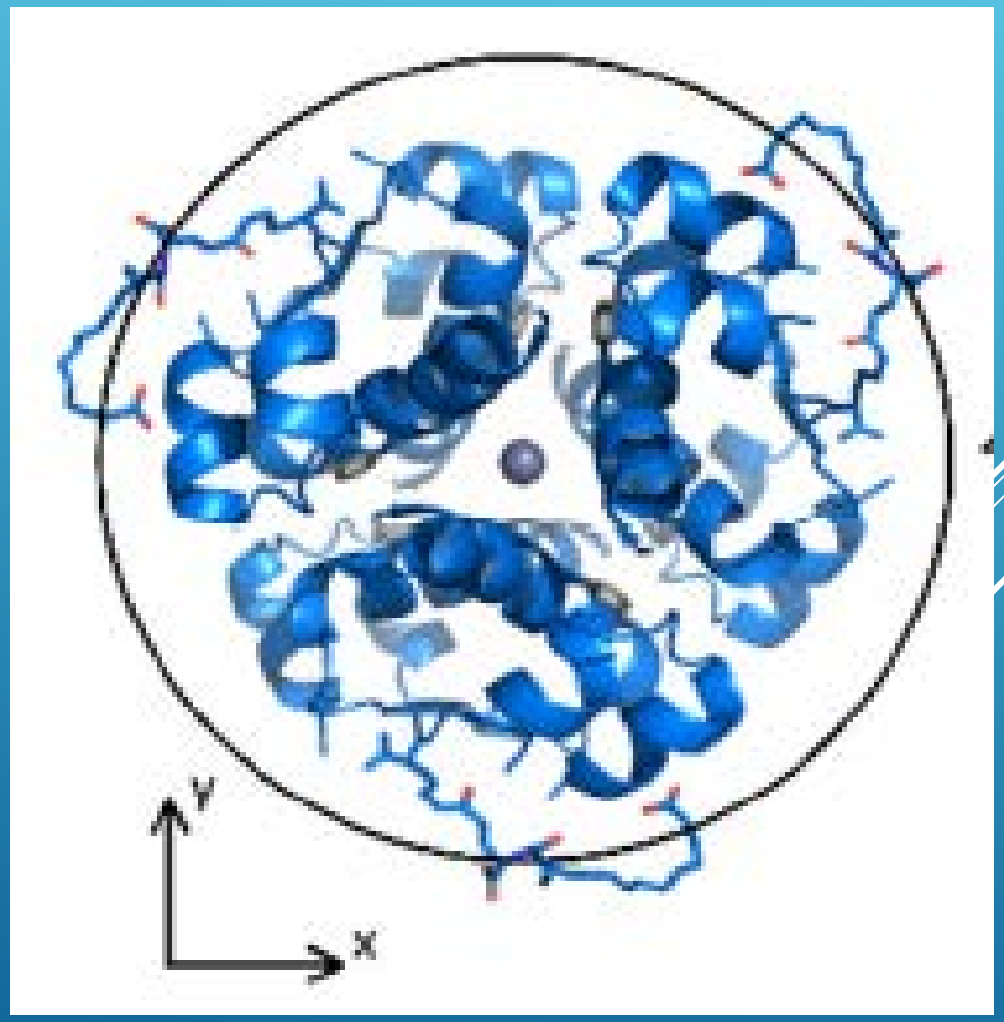
&

NCNR, NIST Gaithersburg, MD

The structures of insulin & analog with attached fatty acids are shown in stick models. The Zn ions are shown as grey spheres in grey stick models.





PREPARATION OF ZINC- CONTAINING INSULIN SOLUTION WITH DIFFERENT CONCENTRATIONS OF SALT

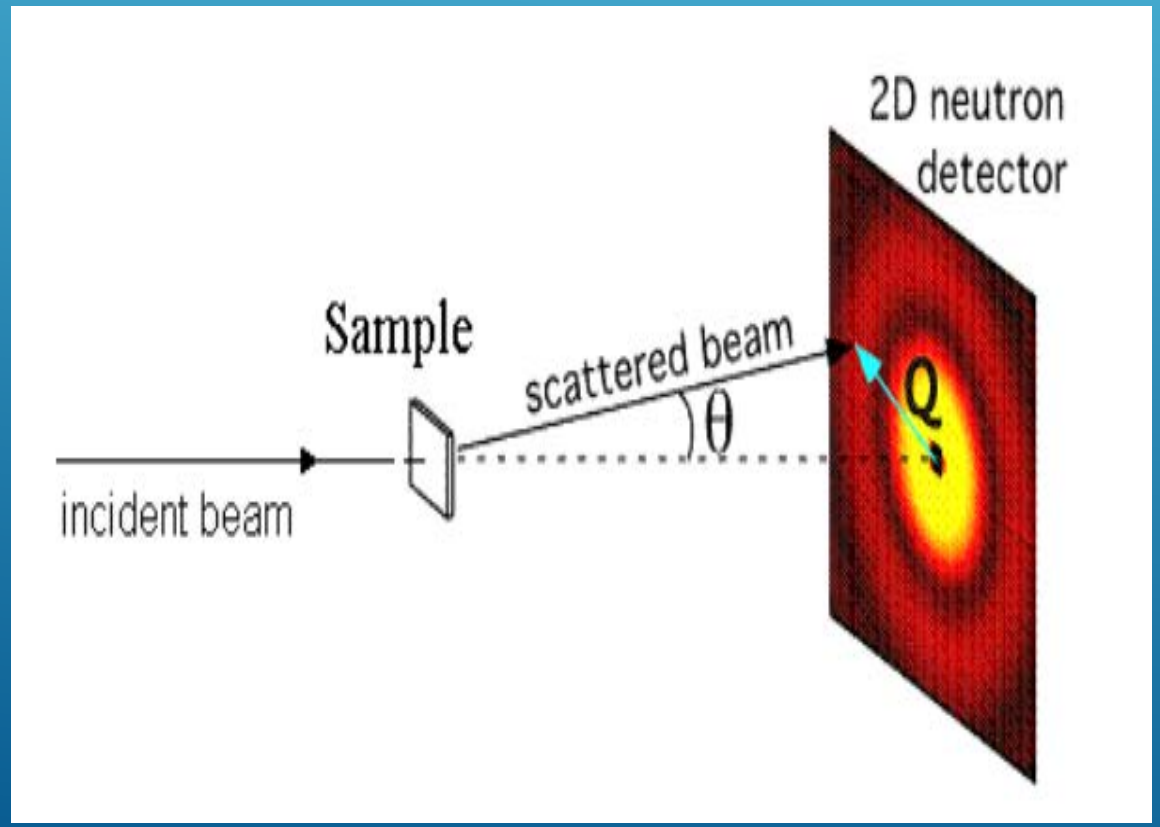
- Prepared stock insulin solution in D_2O and determined insulin concentration by using Spectrophotometer
- Made stock Zn-insulin solution by adding Zn to insulin molar ratio 1:3
- Prepared different concentrations of Zn- Insulin solutions (0.6 mM, 1.2 mM) with 30 mM and 150 mM NaCl
- Additionally, prepared 0.6 mM, 1.2 mM, and 1.8 mM Zn- Insulin solutions with 450 mM NaCl

* Used D_2O as a solvent for the preparation of all stock solutions

MEASUREMENT METHODS USED

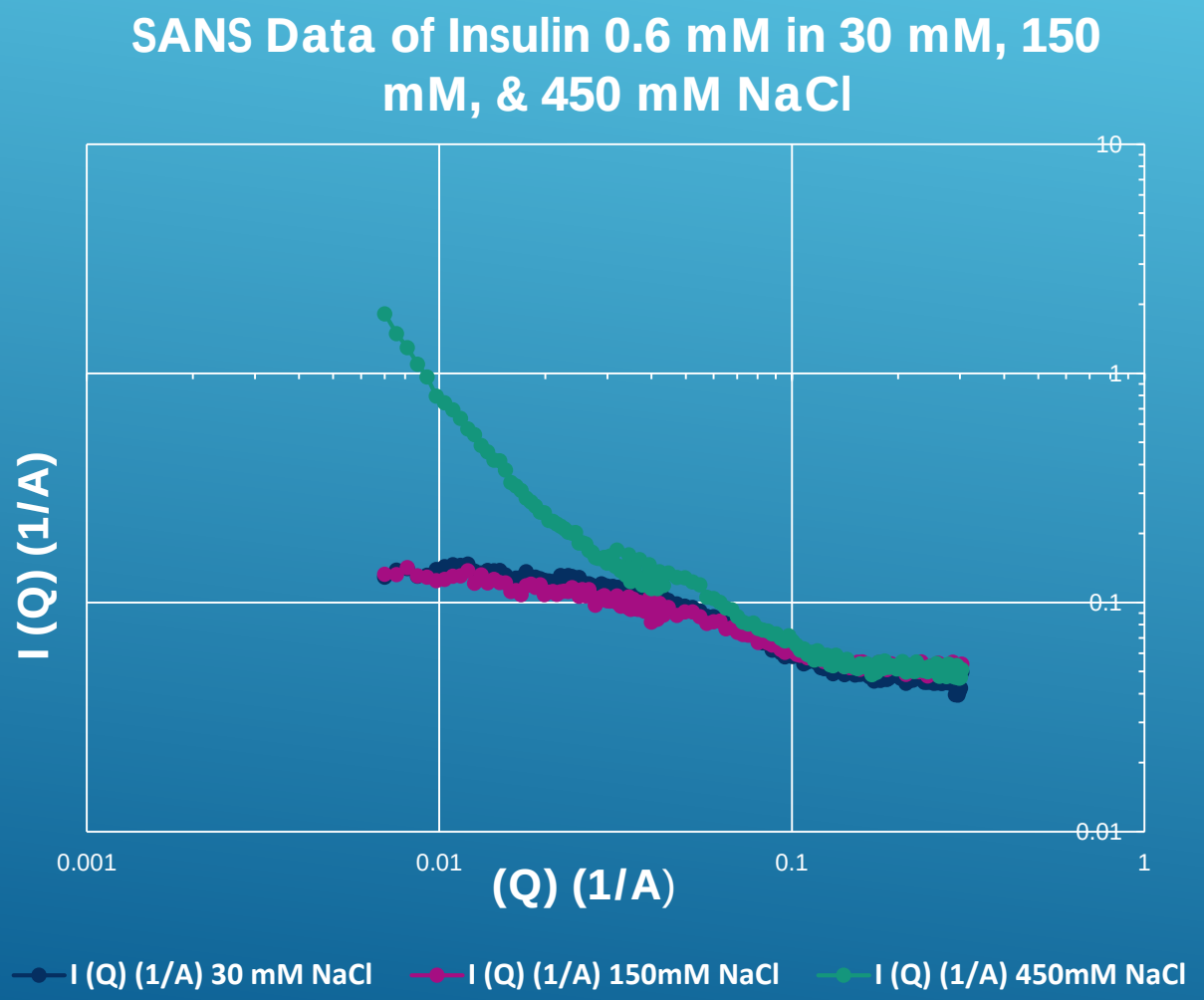
1. Small Angle Neutron Scattering (SANS) measurement
2. Rheological measurement
3. Rheological-Small Angle Neutron Scattering (Rheo-SANS) measurement

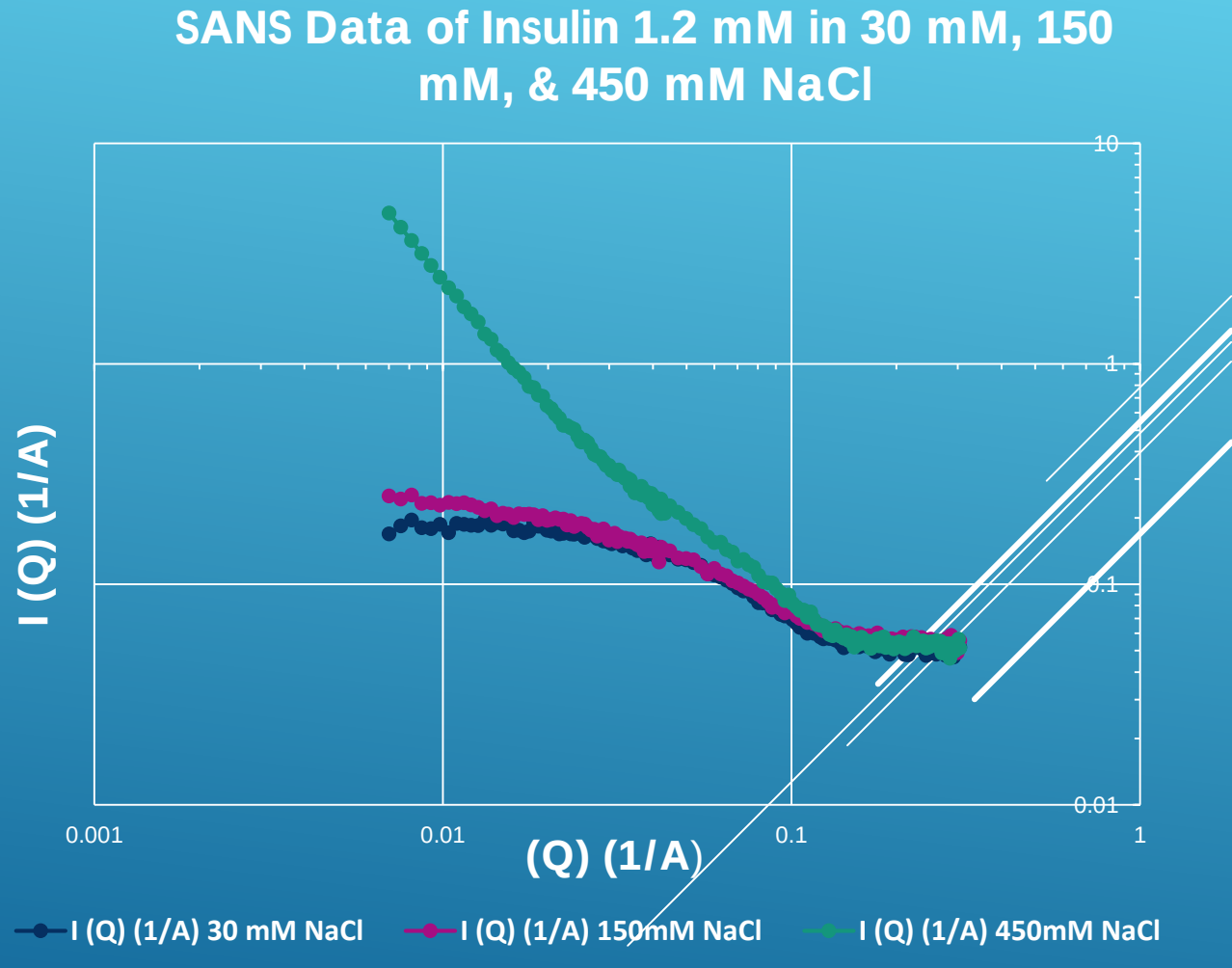
SANS



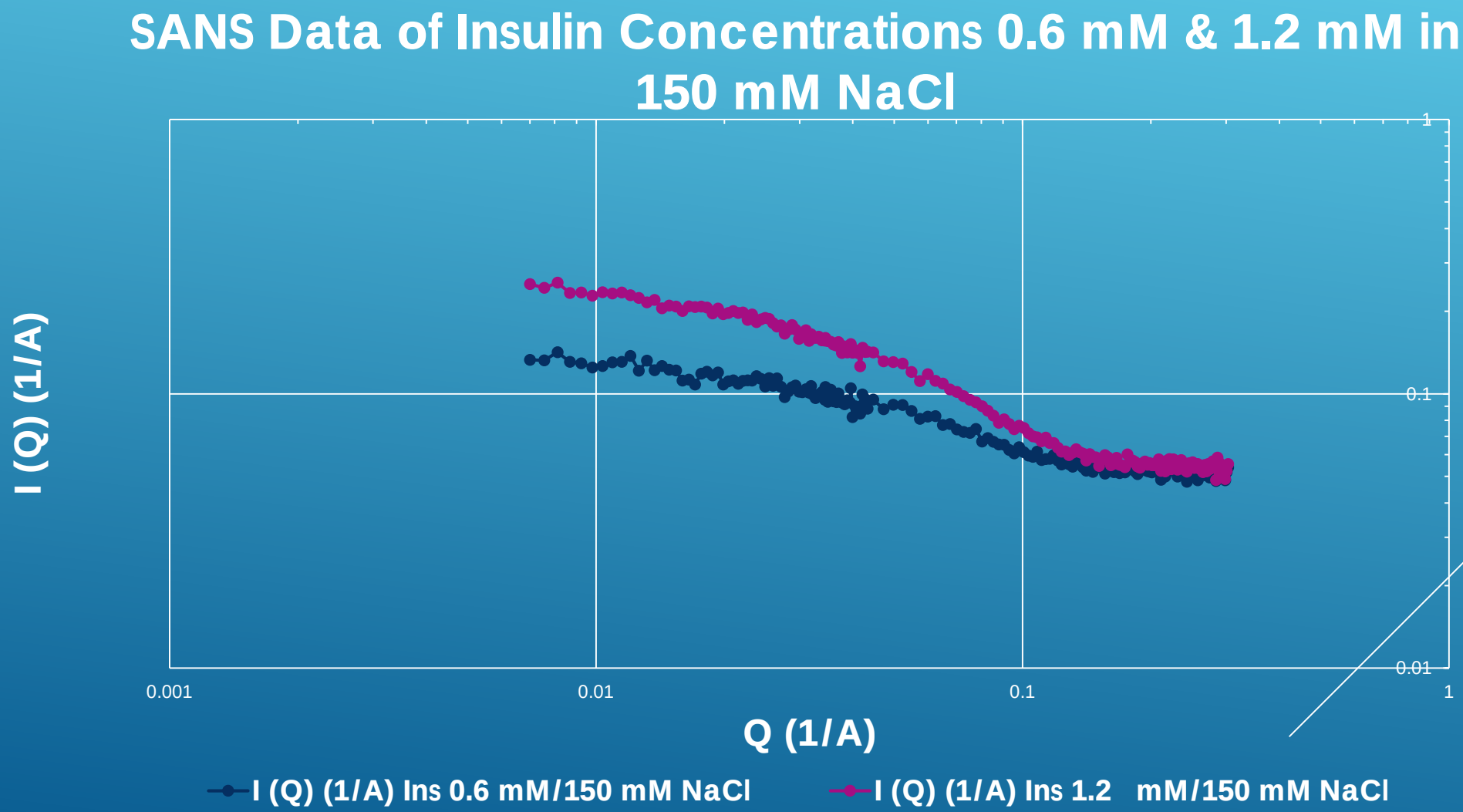
Rheometer

SANS RESULT BY USING ZINC-INSULIN 0.6 & 1.2 MILLIMOLAR WITH DIFFERENT SALT CONCENTRATIONS

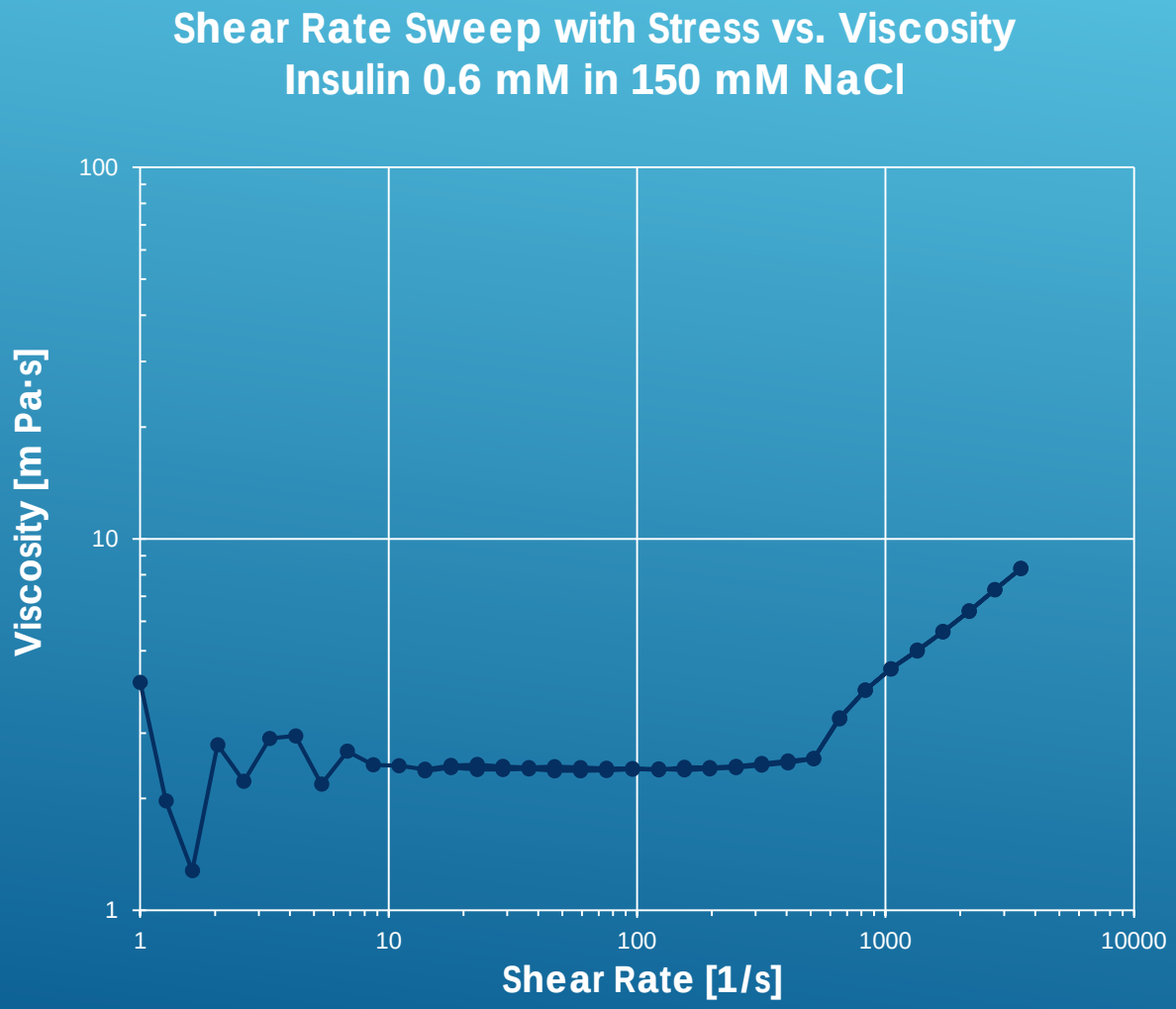


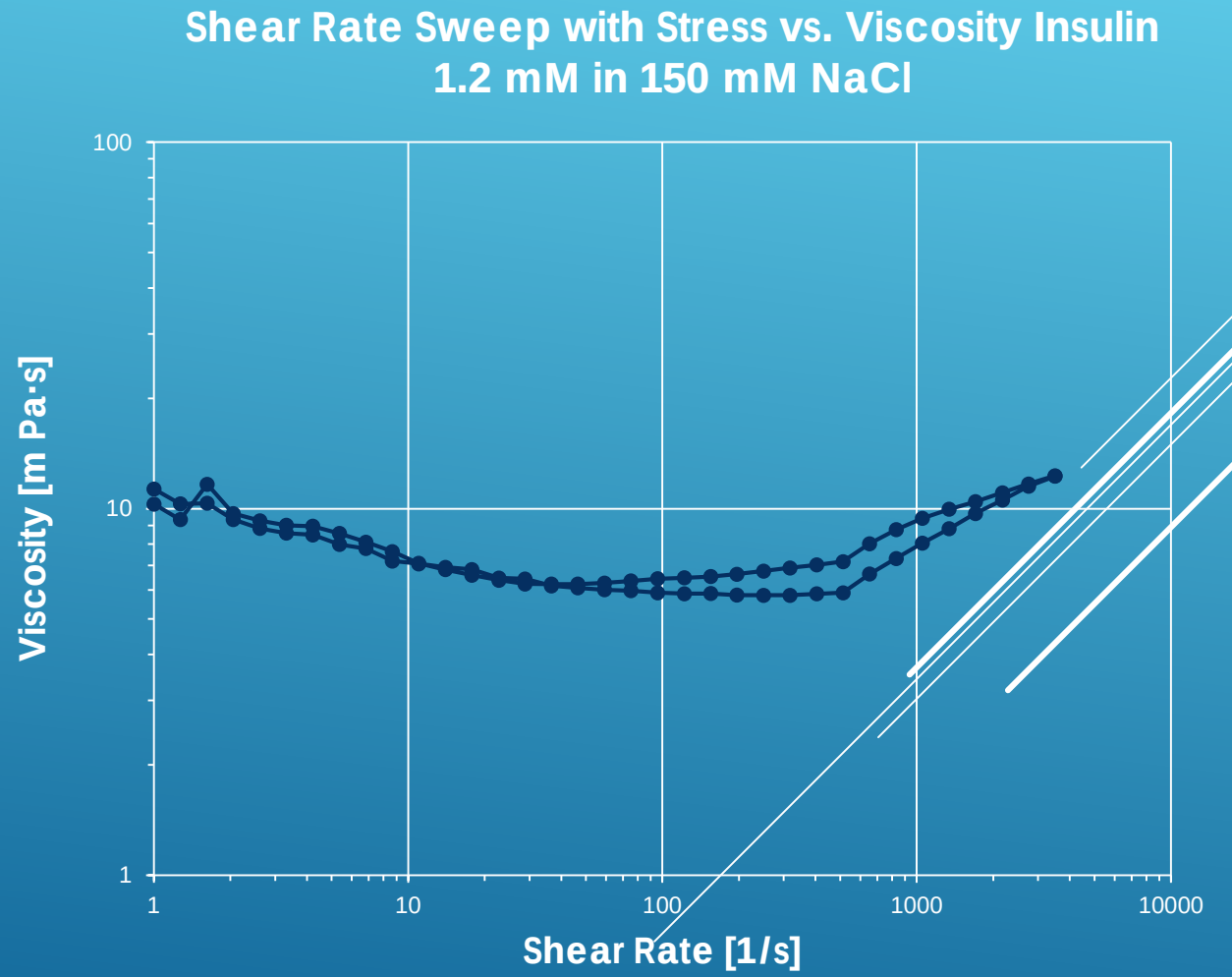


SANS RESULT BY USING ZINC-INSULIN 0.6 & 1.2 MILLIMOLAR WITH 150 MILLIMOLAR OF SALT

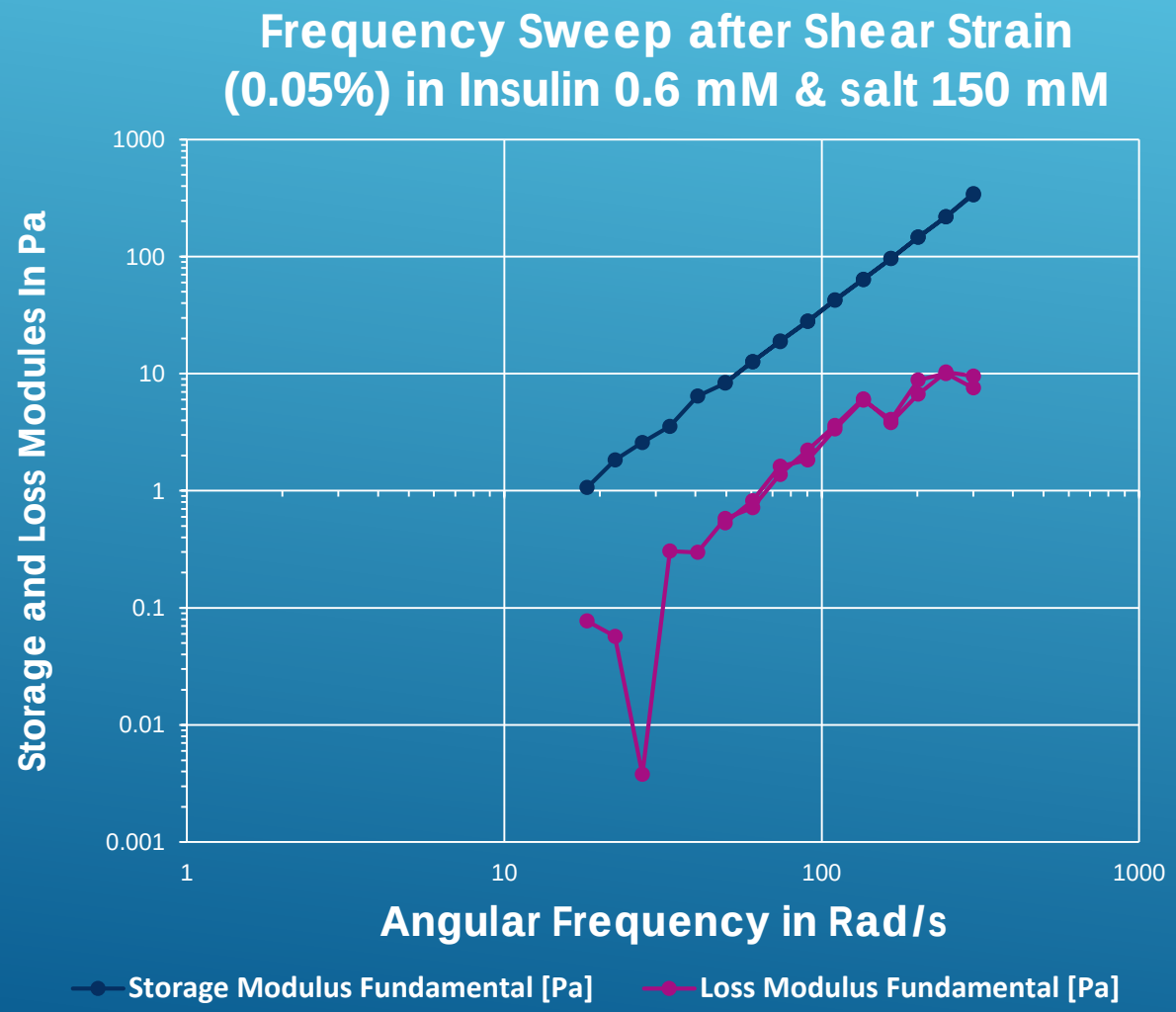


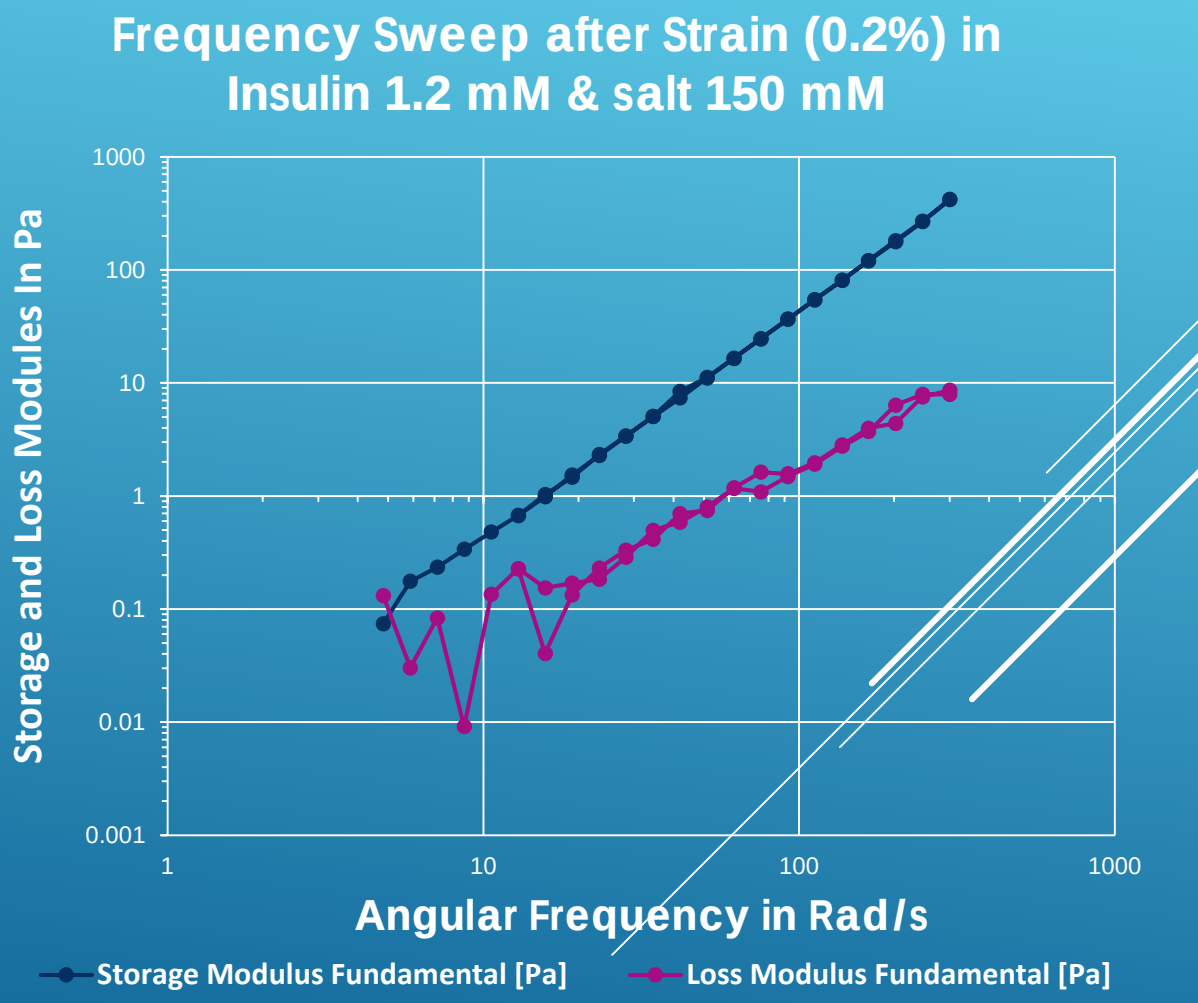
RHEOMETRIC RESULT AFTER SHEAR STRESS BY USING ZINC-INSULIN CONCENTRATIONS OF 0.6 & 1.2 MILLIMOLAR WITH 150 MILLIMOLAR SALT





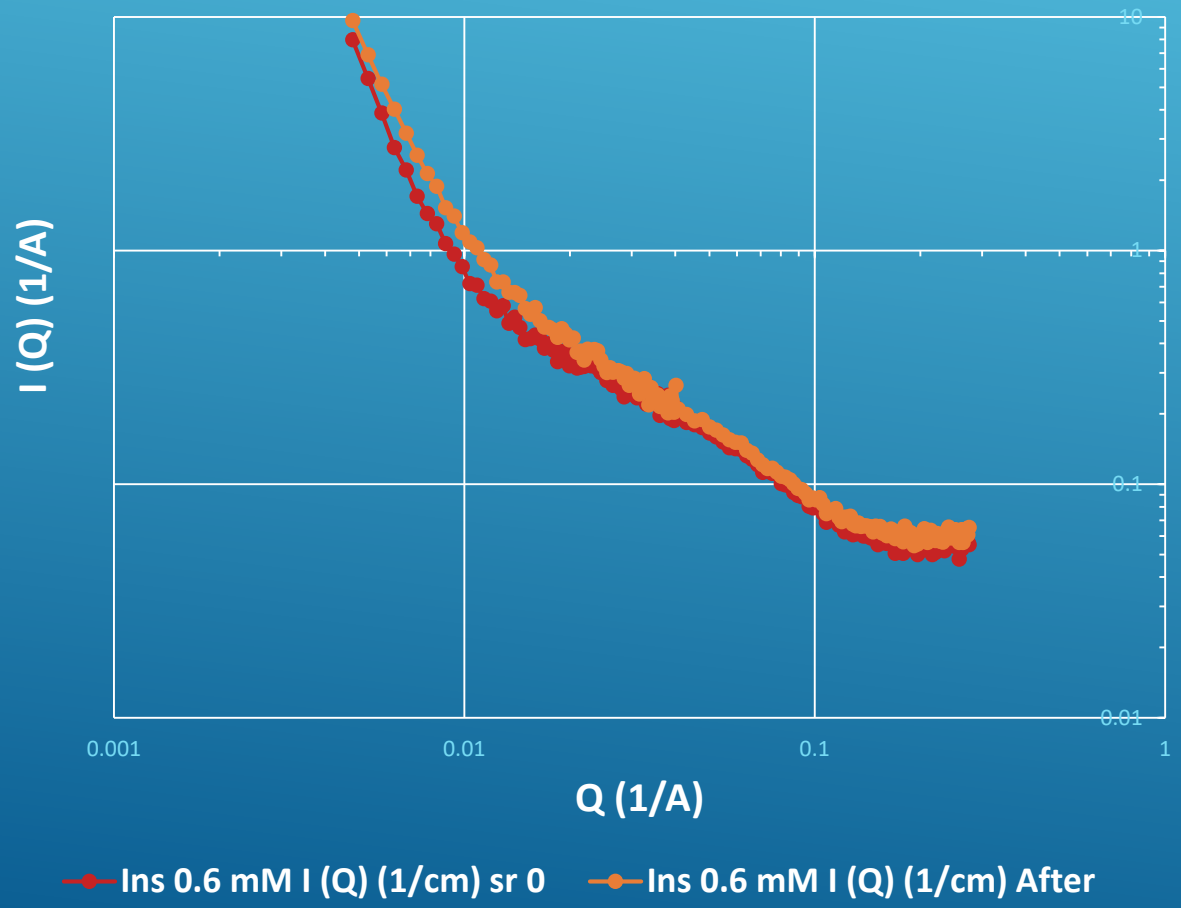
FREQUENCY SWEEP AFTER SHEAR STRAIN BY USING ZINC-INSULIN CONCENTRATIONS OF 0.6 & 1.2 MILLIMOLAR WITH 150 MILLIMOLAR SALT



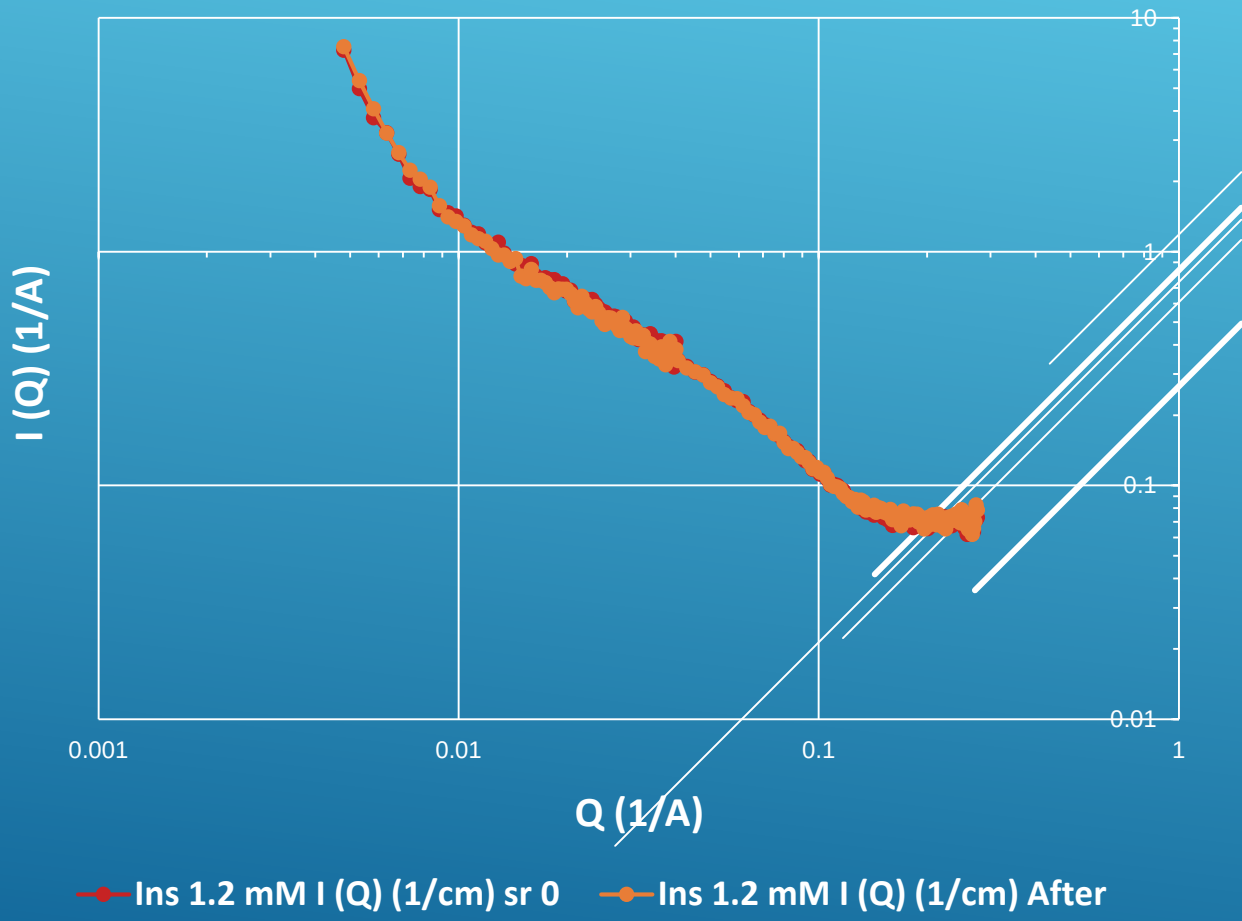


RHEO -SANS RESULT BEFORE & AFTER SHEAR STRAINS BY USING ZINC-INSULIN CONCENTRATIONS OF 0.6 & 1.2 MILLIMOLAR WITH 150 MILLIMOLAR SALT

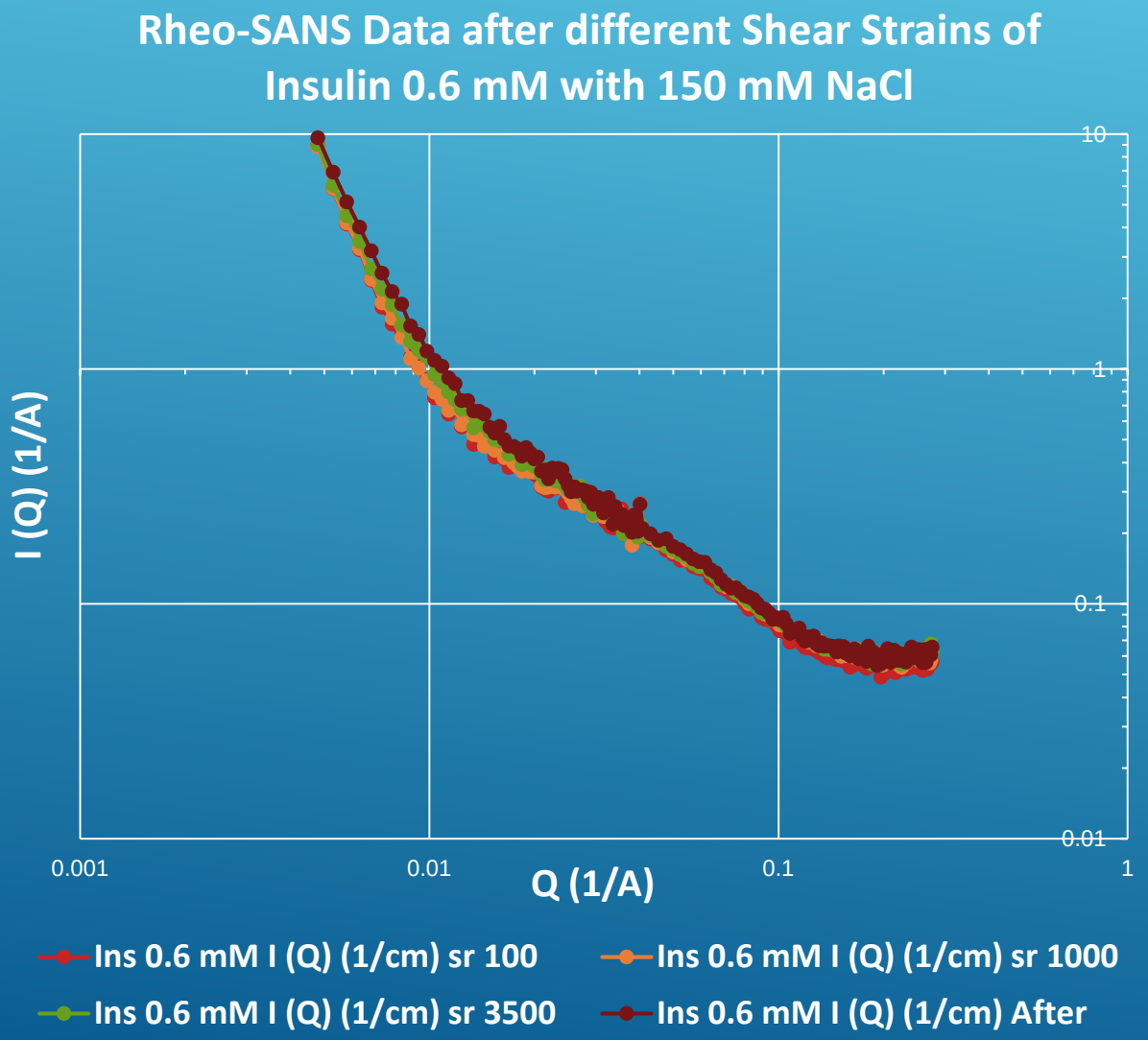
Rheo-SANS Data before and after Shear Strain of Insulin 0.6 mM with 150 mM NaCl

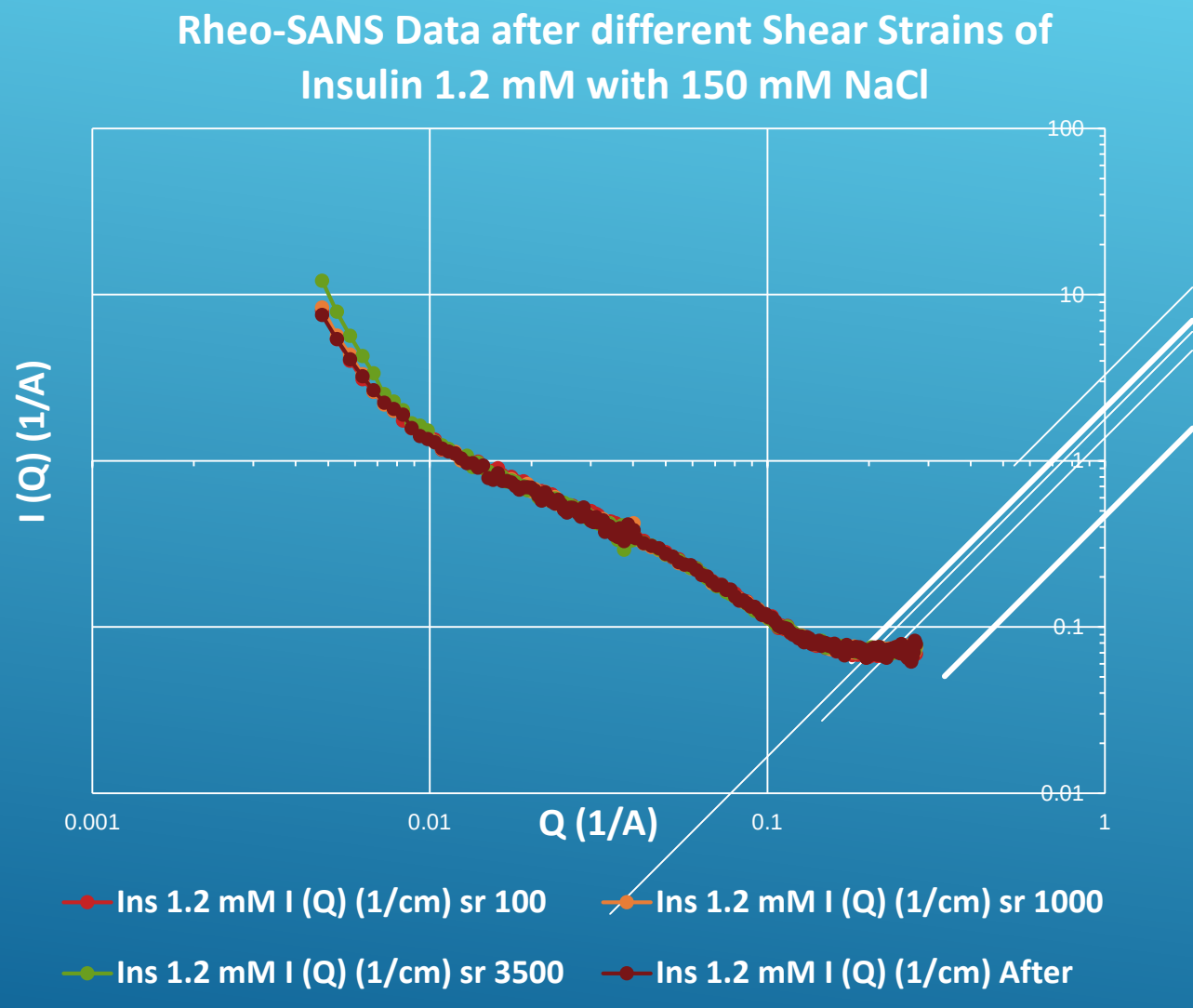


Rheo-SANS Data before and after Shear Strain of Insulin 1.2 mM with 150 mM NaCl

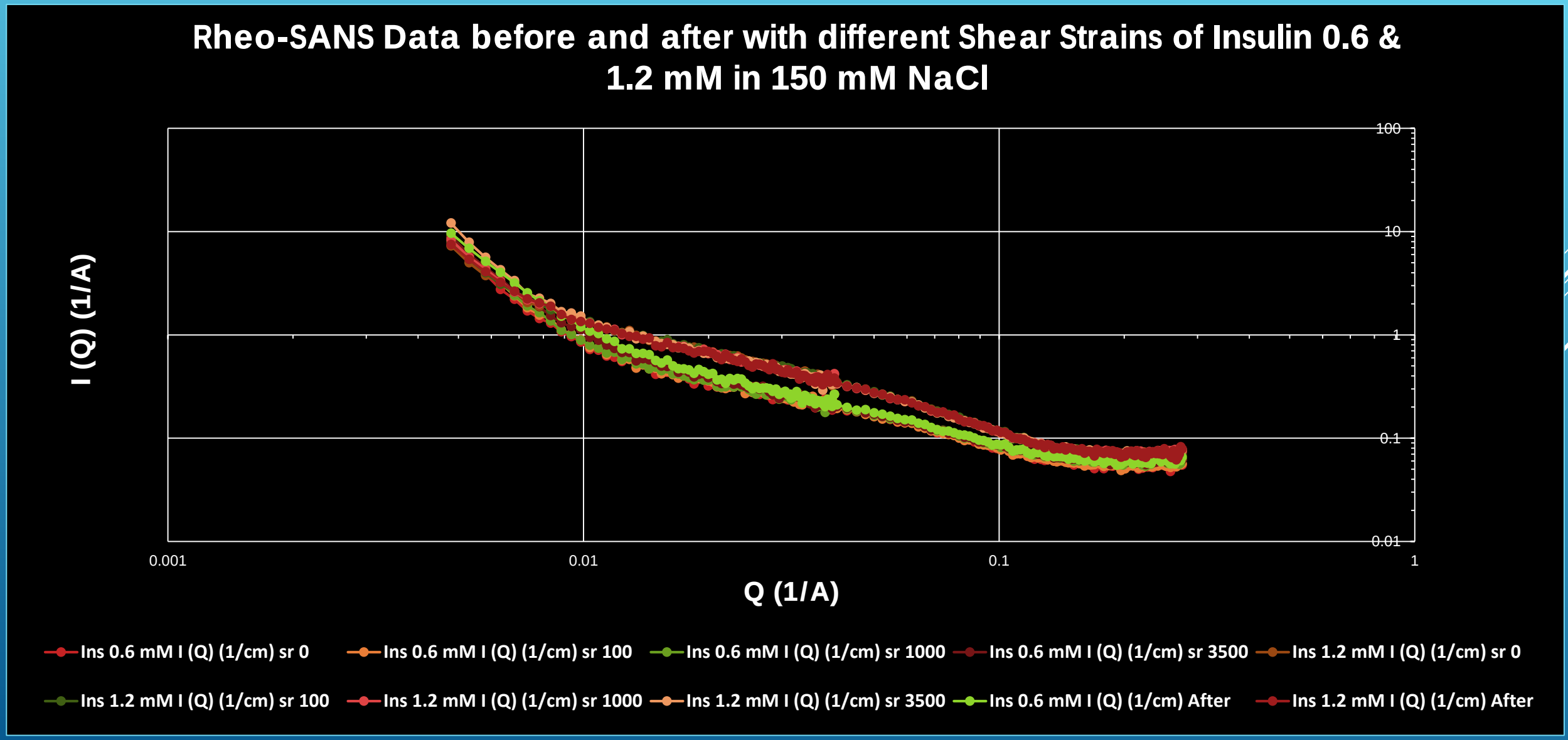


RHEO -SANS RESULT AFTER DIFFERENT SHEAR STRAINS BY USING ZINC-INSULIN CONCENTRATIONS OF 0.6 & 1.2 MILLIMOLAR WITH 150 MILLIMOLAR SALT





RHEO -SANS RESULT BEFORE & AFTER DIFFERENT SHEAR STRAINS BY USING ZINC-INSULIN CONCENTRATIONS OF 0.6 & 1.2 MILLIMOLAR WITH 150 MILLIMOLAR SALT



CONCLUSION

- SANS result indicates the possibility of structural change of insulin is occurring at higher salt concentration compared to lower salt concentrations. It is possible the insulin undergoing structural/conformational change like cylindrical long-rod formation at higher salt.
- Rheological data depicts that it is most likely at higher shear rates, shear thickening transition occurs.
- Rheo-SANS result illustrates the cluster sample and increasing clustering with increasing shear.
- The zinc containing insulin analogs attached with fatty acid may have potential therapeutic effect for diabetes people, as a prolonged duration of insulin supplement.

ACKNOWLEDGEMENTS

I would like to acknowledge and thanks Grethe Jensen from NCNR, NIST for her help regarding this research project

I would also like to extend my thanks to Yamali Hernandez for providing support to continue this project here at NCNR laboratory

Finally, I am thankful to Dr. Briber from UMD, Department of Materials Science & Engineering providing the financial support to continue this research project

Thank you!