Rearranging protein assemblies from 2D layers to 1D fibres by electrospinning

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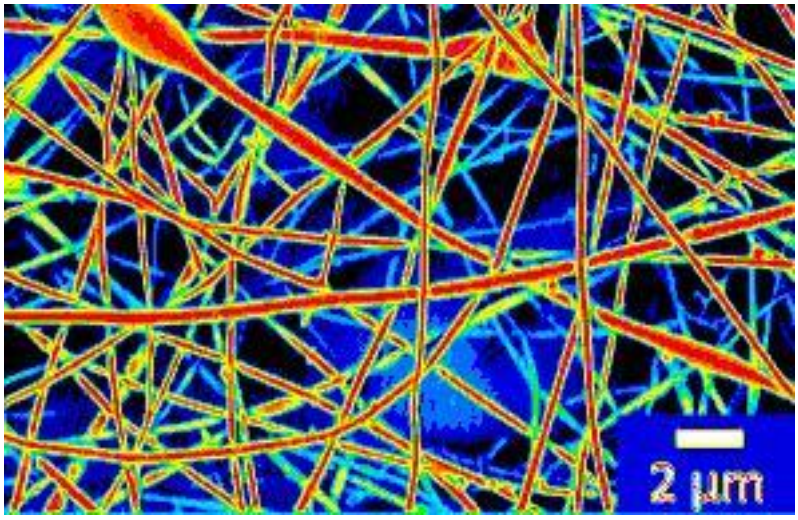
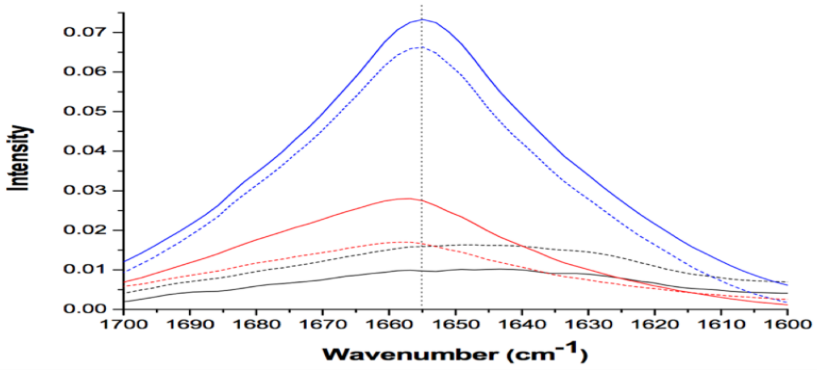
Natural proteins do usually not assemble to regular structures, which very much impedes their characterisation. However, some of them can be crystallised, i.e. they form regular 3D assemblies. Others assemble in 2D, e.g. membrane proteins, and in 1D, e.g. fibrous proteins and some viruses. Conformational changes inside a protein can change its mode of assembly. Some of the best-known examples are prion proteins, which can change to a conformation with increased beta sheet content, which can result in neuropathogenic fibres. Since Alzheimer disease is based on similar mechanisms, 1D assembly is and will remain a very hot topic in protein science [1,2,3].



SEM of electrospun pure hydrophobin. The quasi endless fibre has ca. 300 nm average diameter.

Various proteins, some related to Alzheimer and Parkinson disease, exhibit a surprising in vitro behaviour: Shear forces speed up their assembly to fibres [1,4,5]. We here show that a combination of solvent-induced conformational change and extreme shear induces such severe changes that the assembly mode switches from 2D to 1D. To this end, we dissolved a protein, hydrophobin, at high concentration in an organic solvent, and applied polymer-free (i.e. monomer) electrospinning [6].

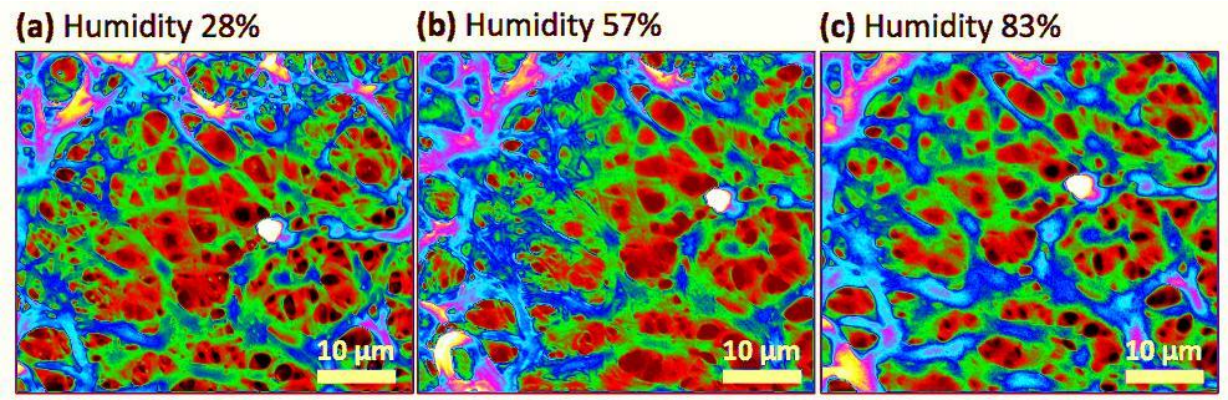


IR spectra of the amide band region of HP samples. The bottom curves (maxima at 1645 cm^{-1}) correspond to the original state, the top curves with maxima at 1655 cm^{-1} point to an increase in alpha helices in organic solvent.

Hydrophobins (HPs) exhibit 2D organisation. They are used in the context of "White Biotechnology" as emulsifiers and surface primers (www.hydrophobin.basf.com). HPs are naturally found on fungi, where they determine the very hydrophobic nature of the fungus cap. They can easily be produced in kg amounts by biotechnological fermentation techniques.

Electrospinning requires highly concentrated solutions of HPs in a solvent with high vapour pressure. We showed with electrophoresis (SDS-PAGE) and Raman, IR, and circular dichroism spectroscopy that already the dissolution changes the conformation of the HPs, different from contact with water. However, HPs did not assemble in solution. Only during electrospinning the high shear force, together with the evaporation, induced 1D alignment, and we were able to spin extremely long microfibers that consist of pure (polymer-free) proteins. We characterised the fibres with SEM, including environmental SEM in water vapour.

We believe that electrospun fibres of pure proteins can have multiple applications in biotechnology, e.g. as highly biocompatible scaffold materials.



Environmental SEM of electrospun hydrophobin. The water vapour pressure increases the diameter of the fibres.

References

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