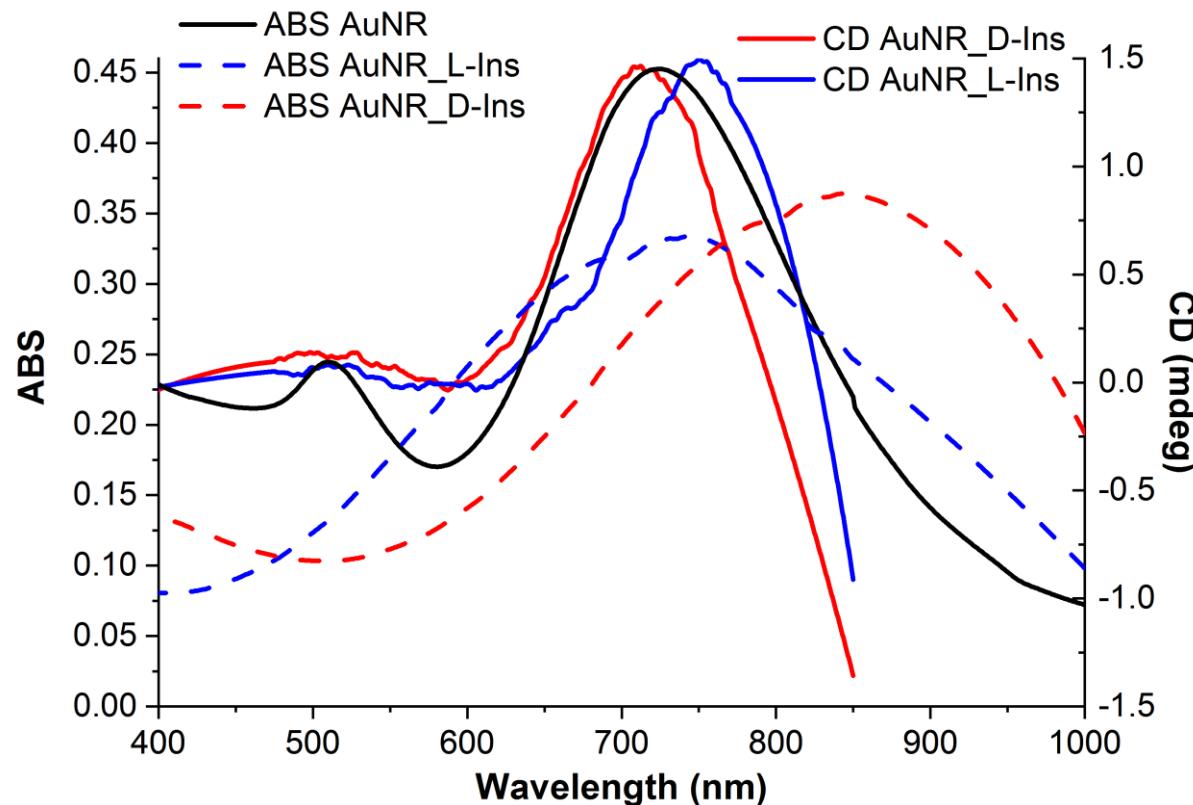


Induced chiral plasmon as a new amyloid sensing method

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1. Introduction – amyloid detection methods

- Amyloids are chiral fibrillar ordered protein aggregates of characteristic cross-beta sheet structure
- They are present in brains of people infected with the neurodegenerative diseases such as Alzheimer's and Parkinson's.
- There is a huge demand for new detection methods of small amounts of amyloid structures
- Gold nanoparticles are promising materials for detection of amyloids due to their biocompatibility and toxicity
- One of the most interesting methods of amyloid detection is based on induction of the chiral plasmon
- It is caused by the chiral arrangement of anisotropic nanoparticles due to connection with a chiral fibril surface

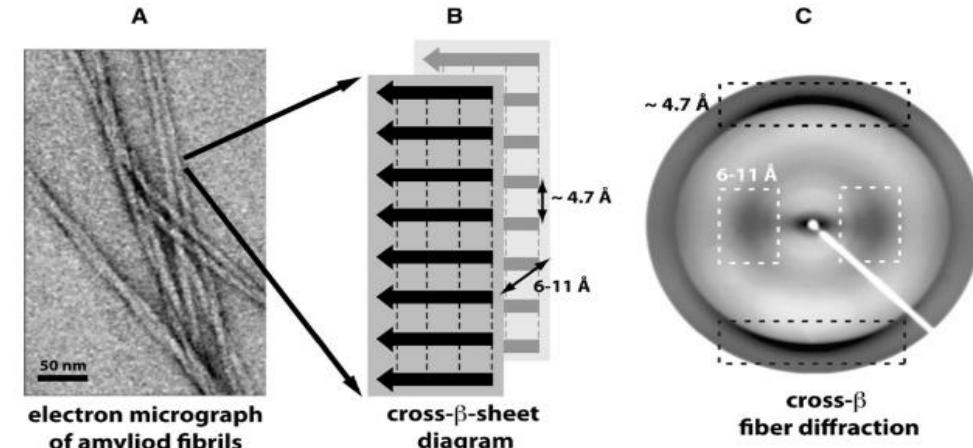


Fig. 1 (A) amyloid fibrils imaged using transmission electron microscope, (B) cross-beta sheet structure and (C) diffraction pattern given by cross-beta sheet structures.

Source: E. Herczenik, “The Biology of Non-Native Proteins.”, Utrecht University, 2007.

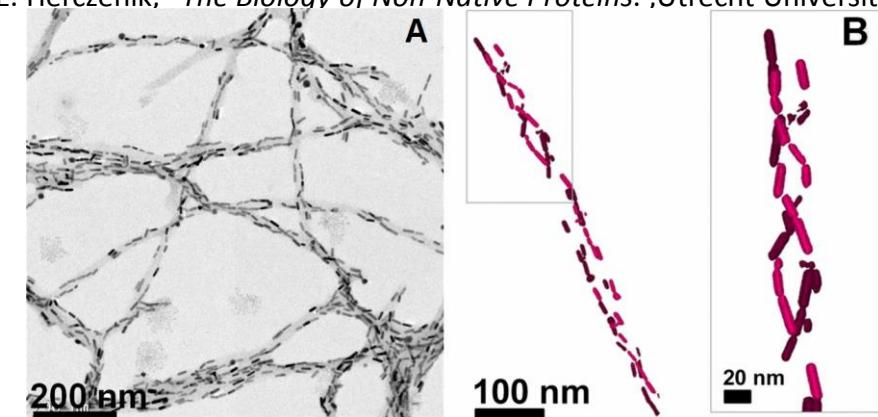


Fig. 2 (A) TEM images of Au NRs in the presence of α -synuclein fibrils, (B) Cryo-TEM tomography reconstruction image of a composite fiber showing the 3D chiral arrangement of Au NRs

Source: Kumar, J., et al., „Detection of amyloid fibrils in Parkinson's disease using plasmonic chirality.”; Proceedings of the National Academy of Sciences, 2018. 115(13): p. 3225.



2. Goals and materials

- The goal of our project was to detect chiral amyloid structures incubated from bovine insulin using mini gold nanorods with aspect ratio 3.2 (24 x 7.5 nm) (Fig. 3) synthesized using a seedless method
- Amyloid aggregates were incubated in different temperatures and pH to obtain samples having both correct fibrillar morphology (Fig. 4A,B) and optical chirality (Fig. 4C)

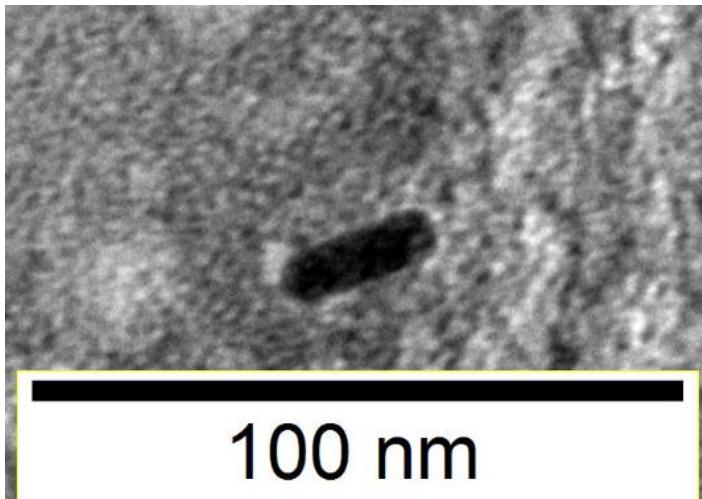


Fig. 3 TEM image of gold nanorod

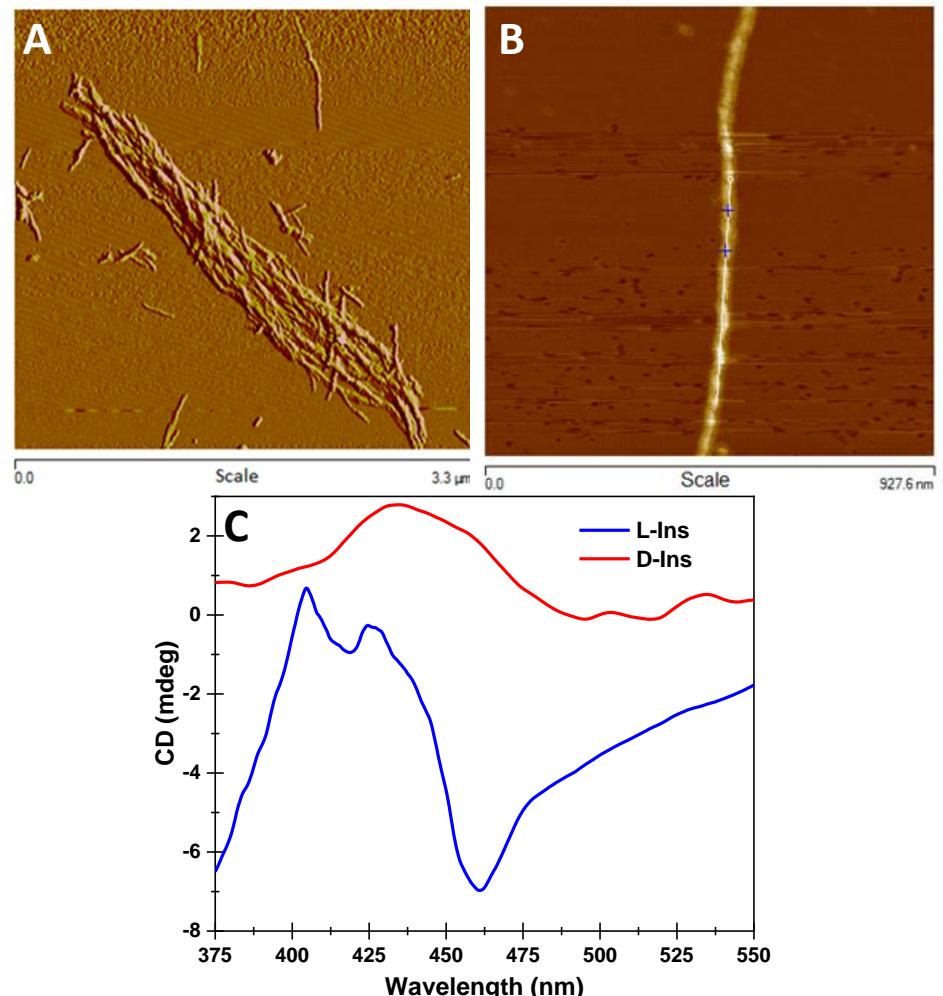


Fig. 4 Two types of insulin amyloid aggregates – L-Ins incubated in pH=1.4 in 40°C, protein conc. 10mg/ml (A); and D-Ins, incubated in pH = 1.5 in 70°C, protein conc. 10mg/ml (B), imaged using atomic force microscope. Circular dichroism spectra of both bovine insulin aggregates stained by amyloid specific dye Thioflavin T (C).



3. Results – extinction and circular dichroism spectra of insulin-nanorods aggregates

- After addition of insulin aggregates to AuNRs the extinction spectra has lowered and LSPR maxima has shifted into different directions for each amyloid (Fig. 5)
- We have successfully induced a chiral plasmon visible in circular dichroism spectra, measured in similar conditions as ABS (Fig. 6)
- The ICP maximum has also shifted, but in opposite directions than LSPR in ABS maxima

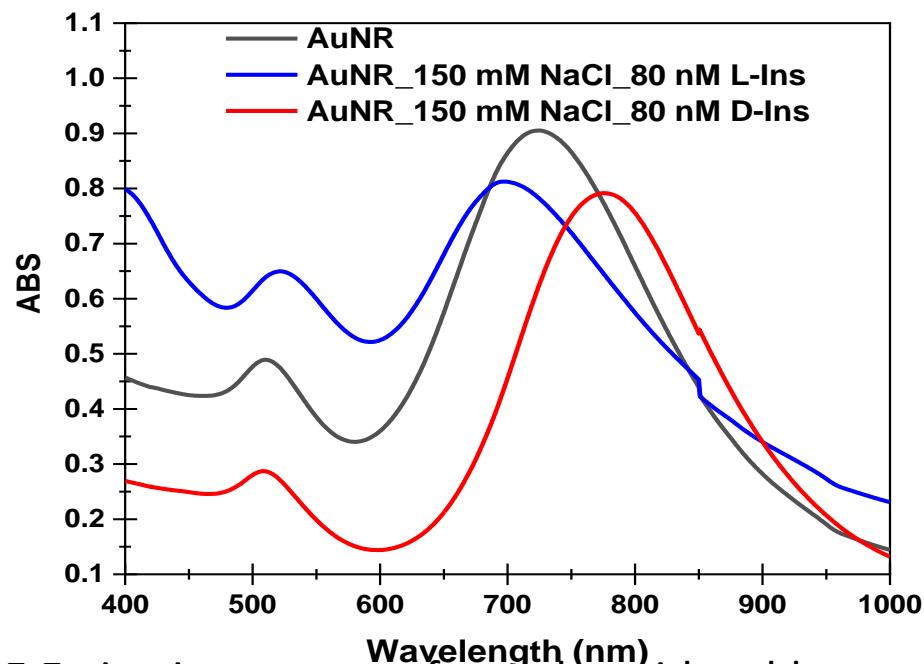


Fig. 5 Extinction spectra of samples with gold nanorods 30 minutes after the addition of insulin aggregates.

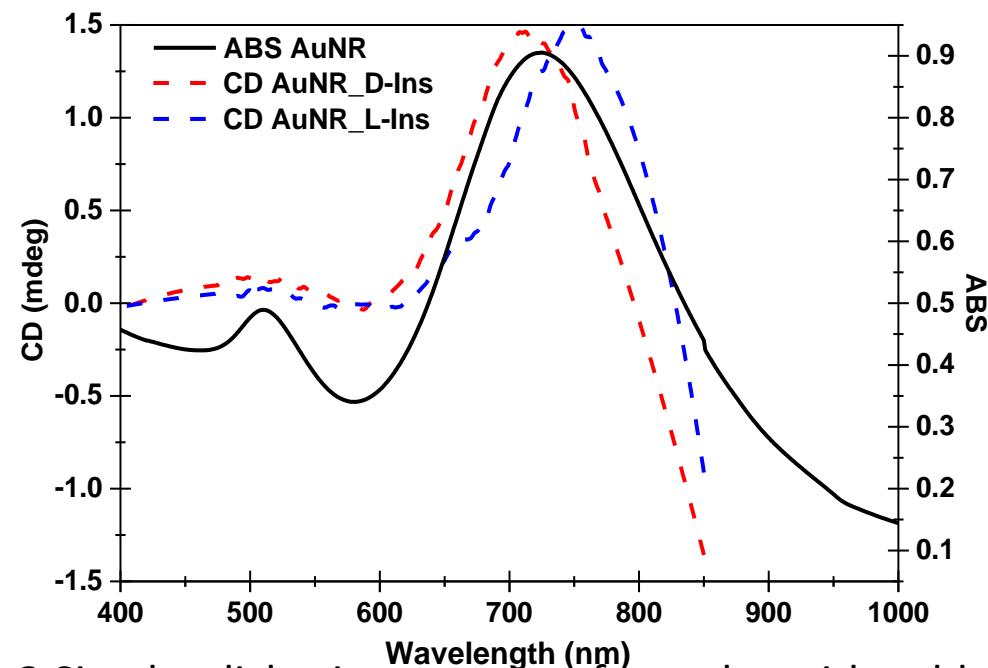


Fig. 6 Circular dichroism spectra of samples with gold nanorods 30 minutes after the addition of insulin aggregates.



4. Results – TEM images of insulin-nanorods aggregates

- Analysis of TEM images have showed that there was no chiral arrangement of gold nanorods connected to amyloid surface in both samples (Fig. 7A,B) indicating that induced plasmon chirality must be of another origin
- However, nanorods were within the range enabling effective plasmon coupling which explains the shifts in extinction spectra
- **Hypothesis:** Origin of the induced chiral plasmon could be induced by nanorods dimers and trimers connected in a chiral manner

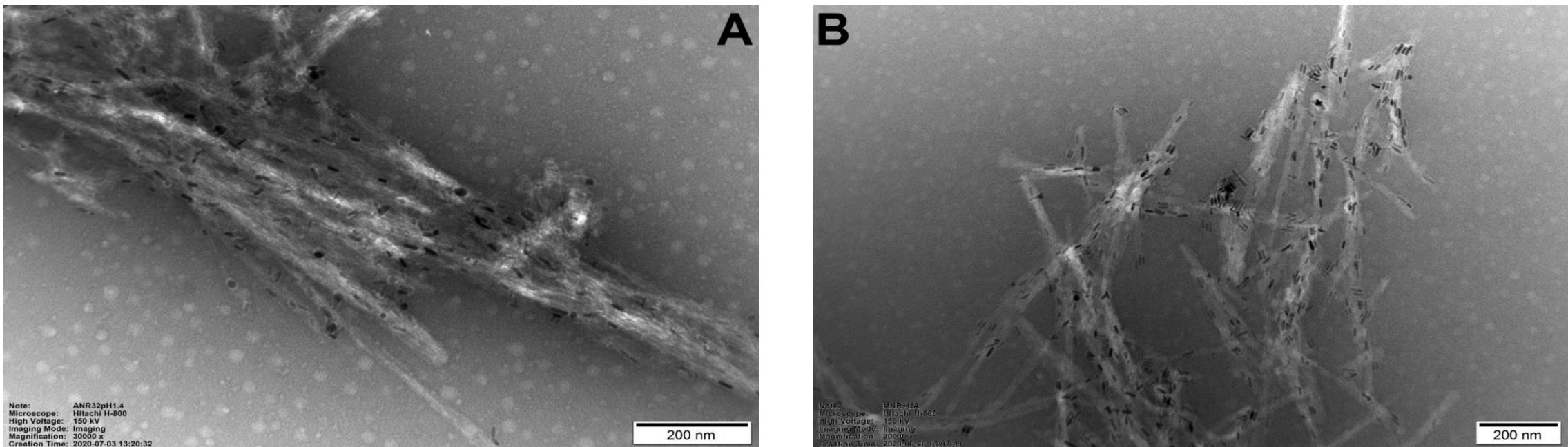


Fig. 7 Images of aggregates created from gold nanorods and amyloids 30 min. after addition of **L-Ins (A)** and **D-Ins (B)**, taken using TEM



5. Conclusions

- The interaction of gold nanorods with the bovine insulin amyloids leads to the **modification of thier optical properties**
- Insulin aggregates of different optical chiralities and morphologies **influence the optical properties of gold nanorods in different ways**
- Both **shifting the extinction maximum** and **induced chiral plasmon**, in respect to the location of LSPR of pure nanoparticles, **could be used as a new detection method of bovine insulin aggregates**