

### Multilayered Thin Films Containing Amyloid-like $\kappa$ -Casein Fibrils Prepared by the Layer-by-Layer Deposition

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The formation of misfold protein aggregates called amyloid fibrils in tissues is known to be associated with diseases such as Alzheimer's disease, Type II diabetes and prion disease. However, highly ordered, long and unbranched thread-like fibrils with robust mechanical strength but still with some flexibility could make them attractive for advanced nanocomposites in many applications. In the present study, we propose a novel strategy to fabricate unique free-standing multilayered thin films containing amyloid fibrils derived from  $\kappa$ -caseins, which are one kind of caseins in bovine milk but not linked to any disease. Prior to the construction of amyloid-based multilayer films, the kinetics of  $\kappa$ -casein fibril formation was monitored under various conditions of physiological pH and temperature by fluorescent assay with thioflavin-T as well as small angle neutron scattering(SANS) analysis. It was noted that the fibrillation kinetics of  $\kappa$ -caseins strongly depends on temperature and also is fast near the isoelectric point of  $\kappa$ -caseins. Free-standing amyloid multilayer thin films were prepared by the sequential deposition of  $\kappa$ -casein fibrils and positively charged polyelectrolytes on a sacrificial layer.