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Sustainable Nanotechnology Organization

Research | Education | Responsibility

Inhibiting Protein Amyloid Aggregation with Nanoparticles

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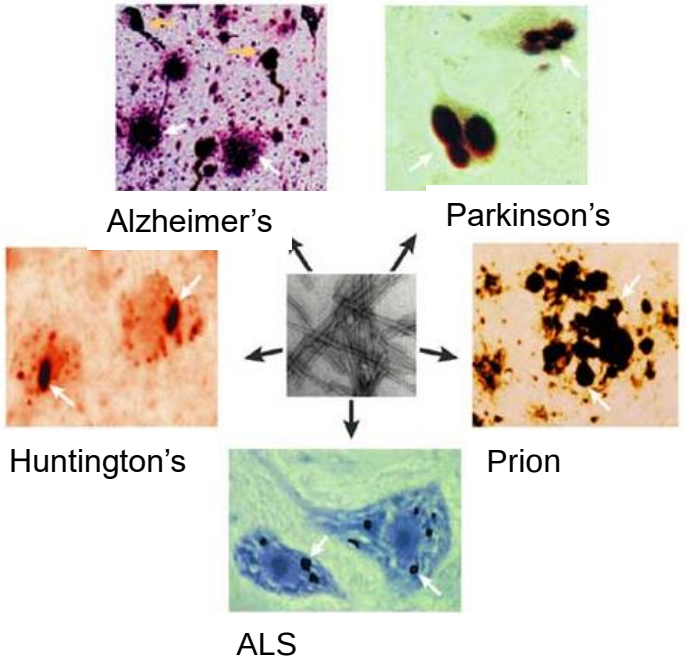
Protein misfolding diseases

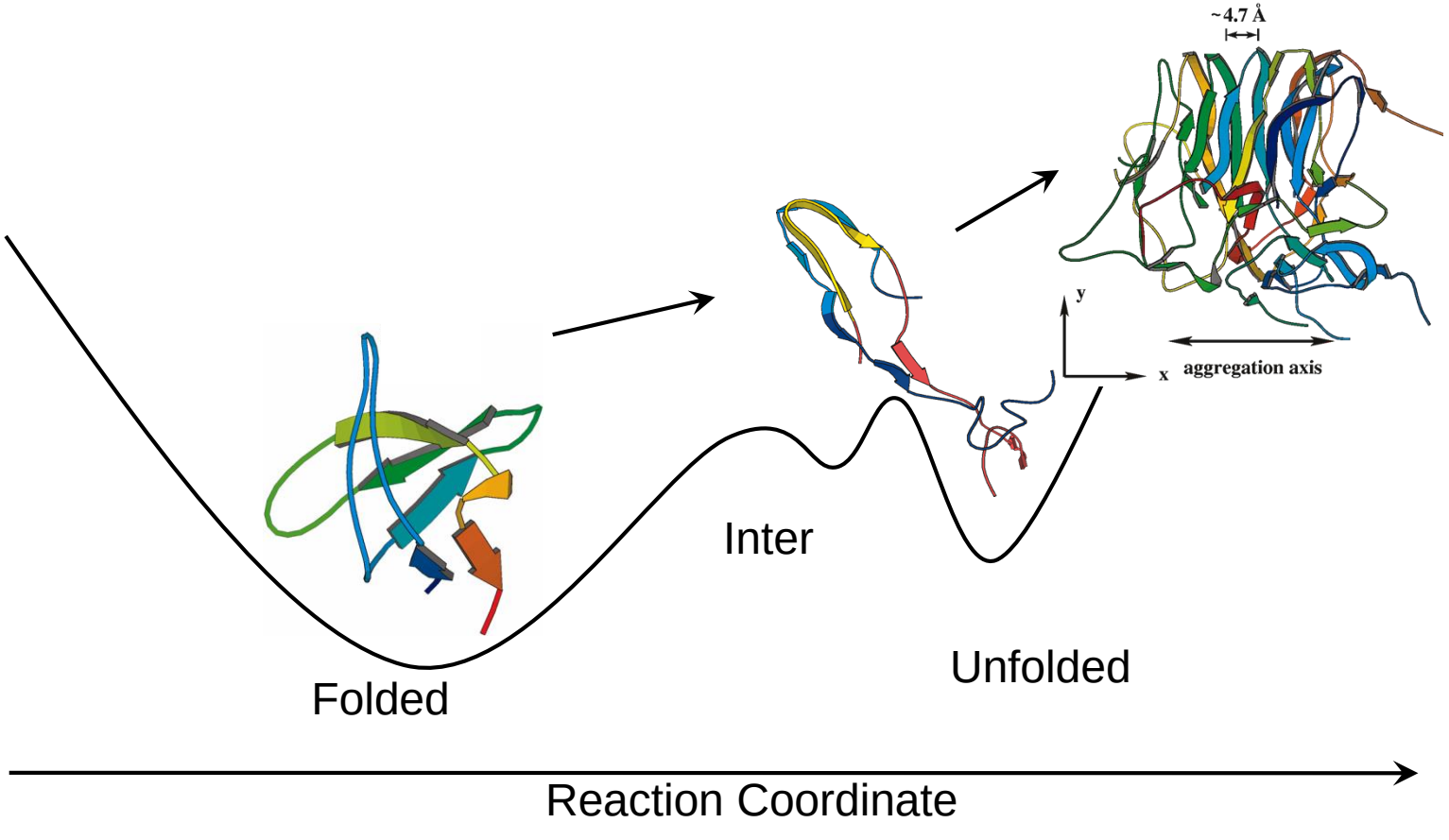
- An increasing list of protein misfolding diseases
  - Alzheimer's disease – Aβ
  - Parkinson's disease – α-synuclein
  - Huntington's disease – huntingtin
  - Type-2 Diabetes – Islet Amyloid Polypeptide
  - Amyotrophic Lateral Sclerosis – SOD1

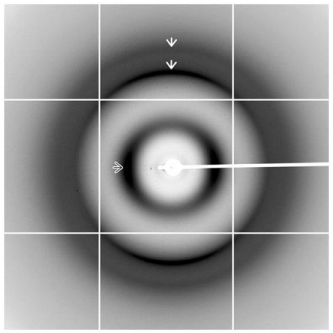
- Common hallmarks
  - Fibrillar aggregates – regular structures from different precursors
  - Long process; rare nucleating events
  - Symptoms typically appear in mid to later life (50-70 years)

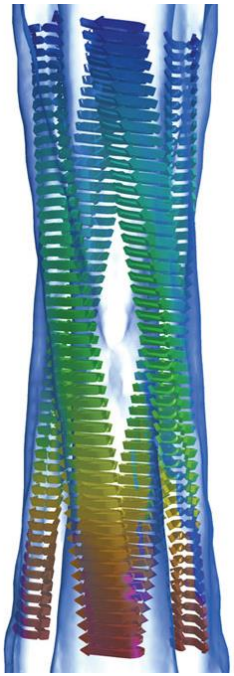


Soto C., Nature Reviews Neuroscience, 4:49-60 (2003)
Carulla, et al *PNAS.* 106(19):7828 (2009)

Amyloid fibril – the common cross-beta structure

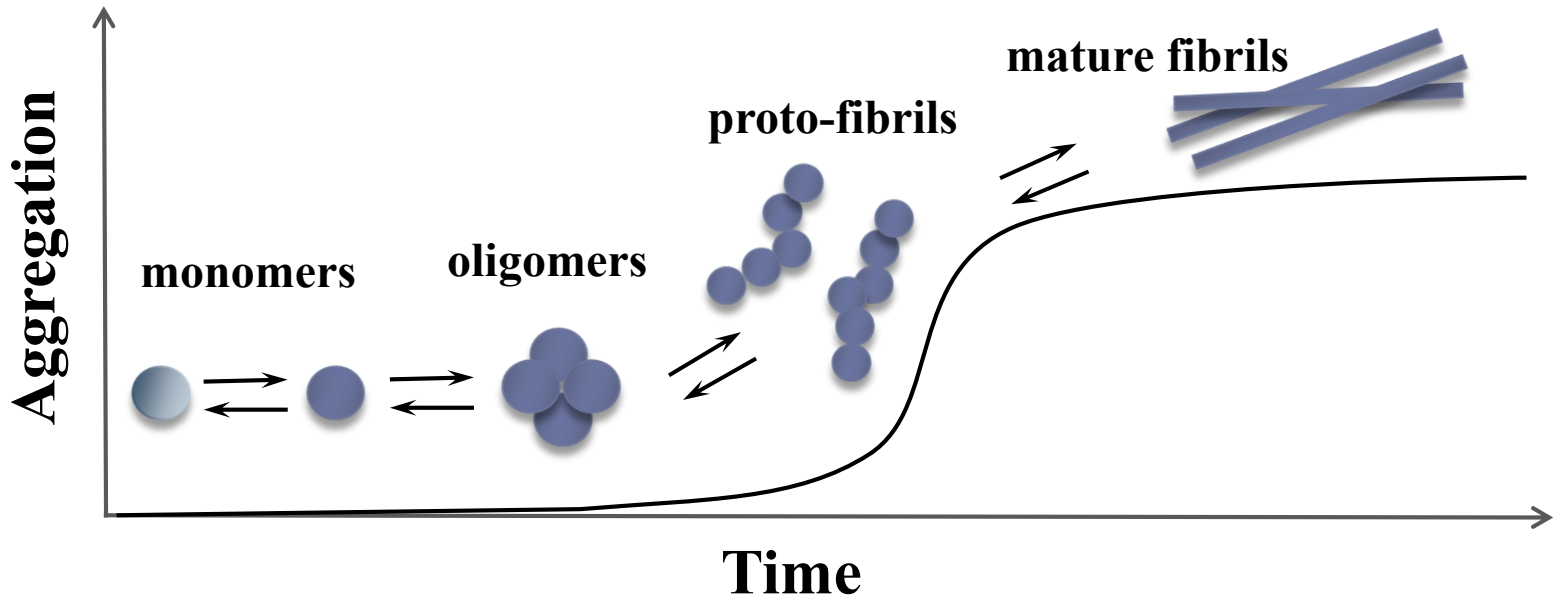






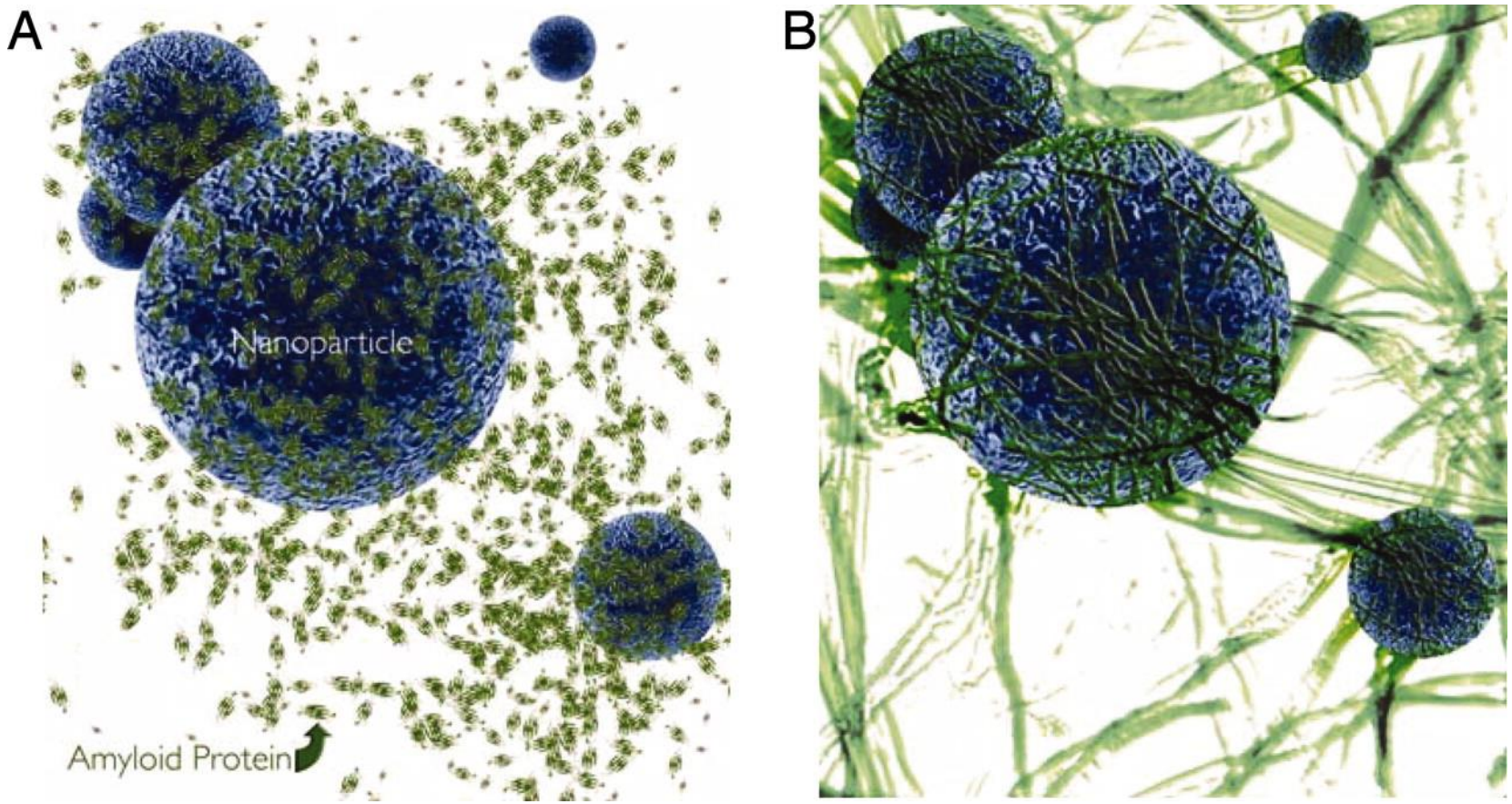
Ding F., Dokholyan N.V., Buldyrev S.V., Stanley H.E. and Shakhnovich E.I., J Mol Biol, 324, 851-857 2002

Nucleation Process – Sigmoidal Kinetics



Inhibitor design – targeting each of the step

Nanoparticles as catalysts for protein fibrillation



Linse S. et al, PNAS 104:8691-6, 2007
Colvin VL and Kulinowski KM, PNAS 104:8679-8, 2007

Aggregation promoting or inhibiting – the contrasting effects of NPs?

Nanoparticles	Proteins	Effects on Amyloid Aggregation
Multi-walled CNT, QDs, Copolymer NP, CeO_2 NP[23]	β-2 microglobulin	Promotion
TiO_2 NP[43]	Aβ	
AuNP[75]	lysozyme	
Graphene oxide[67]	Aβ	Inhibition
AuNP[45]	Aβ	
CNT[42]	$Aβ_{16-22}$	
Carbon Dots[76]	Insulin	
Polymeric NP[68]	Aβ	
Polystyrene NP[37]	Aβ	Either promotion or inhibition

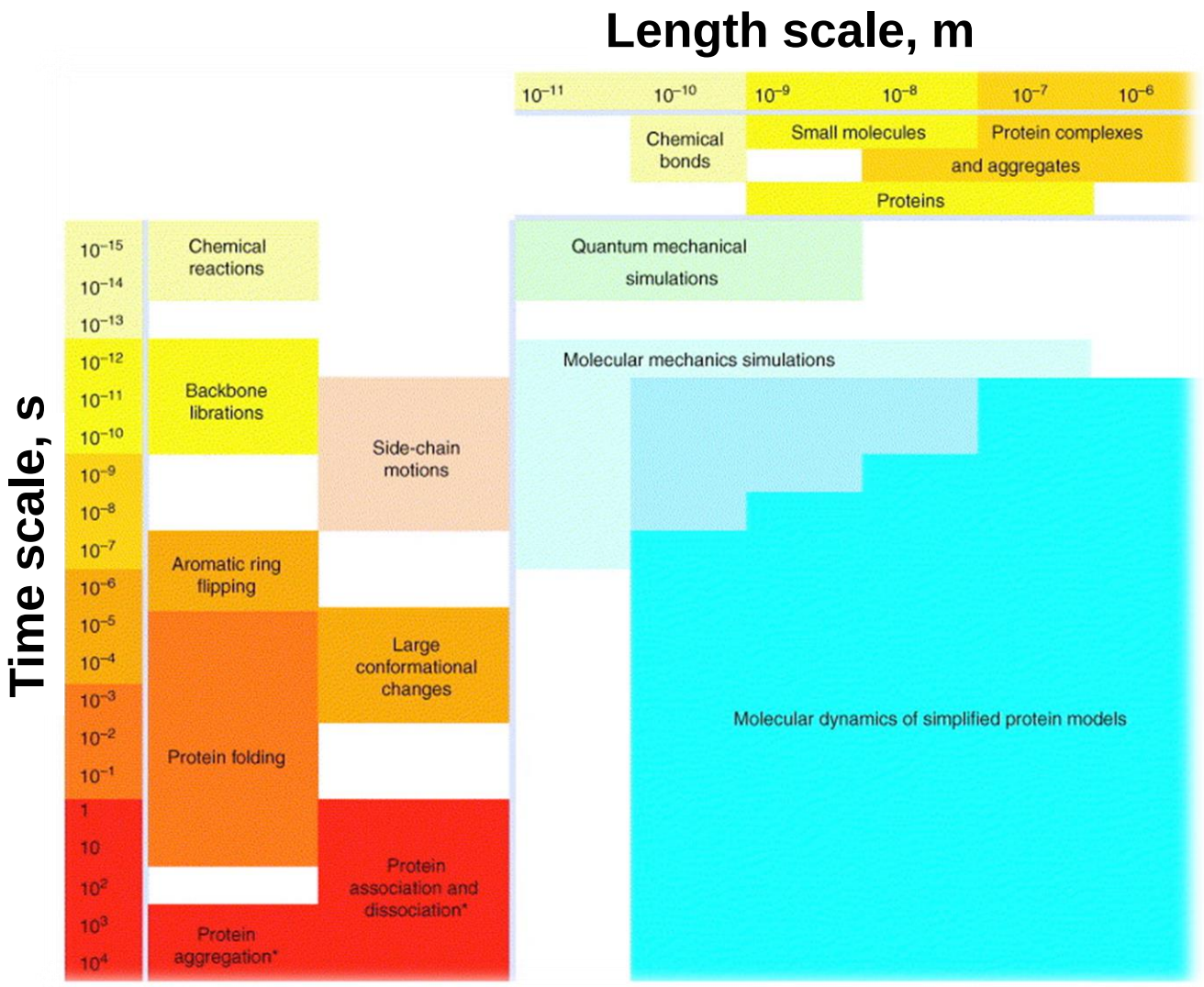
Q: What are the *determinants* of NPs and/or proteins for the complex and seemingly contrasting behaviors?

Objective – Amyloid-inhibiting nanomedicine

Outline

- Multiscale modeling approach
  - DMD simulations
  - Multiscale models
- Uncovering the effects of NPs on protein aggregation
  - Varying NP-Protein attractions
  - Competing aggregation in solution and on NP surface
  - A complete picture of protein aggregation influenced by NPs
- Applications of anti-amyloid Nanomedicine
  - Graphene oxide
  - Dendrimer

Challenges in computational modeling: multiscale modeling



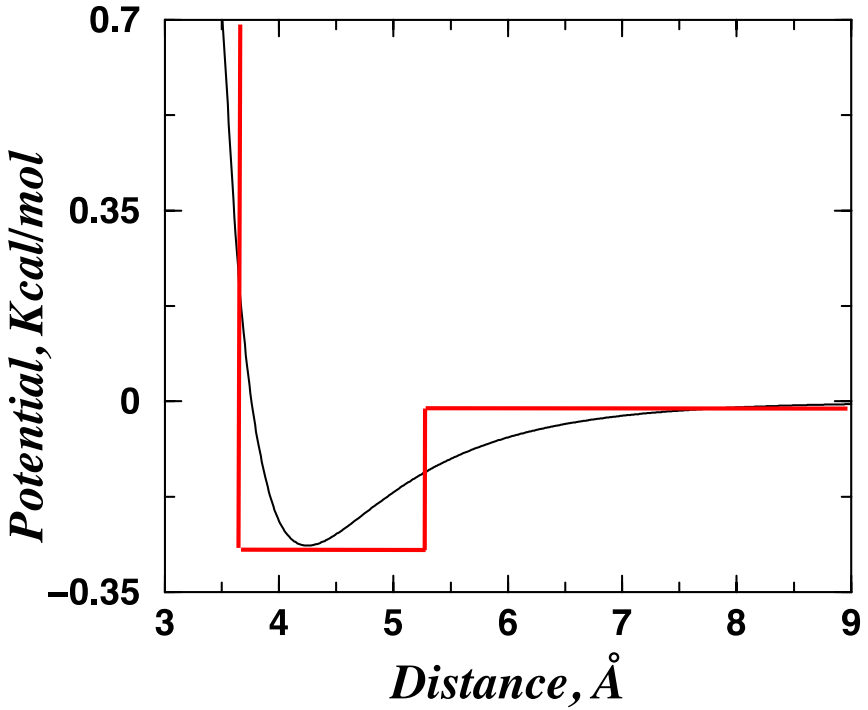
Large **gaps** of time and length scales between experimental observation and the underlying molecule system

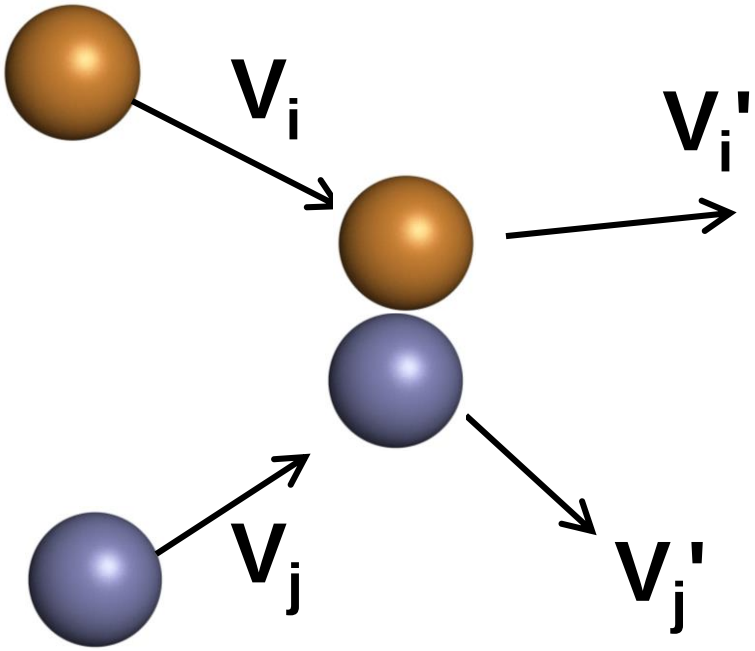
Approaches:

- Enhanced sampling methods
- Simplified protein models

Ding F. and Dokholyan N.V., *Trends in Biotechnology,* (2005)

Enhanced MD method: **DMD**





$$m\vec{a}_i = \sum_j \vec{F}_{ij}$$

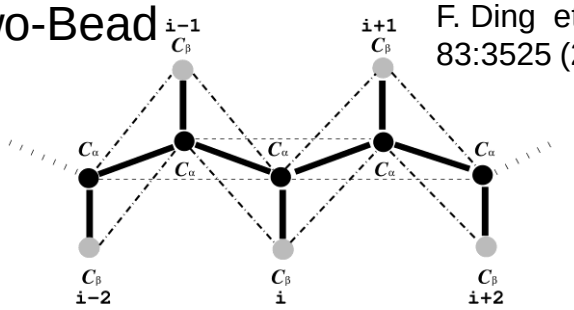
- **Dynamics become event-driven:**
  - **collision prediction,**
  - **sorting for next collisions,**
  - **updating the colliding atoms**

- Alder and Wainwright, J. Chem. Phys. 27:1208 (1957); Zhou Y and Karplus M, PNAS, 94, 14429 (1997); McCammon, J.A., Gelin, B.R. & Karplus, M. Nature 267, 585−590 (1977); Dokholyan NV et al., *Folding & Design,* (1998)

Multi-scale protein models

Two-Bead

F. Ding et al, *Biophys. J.*, 83:3525 (2002)

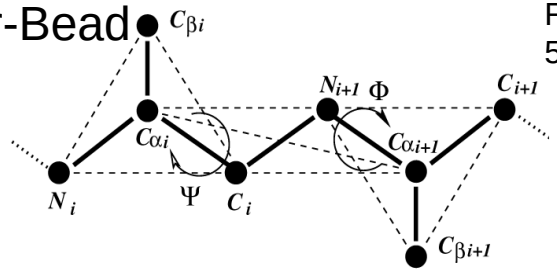


Time scale: ~*seconds-hours*

Applications: Protein folding/misfolding, Protein aggregation,

Four-Bead

F. Ding et al, *Proteins*, 53:220 (2003)

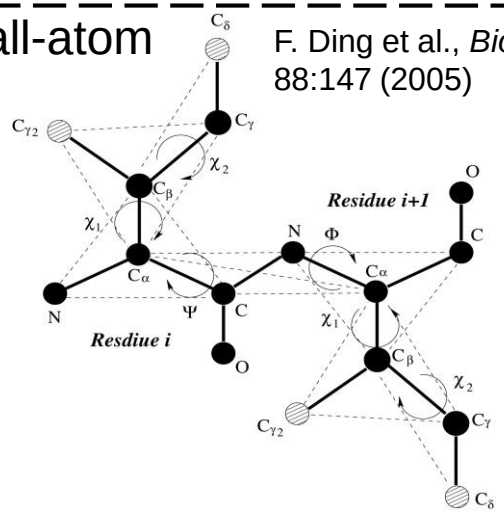


Time scale: ~*seconds*

Applications: 2nd structure transition, Protein folding/misfolding, Protein aggregation

Pseudo all-atom

F. Ding et al., *Biophy. J.*, 88:147 (2005)

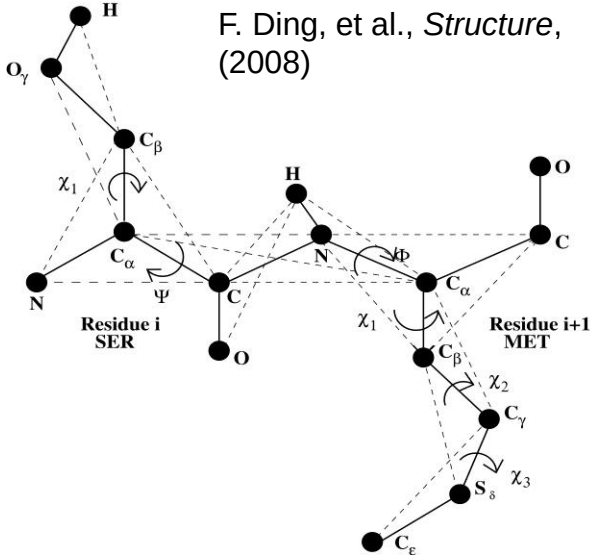


Time scale: ~*μs-ms*

Applications: Protein folding/misfolding, aggregation of short peptides

All-atom

F. Ding, et al., *Structure*, (2008)

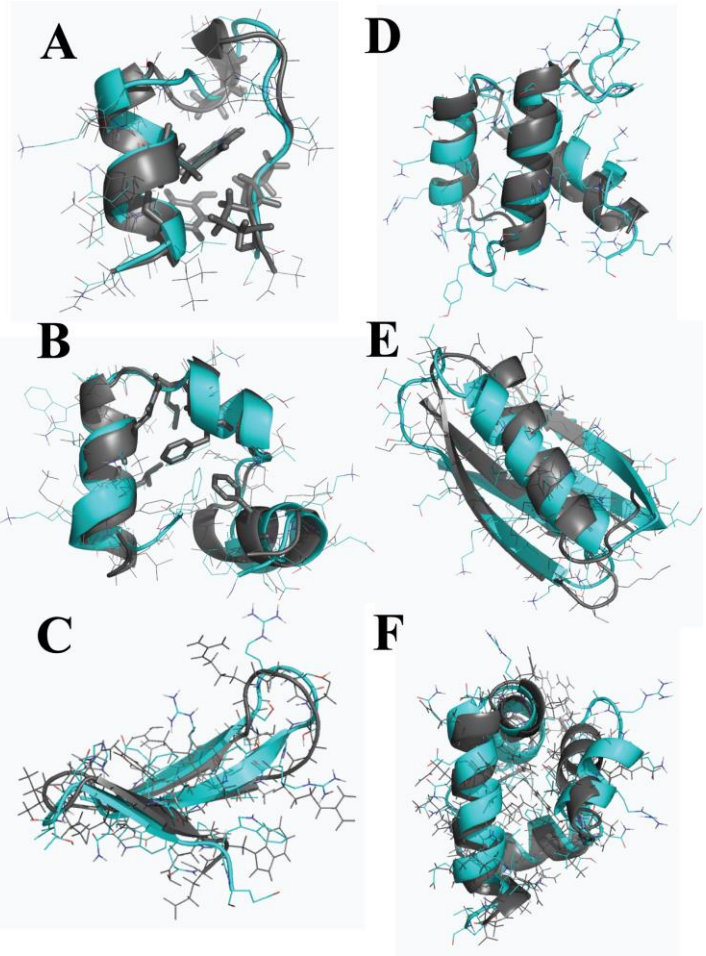


Time scale: ~*μs*

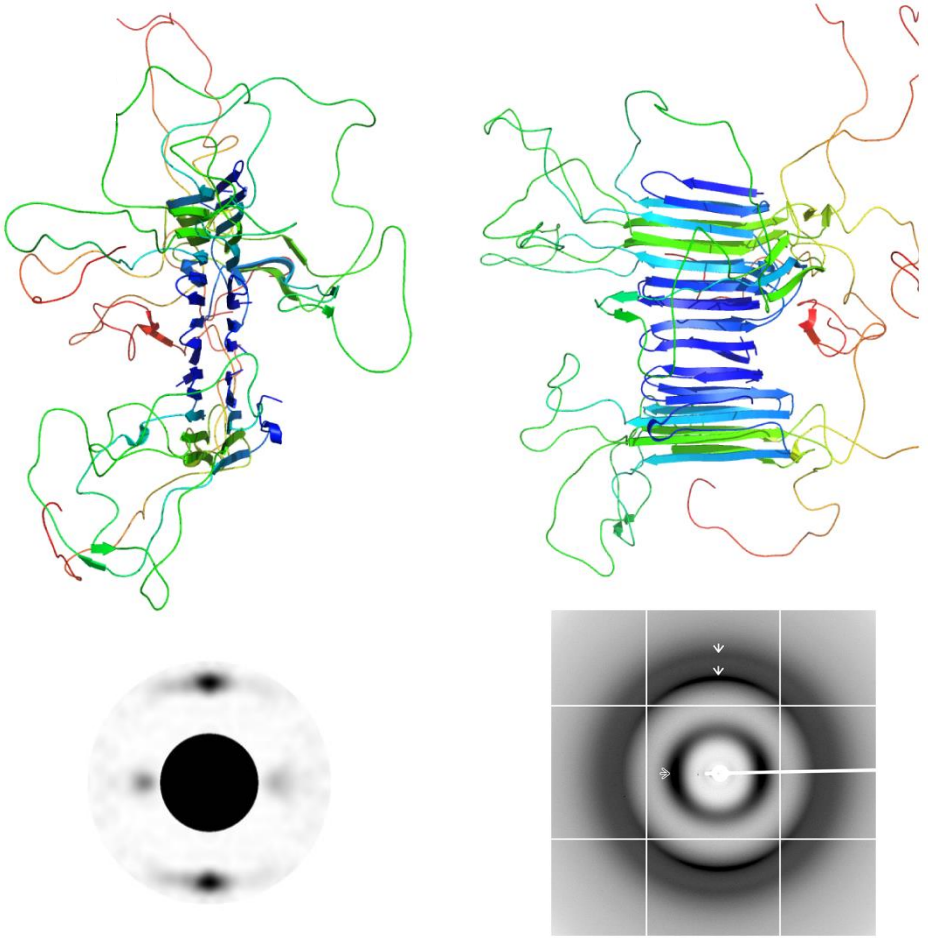
Applications: Folding of small proteins; near-native dynamics; and protein unfolding

Multiscale DMD simulations

Ab initio protein folding



Coarse-grained simulation of protein aggregation



F. Ding, D. Tsao, H. Nie and N.V. Dokholyan, *Structure* (2008)
F. Ding, Y. Furukawa, N. Nukina, and N.V. Dokholyan, *JMB(2012)*

Outline

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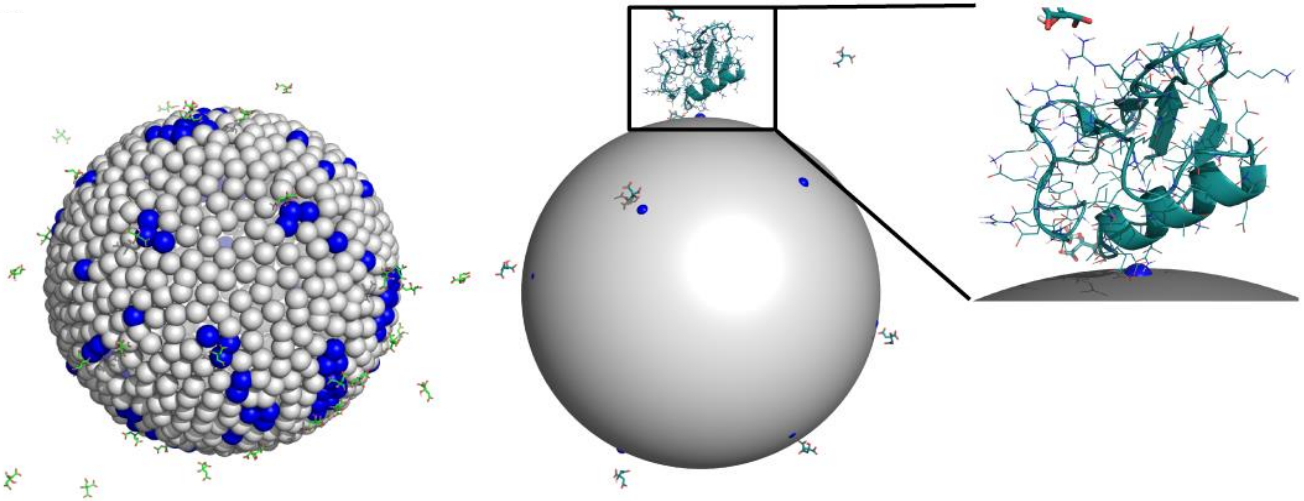
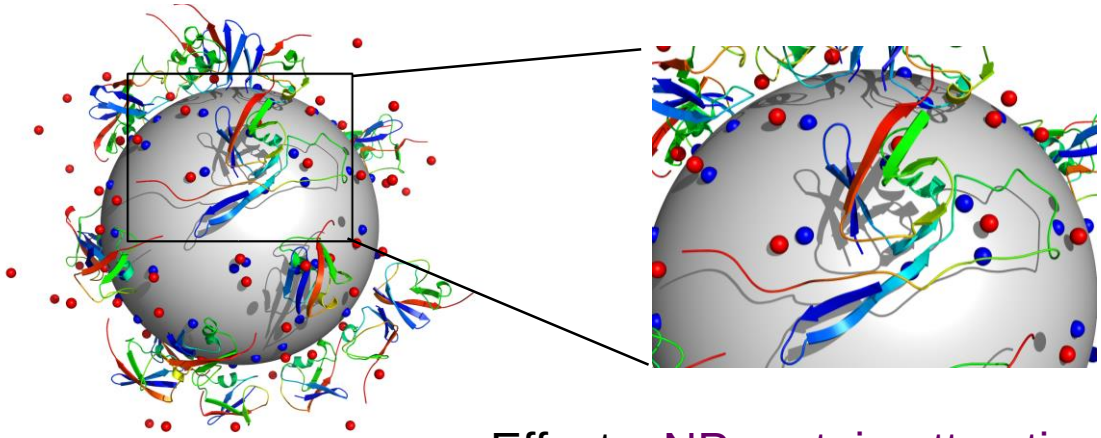
  - Graphene oxide
  - Dendrimer

Coarse-grained modeling of NPs

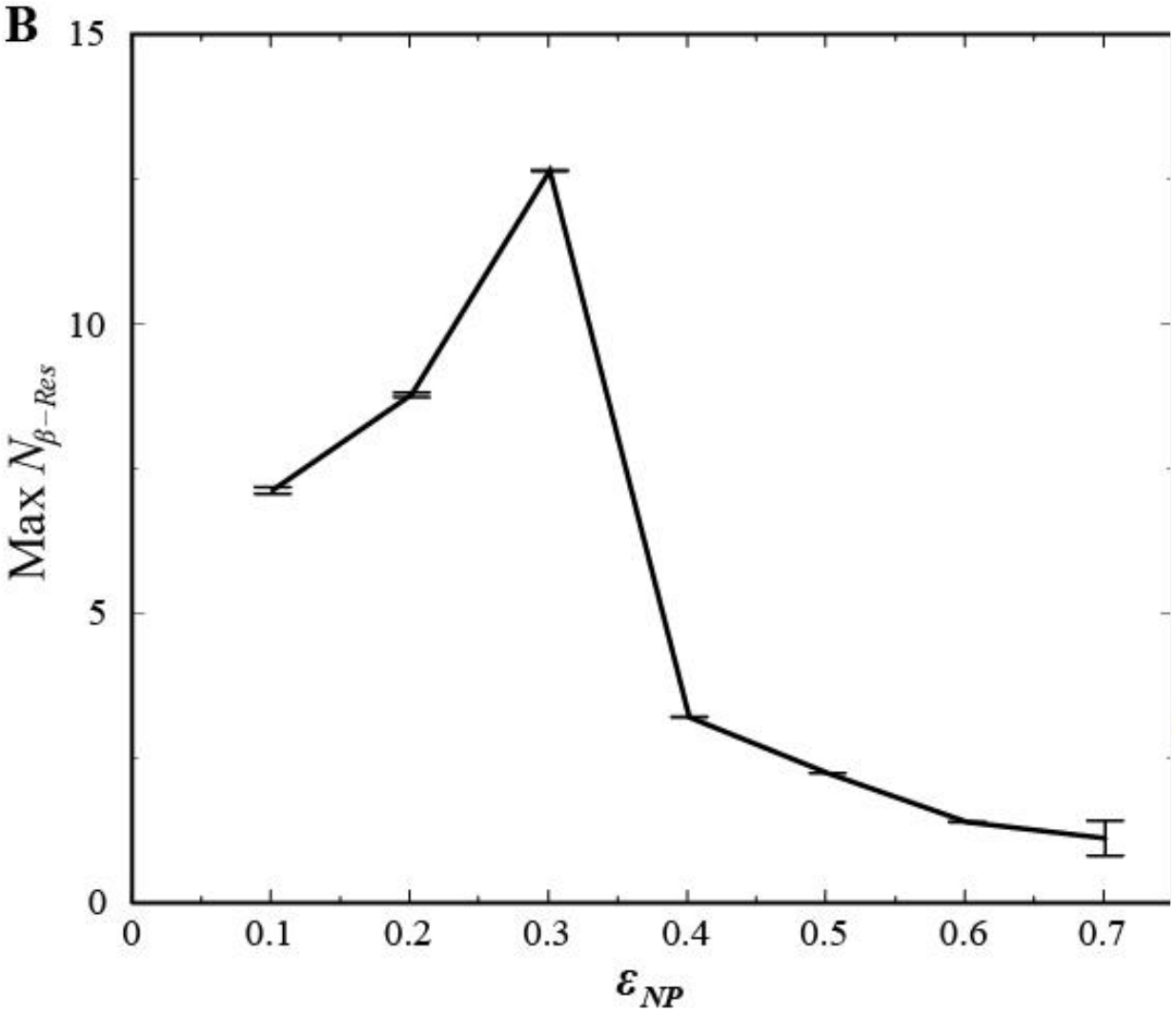


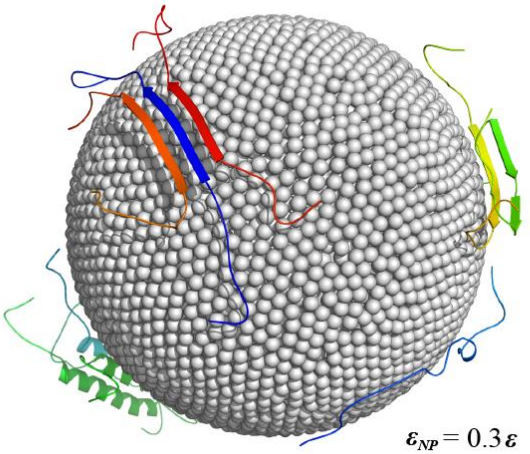


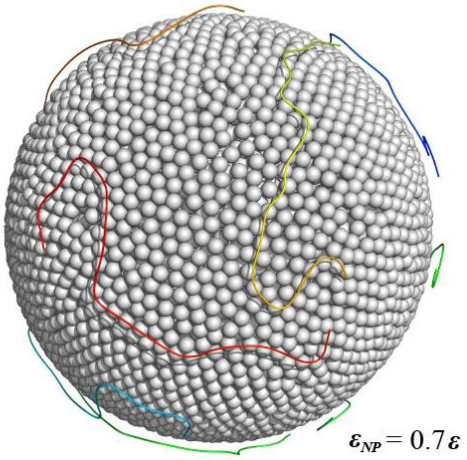
Effects: NP-protein attractions (affinities), relative concentrations, competition between bulk and surface, etc.

F. Ding et al, Nanoscale, 5:9162-9(2013)

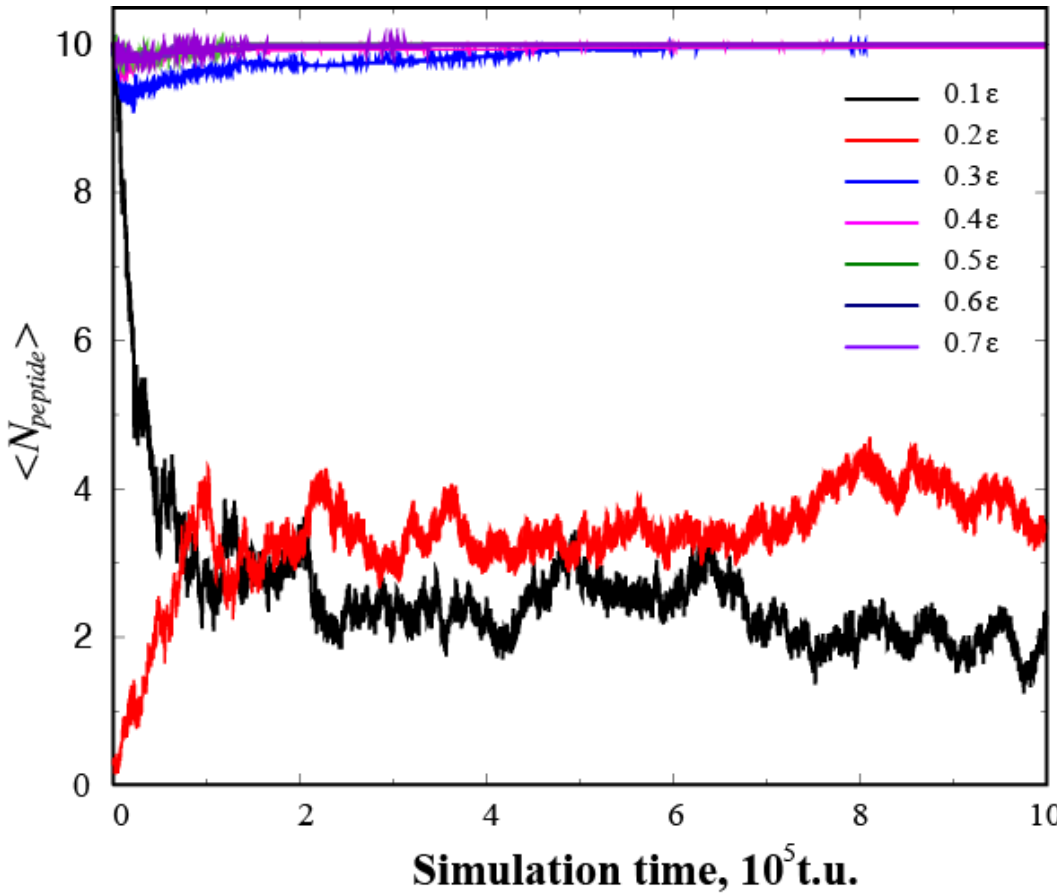
Complex effects of NP-Protein attractions on protein aggregation





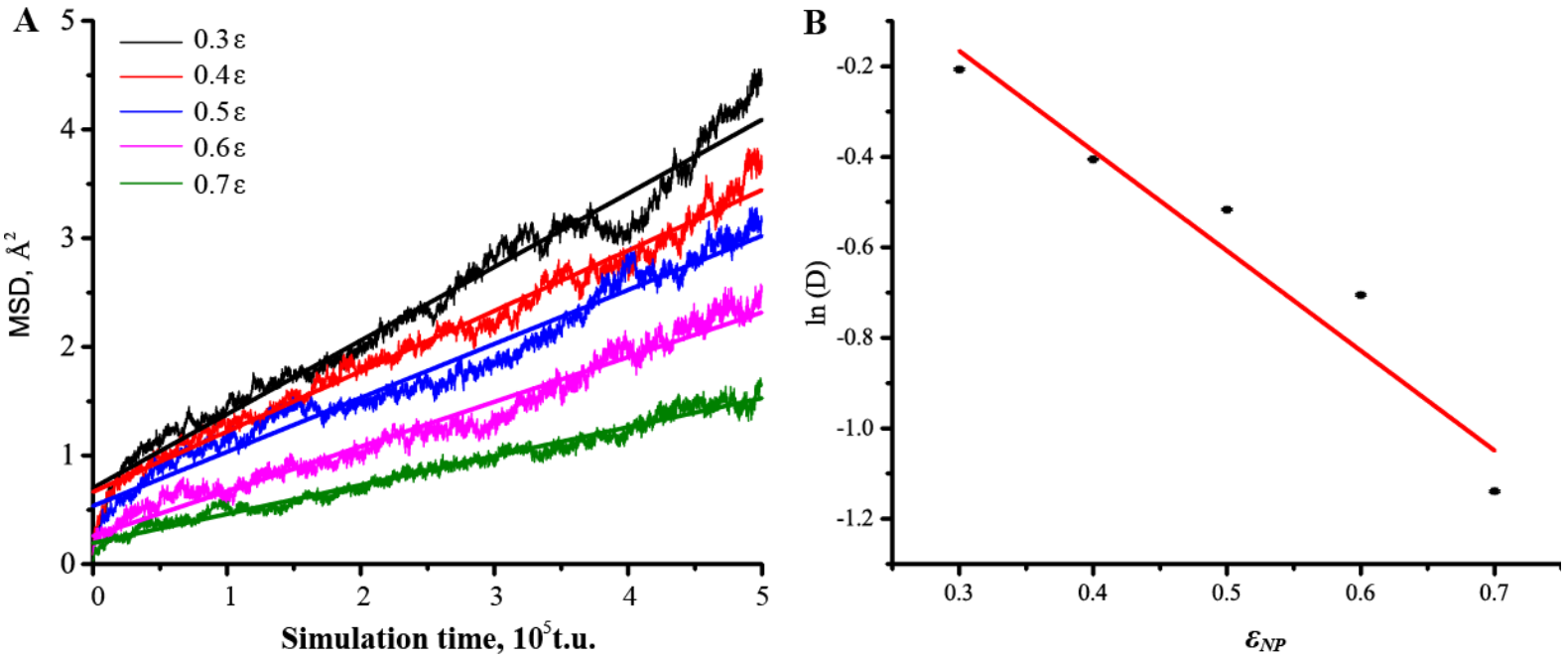


The dependence of protein surface concentration on NP-Protein attractions



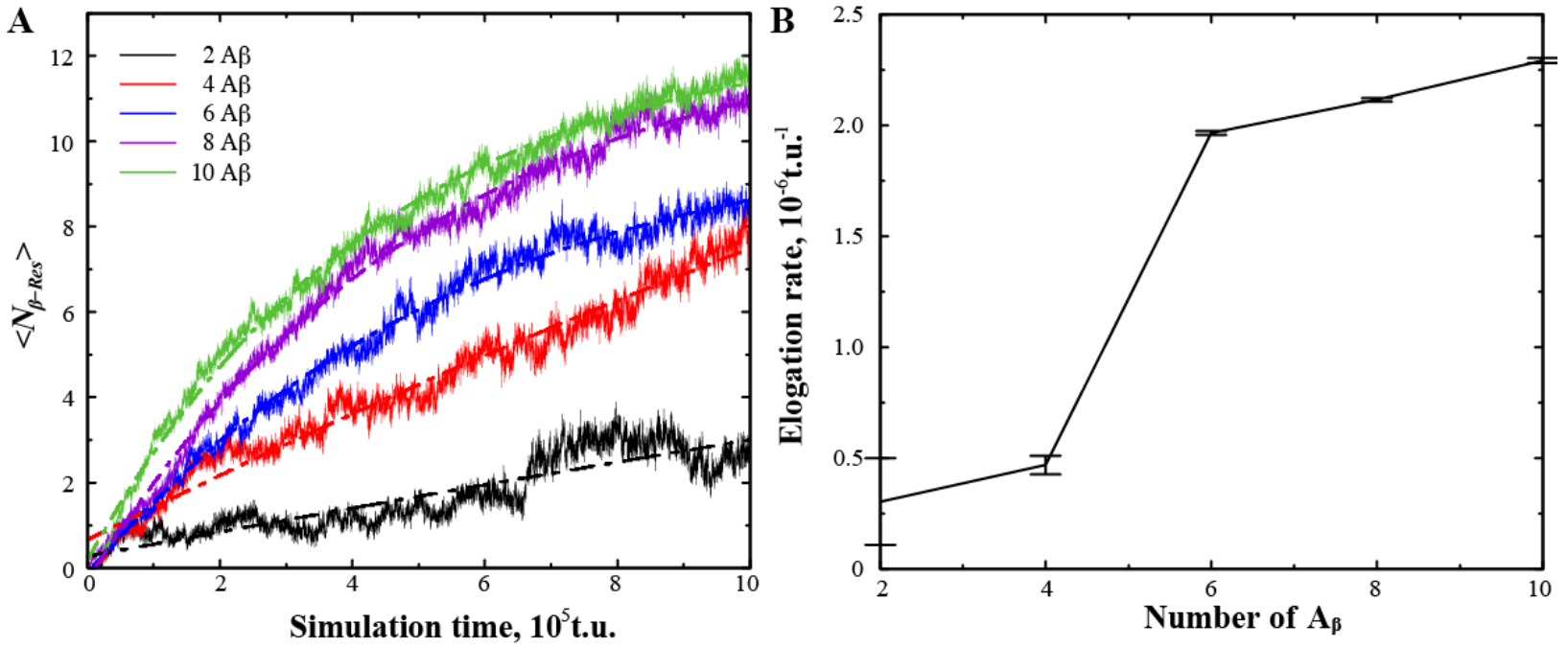
Increasing NP-protein attractions leads to **more proteins** on NP surface

The dependence of diffusion on NP-protein attractions



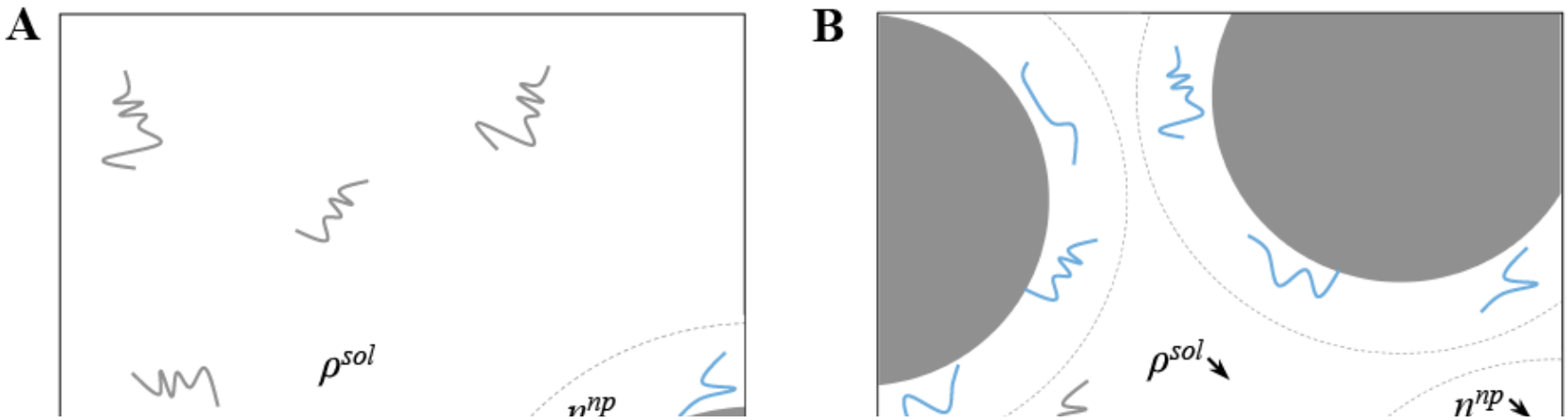
Increasing NP-protein attractions leads to **decreased protein diffusion** on NP surface.

Dependence of protein concentrations (fixed attraction)

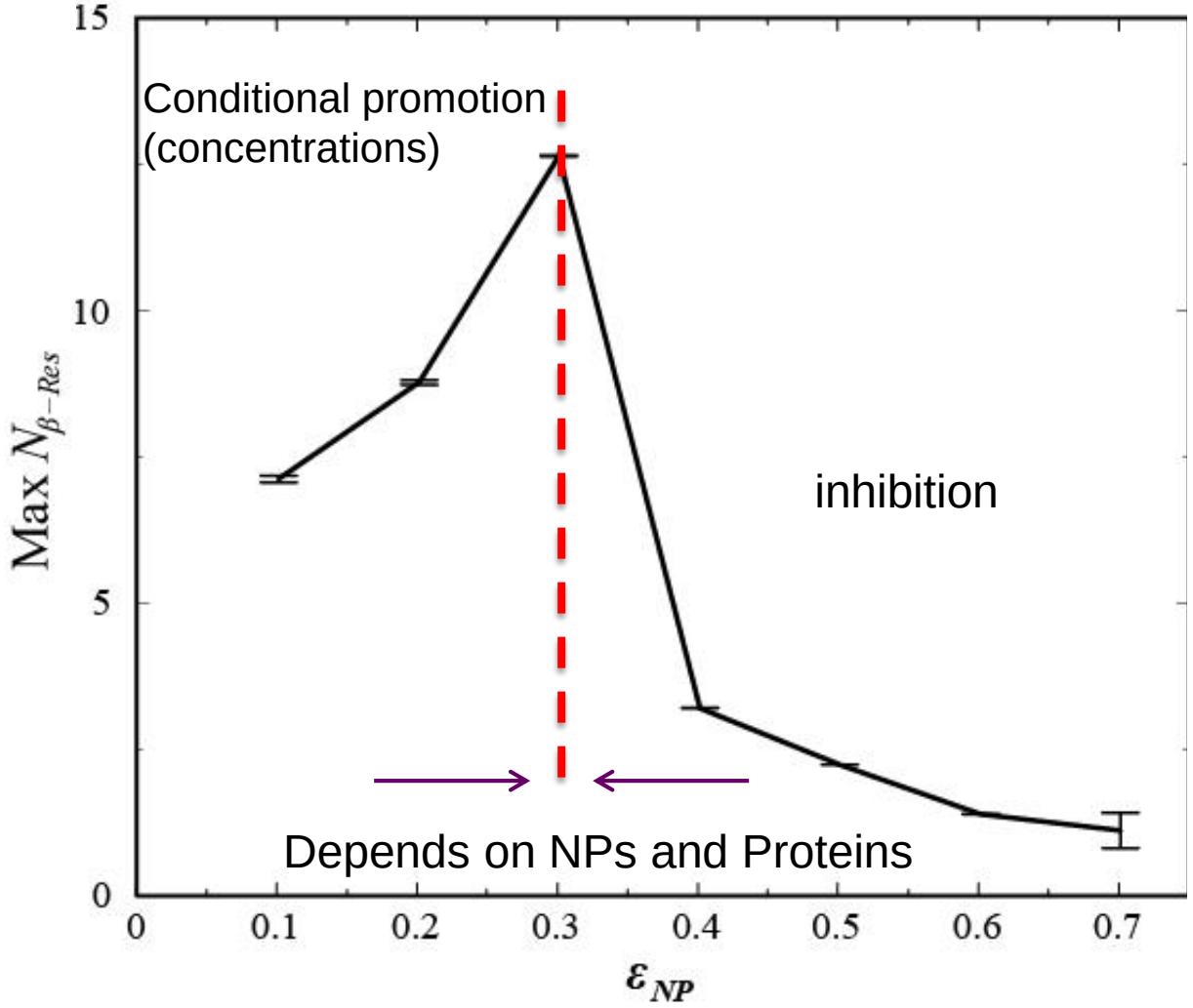


Aggregation on NP surface is concentration dependent

Effect of relative protein/NP concentration



A multi-factorial effects of NPs on aggregation



Radic, S., Ke PC, Davis, TP, Ding F., RSC Adv., 2016
Galvagnion, Nat Chem Biol. 2015 Mar;11(3):229-34.

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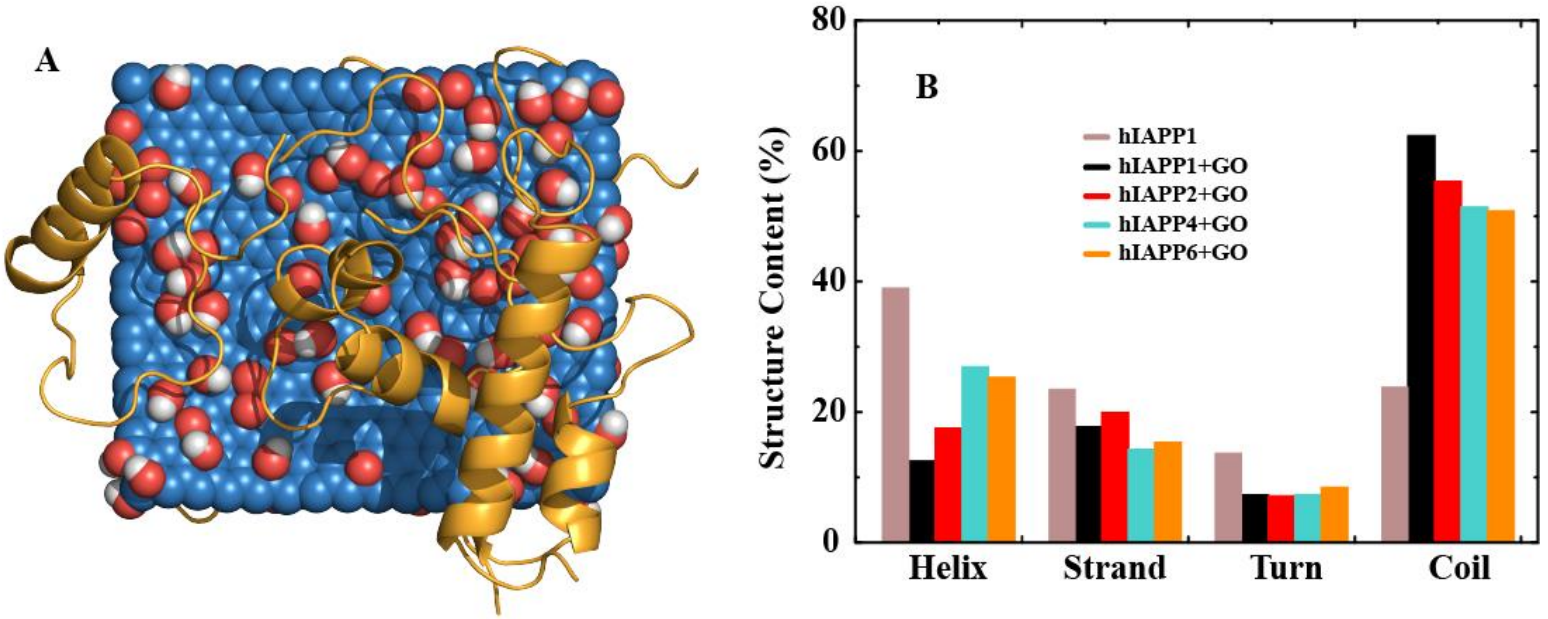
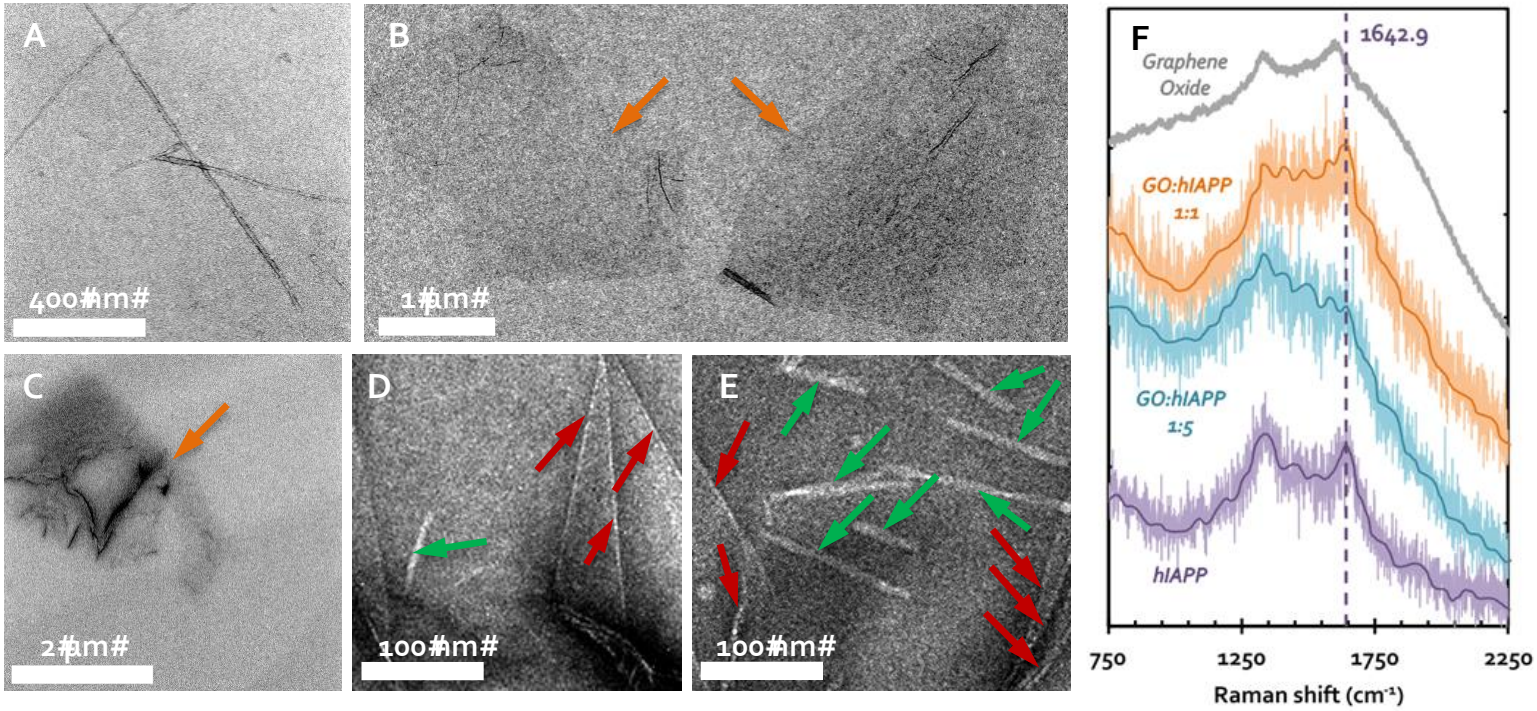
Graphene oxide inhibits IAPP aggregation and cytotoxicity

Graphene oxide sequesters IAPP



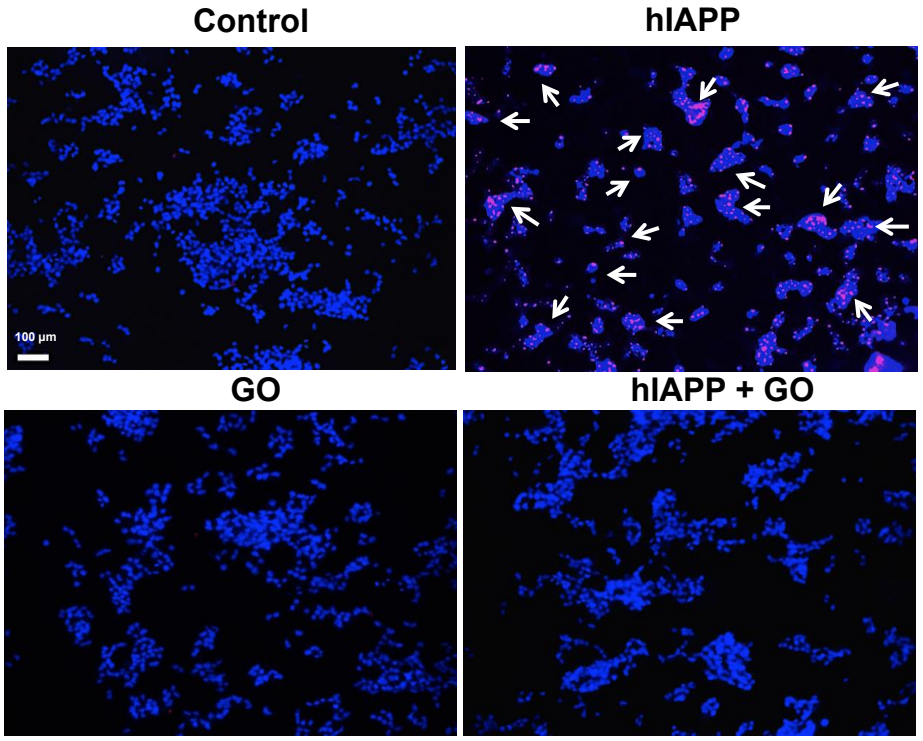
Nedumpully-Govindan et al, PCCP, 18:94-100 (2016)

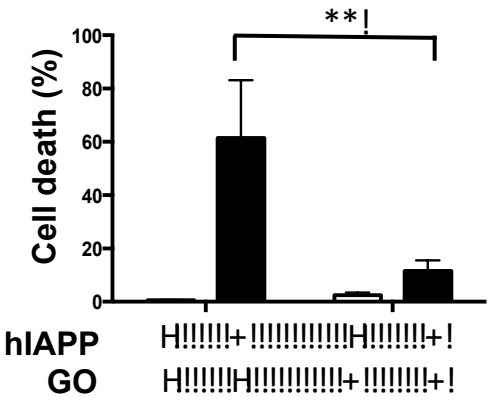
Biophysical characterization of GO-IAPP interaction



Nedumpully-Govindan et al, PCCP, 18:94-100 (2016)

GO reduces cytotoxicity of IAPP



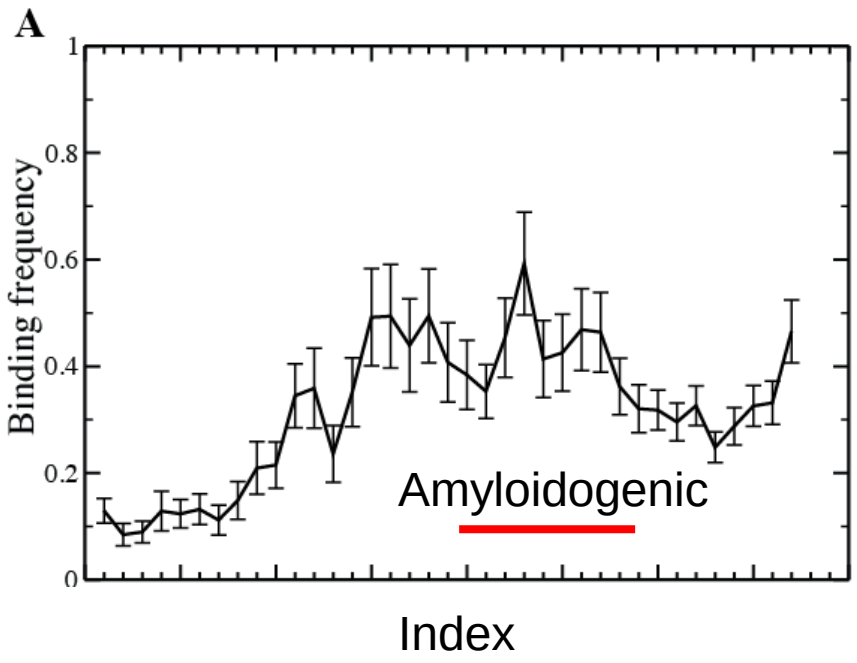


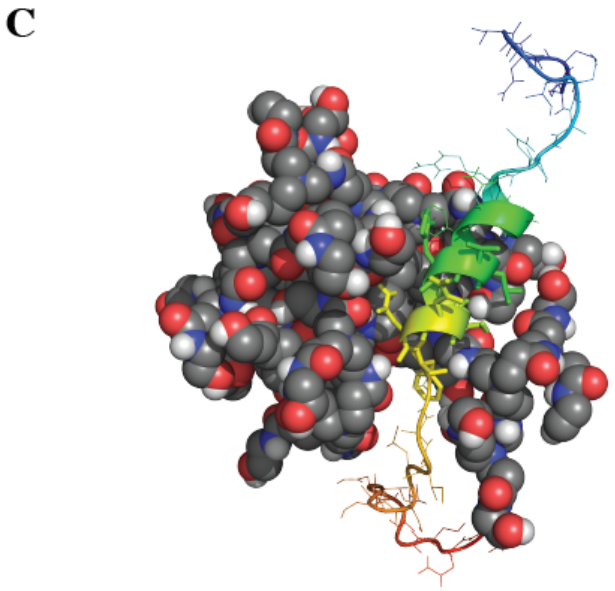
n=4, One–way ANOVA (24 h treatment)

Nedumpully-Govindan et al, PCCP, 18:94-100 (2016)

PAMAM dendrimer inhibits IAPP aggregation and cytotoxicity

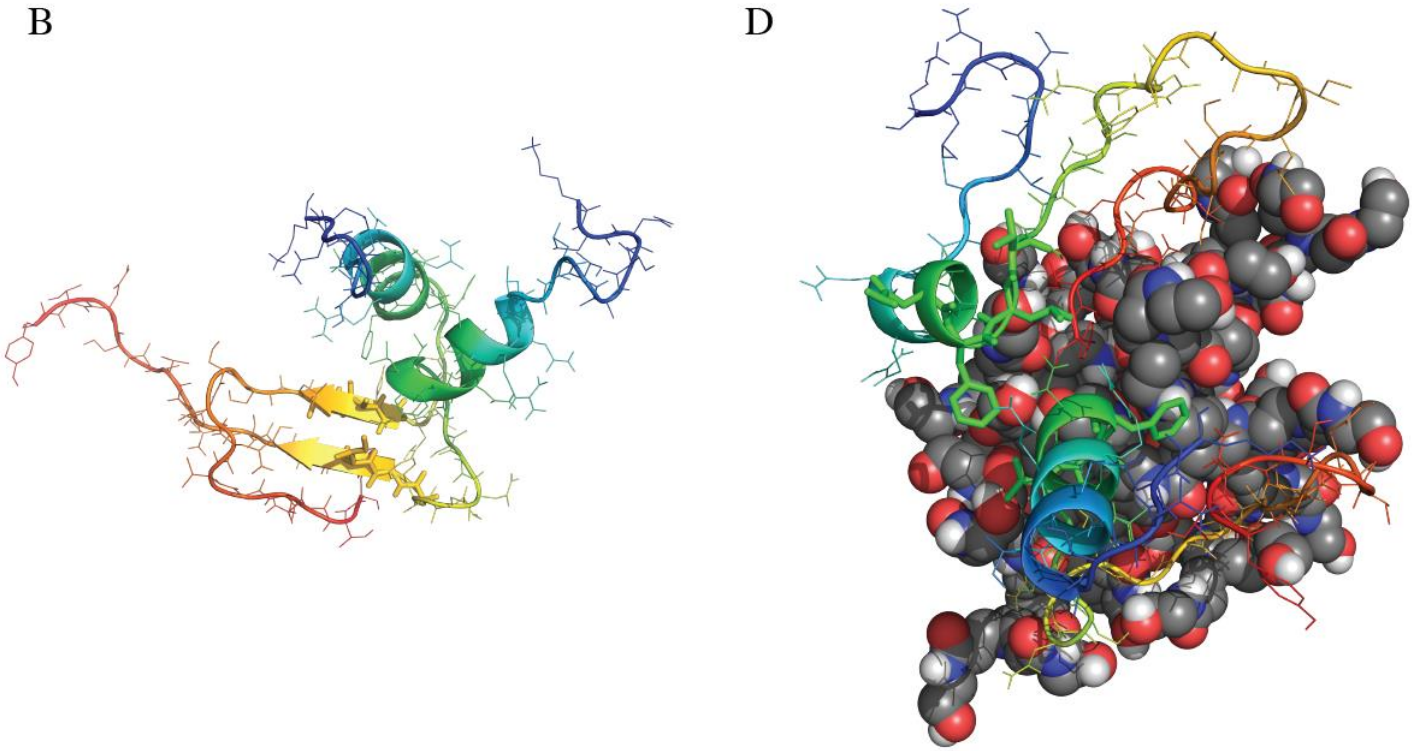
PAMAM dendrimer binds the amyloidogenic region of Amyin monomer





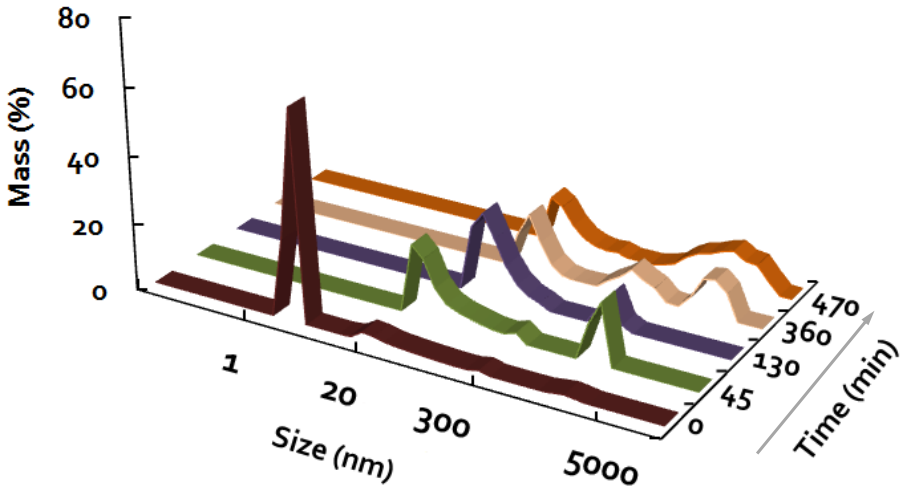
E.N. Gurzov et al, Small, in press (2016)

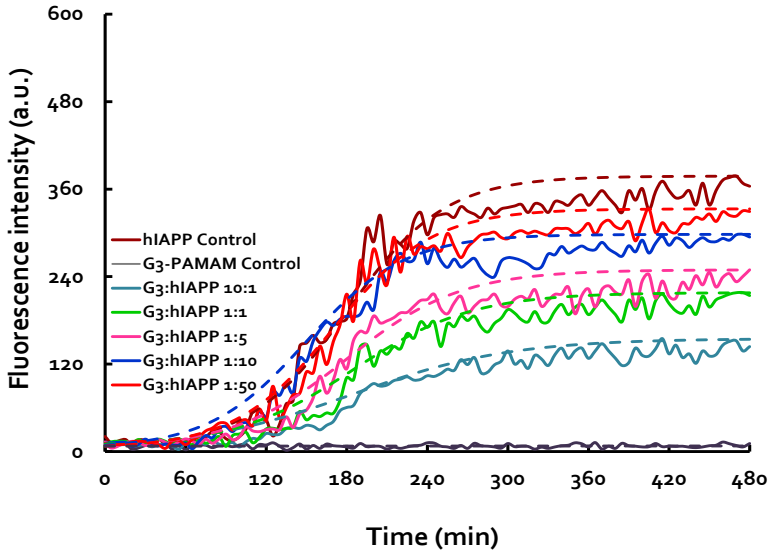
PAMAM dendrimer inhibits dimerization

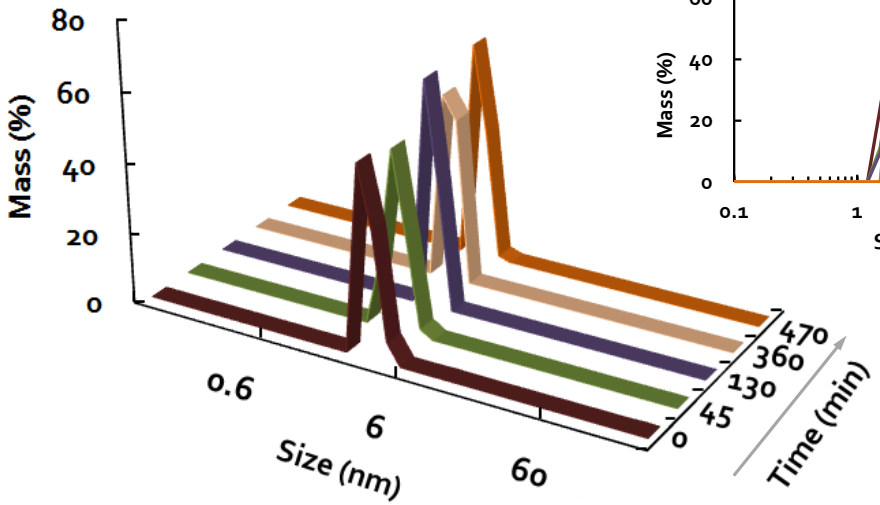


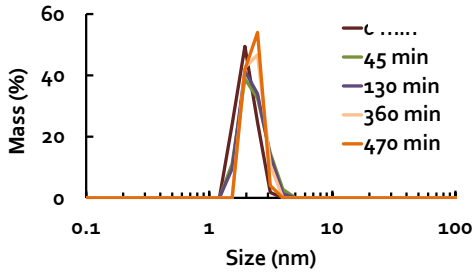
E.N. Gurzov et al, Small, in press (2016)

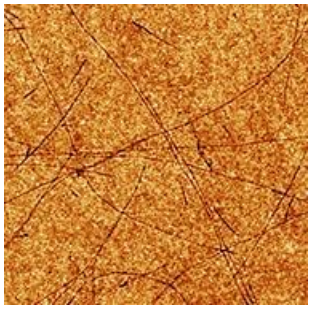
Biophysical characterization of the anti-aggregation effects – DLS, ThT, TEM

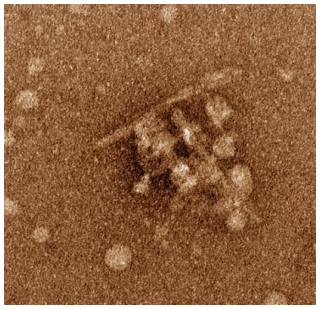






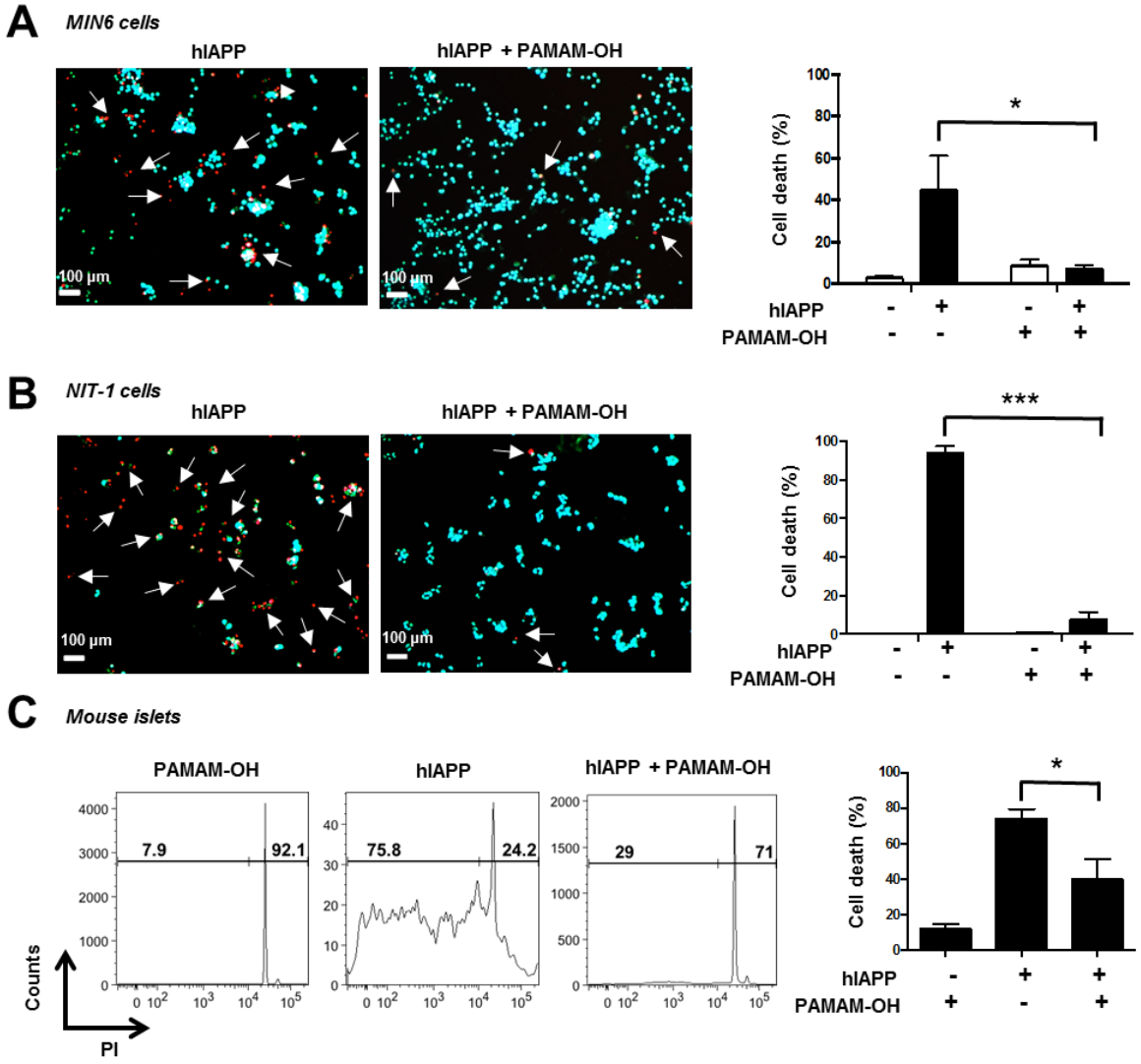






E.N. Gurzov et al, Small, in press (2016)

Inhibition of IAPP cytotoxicity *in vitro* and *ex vivo*



E.N. Gurzov et al, Small, in press (2016)

Summary

- A multiscale approach for modeling protein aggregation at the Nano-Bio interface with long time scales and large system sizes
- A mechanistic insight about the complex and seemingly contrasting effects of NPs on amyloid aggregation
- Utilizing the anti-aggregation effects of NPs for anti-amyloid nanomedicine design.

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