



# Cohesive behavior of soft biological adhesives: Experiments and modeling

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## ABSTRACT

Extracellular proteins play a key role in generating and maintaining cohesion and adhesion in biological tissues. These “natural glues” are involved in vital biological processes such as blood clotting, wound healing and maintaining the structural integrity of tissues. Macromolecular assemblies of proteins can be functionally stabilized in a variety of ways in situ that include ionic interactions as well as covalent crosslinking to form protein networks that can extend both within and between tissues. Within tissues, myriad cohesive forces are required to preserve tissue integrity and function, as are additional appropriate adhesive forces at interfaces both within and between tissues of differing composition. While the mechanics of some key structural adhesive proteins have been characterized in tensile experiments at both the macroscopic and single protein levels, the fracture toughness of thin proteinaceous interfaces has never been directly measured. Here, we describe a novel and simple approach to measure the cohesive behavior and toughness of thin layers of proteinaceous adhesives. The test is based on the standard double-cantilever beam test used for engineering adhesives, which was adapted to take into account the high compliance of the interface compared with the beams. This new “rigid double-cantilever beam” method enables stable crack propagation through an interfacial protein layer, and provides a direct way to measure its full traction–separation curve. The method does not require any assumption of the shape of the cohesive law, and the results provide abundant information contributing to understanding the structural, chemical and molecular mechanisms acting in biological adhesion. As an example, results are presented using this method for thin films of fibrin—a protein involved in blood clotting and used clinically as a tissue bio-adhesive after surgery—with the effects of calcium and crosslinking by Factor XIII being examined. Finally, a simple model is proposed, demonstrating how a bell-shaped cohesive law forms during the failure of the fibrin interface based on an eight-chain model whose structure degrades and changes configuration with stress.

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## 1. Introduction

High-performance biological adhesives composed of proteins and/or polysaccharides are found in essentially all organisms [1]. These “bio-glues” vary extensively in structure and capabilities according to their function and performance requirements [2,3]. In general, bio-glues are deformable and can dissipate a significant amount of energy [4,5], and they can adhere to a variety of surfaces [1]. For instance, in humans, a fibrin network forms after bleeding following tissue trauma. Fibrin is a natural adhesive which can adhere to soft and hard tissues and withstand the stress imposed by pulsating blood pressure and ultimately stop bleeding [6]. Other examples include mussel, which can secrete a high-performance proteinaceous adhesive (3,4-dihydroxyphenylalanine) to anchor

themselves to rocks in order to resist the shear forces of tidal currents and waves [7]. Biological adhesives also ensure the intrinsic cohesion of key structural tissues such as bone, tooth enamel, dentin and cementum, and seashells. These tissues are composite materials consisting of both organic and inorganic phases, often with fibrillar elements such as collagen forming a scaffold within which mineral nano-crystallites are deposited, all of which are held together by specific non-collagenous proteins. For instance, in bone, the cohesion of collagen fibrils is ensured by bio-adhesive non-collagenous proteins [8]. Optimal deformability, strength and toughness of these bio-glues are therefore critical to the overall mechanical performance of mineralized tissues such as bone. While little is known about which protein molecules maintain the cohesion of bone, recent experiments have suggested that osteopontin may be such a key adhesive protein [9]. Transgenic mice lacking osteopontin have bones with a 30% decrease in fracture toughness independent of bone mineral density, a traditionally

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