Phase Separation of Intrinsically Disordered Protein Polymers Mechanically Stiffens Fibrin Clots

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Abstract

Fibrin (Fb) networks self-assemble through the coagulation cascade and serve as the structural foundation of blood clots. Following severe trauma or drug therapy, reduced integrity of Fb networks can lead to formation of clots with inadequate mechanical properties. A key feature of therapeutic interventions for hemostasis is therefore the ability to restore mechanical strength to clots formed under coagulopathic conditions. Here, we describe an intrinsically disordered protein based on an elastin-like polypeptide (ELP) sequence that specifically binds Fb and modulates its mechanical properties. We designed hemostatic ELPs (hELPs) containing N- and C-terminal peptide tags that were selectivity recognized by human transglutaminase factor XIIIa, and covalently linked into fibrin networks via the natural coagulation cascade. Phase separation of hELPs above their lower critical solution temperature (LCST) led to stiffening and rescue of clot biophysical properties under simulated conditions of dilutive coagulopathy. In addition to phase-dependent stiffening, the resulting hELP-Fb networks exhibited resistance to plasmin degradation, reduced pore sizes, and accelerated gelation rate following initiation of clotting. These results demonstrate the ability of protein-based phase separation to modulate the physical and biochemical properties of blood clots and suggest protein phase separation as a new mechanism for achieving hemostasis in clinical settings.[1]

[1] Urosev, I., Lopez Morales, J. & Nash, M. A. Phase separation of intrinsically disordered protein polymers mechanically stiffens fibrin clots. *Adv. Funct. Mater.* 2005245 (2020).