

Emergent intercellular junction stability during cyclic tissue loading

Eleni Papafilippou,^{1,*} Alessandra Bonfanti,^{2,*} Guillaume Charras,^{3,4,*} and Alexandre Kabla^{1,*}

¹Department of Engineering, University of Cambridge, Cambridge, UK; ²Department of Civil and Environmental Engineering, Politecnico di Milano, Milan, Italy; ³London Centre for Nanotechnology, University College London, London, UK; and ⁴Department of Cell and Developmental Biology, University College London, London, UK

ABSTRACT Epithelial tissues are often exposed to cyclic deformations in their physiological environment. Maintenance of mechanical integrity relies on intercellular adhesion proteins that link neighboring cells and transmit forces across the cellular network. The stability of these intercellular adhesion complexes is therefore critical for tissue strength. Under sustained stress, failure of intercellular adhesion complexes leads to damage accumulation that progressively weakens the material and ultimately causes failure at the tissue scale. Although the collective behavior of adhesion complexes under static loading has been characterized to some extent, their response to dynamic loading remains largely unknown, despite its physiological importance. Here, we combine quantitative experiments on Madin-Darby Canine Kidney monolayers with modeling of intercellular adhesion complexes with force-dependent detachment rates to investigate tissue resilience under cyclic loading. We find that cyclic loading significantly prolongs tissue lifetime and increases the maximum deformation the tissue can withstand before failure compared with constant tension, thanks to repair occurring during low-tension phases. Our model identifies intrinsic rupture and repair timescales governing adhesion stability, revealing three regimes of tissue behavior: rupture, slow damage accumulation, and stable equilibrium. Normalizing loading parameters by the intrinsic material timescales collapse experimental and simulated data into universal stability maps. These findings demonstrate that epithelial resilience emerges from stochastic adhesion bond dynamics, providing a predictive framework linking adhesion complex turnover to macroscopic tissue mechanics under physiological cyclic forces.

SIGNIFICANCE Epithelial tissues, the body's primary mechanical barriers, experience continuous mechanical stress from physiological movements, such as breathing or peristalsis. Through experiments and modeling, we show that a tissue's ability to resist and repair damage under cyclic loading comes from the natural binding and unbinding of the intercellular adhesion complexes that link neighboring cells. We identify fundamental timescales governing rupture and recovery in cell-cell junctions, showing that tissue stability can be universally predicted from intercellular adhesion kinetics. This work establishes a quantitative framework connecting microscopic adhesion kinetics to macroscopic tissue mechanics. It provides a tool for studying resilience in diverse biophysical soft matter systems and for understanding how living materials maintain integrity under fluctuating forces.

INTRODUCTION

Tissue integrity and barrier strength are fundamental to organ health, yet epithelial tissues are continually challenged by their dynamic mechanical environments. Physiological processes such as breathing, peristalsis, and limb movement impose cyclic loads that vary over seconds, while growth, pregnancy, and tumor expansion generate slow, sustained

loads lasting months or years.^{1–3} While moderate cyclic loading can enhance cytoskeletal organization and intercellular adhesion,⁴ irregular or high-frequency oscillations can destabilize adhesion and compromise barrier function, contributing to diseases like irritable bowel syndrome.^{5–8} Experiments and theory indicate that soft epithelial tissues often develop microlesions or cracks under constant loads.⁹ In some contexts, such defects are homeostatic, as seen in goblet-associated fractures of the gut epithelium, which represent regulated, localized remodeling.⁸ In other cases, microcracks can nucleate and propagate, leading to tissue-scale rupture.¹⁰ Together, these findings suggest that

Submitted December 29, 2025, and accepted for publication June 30, 2026.

*Correspondence: ep670@cam.ac.uk or alessandra.bonfanti@polimi.it or g.charras@ucl.ac.uk or ajk61@cam.ac.uk

Editor: Gijsje Koenderink.

<https://doi.org/10.1016/j.bpj.2026.06.038>

© 2026 The Authors. Published by Elsevier Inc. on behalf of Biophysical Society.

This is an open access article under the CC BY license (<http://creativecommons.org/licenses/by/4.0/>).



epithelial tissues exist in a delicate balance between applied loading patterns and intrinsic mechanical responses. This raises a central question: how do epithelial tissues maintain their integrity when exposed to periodic mechanical loads?

In epithelia, cohesion is maintained by intercellular adhesion complexes that form dynamic links between cells, such as adherens junctions or desmosomes.¹¹ The dynamics of binding and unbinding of these adhesive complexes present intrinsic timescales.⁵ A variety of cellular mechanisms, ranging from cytoskeletal remodeling to mechanotransductory and biochemical signaling, contribute to the regulation of intercellular junctions.^{12–14} Intercellular adhesion proteins within intercellular complexes can display a range of behaviors: slip bonds, which become weaker and more likely to break as force increases,¹⁵ and catch bonds, which are paradoxically stabilized by intermediate levels of force.¹⁶ Yet, it is challenging to determine whether epithelial resilience can be explained purely by the emergent kinetics, or if active feedback and signaling pathways also contribute.^{17,18} To what extent, then, does the ability of a tissue to retain its integrity in the face of repeated cycles of loading depend on the emergent mechanisms of rupture and repair of intercellular complexes?

Our theoretical understanding of the mechanics of intercellular adhesion complexes largely stems from modeling efforts. Building on Bell's seminal model of force-dependent bond dissociation,¹⁵ many studies have used the slip-bond framework to predict junctional integrity under constant or monotonically increasing loads. Multi-bond stochastic models have shown how parallel bonds rupture probabilistically, producing exponentially distributed lifetimes and collective failure events.^{19,20} These models provide insight into how junction strength scales with bond number, how variability emerges from stochastic rupture events, and how catch-bond kinetics can help to stabilize intercellular junctions. However, most applications to date have either focused on zero-load equilibrium states or on sustained loads and force ramps, where the probability of rebinding becomes negligible.^{10,21,22} In contrast, cyclic loading introduces alternating phases of high stress, during which bond rupture occurs, and phases of low stress, when rebinding is more likely to take place. Using cyclic loading, we can investigate whether periodic relief from loading enables recovery, thereby shaping epithelial resilience over longer timescales. More generally, how do rupture and repair events determine junctional integrity, and what periodic loading characteristics define different regimes of epithelial stability?

Here, we use experiments and modeling to frame a novel approach to test tissue resilience and illustrate the subtle interactions between mechanical loading patterns and junction stability. We compare constant and cyclic stress experiments on Madin-Darby Canine Kidney (MDCK) epithelial monolayers and interpret the results with analytical and stochastic models to explore how intercellular

junctions accumulate damage and repair over time. Under sustained high stress, we identify a characteristic timescale that sets the limit of the intercellular junction's lifetime. At lower stress levels, we uncover a distinct repair timescale that captures how intercellular adhesion complexes reform to restore junctional integrity. Together, these emergent timescales encapsulate how tissues resist stress across a broad range of loading conditions. Building on this framework, we examine tissue behavior under cyclic loading, where alternating phases of high and low tension establish a threshold between rupture and stable recovery. Our results reveal distinct regimes: rupture, slow damage accumulation, and full recovery, whose boundaries are determined not by absolute load amplitude, but by how the durations of force application compare to intrinsic rupture and repair timescales. This unifying framework demonstrates that epithelial resilience arises as an emergent property of adhesion complex kinetics, offering a tool for understanding how tissues withstand physiological forces over the lifetime of the organism.

MATERIALS AND METHODS

Experimental *in vitro* model

Cell culture

MDCK wild-type cells were cultured at 37°C in an atmosphere of 5% CO₂ in DMEM (1×) (Thermo Fisher) supplemented with 10% fetal bovine serum (FBS, Sigma-Aldrich), 2.5% of 1 M HEPES buffer (Sigma-Aldrich), and 1% penicillin-streptomycin (Thermo Fisher). Cells were passaged at 1:8 ratio every 4 days using standard cell culture protocols and disposed of after 15 passages. Mechanical experiments and imaging were performed in Leibovitz's L15 (Thermo Fisher) supplemented with 10% FBS (Sigma-Aldrich) and 1% penicillin-streptomycin (Thermo Fisher).

Stretching devices

The stretching devices were microfabricated following the protocol described in Harris et al.,²³ with some adaptations. The materials required to make each device are borosilicate glass, nitinol wire (two thicknesses), Tygon rings (two thicknesses), and UV glue. Each assembled device consists of one stiff rod covered by a glass capillary and one compliant, soft rod. Two rings of Tygon of different thicknesses cover the extremities of the two wires, such that epithelial cell monolayers can grow between the test rods. The stepwise assembly involves (1) local melting of the borosilicate glass capillaries into a U-shape using a micro-pen blow torch (Inoda PT-200 Pro torch), (2) using pliers to cut the bent capillary into one U-shape (5 mm) and one straight section (25 mm), cutting the thick wire (30mm), and the thin wire (30 mm), using scissors to cut the thick Tygon (5 mm) and the thin Tygon (5 mm), (3) connecting the parts of the devices and securing in place using UV curing glue, and

(4) curing for 30 min. Each device was secured to the bottom of a 50-mm Petri dish by applying biocompatible adhesive glue on the U-shape glass base, minimizing any lateral movement during the experiment. During the gluing step, a short glass capillary was placed beneath the Tygon tips to slightly elevate the device, preventing contact with the bottom of the dish.

Preparation of suspended MDCK monolayers

The protocol for generating suspended MDCK monolayers is based on the work of Harris (2013) and Duque (2024).^{10,23} Cells were cultured on a sacrificial collagen substrate. Collagen type 1A was reconstituted on ice, by mixing 50% collagen (Cellmatrix type I-A, Nitta Gelatin), 20% sterile Milli-Q, 20% of 5× DMEM, and 10% reconstitution buffer 50 mM NaOH solution in sterile water, 200 mM HEPES, and 262 mM of NaHCO₃. A 10-μL droplet was placed between the device's test rods. The devices were then placed in an incubator for drying over 60–75 min at 37°C. This was followed by a rehydration step with 10 μL of culture medium for 30 min at 37°C. Approximately 40,000 cells were seeded on the rehydrated collagen and incubated at 37°C and 5% CO₂ for 30–45 min. Lastly, 55 mL of culture medium was added to completely submerge the test rods, and the monolayers were left in the incubator to grow for approximately 72 h, or until the monolayer extended from one rod to the other, covering the collagen substrate as well as each rod partially. Before mechanical testing, the collagen was digested enzymatically, using collagenase type-II (Worthington Biochemical) with imaging medium to reach a final concentration of 250 units per mL. The culture medium was exchanged for the collagenase solution and was placed at 37°C for 1 h. Before testing, the collagenase solution was exchanged for the imaging medium. Under these conditions, the cells remained healthy and retained their characteristic epithelial apico-basal polarity for 3 h.

Force measurement

A Petri dish containing a stretching device and suspended monolayer was placed on the stage of an inverted bright-

field microscope (Olympus IX71). The stiff rod was attached to a motorized micromanipulator (M126-DG1) controlled through a C-863 controller (Physik Instrumente) via a LabVIEW code (National Instruments). During experiments, force was applied to the tissue by stretching it through controlled movements of the micromanipulator.^{10,12,23} The soft rod was attached to the tip of a force transducer (SI-KG7A, World Precision Instruments), which deflects relative to the deflection of the soft wire. The deflection is converted into a digital signal and recorded as voltage data (USB-1608G, Measurement Computing). Images were taken every 6 s using a ×2 objective (PLN2×, Olympus) and a camera (Point Gray Grasshopper3 U3, GS3-U3-60QS6M-C). The applied force can be measured by monitoring the bending of the soft test rod, detected by the force transducer tip. At the end of each experiment, the monolayers were broken, and the rod's position after failure was used to define the zero-force geometry. Increments of known deformation were applied manually to obtain a set of six voltage-deformation values. The voltage-to-stress conversion was done using a standardized calibration procedure from Duque (2024)¹⁰ (see [Method S1](#), force measurement: calibration procedure).

Stress controller

Constant stress was applied by tuning the position of the motorized micromanipulator, using a PID controller ([Figure 1](#)). Before each creep experiment, a pre-calibration was performed on each device, to obtain a voltage-displacement relationship and a unique conversion factor, α . The target voltage was computed by taking the difference between the target stress and prestress divided by the calibration coefficient. The error signal, $E(t)$, was computed by subtracting the measured voltage from the target voltage and was fed into the PID controller. The controller minimizes this signal by adjusting the strain applied to the sample via the motorized micromanipulator. The PID controller adjusts the micromanipulator's position at a maximum rate of 0.1 mm/s, which is equivalent to a maximum strain rate of 10% s⁻¹. This rate cap was chosen to minimize the effects of

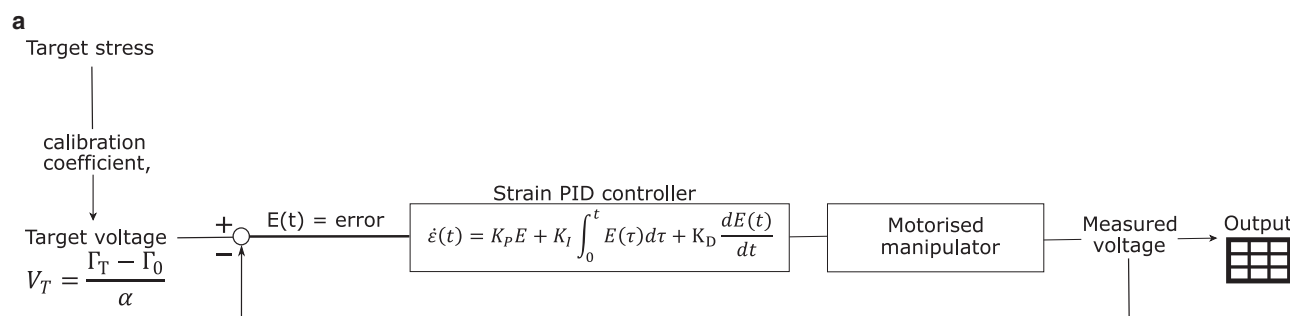


Figure 1. PID controller logic. The target voltage is calculated using the target stress, Γ_T , the prestress, Γ_0 and the pre-calibration coefficient, α . The error signal, given by subtracting the measured voltage from the target voltage, is then minimized through the PID controller, which adjusts the strain by adjusting the motorized manipulator position. At each timestep the data output is recorded in a text file.

strain-rate-dependent rupture that have been reported in the literature.¹⁰ The controller parameters, K_P , K_I , K_D , were empirically tuned for stable convergence to the target stress. The optimized controller parameters are found in [Table 1](#). To assess the controller's performance, a post-experiment calibration was performed following each test. Calibration coefficients before and after the experiment were compared by plotting them against each other. The resulting data closely aligned with the identity line, which fell within the confidence interval of the data, indicating that the pre-experiment calibration coefficients were reliable for computing the target voltage (see in [Method S1](#), force measurement: calibration procedure, [Figure SM 1](#)).

Analytical model

The analytical slip-bond model is based on Bell's theoretical framework of cell adhesion.¹⁵ The governing analytical [Equation 4](#) is solved in Julia by creating an ordinary differential equation problem, using the `Tsit5()` Runge-Kutta equation solver from the `OrdinaryDiffEq` package.²⁴ The relative and absolute tolerances are set to 10^{-8} , and the step, dt , is set to 10^{-2} s. The rupture timescale, t^* was initialized at the stable bond ratio for zero tension, [Equation 1](#), where k_{on} is the association constant rate, and $k_{off,0}$ is the dissociation rate at the start of the simulation. An alternative initialization considered was starting at 100% of bonds closed; however, this is not a physiologically representative condition, as there is continuous turnover of intercellular adhesion complexes and would likely lead to an overestimation of the rupture timescale. The repair timescale, t_e , was initialized at the critical ratio of bonds, which corresponds to the ratio of bonds at critical tension, Γ_c , and was computed by calculating the characteristic time of the exponential. For the cyclic loading condition, the model was also initialized at the critical ratio of bonds.

$$\beta(0) = \frac{k_{on}}{k_{on} + k_{off,0}}, \quad (1)$$

The slip bond model parameters ([Table 2](#)) were obtained by fitting the model's fracture timescales to experimental fracture timescales using the root-mean-squared error in tension. Optimization was performed using the NelderMead algorithm in Julia's `Optim` package.²⁵ More details on the fitting can be found in [Method S1](#), slip bond parameters: fitting and optimisation details.

Table 1. Stress controller parameters

Parameter	Value
K_P	1.2E-1
$K_I(1/s)$	0
$K_D(s)$	5E-5
Control velocity (mm/s)	1E-1

Table 2. Slip bond model parameters

Parameter	Value
$k_{on}(1/s)$	3.47E-4
$k_{off,0}(1/s)$	2.68E-4
$f_0(N/m)$	2.49E-1

Stochastic model

The stochastic model is based on the same governing equations as the analytical model, applied to a population of bonds with slip-bond properties. A junction is defined with $N = 100$ bonds, and each bond can independently associate or dissociate with probabilities defined by [Equations 2](#) and [3](#). The choice of $N = 100$ is based on prior work¹⁰ in which the number of effective bonds, N , was calibrated to reproduce the standard deviation in experimental rupture data. By matching the standard deviation of stochastic model predictions to the observed spread in rupture times, a value of $N = 100$ was found to provide the best agreement, with no systematic dependence on loading rate. The junction dynamics are simulated using a Monte Carlo scheme, initialized with a random distribution of bond states evolving in discrete probability steps. Each run results in either complete rupture of the junction (all bonds dissociate) or in reaching the maximum duration (three times longer than the largest repair timescale), in which case the junction is classified as healed. All stochastic simulations were performed using the open-source Julia package `CellAdhesion.jl`.²⁶ Initialization conditions were set by assigning bond states randomly across a network of 100 bonds, each representing an individual molecular connection. For each condition (rupture, healing, or cyclic loading) the proportion of initially closed bonds was determined based on the corresponding ratio used in the analytical model. In each case, the number of closed bonds at the start of the simulation was calculated by multiplying the total number of bonds by the corresponding ratio, with the positions assigned randomly across the network to avoid spatial bias.

Phase map generation

Phase maps were generated for different combinations of Γ_{high} and Γ_{low} each with 40×40 $t_{high} \times t_{low}$ combinations. The range of 40 possible t_{high} values was scaled from 0.01 to 4.5 times the rupture timescale. Similarly, the range of 40 possible t_{low} values was scaled from 0.01 to 1.5 times the repair timescale. For the stochastic model phase maps, the same coordinates were selected, but 100 model simulations were done at each coordinate, and the mean was plotted. The maximum simulation duration was set to 10 times the longest loading cycle period, corresponding to 10 times the sum of one and a half times t_{high} and one and a half times t_{low} . This design choice ensured that any events classified as healing were sufficiently different from rupture or slow damage accumulation regimes.

The simulation timestep, dt , was set to one-hundredth of the smaller of t_{high} and t_{low} , providing adequate temporal resolution across all tested regimes.

RESULTS

Evidence for healing of intercellular junctions in epithelial monolayers under cyclic loading

As a baseline for understanding tissue failure under cyclic loading, we first consider the limit case of creep loading where the material is brought to a chosen stress, which is then kept constant. Previous studies have shown that epithelial sheets fail within a narrow range of tensions, corresponding to the mechanical limit of intercellular adhesion.¹⁰ We selected a load close to this range to challenge tissue integrity without inducing immediate catastrophic failure. We applied a step in stress implemented through proportional-integral-derivative (PID)-controlled

feedback (materials and methods, stress controller) and monitored the load using a force transducer (Method S1, force measurement: calibration procedure). We used an *in vitro* model of MDCK monolayers (materials and methods, cell culture) suspended between the two rods of a custom-built stretching device (Figures 2a and 2b), previously described in Harris et al.²³ We applied constant tensions (Figure 2c) with a mean centered around 0.15 Nm^{-1} ($\pm 0.05 \text{ Nm}^{-1}$). This value was chosen to lie at the lower end of the range of tensions reported to cause rupture in monolayers.¹⁰ This placed the system in a regime where rupture is reliably induced, but not instantaneous, allowing its onset to be systematically characterized. Our experimental data also show that, prior to crack formation, there is very little stress heterogeneity at the tissue scale. Previous work has demonstrated that local cell deformations follow the imposed global strain.^{23,27} This implies that strain is distributed uniformly across the monolayer prior to rupture, supporting our assumption of a globally applied load on

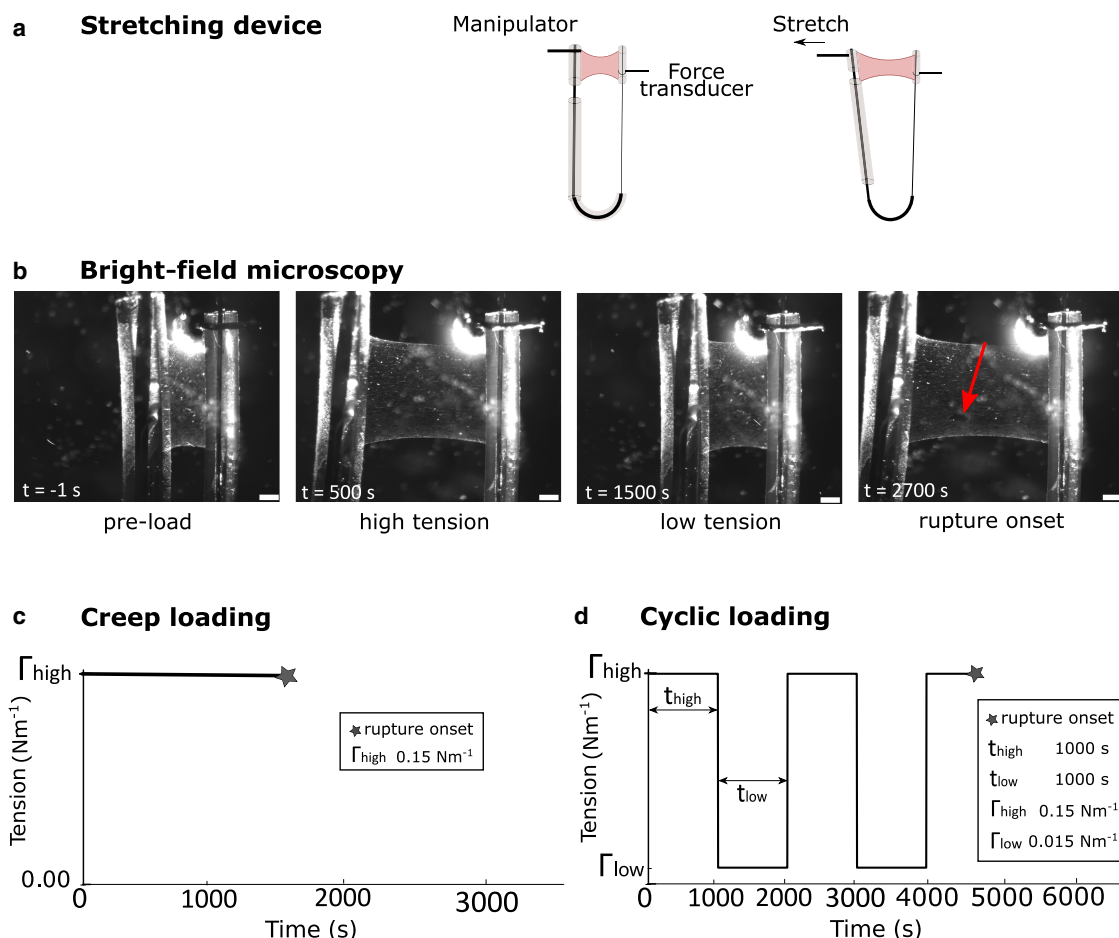


Figure 2. Experimental setup and loading protocol. (a) Tissue stretching device consisting of a monolayer suspended between a thick wire attached to a motorized micromanipulator and a thin pliable wire hooked onto a force transducer. (b) Bright-field microscopy time series of an MDCK monolayer during tension cycles. The rupture time, t^* , is defined as the time of the first defect, which is indicated with a red arrow. Scale bars, $500 \mu\text{m}$. (c) Idealized schematic of creep loading at tension Γ_{high} , where the black star indicates rupture onset. (d) Idealized schematic of cyclic loading between high tension, Γ_{high} and low tension, Γ_{low} . The durations of high and low tension are t_{high} and t_{low} respectively, and the black star indicates rupture onset.

junctions. We note, however, that our imaging resolution does not allow us to resolve subcellular defects or tricellular junctions, which may act as local stress concentrators at smaller length scales. The timing of the first detectable cellular-scale defect within the tissue was defined as the rupture time t^* . Rupture events predominantly initiate within the interior of the monolayer rather than at boundaries (see Figure SM 5), indicating that failure is not caused by edge-induced stress concentrations. For a given tension value, we observed stochastic variation in the rupture times between samples (Figure 3a), but no systematic differences (see Figure SM 6). Higher tension is associated with a faster characteristic rupture time, t^* (Figure 3c). The range of rupture times under creep indicates that failure could occur stochastically even under identical sustained loads. The data

therefore provide an average material timescale for a given tension value, beyond which epithelia cannot maintain cohesion. This observation raises a number of important questions: Is the damage accumulated during constant loading reversible? Can rupture be delayed by intermittently relieving tension to allow for repair?

To explore these questions, we subjected monolayers to cycles alternating periods of high and low tension. The high tension (Γ_{high}) was applied for a duration t_{high} , and the low tension (Γ_{low}) was applied for a duration t_{low} (Figure 2d). The high-tension target was 0.15 Nm^{-1} ($\pm 0.05 \text{ Nm}^{-1}$), matching the constant loading experiments. The target low tension was set to a small nonzero value, to ensure stable control by the PID feedback loop while maximizing the mechanical contrast between

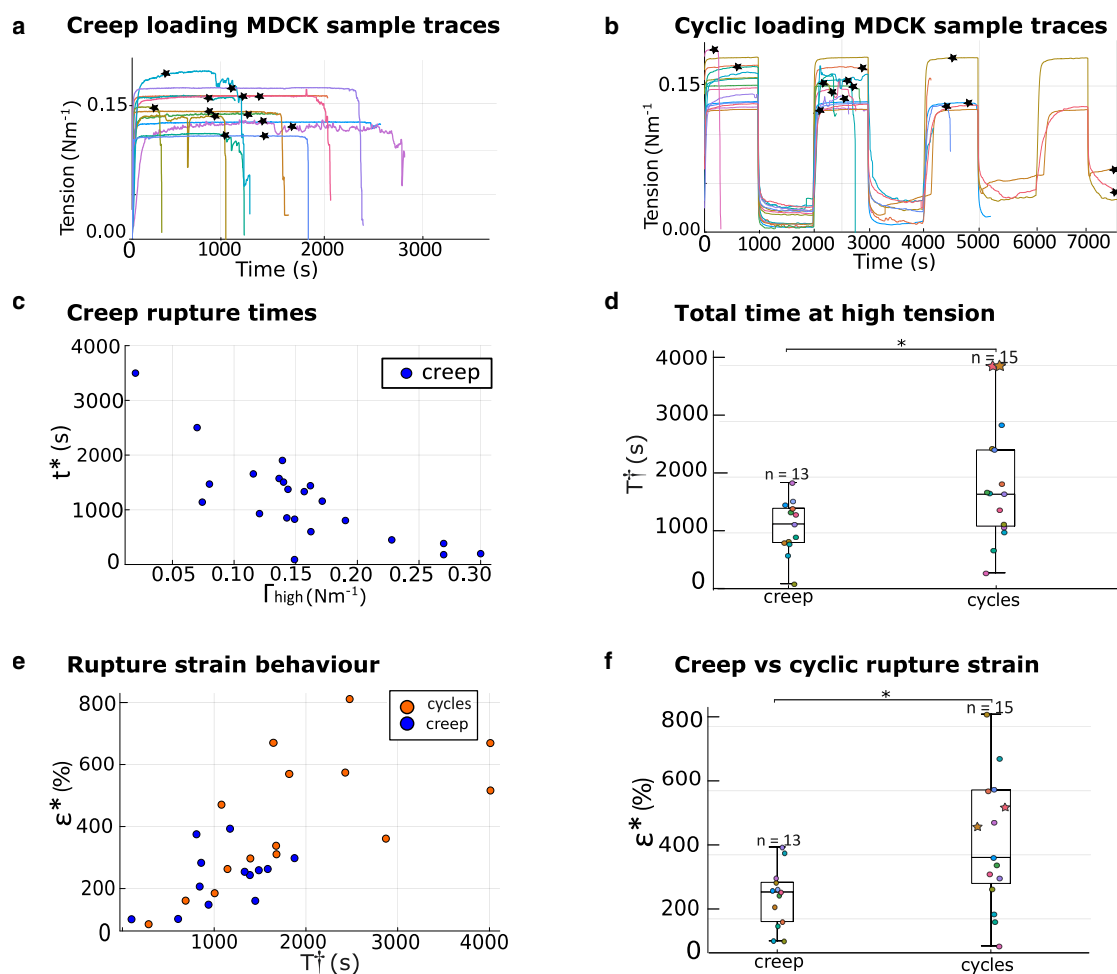


Figure 3. MDCK epithelial monolayers are more resilient to rupture when subjected to cyclic loading than creep loading. (a) Experimental creep loading data. Curves of different colors represent different samples, and the stars denote rupture. (b) Experimental cyclic loading data with Γ_{high} centered at 0.15 Nm^{-1} and low tension and Γ_{low} centered at 0.015 Nm^{-1} . Both t_{high} and t_{low} durations are 1,000 s. Curves of different colors represent different samples, and the stars denote rupture. (c) Creep rupture time t^* for tension across two orders of magnitude. (d) Boxplots comparing the total time at Γ_{high} , $T^\dagger$, under creep and cyclic loading. The central mark indicates the median, the top and bottom edges indicate the 25th and 75th percentiles, respectively, and the whiskers indicate the range excluding outliers. Stars indicate monolayers that did not break by the end of the experiment. $*p = 0.0382$ for Wilcoxon rank-sum test. (e) Relationship between rupture strain, ϵ^* , and time at high tension, $T^\dagger$. (f) Boxplot comparing ϵ^* for creep and cyclic loading; $*p = 0.0146$ for Wilcoxon rank-sum test.

high- and low-tension phases. Experiments (Figure 3b) were limited to 8,000 s, beyond which cells progressively lose apico-basal polarity and monolayers start accumulating defects even without loading. Remarkably, MDCK epithelial monolayers showed, on average, enhanced mechanical resilience under cyclic loading relative to those subjected to constant creep (Figure 3d). The total amount of time spent at high tension, $T^{\dagger}$, was approximately 50% longer in the cyclic loading condition (creep mean lifetime: 1,107 s, cycles mean time spent at high tension: 1,885 s, Wilcoxon rank-sum test for independent samples: $*p = 0.0382$). Furthermore, 5/15 cyclic loading experiments exhibited markedly longer lifetimes when compared with the constant loading test data and were statistical outliers. Notably, in two of these cases, the monolayer did not rupture within the experimental window (starred data points, Figure 3d), implying that their actual rupture times would extend even further. Thus, monolayers can endure high tension for substantially longer when subjected to cyclic loading compared with constant loading, and the reported mean time spent at high tension for cyclic loading should be interpreted as a conservative lower bound.

Next, we compared the rupture strain under cyclic and monotonic loading to determine if rupture might be a strain-controlled mechanism. Under constant stress, soft tissues typically display an initial regime of slow creep, followed by accelerated nonlinear deformation in which strain growth becomes increasingly rapid and ultimately diverges as the material approaches fracture.²⁸ During cyclic loading, the global strain response reflects this progression (see Figure SM 7). We quantified the strain in the monolayer and compared the strain at rupture, ϵ^* , across experimental conditions (Figure 3e). For both monotonic and cyclic loading, strain accumulated progressively during periods of applied tension, consistent with the viscoelastic nature of epithelial monolayers. Importantly, the rate of strain accumulation during high-tension phases was comparable across loading conditions (Figure 3f). As a result, cyclic loading, which on average maintained monolayers at high tension for longer durations, led to a greater cumulative deformation, ϵ^* , prior to rupture. Therefore, even though the tissue experienced similar stress levels and deformed at the same rate, the monolayers that were stretched in cycles could deform more before rupture, indicating that failure is not determined solely by reaching a critical strain threshold. This observation argues against a strain-controlled failure mechanism. Interestingly, this also suggests that periodic unloading in the low-tension regime may allow the monolayer to reach greater cumulative deformation before failure. Introducing cycles appears to facilitate continued viscoelastic flow, highlighting that rupture occurs in a stress-limited regime. In our experimental conditions, the absence of a substrate signifies that the sustained load is entirely borne by cell-cell junctions, where the gradual

accumulation of damage may be taking place.^{17,29} To gain insight into the physical origin of this phenomenon, we examined whether junctional bond dynamics could underlie the delayed failure observed under cyclic loading conditions.

Intrinsic timescales of intercellular adhesion complexes control the rupture and equilibrium dynamics of cell-cell junctions

A common approach to modeling intercellular dynamics is to represent intercellular links by slip bonds, whose lifetime decreases exponentially with increasing force (Figure 4a).^{15,21,30,31} Bell's classical model of receptor-ligand binding describes the force-lifetime relationship of a slip bond, which can exist in one of two states: bound or unbound. The dynamics of a slip bond are governed by a force-dependent dissociation rate and a constant association rate, Equations 2 and 3, where $k_{\text{off},0}$ is the dissociation rate constant in the absence of force, f is the externally applied force, and f_0 is a characteristic force scale primarily determined by the difference in free energy between the bound and unbound states.¹⁵ The application of tensile force lowers the energy barrier for bond rupture, thereby increasing the unbinding rate of individual slip bonds.

$$k_{\text{off}} = k_{\text{off},0} e^{\frac{f}{f_0}}, \quad (2)$$

$$k_{\text{on}} = \text{constant} \quad (3)$$

In the present work, we employ Bell's model as an effective description of intercellular adhesion. In epithelial tissues, intercellular adhesion is mediated by large, complex structures, such as desmosomes, which are clusters of desmosomal cadherins acting collectively.³² These adhesion complexes span the entire force transmission pathway between cells, from extracellular cadherin ectodomains through intracellular anchor proteins to the cytoskeleton. This force chain would represent E-cadherin, beta-catenin, and alpha-catenin for adherens junctions and desmoglein-desmocollin, plakoglobin, plakophilin, and desmoplakin for desmosomes. Importantly, the bonds in our model represent effective adhesion units, which can be interpreted as intercellular adhesion complexes, rather than individual intercellular adhesion proteins. The model is therefore agnostic to specific molecular details. While junctions in living tissues can be arranged both perpendicular and parallel to the loading direction, in our model, we assume that the junctional interface is normal to the loading direction. This choice was made because these are the junctions that are observed to rupture first, both in previous work¹⁰ and in our experiments, suggesting that these control the onset of rupture. In contrast, transversely oriented junctions are less likely to fail.

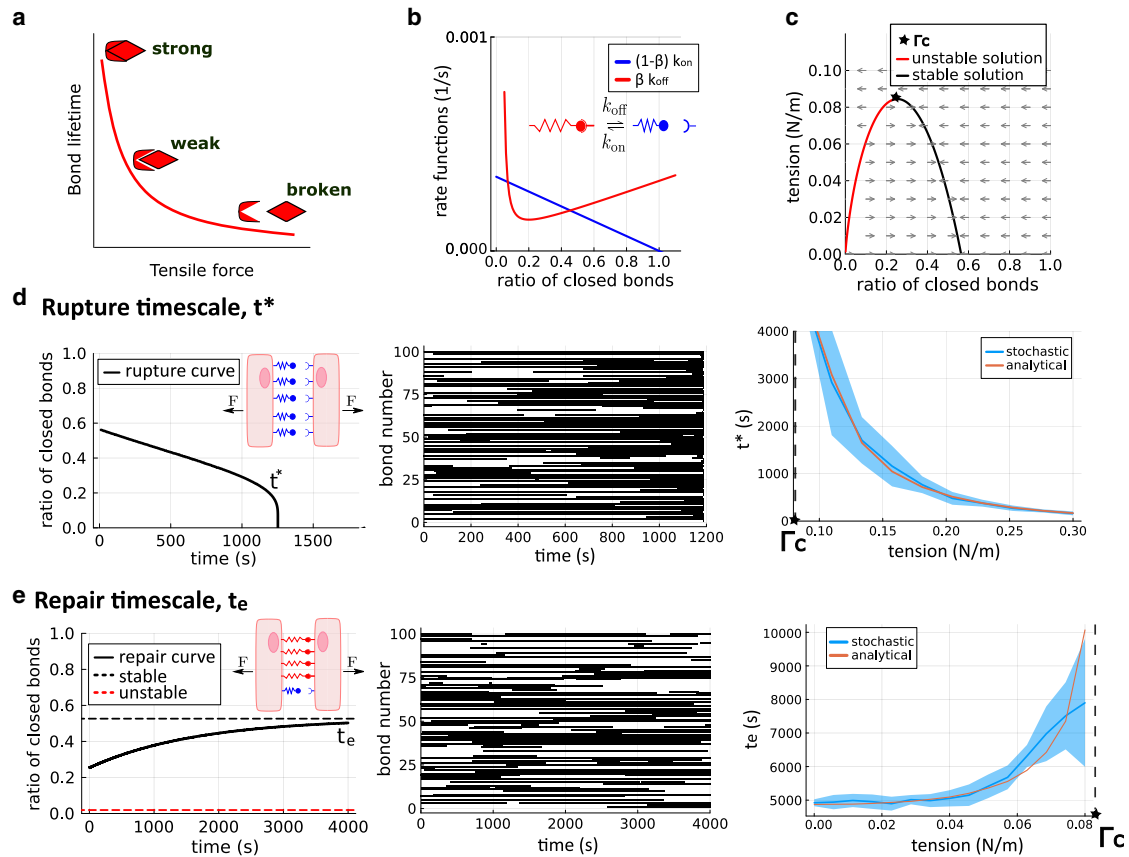


Figure 4. Theoretical framework for modeling an intercellular junction using a population of slip bonds under force. (a) A slip bond's lifetime exponentially decays with increasing tensile force. (b) Rate functions of bond association (blue) and dissociation (red) as a function of the fraction of closed bonds, β , for a given tension (here, 0.04 N/m). Points of equilibrium occur at the intersection between the two curves, where the rates of association and dissociation are equal. The stability of these points is determined by the local slopes of the two curves: a point is stable if small increases or decreases in the fraction of closed bonds produce net rates that drive the system back toward the intersection, and unstable if such deviations push it away. The values for the slip bond parameters, k_{on} and $k_{off,0}$, are found in the [materials and methods](#), Table 2. (c) Quiver plot depicting stable (black) and unstable (red) analytical solutions for the ratios of closed bonds at different values of tension. The critical tension, Γ_c , is the maximum tension value for which a solution exists. (d) Determination of the rupture timescale t^* . Left: rupture timescales, t^* , are defined for tension values larger than Γ_c , initialized at the stable zero tension ratio of closed bonds. t^* is calculated analytically by tracking the ratio of closed bonds over time. Middle: t^* is calculated stochastically by tracking the states of 100 bonds over time (black is an open bond, and white is a closed bond). Right: analytical (orange) and stochastic (blue) rupture timescales as a function of applied tension decrease sharply with increasing tension and diverge near the critical tension Γ_c . The blue shaded region represents the standard deviation of the stochastic model data. (e) Determination of the repair timescale t_e . Left: repair timescales, t_e , are defined for tension values smaller than Γ_c , initialized at the stable ratio of closed bonds for a given tension value. t_e is calculated analytically by tracking the ratio of closed bonds over time. Middle: t_e is calculated stochastically by tracking the states of 100 bonds over time (black is an open bond, and white is a closed bond). Right: analytical (orange) and stochastic (blue) repair timescales as a function of applied tension remain relatively constant at low tensions but increase sharply as the applied tension approaches Γ_c . The blue shaded region represents the standard deviation of the stochastic model data. Close to the transition point, there is an increase in standard deviation for the stochastic model's t_e .

Considering a population of N such bonds allows us to analytically describe how collective bond kinetics govern the stability of cell-cell junctions under mechanical stress. We define the fraction of closed bonds at any given time as β . The dynamics of β are governed by the balance between bond association and dissociation, Equation 4, where the first term represents the rate of bond formation, which decreases linearly as β approaches unity, and the second term describes the rate of bond dissociation, which increases exponentially with force. Γ denotes the applied tension (force per unit length), and $\rho = \frac{N}{l}$ is the density of available bonds along a junction

of length l . We assume tension is equally distributed across all closed bonds.

$$\frac{d\beta}{dt} = \underbrace{(1 - \beta) k_{on}}_{\text{association}} - \underbrace{\beta k_{off,0} e^{\frac{\Gamma}{\rho \rho \rho}}}_{\text{dissociation}} \quad (4)$$

The rates of bond formation and bond loss can be written as functions of β (Figure 4b). At the intersection points of the two functions, the junction is in equilibrium, meaning that there is no net loss or gain of bonds over time, such that the rate of change of β is zero. The stability of each intersection point depends on the relative slopes of the two functions. For instance, if a small increase in β causes

dissociation to dominate and a small decrease causes association to dominate, the system is driven back toward the intersection, making the equilibrium stable. In contrast, if deviations from the intersection are amplified, such that both an increase and decrease in β move the system away from the intersection point, then the fixed point is unstable. Increasing tension shifts the dissociation curve upward (in red, Figure 4b), which alters the position and eventually the number of intersection points. If there are no intersection points, there is no steady-state solution to Equation 4, and therefore the cell-cell junction inevitably fails. Pairs of stable and unstable fixed points exist for tension values ranging from zero up to a maximum tension, which we define as the critical tension value, Γ_c (Figure 4c). Conceptually, Γ_c represents the maximum mechanical load that an intercellular junction can bear. As such, Γ_c may also serve as a mechanical threshold for tissue integrity, beyond which stable cell adhesion cannot persist indefinitely.

The slip bond model parameters (see materials and methods, analytical model) were obtained by fitting the model's fracture timescales to experimental fracture timescales using the experimental creep data. Further details on the fitting can be found in Methods S1, slip bond parameters: fitting and optimisation details. For convenience, we treat the product $f_0\rho$ as a single effective parameter, so that the influence of bond density is absorbed into an empirical fitting constant. The fitted parameters capture the experimental fracture dynamics, with simulated fracture times closely matching the experimental measurements across the tested range (see Figure SM 8). By varying model parameters such as the bond kinetics or the applied tension, we can now study how these changes affect the mechanical stability of intercellular junctions.

Our steady-state analysis identifies stable and unstable adhesion regimes determined by the applied tension. Having delineated these regimes, we then analyze the transitions across them. We begin by considering the characteristic time to failure, which provides a direct comparison to the experimentally measured rupture time. For tension values larger than the critical tension Γ_c , we define a rupture timescale, t^* , as the time required for all bonds to dissociate under constant tension, leading to junction failure (Figure 4d). For tension values below Γ_c , we define a repair timescale, t_e , as the time required for the junction to recover a stable equilibrium of connected bonds, under constant tension (Figure 4e), which can be calculated through the exponential half-life. Both timescales depend on the way the population is initialized; for the rupture timescale measurements, $\beta(t=0)$ was set at the zero-tension equilibrium fraction of closed bonds, while for the repair timescale, $\beta(t=0)$ was initialized at the closed-bond fraction at Γ_c . Further details can be found in materials and methods, analytical model.

While the analytical model provides valuable insights into the average behavior of adhesion at intercellular junctions, it inherently describes an idealized, deterministic

system. In reality, adhesion complexes operate in a noisy environment with fluctuations that cannot be fully captured by a purely deterministic framework. Near the critical tension, stochastic behavior can be the difference between a junction staying stable or failing catastrophically. To account for these effects, our analysis can be performed using a stochastic multi-bond framework governed by Equations 2 and 3. A cell-cell junction is defined as an interface with a number of bonds, N , which can be in one of two states: bound or unbound. The binding and unbinding of individual bonds with slip bond properties are treated as probabilistic events. The dynamics of a population of bonds can be simulated using a kinetic Monte Carlo scheme, initialized with a random distribution of states. We explored different values of N over three orders of magnitude (see Figures SM 9 and SM 10) and chose $N = 100$, a value also used to capture MDCK monolayer dynamics and experimental noise levels in prior studies.¹⁰ For the stochastic model, t^* was defined for $\Gamma > \Gamma_c$, as the average time to failure across 100 creep simulations, each initialized at the zero-tension equilibrium fraction of closed bonds (see materials and methods, stochastic model). t_e was defined for $\Gamma < \Gamma_c$, starting from the steady-state bond fraction at Γ_c . We estimated t_e by tracking the states of 100 bonds over time and taking an average. A further criterion was implemented, requiring that the starting point lie at least one standard deviation away from the stable fixed point, computed from the bond ratios across the 100 simulations at steady state. This criterion ensures that the repair timescale is clearly distinguished from stochastic fluctuations at steady state.

Rupture and repair timescales can be computed for a range of tensions. As tension increases from zero to Γ_c , t_e increases exponentially, indicating slower recovery dynamics and increased variability near the transition point. In contrast, for tensions exceeding Γ_c , t^* decays exponentially, reflecting faster dissociation and mechanical failure under larger loads. When comparing the analytical and stochastic models, we find strong agreement across a wide tension range. The main difference arises for the repair timescale t_e close to Γ_c . For these tensions, a large portion of stochastic trajectories fractured instead of repairing, as reported in Figure SM 11. Trajectories that ruptured were included only up to failure, to capture the initial recovery dynamics. This causes an underestimate of the analytical repair timescale close to the critical tension. An alternative definition of the stochastic repair timescales where trajectories that ruptured are assigned an infinite repair time is considered in Figure SM 12. This alternative definition does not qualitatively alter the results, and the same trends and conclusions are preserved. Therefore, the stochastic model of junctions predicts a gradual shift in the balance of rupture and repair probabilities, while the analytical model exhibits an absolute boundary between stable and unstable behavior. In

the following sections, we explore the interplay between the intrinsic timescales of bond dynamics and the extrinsic timescales of cyclic loads in tuning epithelial tissue behavior.

Intercellular junction stability under cyclic mechanical loads

We now investigate how cyclic loading affects the integrity of an intercellular junction. The applied load alternates between a high tension, Γ_{high} , and a low tension, Γ_{low} , sustained for durations of t_{high} and t_{low} , respectively. For cyclic loading to meaningfully engage both the rupture and healing processes, Γ_{high} must exceed the critical tension Γ_c , while Γ_{low} must remain below Γ_c . To systematically explore the dynamic range of a junction's response, we test a broad range of loading frequencies and duty ratios (the relative durations of t_{high} and $t_{\text{high}} + t_{\text{low}}$) (materials and methods, phase map generation). The results can be visualized through a map for the analytical (Figure 5a) and stochastic (Figure 5b) models. The color bar quantifies the stability of the junction relative to the creep case, using the stability index, Θ , defined in Equation 5. $T^\dagger$ is the total time spent at high tension until rupture (the same quantity previously used to analyze experimental data), and t^* is the rupture timescale under constant high tension, as previously defined. For experimental data, we obtain Θ by averaging over independent repeats of an experimental condition, while for the stochastic model, we average over 100 simulated trajectories. Average quantities are indicated by a bar in Equation 5.

$$\langle \Theta(\Gamma_{\text{high}}, \Gamma_{\text{low}}) \rangle = \frac{\bar{T}^\dagger - \bar{t}^*}{\bar{T}^\dagger} \quad (5)$$

When the time to rupture $T^\dagger$ is identical to the creep case t^* , $\Theta = 0$. When rupture doesn't happen over the duration of the simulation, $\Theta = 1$. Intermediate values (green to blue) are defined by the fractional delay in failure relative to t^* . Higher values indicate greater prolongation of junction lifetime compared with t^* , while lower values reflect only marginal extension. In this intermediate regime, junctional failure is not triggered by a single supercritical stress event, but by the progressive build-up of damage over many cycles. Each loading cycle gradually pushes the junction closer to rupture leading to a time to rupture $T^\dagger$ much longer than a single loading period. This regime reveals a fundamentally different pathway to failure that cannot be captured under monotonic or steady loading. Experimental observations from both creep and cyclic loading conditions can be mapped onto this phase diagram: creep-induced failure may be represented along the t_{high} axis and defines the t^* , $\Theta = 0$, for a given tension (red cross Figure 5a), while cyclic loading resulted in delayed failure (orange cross, Figure 5a). Our analytical and stochastic models predicted values of Θ , close to one another (an analytical $\Theta = 0.28$ and a stochastic $\Theta = 0.23 \pm 0.18$) and close to the experimentally observed value of $\Theta = 0.32 \pm 0.27$.

Overall, our map reveals three distinct regimes of junction behavior in response to cyclic loading: (1) stable repair, (2) slow damage accumulation, and (3) rupture. Moreover, the model demonstrates that transitions between regimes can be induced by tuning either the loading frequency or the duty ratio of t_{high} and t_{low} (Figure 5). These observations

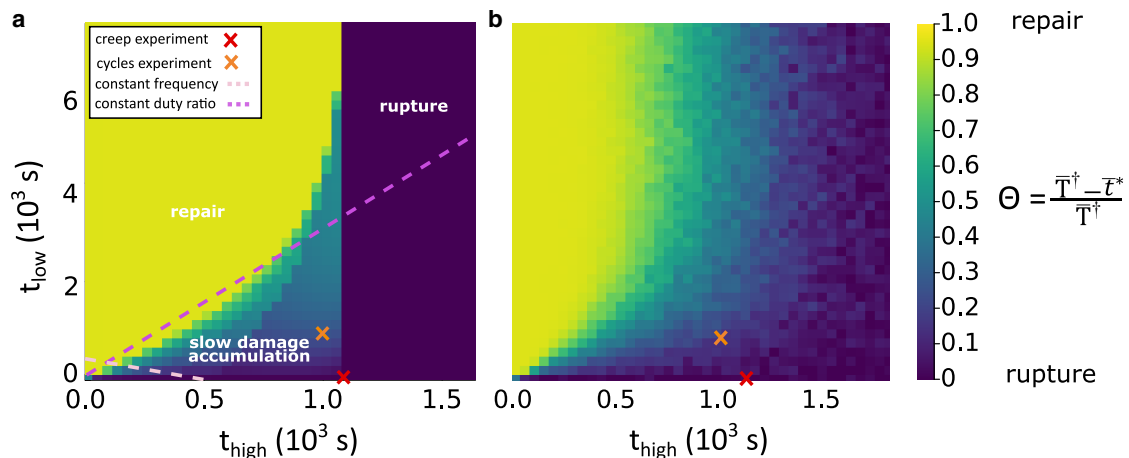


Figure 5. Models of intercellular dynamics reveal three distinct cell-cell junction behaviors in response to cyclic loading. (a) Analytical heatmap illustrating junction behavior under cyclic loading, defined between $\Gamma_{\text{high}} = 0.15 \text{ Nm}^{-1}$ and $\Gamma_{\text{low}} = 0.015 \text{ Nm}^{-1}$ (matching prior experimental conditions) for a broad range of combinations of t_{high} and t_{low} . The color scale encodes the normalized metric $\frac{\bar{T}^\dagger - \bar{t}^*}{\bar{T}^\dagger}$ with values 1.0 (yellow) indicating full healing, 0 (purple) indicating immediate rupture, and intermediate values representing a slow damage regime where bonds are progressively lost due to incomplete recovery during the low tension phase. The experimental data points can be overlaid on the map. The red cross indicates creep experiments leading to rupture at the critical time t^* , and the orange represents cyclic loading experiments ($t_{\text{high}} = t_{\text{low}} = 1,000$ s) that lie within the slow damage regime. The pink dashed line denotes a constant frequency of 0.001 Hz, and the violet dashed line represents a constant duty ratio, $\frac{t_{\text{high}}}{t_{\text{high}} + t_{\text{low}}}$. (b) Stochastic heatmap defined with identical cyclic loading conditions.

raise several key questions: how does the balance of bond loss and gain explain the different regimes of junctional stability? And, how do changes in the loading parameters Γ_{high} , Γ_{low} , t_{high} , and t_{low} influence the structure of the maps?

Discrete transitions in adhesion complex dynamics govern junctional resilience under cyclic loading

Considering both stochastic simulations (Figures 6a–6f, blue) and analytical solutions (Figures 6g–6i, red), we

resolve how distinct regimes of cell-cell junction behavior emerge from the balance between bond association and dissociation under cyclic loading. In the rupture regime (left column: Figure 6), bond dissociation dominates under sustained high tension, leading to monotonic accumulation of bond ruptures and junction failure within a time shorter than t_{high} . In the full recovery regime (right column: Figure 6), all broken bonds re-associate during each low-tension phase, fully restoring the equilibrium population of bonds and maintaining junction integrity over repeated cycles. This explains why we observe a saturation at large

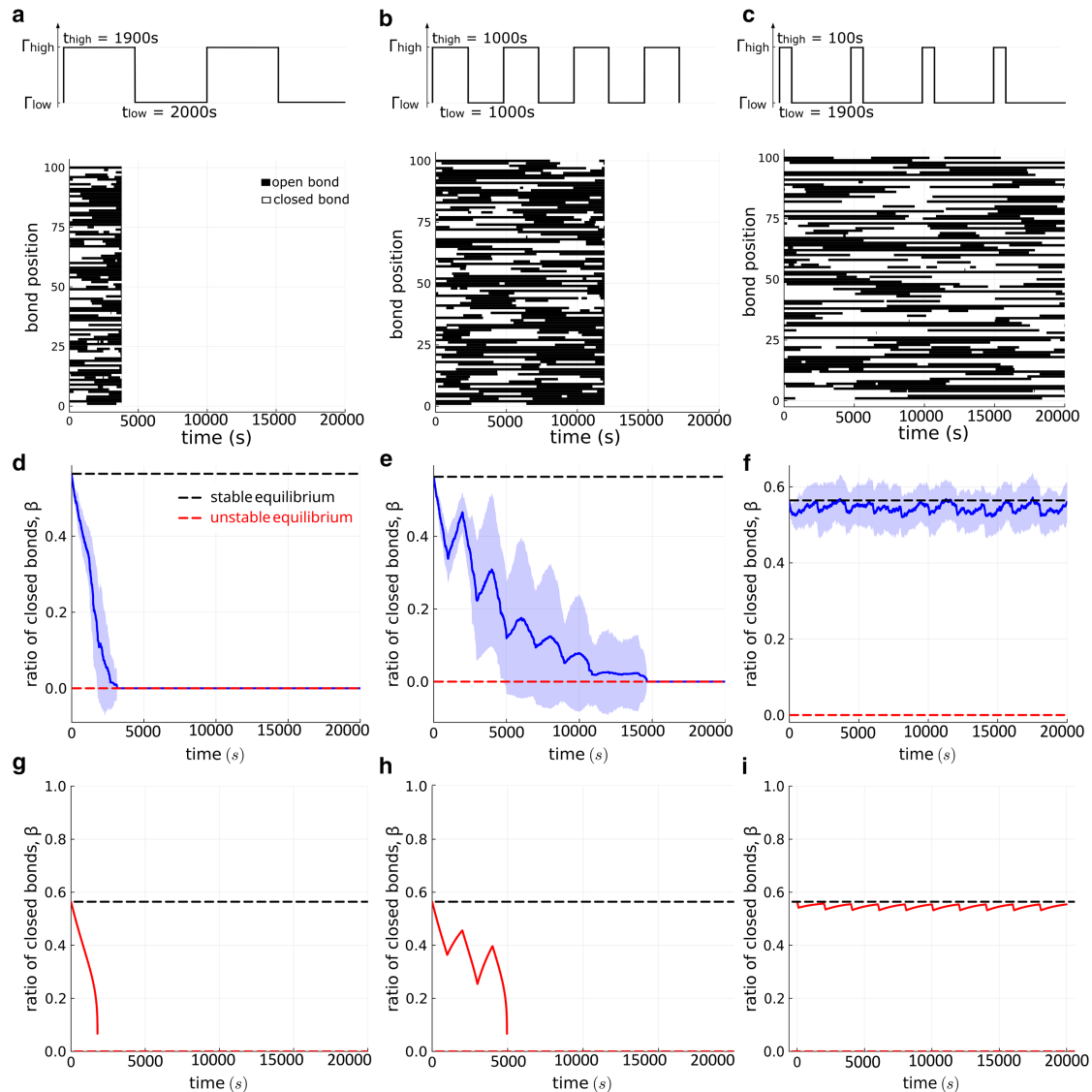


Figure 6. Balance of bond association and dissociation predicts cell-cell junction response to cyclic loading. Left column: $t_{\text{high}} > t^*$, such that all bonds monotonically dissociate within the first cycle. Middle column: $t_{\text{high}} \approx t_{\text{low}}$, such that the relative durations at high and low tension lead to slow damage accumulation, resulting in delayed rupture. Right column: $t_{\text{high}} \ll t_{\text{low}}$, such that all accumulated damage is fully recovered, and the fraction of closed bonds returns to equilibrium after each cycle. (a–c) Top row: loading conditions for each regime: (a) rupture, (b) slow damage accumulation, and (c) recovery. Bottom row: temporal maps of junction behavior from stochastic simulations. A population of 100 bonds is shown. Black represents an open bond and white a closed bond. (d–f) Temporal evolution of the fraction of closed bonds from stochastic simulations. Solid lines indicate the mean, and shaded regions denote stochastic variability across repeated simulations. The red dashed line indicates the unstable equilibrium and the black dashed line the stable equilibrium. (g–i) Temporal evolution of the fraction of closed bonds from the analytical model. The black dashed line indicates the stable equilibrium.

t_{low} on the map of [Figure 5](#): once the junction has relaxed to its low-tension equilibrium, additional time at Γ_{low} does not further increase the fraction of closed bonds. In the slow damage accumulation regime (middle column: [Figure 6](#)), the junction exhibits partial recovery between cycles, but the cumulative net loss of bonds progressively shifts the junction toward failure. A phase space representation of the loss and gain of bonds can be found in [Figure SM 15](#).

Furthermore, there is a conserved pattern within the slow damage accumulation regime that can be distinguished by the presence of distinct fringe-like bands in the analytical model. These bands mark discrete transitions in rupture time, corresponding to failure occurring after the first, second, third, or subsequent cycles (see [Figure SM 16](#)). Mechanistically, this layered structure reflects the discrete nature of bond reformation. Each cycle at low tension provides a quantized opportunity for bonds to re-associate, incrementally offsetting accumulated damage and thereby shifting the rupture event to a later cycle. As the number of cycles to failure increases, the spacing between successive fringes becomes progressively smaller, and the junction approaches an asymptotic limit beyond which it transitions to the stable recovery regime. Although this structure is clearly expressed in the analytical model, it does not appear in the stochastic map, where fluctuations blur these discrete transitions. These results highlight that the observed macroscopic stability regimes are rooted in the emergent kinetics of intercellular adhesion complexes.

These observations show that the slow damage accumulation regime arises from the interplay between damage and recovery within each loading cycle. To capture this balance quantitatively, we consider how the relative durations of the high- and low-tension phases compare with intrinsic timescales of rupture and repair. Specifically, the extent of damage accumulated per cycle can be estimated from the ratio of the duration of application of high tension, t_{high} , to the rupture timescale, t^* , while the degree of recovery depends on the ratio of the duration of low-tension application, t_{low} , to the repair timescale, t_c . Additionally, the high- and low-tension amplitudes, Γ_{high} and Γ_{low} , can be normalized by the critical tension Γ_c . Looking at the data through this lens removes any effects arising from the choice of specific loading parameters, allowing for comparisons across different loading conditions.

Timescale normalization reveals a universal master stability map

Normalizing the loading durations (t_{high} and t_{low}) by the intrinsic rupture (t^*) and repair (t_c) timescales reveals that the same three regimes, rupture, slow damage accumulation and recovery, emerge across a wide range of absolute tension amplitudes ([Figure 7](#)). The general shapes of the boundaries between different regimes are qualitatively conserved across

different combinations of loading parameters. When both Γ_{high} and Γ_{low} are far from Γ_c , the system exhibits well-defined and robust regimes. This demonstrates that epithelial stability is governed by the relative interplay between intrinsic adhesion kinetics and extrinsic loading timescales, rather than by the absolute magnitude of stress. For example, a tissue may persist longer under a high peak stress that is periodically relieved by low-tension intervals than under a continuous intermediate stress, highlighting that higher loads can be more sustainable when the loading pattern aligns with the intrinsic timescales. Despite differences in noise and individual bond variability, both the analytical and stochastic models produce equivalent regime boundaries when expressed in dimensionless timescales. This convergence indicates that the normalized map captures the fundamental physics of rupture-repair competition, independent of model implementation. Moreover, the slope of the initial linear region between healing and slow damage accumulation was found to be robust to changes in tension. When perturbing bond kinetics by varying k_{on} and $k_{\text{off},0}$ while preserving their ratio, the structure of the normalized stability maps also remained robust (see [Figure SM 14](#)). This highlights a key strength of the framework: by normalizing mechanical inputs by intrinsic molecular timescales, all loading conditions collapse onto a single master stability map.

Another key observation is that as either value approaches Γ_c , the phase boundaries become less distinct and more sensitive to stochastic fluctuations. Notably, the closer Γ_{low} is to Γ_c , the broader and more diffuse the transition between the rupture, slow damage, and recovery regimes becomes. We find that increasing Γ_{low} while keeping Γ_{high} constant progressively reduces the size of the healing region. In this case, the effective window for rebinding decreases, especially as the repair timescale t_c becomes much longer relative to t_{low} . Consequently, junctions spend a larger fraction of the cycle under damaging conditions, shifting the balance from reversible to irreversible failure. Interestingly, when Γ_{low} is set to nonzero values, we identify a fourth stability regime (white arrow, [Figure 7](#)), which appears as a rectangular band within the slow damage accumulation regime and extends into the healing phase space for increasing values of Γ_{low} . In this regime, the bond loss during the first high-tension phase drives the system across the unstable fixed point, placing it within the diverging regime of the model. In this case, even when the tension is reduced to Γ_{low} , the junction continues to accumulate damage, albeit at a slower rate. In this fourth regime, initial conditions become especially important, as small differences in the initial bond population can determine whether the junction crosses the unstable solution or not (see [Figure SM 13](#)).

The normalized framework enables direct comparison across tissues or conditions by mapping experimental measurements (e.g., cycle frequency and duty ratio) onto the same dimensionless axes. Tissue stability can therefore be predicted independently of specific parameter choices,

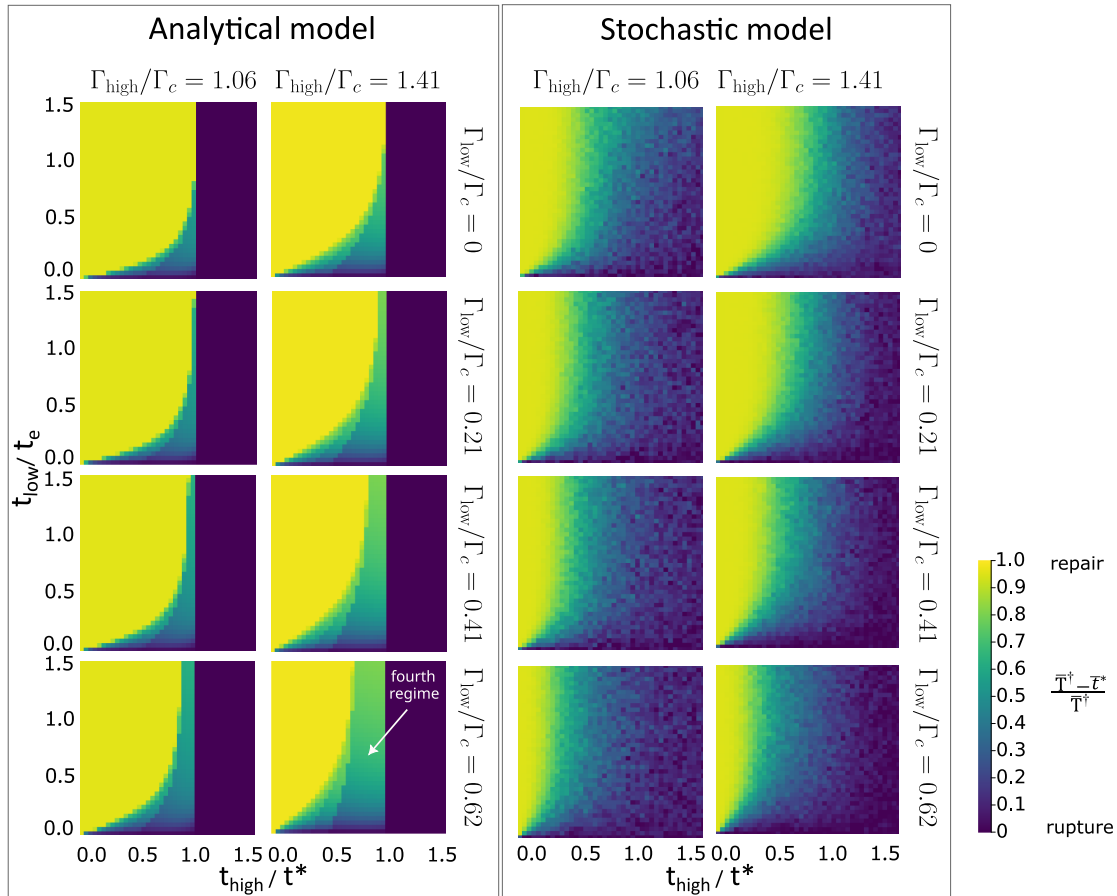


Figure 7. Distinct regimes of epithelial stability, with conserved characteristics. Different combinations of Γ_{high} ranging from 0.09 Nm^{-1} to 0.12 Nm^{-1} and Γ_{low} ranging from 0 Nm^{-1} to 0.0525 Nm^{-1} . For each combination, the stability graph is computed for different ranges of t_{low} and t_{high} . In these graphs, t_{low} is normalized by the repair timescale, t_e , and t_{high} is normalized by the rupture timescale, t^* , of the corresponding tension values. Columns one and two capture the dynamics of the analytical model, and columns three and four capture the dynamics of the stochastic model. The white arrow points to a fourth regime that emerges for nonzero Γ_{low} values.

making the framework broadly applicable to biological systems with diverse molecular dynamics and different loading conditions.

Building on the generality of the mechanical response, to what extent do these phase maps remain invariant to the details of force transmission and bond kinetics complexity? We extended the model to include local force redistribution across adhesive units, generating local stress concentrations. As a result, the transition from recovery to rupture happens at a lower critical tension. However, this does not alter the emergent dynamics (Method S1, local force redistribution model implementation). Rupture and repair timescales follow the same dependence on tension as in the global load-sharing model, differing only by a horizontal shift to lower critical tension. Importantly, the qualitative structure of the cyclic loading phase maps remains unchanged. We also modified the intrinsic bond kinetics, to capture a more complex bond model. Catch-bond-like behavior was incorporated by introducing transient stabilization at intermediate tension. When the rupture timescale t^* is defined relative to the steady-state

bond fraction at the corresponding low-tension condition, the same cyclic loading phase maps are recovered (see Method S1, catch-bond-like behaviour model implementation). Both local stress redistribution and more complex bond kinetics rescale model parameters without modifying the underlying dynamics, reinforcing the robustness of the identified regimes.

DISCUSSION

Our experiments showed that when epithelial monolayers are held under a sufficiently high constant tension, they progressively deform and ultimately rupture, with higher loads producing faster failure and smaller strains at the rupture point. We then investigated the reversibility of damage by subjecting the monolayer to cycles of controlled supercritical and subcritical tension. Under repeated cycles of high and low tension, the monolayers' lifetime at high load and their overall deformation increased markedly, compared with constant loading. To determine whether this enhanced

resilience may arise from emergent physical behavior rather than active adaptation, we turned to a simple micromechanical model of a population of intercellular adhesion complexes with slip-bond kinetics. Under cyclic loading, the model predicted a rich range of behaviors, ranging from fast rupture to stable adhesion. When a junction approached rupture, damage accumulated continuously as intercellular bonds weakened and separated. Periods of low tension appeared to allow partial recovery of bonds, enabling the tissue to withstand high stresses for longer. A distinct regime of slow damage accumulation accounts in particular for our experimental observation of delayed rupture under cyclic loading. The key to understanding these dynamics lies in quantifying two timescales: the rupture timescale at high load, corresponding to the time required for all the bonds in a population to dissociate, and the repair timescale at low or zero load, over which the bond population returns to a kinetic equilibrium. The magnitudes of these timescales, relative to the durations of loading and rest, control the outcome of exposure to cyclic loading.

Our results reveal emergent damage accumulation and healing behaviors arising from the coupled dynamics of cellular mechanics and junctional rupture and repair. In response to deformation, the rheology of the tissue sets the stress magnitude to which cell-cell junctions are subjected. Although the intercellular adhesion complexes (cadherin-catenin, desmosomal cadherins-plakoglobin-desmoplakin) linking the cytoskeleton of adjacent cells contribute negligibly to bulk deformation, the dynamic remodeling of these intercellular adhesive units under force plays a critical role in setting tissue strength and controlling the onset of failure. This coupling highlights that studying the mechanical behavior of epithelial tissues requires a holistic approach, encompassing the entire force transmission pathway, from the complexes that mediate adhesion to the collective deformation of all the cells in the tissue. The stochastic turnover of intercellular adhesion complexes shapes the transition between reversible and catastrophic damage accumulation. Importantly, the emergent rupture and repair behavior can arise naturally from the binding and unbinding kinetics of intercellular adhesion complexes under load. Our results also highlight the broader significance of cellularization. Tissue strength is primarily set by intercellular adhesion complexes that bear load across junctions, while rheological behavior is governed by intracellular cytoskeletal networks that determine the viscoelastic response. This separation enables independent tuning of strength and deformation dynamics.³³

When tissues operate near the boundaries separating rapid rupture, repair, and slow damage accumulation, their response becomes highly sensitive to bond association kinetics, making the repair timescale a key determinant of tissue integrity. This repair timescale is distinct from the cellular processes involved in wound healing, such as lamellipodial protrusion or purse string closure³⁴; rather, it arises from the reassociation of intercellular adhesion links once

tissue tension becomes subcritical. Our experiments support such a hypothesis because they are performed on freely suspended monolayers, a condition with no cell-substrate interactions, in which migration-driven neighbor exchanges are not observed.^{27,35}

Our findings outline a strategy to probe the dynamic response of tissues under cyclic loading, providing a means to address how epithelial tissues preserve their integrity under repeated mechanical perturbations. Systematic variation of Γ_{high} , Γ_{low} , t_{high} , and t_{low} enables direct measurement of rupture onset and of the gradual accumulation of damage over successive cycles. However, direct experimental characterization of the repair timescale remains challenging, as it involves molecular processes occurring below optical resolution and over a wide range of timescales.³⁶ In principle, the repair timescale, t_e , can be estimated indirectly by taking advantage of the broadly conserved normalized stability maps. The experimentally measured t^* and applied t_{high} and t_{low} can be used to locate the loading protocol on the map, from which t_e can be inferred relative to the low-tension phase. In the case of our MDCK monolayer experiments, the minute-scale t_{low} confined the tissue to a partial-repair regime. This observation suggests that the repair timescale t_e in MDCK monolayers is likely on the order of tens of minutes. While direct measurements of junctional repair timescales remain scarce, prior studies of E-cadherin and associated p120-catenin complexes show turnover and membrane re-incorporation occurring on timescales of tens of minutes.^{12,37,38} Photobleaching experiments of keratins and desmosomes indicate turnover times on the order of hours.^{39–42} This indicates that the timescale for mechanical stability is set by the persistence of the whole adhesion complex, rather than by the turnover of individual molecular components. This raises an important mechanistic question. How can we understand the effective adhesion properties of larger adhesion units in terms of the molecular turnover? We anticipate that advances in imaging of single-molecules and of protein-protein interactions may enable direct measurement of the lifetime of individual adhesion proteins as well as their open or closed configuration under controlled mechanical load.^{36,43} Such measurements would allow quantitative validation of the predicted coupling between junction dynamics and tissue-scale forces.

Our analysis focuses on a single population of intercellular adhesion complexes with uniform kinetics, providing a simple approach to connect junctional dynamics to macroscopic mechanical behavior. In reality, healthy tissues draw on a broad range of adhesion complexes, made of different proteins (cadherins, desmosomal cadherins, nectins, tight junction proteins, JAMs, etc.), with different properties. As a result, junctional complexes can vary in stiffness and compliance and therefore carry different fractions of the total load rather than sharing it equally. Junctions can therefore form and stabilize even under minimal or oscillatory loads,⁴⁴ and their specific characteristics may provide a

basis for tissue resilience, allowing different intercellular adhesion complexes to contribute to cell adhesion.⁴⁵ Incorporating heterogeneity into the model, offers a testbed for this hypothesis while paving the way toward a multiscale description that captures the physiological complexity and reveals how intercellular dynamics shape tissue strength.

The recovery response we highlight is relevant beyond biological tissues, offering direct analogies to a broad class of soft materials such as self-healing polymers and dynamic hydrogels.^{46–49} Although these materials have different microscopic components, they rely on comparable reversible binding mechanisms. In these systems, the introduction of reversible linkers, such as dynamic covalent bonds, hydrogen bonds, and metal-ligand complexes, enables materials to dissipate energy and recover after damage.⁴⁶ In our system, the mechanical work up to rupture onset can be estimated by integrating the stress-strain curve up to the first detectable defect (see Figure SM 17), providing a measure of pre-failure dissipation associated with changes in junction integrity. However, in soft materials, fracture is more appropriately understood in terms of energy dissipation during crack propagation, which is beyond the scope of the present model. Recent work on hydrogels with sacrificial bonds has shown that resistance to rupture is not governed solely by bond strength, but also by the interplay between bond breaking, bond reformation, and stress redistribution.^{50–52} These studies demonstrate that reversible bonds can delay or arrest crack propagation by redistributing stress. Similarly, recent work on epithelial junctions highlights the role of cortical dissipation in regulating rupture by modulating local stress concentrations and failure thresholds.⁵³ Our model captures the same underlying principle at the level of junctional dynamics, where macroscopic resilience arises from the collective behavior of reversible adhesion complexes and load redistribution, rather than from single-bond energies. The adhesion dynamics seen in epithelial junctions may therefore inform the design of bioinspired materials with tunable strength and reversible adhesion.⁵⁴ For instance, tuning the force sensitivity or timescales of adhesive interactions could enable both rapid healing and long-term durability. Similar design strategies are now being explored in self-healing polymer networks and hybrid hydrogels, where hierarchical or multi-bond architectures enhance performance under cyclic loading.^{47,48} Ultimately, the physics of epithelial cohesion offers a foundation for engineering synthetic materials that are simultaneously strong and self-repairing.

The cyclic loading periods used in this study, on the order of 1,000 s on and 1,000 s off, correspond to those of processes such as intestinal peristalsis or bladder filling and voiding, which occur over tens of minutes and involve substantial mechanical stress.⁵ Physiologically, our experiments and simulations indicate that soft biological tissues can tolerate extended periods of overload without rupture, provided these are interspersed with phases of low tension

that permit recovery. *In vivo*, loading is unlikely to present step transitions, as tissue viscoelasticity smooths stress variations, reducing peak stresses and enhancing dissipation. Such relaxation shifts the system toward more stable regimes, making the square-wave loading considered here a conservative worst-case scenario for junctional stability. Determining which regimes tissues operate in, and whether they are close to boundaries that can shift tissues to loss of integrity, may therefore help assess their susceptibility to mechanical failure and disease.^{8,55–58} Tissues positioned close to the recovery regime boundaries may be particularly vulnerable, as modest random fluctuations in tension or junctional stability can tip the system into failure. Such mechanical vulnerability could contribute to multifactorial diseases, such as Crohn's disease and epidermolysis bullosa.^{59,60} Comparison of the predictions of our simulations to experimental data may highlight the presence of active cellular responses to damage. Indeed, enhanced recovery, adaptive adhesion, or delayed failure likely reflect the activation of active feedback and complex signaling pathways that modulate intercellular adhesion or cytoskeletal rheology in response to load. Therefore, the subtle balance between damage accumulation and healing may be physiologically relevant and important for assessing the health of tissues.

CONCLUSION

Overall, contrary to the notion of stable and static intercellular junctions conveyed by their continued presence in time-lapse videos of epithelia, the intercellular adhesion complexes that underlie these junctions are inherently dynamic, with the number of closed adhesion complexes fluctuating as a function of applied load and time. The interplay between applied force cycles and intrinsic binding kinetics of intercellular adhesive units governs the transition between rupture and repair. These insights pave the way for a more predictive understanding of epithelial tissue mechanics in which adhesion dynamics and loading timescales jointly determine tissue stability.

DATA AVAILABILITY

All code and data used in this study are archived on Zenodo, DOI: [10.5281/zenodo.21777729](https://doi.org/10.5281/zenodo.21777729).

ACKNOWLEDGMENTS

We thank the past and present members of the Charras and Kabla laboratories for discussions. We wish to acknowledge Nargess Khalilgharibi and Jonathan Fouchard for technical support with LabVIEW development and Lucia Baldauf for her important contribution in laboratory training and insightful discussions. E.P. was supported by the Engineering and Physical Sciences Research Council Centre for Doctoral Training in Sensor Technologies for a Healthy and Sustainable Future (EP/S023046/1), and G.C. was supported by an sLoLa grant from the British Biotechnology and Biological

Sciences Research council (BBSRC, grant no. BB/V019015/1). A.B. was supported by the European Research Executive Agency via the MSCA Postdoctoral Fellowships (BIOFRAC, project number: 101105376).

AUTHOR CONTRIBUTIONS

All authors conceived the project and wrote the paper. E.P. performed the experiments, imaging, and analysis. G.C. advised on the analysis of experimental data. E.P. implemented the analytical and stochastic modeling. A.B. and A.K. designed the stochastic model and advised on the analysis of computational data. A.B., G.C., and A.K. oversaw the entire project. All authors discussed the results and the paper.

DECLARATION OF INTERESTS

The authors declare no competing interests.

SUPPLEMENTAL INFORMATION

Supplemental information can be found online at <https://doi.org/10.1016/j.bpj.2026.06.038>.

REFERENCES

- Aragona, M., A. Sifrim, ..., C. Blanpain. 2020. Mechanisms of stretch-mediated skin expansion at single-cell resolution. *Nature*. 584:268–273.
- Houtekamer, R. M., M. C. van der Net, ..., M. Gloorich. 2022. Mechanical forces directing intestinal form and function. *Curr. Biol.* 32:R791–R805.
- Guillot, C., and T. Lecuit. 2013. Mechanics of epithelial tissue homeostasis and morphogenesis. *Science*. 340:1185–1189.
- Lee, L.-W., G.-H. Lee, ..., M.-J. Tang. 2025. Mechanobiological mechanism of cyclic stretch-induced cell columnarization. *Cell Rep.* 44:115662.
- Papafiliippou, E., L. Baldauf, ..., A. Bonfanti. 2025. Interplay of damage and repair in the control of epithelial tissue integrity in response to cyclic loading. *Curr. Opin. Cell Biol.* 94:102511.
- Clemens, C. H. M., M. Samsom, ..., A. J. P. M. Smout. 2003. Abnormalities of left colonic motility in ambulant nonconstipated patients with irritable bowel syndrome. *Dig. Dis. Sci.* 48:74–82.
- Duboc, H., B. Coffin, and L. Siproudhis. 2020. Disruption of circadian rhythms and gut motility: an overview of underlying mechanisms and associated pathologies. *J. Clin. Gastroenterol.* 54:405–414.
- Creff, J., Y. He, ..., D. Krdija. 2025. Goblet cells mechanically breach the epithelial barrier in gut homeostasis. Preprint at bioRxiv. <https://doi.org/10.1101/2025.04.01.646617>.
- Liu, J., G. H. Koenderink, ..., D. A. Weitz. 2007. Visualizing the Strain Field in Semiflexible Polymer Networks: Strain Fluctuations and Nonlinear Rheology of F-Actin Gels. *Phys. Rev. Lett.* 98:198304.
- Duque, J., A. Bonfanti, ..., G. Charras. 2024. Rupture strength of living cell monolayers. *Nat. Mater.* 23:1563–1574.
- Bonfanti, A., J. Duque, ..., G. Charras. 2022. Fracture in living tissues. *Trends Cell Biol.* 32:537–551.
- Khalilgharibi, N., J. Fouchard, ..., G. Charras. 2019. Stress relaxation in epithelial monolayers is controlled by the actomyosin cortex. *Nat. Phys.* 15:839–847.
- Baghdadi, M. B., R. M. Houtekamer, ..., T. H. Kim. 2024. PIEZO-dependent mechanosensing is essential for intestinal stem cell fate decision and maintenance. *Science*. 386:eadj7615.
- Borghi, N., M. Sorokina, ..., A. R. Dunn. 2012. E-cadherin is under constitutive actomyosin-generated tension that is increased at cell–cell contacts upon externally applied stretch. *Proc. Natl. Acad. Sci. USA*. 109:12568–12573.
- Bell, G. I. 1978. Models for the specific adhesion of cells to cells: a theoretical framework for adhesion mediated by reversible bonds between cell surface molecules. *Science*. 200:618–627.
- Rakshit, S., Y. Zhang, ..., S. Sivasankar. 2012. Ideal, catch, and slip bonds in cadherin adhesion. *Proc. Natl. Acad. Sci. USA*. 109:18815–18820.
- Eckert, J., V. Viasnoff, and A. S. Yap. 2025. New directions in epithelial mechanoadaptation. *Curr. Opin. Cell Biol.* 95:102536.
- Banerjee, S., M. L. Gardel, and U. S. Schwarz. 2020. The actin cytoskeleton as an active adaptive material. *Annu. Rev. Condens. Matter Phys.* 11:421–439.
- Erdmann, T., and U. S. Schwarz. 2004. Stochastic dynamics of adhesion clusters under shared constant force and with rebinding. *J. Chem. Phys.* 121:8997–9017.
- Smith, A. S., and U. Seifert. 2005. Force-induced de-adhesion of specifically bound vesicles: strong adhesion in competition with tether extraction. *Langmuir*. 21:11357–11367.
- Mulla, Y., G. Oliveri, ..., G. H. Koenderink. 2018. Crack initiation in viscoelastic materials. *Phys. Rev. Lett.* 120:268002.
- Sonam, S., L. Balasubramaniam, ..., B. Ladoux. 2023. Mechanical stress driven by rigidity sensing governs epithelial stability. *Nat. Phys.* 19:132–141.
- Harris, A. R., J. Bellis, ..., G. T. Charras. 2013. Generating suspended cell monolayers for mechanobiological studies. *Nat. Protoc.* 8:2516–2530.
- Rackauckas, C., and Q. Nie. 2017. OrdinaryDiffEq.jl: High-Performance Ordinary Differential Equation Solvers for Julia. *J. Open Res. Software*. 5:15.
- Mogensen, P., and A. Riseth. 2018. Optim: A mathematical optimization package for Julia. *J. Open Source Softw.* 3:615.
- ComputationalMechanobiology. 2025. CellAdhesion.jl: Modelling Dynamics of Binding–Unbinding of Cell-Adhesion Molecules. GitHub repository. <https://github.com/ComputationalMechanobiology/CellAdhesion.jl>.
- Harris, A. R., L. Peter, ..., G. T. Charras. 2012. Characterizing the mechanics of cultured cell monolayers. *Proc. Natl. Acad. Sci. USA*. 109:16449–16454.
- Bonfanti, A., J. Fouchard, ..., A. Kabla. 2020. A unified rheological model for cells and cellularised materials. *R. Soc. Open Sci.* 7:190920.
- Charras, G., and A. S. Yap. 2018. Tensile forces and mechanotransduction at cell–cell junctions. *Curr. Biol.* 28:R445–R457.
- Kechagia, J. Z., J. Ivaska, and P. Roca-Cusachs. 2019. Integrins as biomechanical sensors of the microenvironment. *Nat. Rev. Mol. Cell Biol.* 20:457–473.
- Rogers, J., A. T. Bajur, ..., K. M. Spillane. 2024. Mechanical control of antigen detection and discrimination by T and B cell receptors. *Biophys. J.* 123:2234–2255.
- He, W., P. Cowin, and D. L. Stokes. 2003. Untangling desmosomal knots with electron tomography. *Science*. 302:109–113.
- Zinner, M., I. Lukonin, and P. Liberali. 2020. Design principles of tissue organisation: How single cells coordinate across scales. *Curr. Opin. Cell Biol.* 67:37–45.
- Brugués, A., E. Anon, ..., X. Trepat. 2014. Forces driving epithelial wound healing. *Nat. Phys.* 10:683–690.
- Wyatt, T. P. J., A. R. Harris, ..., B. Baum. 2015. Emergence of homeostatic epithelial packing and stress dissipation through divisions oriented along the long cell axis. *Proc. Natl. Acad. Sci. USA*. 112:5726–5731.
- Janssen, V., and S. Huveneers. 2024. Cell–cell junctions in focus: imaging junctional architectures and dynamics at high resolution. *J. Cell Sci.* 137:jcs262041.

37. de Beco, S., J.-B. Perney, ..., F. Amblard. 2015. Mechanosensitive adaptation of E-cadherin turnover across adherens junctions. *PLoS One*. 10:e0128281.
38. Iyer, K. V., R. Piscitello-Gómez, ..., S. Eaton. 2019. Epithelial viscoelasticity is regulated by mechanosensitive E-cadherin turnover. *Curr. Biol.* 29:578–591.e5.
39. Windoffer, R., M. Borchert-Stuhlträger, and R. E. Leube. 2002. Desmosomes: interconnected calcium-dependent structures of remarkable stability with significant integral membrane protein turnover. *J. Cell Sci.* 115:1717–1732.
40. Holthöfer, B., R. Windoffer, ..., R. E. Leube. 2007. Structure and function of desmosomes. *Int. Rev. Cytol.* 264:65–163.
41. Gloushankova, N. A., T. Wakatsuki, ..., S. M. Troyanovsky. 2003. Continual assembly of desmosomes within stable intercellular contacts of epithelial A-431 cells. *Cell Tissue Res.* 314:399–410.
42. Fülle, J. B., H. Huppert, ..., C. Ballestrem. 2021. Desmosome dualism—most of the junction is stable, but a plakophilin moiety is persistently dynamic. *J. Cell Sci.* 134:jcs258906.
43. Esfahani, A. M., J. Rosenbohm, ..., R. Yang. 2021. Characterization of the strain-rate-dependent mechanical response of single cell-cell junctions. *Proc. Natl. Acad. Sci. USA*. 118:e2019347118.
44. Mangeol, P., D. Massey-Harroche, ..., P.-F. Lenne. 2024. The zonula adherens matura redefines the apical junction of intestinal epithelia. *Proc. Natl. Acad. Sci. USA*. 121:e2316722121.
45. Rikitake, Y., K. Mandai, and Y. Takai. 2012. The role of nectins in different types of cell–cell adhesion. *J. Cell Sci.* 125:3713–3722.
46. Zhou, S., N. Qi, ..., X. Su. 2025. Recent progress in intrinsic self-healing polymer materials: Mechanisms, challenges and potential applications in oil and gas development. *Chem. Eng. J.* 511:161906.
47. Xiao, Y., Q. Li, ..., C. Yang. 2022. Fatigue of amorphous hydrogels with dynamic covalent bonds. *Extreme Mech. Lett.* 53:101679.
48. Xue, B., Z. Bashir, ..., Y. Cao. 2023. Strong, tough, rapid-recovery, and fatigue-resistant hydrogels made of picot peptide fibres. *Nat. Commun.* 14:2583.
49. Devi V K, A., R. Shyam, ..., A. J. Nathanael. 2021. Self-healing hydrogels: preparation, mechanism and advancement in biomedical applications. *Polymers*. 13:3782.
50. Shen, T., and F. J. Vernerey. 2020. Rate-dependent fracture of transient networks. *J. Mech. Phys. Solid.* 143:104028.
51. Tang, Z., W. Xian, ..., Y. Li. 2025. From bonds to breaks: multiscale perspectives on polymer network fracture. *Int. J. Smart Nano Mater.* 16:779–844.
52. Mohanty, S., and W. Cai. 2026. From Connectivity to Rupture: A Coarse-Grained Stochastic Network Dynamics Approach to Polymer Network Mechanics. Preprint at arXiv. <https://doi.org/10.48550/arXiv.2602.08089>.
53. Arora, A., M. S. Rizvi, ..., V. Viasnoff. 2025. Viscous dissipation in the rupture of cell–cell contacts. *Nat. Mater.* 24:1126–1136.
54. Mulla, Y., M. J. Avellaneda, ..., G. H. Koenderink. 2022. Weak catch bonds make strong networks. *Nat. Mater.* 21:1019–1023.
55. Navis, A., and C. M. Nelson. 2016. Pulling together: Tissue-generated forces that drive lumen morphogenesis. *Semin. Cell Dev. Biol.* 55:139–147.
56. Tang, Z., Y. Hu, ..., N. Tang. 2018. Mechanical forces program the orientation of cell division during airway tube morphogenesis. *Dev. Cell.* 44:313–325.e5.
57. Ruiz, W. G., D. R. Clayton, ..., G. Apodaca. 2024. The rat bladder umbrella cell keratin network: Organization, dependence on the plectin cytolinker, and responses to bladder filling. *Mol. Biol. Cell.* 35:ar139.
58. Pietramaggiore, G., P. Liu, ..., D. P. Orgill. 2007. Tensile forces stimulate vascular remodeling and epidermal cell proliferation in living skin. *Ann. Surg.* 246:896–902.
59. Li, J., A. J. Simmons, ..., Q. Liu. 2024. Identification and multimodal characterization of a specialized epithelial cell type associated with Crohn's disease. *Nat. Commun.* 15:7204.
60. Nyström, A., and L. Bruckner-Tuderman. 2018. Injury-and inflammation-driven skin fibrosis: the paradigm of epidermolysis bullosa. *Matrix Biol.* 68–69:547–560.

Biophysical Journal, Volume 125

Supplemental information

Emergent intercellular junction stability during cyclic tissue loading

Eleni Papafilippou, Alessandra Bonfanti, Guillaume Charras, and Alexandre Kabla

SUPPLEMENTARY MATERIAL

1 Force measurement: calibration procedure

Force can be calculated using the cantilever beam equation, Eq. 5, where k is the rod's stiffness (which depends on the material and geometrical properties of the wire) and dw is the deflection. The value of k can be computed using Eq. 6, using the Young's modulus, E , the second moment of area, I , and the length, L , of the wire.

$$F = k dw, \quad (5)$$

$$k = \frac{3EI}{L^3}, \quad (6)$$

Using six pairs of voltage and deflection (V , dw) a linear fit is used to determine the conversion factor from Volts to Newtons, given by Eq. 7.

$$\alpha = \frac{1}{V} \frac{3EI}{L^3} dw, \quad (7)$$

Using a 2D thin sheet approximation and Eq. 8, the force of each monolayer was normalised by the average width, w_0 , to calculate tissue tension. The thickness of the monolayer is $\sim 10\mu\text{m}$ which is two orders of magnitude smaller than the length and width of the monolayer, justifying our approximation. The cross-sectional area was computed by multiplying the initial length by the average width. The average width was found by averaging the width at each of the arms of the device and in the middle of the monolayer.

$$\frac{F}{w_0} = \frac{\alpha V}{w_0}, \quad (8)$$

Finally, the monolayer's pre-tension was calculated using two bright-field microscopy images, one from the start and one from the end of the experiment. A $250 \times 150 \text{ pixel}^2$ area was cropped around the flexible rod to measure its displacement Δx that can be used to find the pre-tension, using Eq. 9. The actual tension values were computed by subtracting the pre-tension offset.

$$\frac{F}{w_0} = -k \frac{\Delta x}{w_0}, \quad (9)$$

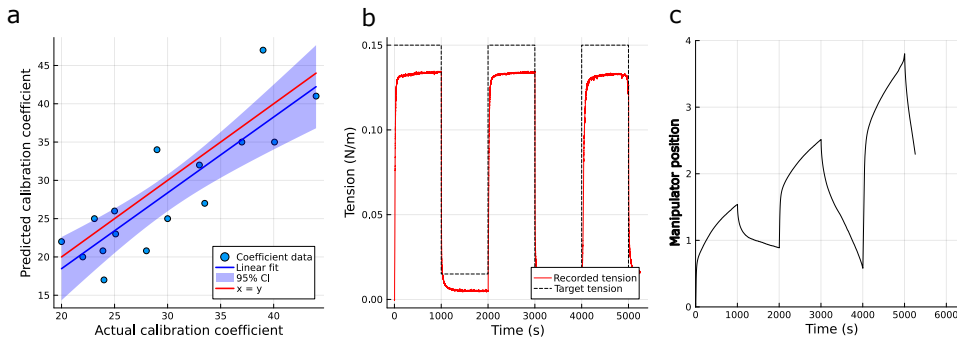


Figure SM 1: PID controller performance. a) Comparison of pre- vs post-experiment calibration coefficients. Data points fall close to the identity line ($x=y$, red), and the linear fit lies within the 95% confidence interval (shaded, blue), indicating that pre-experiment coefficients reliably estimate post-experiment values. b) Target tension (black, dashed) compared to recorded tension (red, solid) during cyclic loading, showing that the controller accurately tracks the target setpoints across cycles. c) Manipulator position over time, illustrating dynamics adjustment of the deformation in each cycle to achieve the target tension, demonstrating active feedback control during repeated loading.

2 Slip bond parameters: fitting and optimisation details

Fracture times were not used directly, as they diverge near the critical tension and would yield undefined RMSE values. For each experimentally measured fracture time, the analytical model was used to find the tension value producing the same rupture timescale. The squared difference between this predicted tension and the experimental value was computed, and the square root of the mean of these squared differences gave the first RMSE value. To enforce accuracy at the main experimental condition, we identified the mean fracture time corresponding to 0.15Nm^{-1} , used the analytical model to predict the tension yielding the same fracture time and added the deviation between this predicted tension value and 0.15Nm^{-1} as a second error term. The combined objective function was defined using Eq. 10, where the factor of three balances accurate reproduction at 0.15Nm^{-1} , with fidelity across the full fracture-time profile.

$$\text{Error} = \text{RMSE} + 3 * E(0.15\text{Nm}^{-1}) \quad (10)$$

The analytical model was evaluated for 100 logarithmically spaced tension values between 10^{-5} and 10N/m , capturing rupture timescales from near zero to 4000s. This range ensured stable optimisation and uniform sampling of the model's exponential decay behaviour. Multiple initialisations were tested and the parameter set yielding the lowest error was selected. Initial values for k_{on} and k_{off} were $3 \cdot 10^{-3}$, $3 \cdot 10^{-4}$, $3 \cdot 10^{-5}$, and for f_0 initial values were 0.0055N/m , 0.055N/m , 0.55N/m . These ranges include parameters previously obtained from epithelial monolayer strain-ramp loading experiments (10) and cover three orders of magnitude. Parameters were optimised in logarithmic form to enforce positivity without explicit bounds.

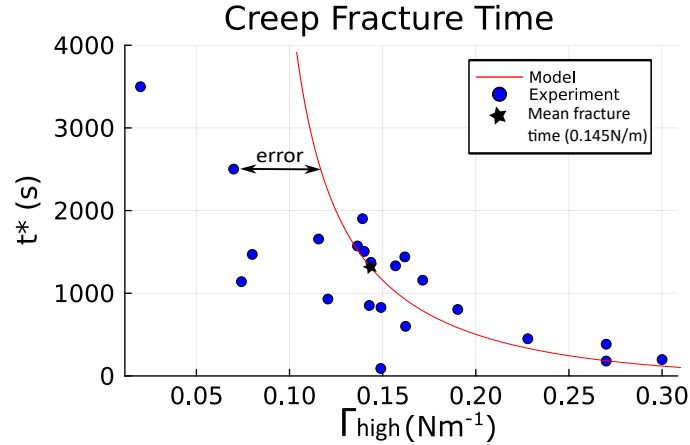


Figure SM 2: **Creep fracture time as a function of applied tension.** Blue points show experimental data, the red curve shows the model, and the black star marks the mean fracture time at 0.15Nm^{-1} . The horizontal arrow illustrates the definition of error in tension, which forms the basis of the RMSE calculation. Overall model error is computed as RMSE plus a weighted term ensuring accurate reproduction of the 0.15Nm^{-1} condition.

3 Local force redistribution model implementation

To assess the effect of spatially heterogeneous force transmission, we implemented a local load-sharing variant of the model. The load is applied non-uniformly across all the closed bonds of the junction, following the local load redistribution assumptions described in (21). For each closed bond, the nearest closed neighbours are identified and weighting factors are assigned to determine the fraction of load carried locally. Bonds with fewer nearby closed neighbours receive larger weights and therefore bear a larger proportion of the applied force, producing local force concentrations. The weights are normalised such that the sum of forces across all closed bonds equals the externally applied tension, ensuring mechanical force balance throughout the simulation. As bonds rupture and reform, the junction evolves dynamically, leading to time-dependent force heterogeneity. The resulting force redistribution can be visualised using colour-scaled salt-and-pepper maps, where colour intensity represents the instantaneous force carried by each bond over time. Compared with the global redistribution model, the local redistribution model generates pronounced force heterogeneity and transient force hot-spots along the junction. However, we find that this more complex, heterogeneous loading assumption does not qualitatively alter the behaviour of the system: rupture and recovery timescales follow the same functional dependence on tension, simply with an adjusted transition tension, Γ_c . Both absolute and normalised maps also show that the different regimes of epithelial stability are qualitatively preserved.

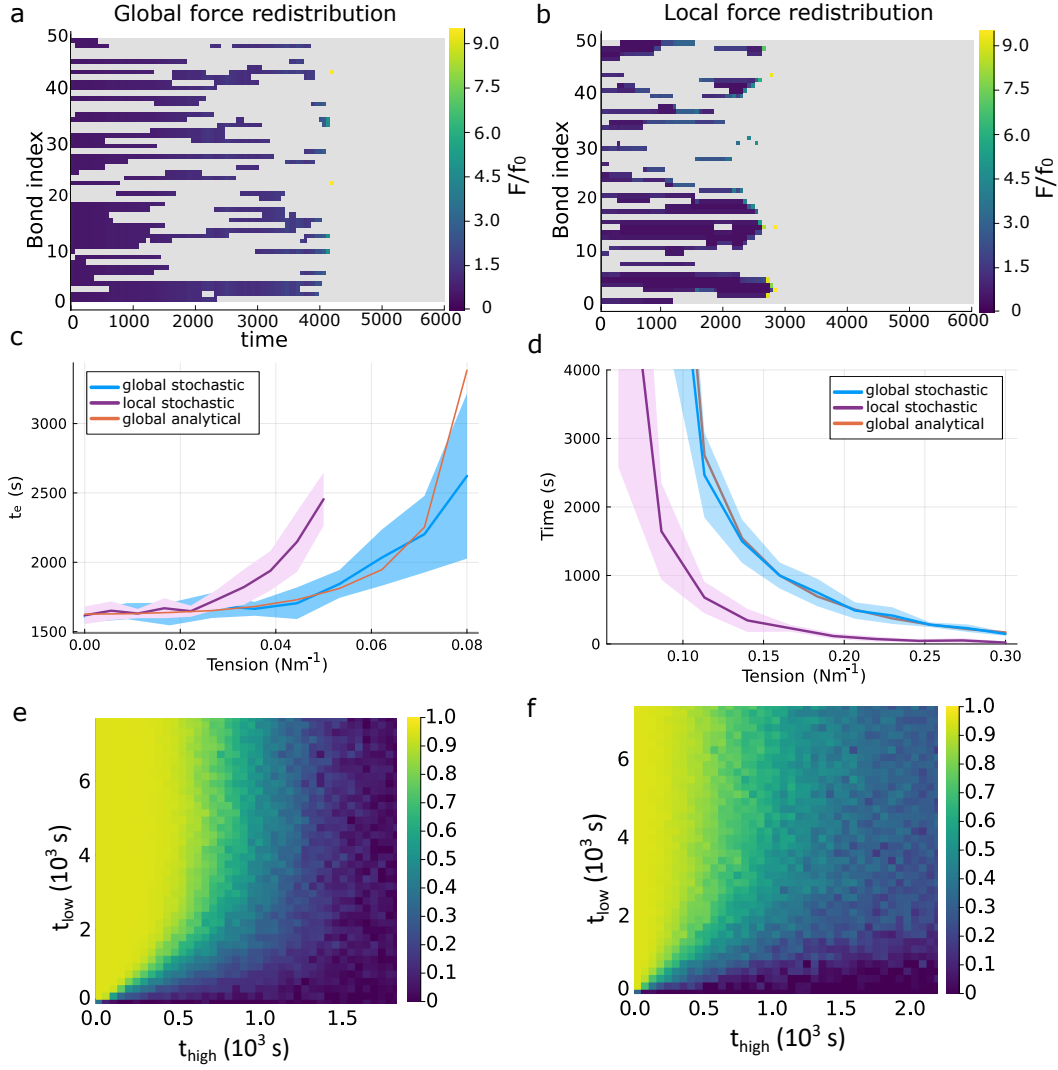


Figure SM 3: Effect of local force redistribution on junctional bond dynamics. Representative bond dynamics until rupture, under a) global (mean-field) and b) local force redistribution, showing heterogeneous load sharing with force hot-spots along the junction in the local case. The force scale colour is given by the force per bond divided by f_0 . c) Rupture d) repair timescales as a function of applied tension. The local redistribution model (orange) exhibits the same functional dependence as the global stochastic (blue) and analytical (green) results, with a shift to lower effective tensions corresponding to a reduced critical tension. e,f) Phase diagrams under cyclic loading for (e) global and (f) local force redistribution models for $\Gamma_{\text{high}} = 0.09\text{Nm}^{-1}$ and $\Gamma_{\text{low}} = 0.0175\text{Nm}^{-1}$ demonstrating that the maps are qualitatively conserved with robust features regardless of details of force redistribution. The colour bar in (e) and (f) encodes the normalised metric $\frac{\bar{T}^\dagger - \bar{T}^*}{\bar{T}^\dagger}$.

4 Catch-bond-like behaviour model implementation

We have also extended our model to include load-bearing complexes with catch-bond-like behaviour, introducing a transient stabilisation at intermediate tension. We have used the following equation, introducing two additional parameters in the dissociation rate term:

$$k_{\text{on}} = \text{constant} \quad (11)$$

$$k_{\text{off}} = k_{\text{off},0} e^{\frac{f}{f_0}} + k_{\text{off},\text{catch}} e^{\frac{-\Gamma}{\beta f_{0,\text{catch}}}} \quad (12)$$

The specific parameters were defined as $k_{\text{off},\text{catch}} = 2.68 * 10^{-3}$ and $f_{0,\text{catch}} = 2.49 * 10^{-1}$ to illustrate the catch-bond like behaviour. We find that, despite the modified bond kinetics, the overall dynamical behaviour remains unchanged. In particular, the same regimes of rupture, slow damage accumulation, and recovery are observed under cyclic loading and the maps retain the same structure as with the slip-bond model.

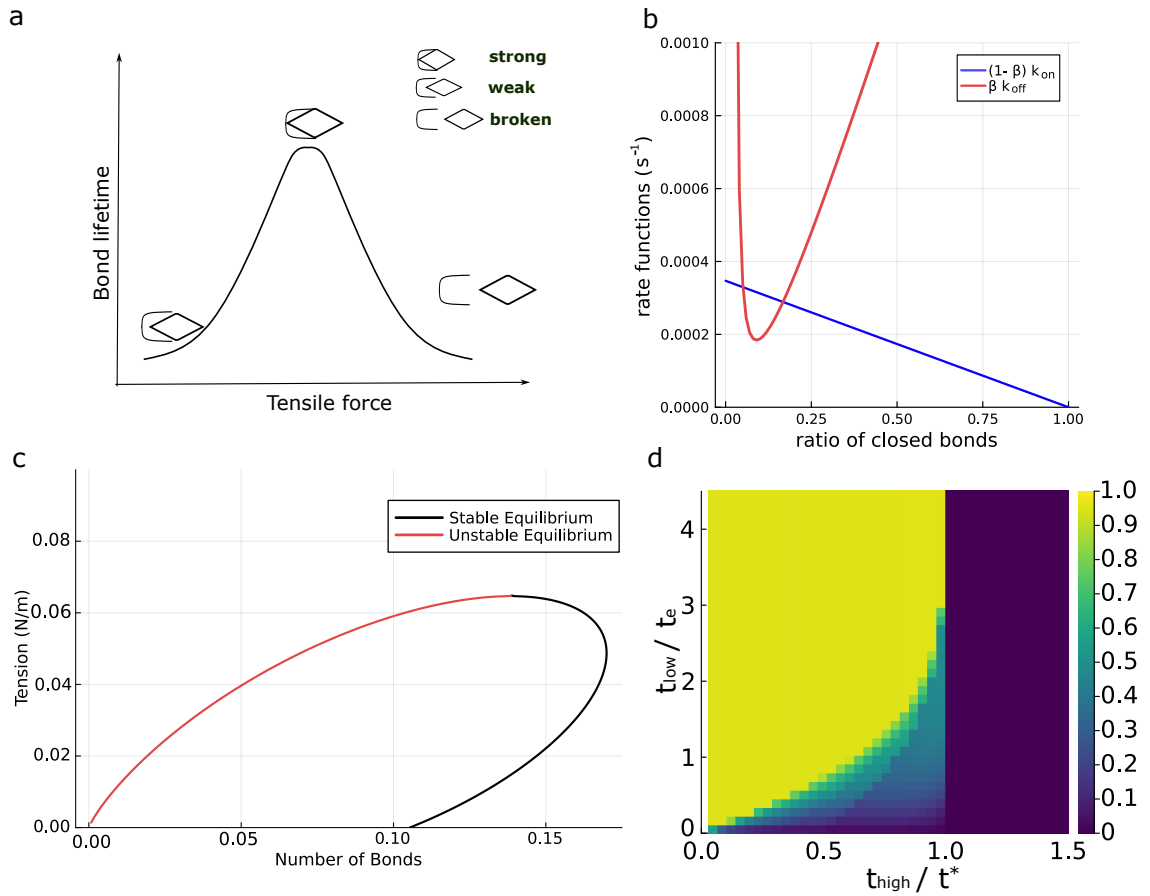


Figure SM 4: **Effect of catch bond-like behaviour on junction dynamics.** a) Schematic illustration of catch bond-like behaviour, showing non-monotonic dependence of bond lifetime on tensile force, with bond strengthening at intermediate tension values. b) Rate functions for bond association (blue) and dissociation (red) as a function of closed bonds, β for a given tension (here, 0.04 N/m). Points of equilibrium occur at the intersection between the two curves, where the rates of association and dissociation are equal. c) Stable (black) and unstable (red) analytical solutions for the ratios of closed bonds at different values of tension. d) Normalised cyclic loading map for the catch bond model, for $\Gamma_{\text{high}} = 0.1 \text{ Nm}^{-1}$ and $\Gamma_{\text{low}} = 0.04 \text{ Nm}^{-1}$, showing the same qualitative regions and robust features regardless of the details of model complexity. The colour bar encodes the normalised metric $\frac{\bar{T}^\dagger - \bar{T}^*}{\bar{T}^\dagger}$.

Supplementary figures

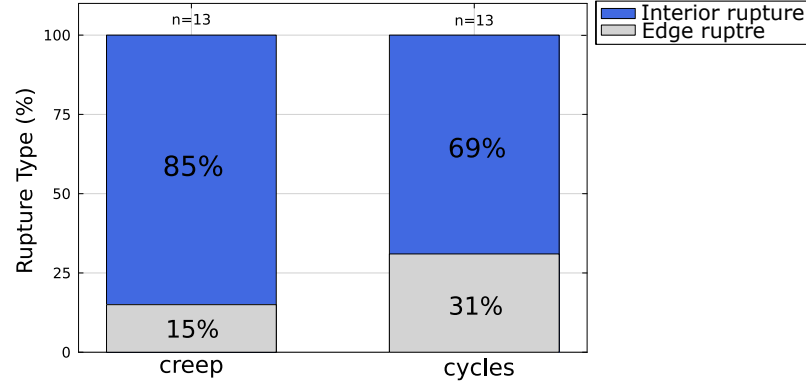


Figure SM 5: **Distribution of rupture initiation sites for creep and cyclic loading conditions.** In both loading settings, rupture occurs predominantly within the interior of the monolayer (85% for creep and 69% for cyclic loading). These observations support the model's assumption of globally distributed loading prior to failure and indicate that rupture is not primarily driven by edge-induced stress concentrations.

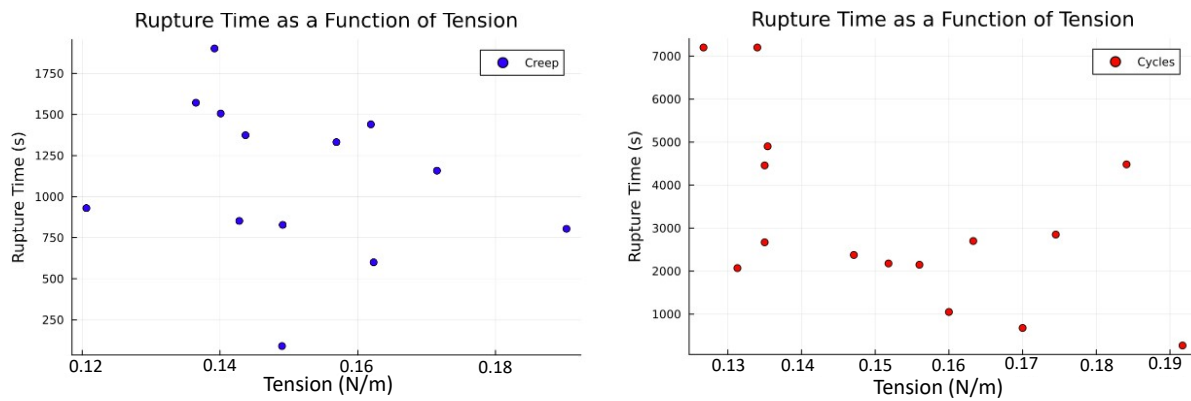


Figure SM 6: **Rupture time as a function of applied tension (Γ_{high}) for creep (left) and cyclic loading (right) tests.** For creep, rupture times show no clear monotonic dependence across the examined tension range (0.12 - 0.195 N/m). In the cyclic loading case, a slight decreasing trend with increasing tension is observed, though variability remains large. These observations support including tests spanning this full tension range.

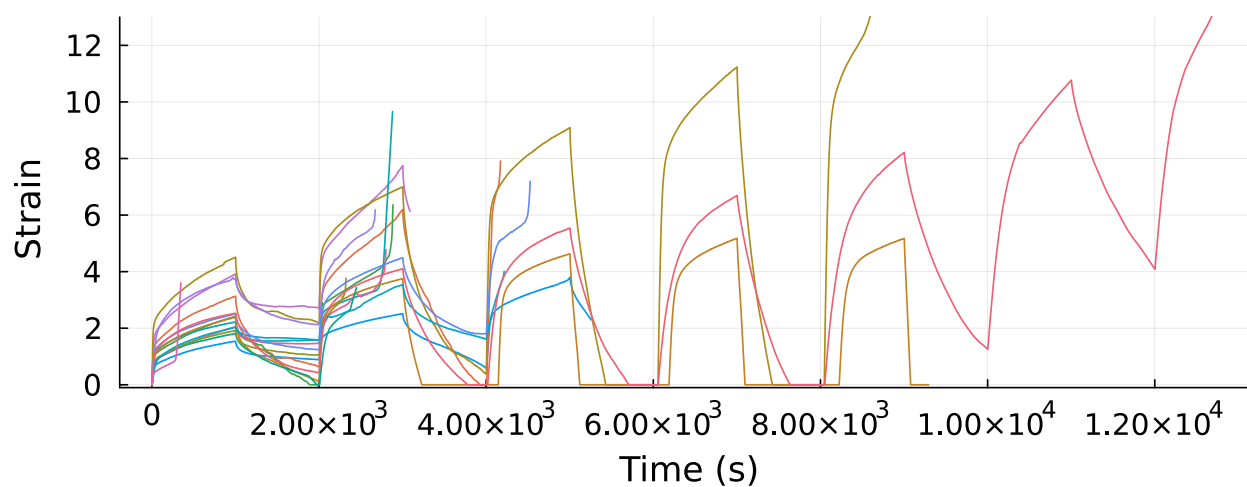


Figure SM 7: **Experimental strain response during cyclic creep loading.** Each curve represents an individual loading–unloading cycle measured under constant applied stress (moving average smoothing, window size 3.5s). Later cycles diverge markedly, indicating the onset of irreversible deformation and rupture within the epithelial monolayers.

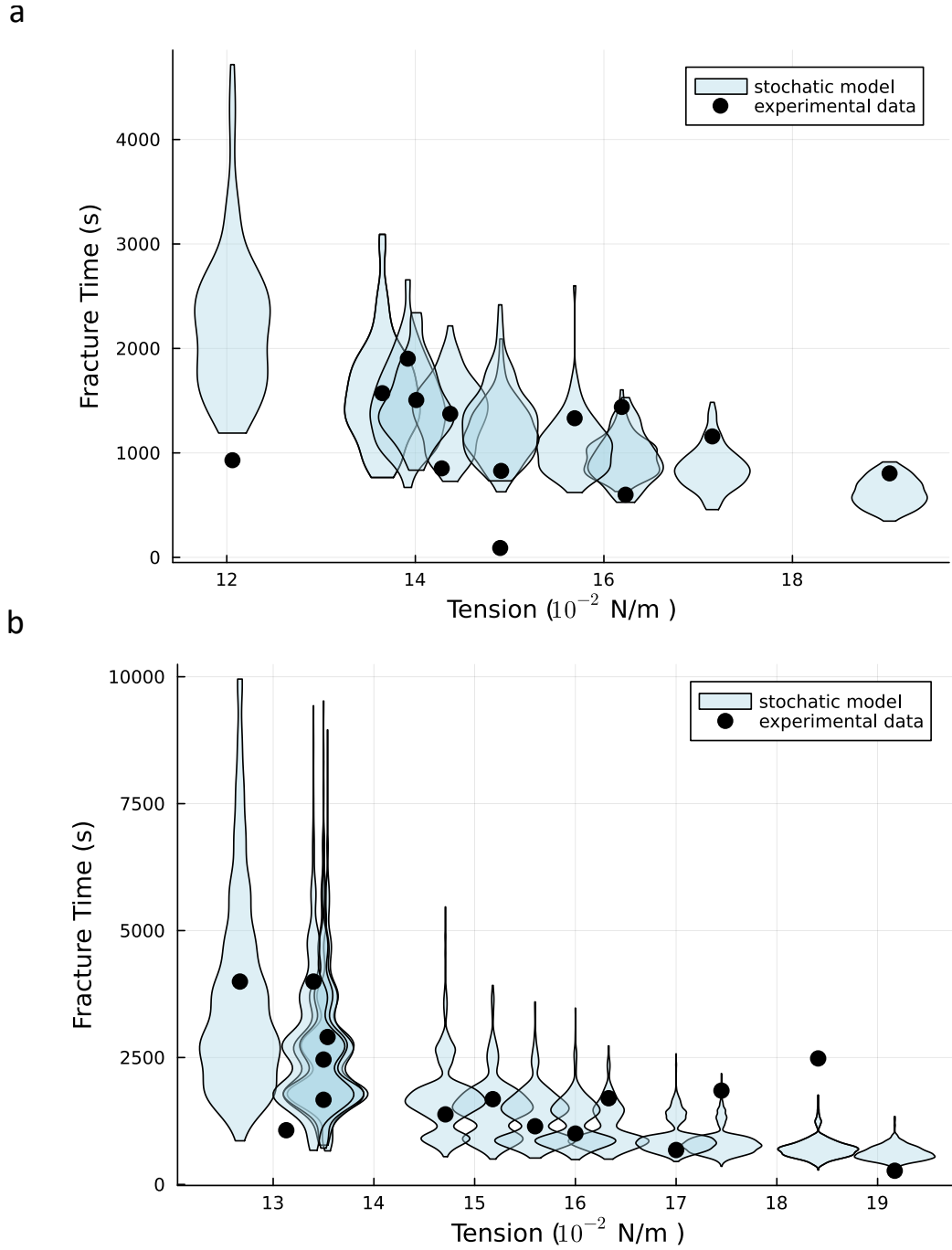


Figure SM 8: Comparison of stochastic model predictions and experimental rupture times for tissues subjected to creep and cyclic loading a) Fracture times as a function of applied tension under creep loading. b) Fracture times under cyclic loading. In both panels, light blue violin plots show the distribution of rupture times obtained from 100 stochastic simulations at each tension value, highlighting variability in the model response. Solid black dots represent individual experimental measurements. Overall, the model captures both the trend and variability of rupture times across loading conditions, while reflecting the broad distribution of failure times observed experimentally.

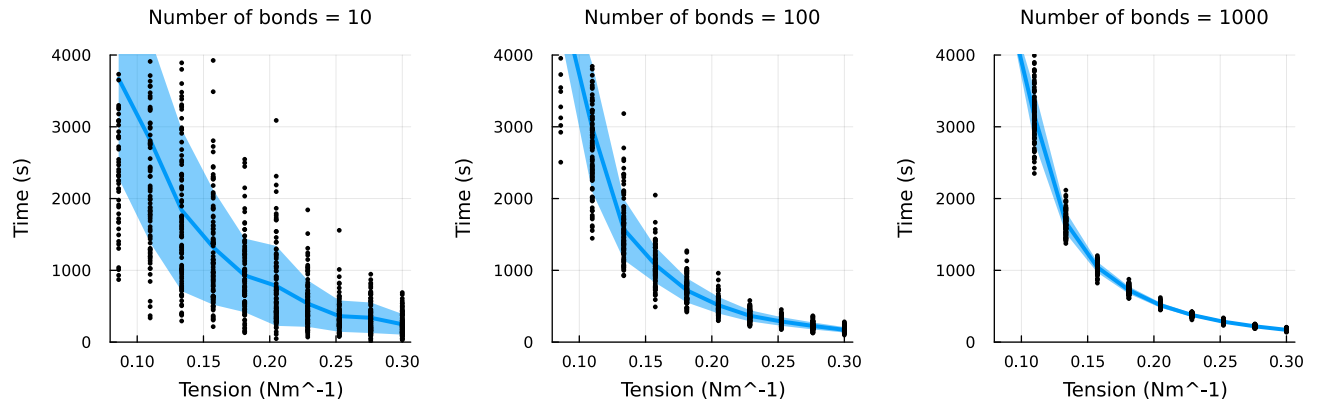


Figure SM 9: **Rupture time as a function of applied tension** for 10, 100, and 1000 bonds. Each black point represents an individual stochastic simulation, and the shaded blue regions indicate the standard deviation across runs. Increasing the number of bonds decreases the variability. N=100 bonds matches the noise of previous experimental data (10).

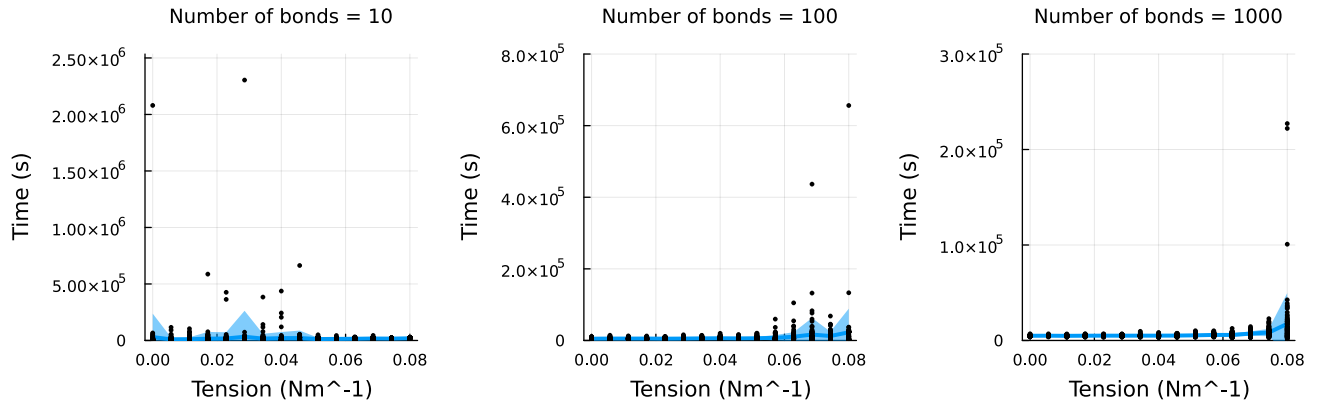


Figure SM 10: **Repair time as a function of applied tension** for 10, 100, and 1000 bonds. Each black point represents an individual stochastic simulation, and the shaded blue regions indicate the standard deviation across runs. Repair is more variable with fewer bonds with certain points exceeding tenfold slower repair times for N=10. Increasing the number of bonds decreases the variability.

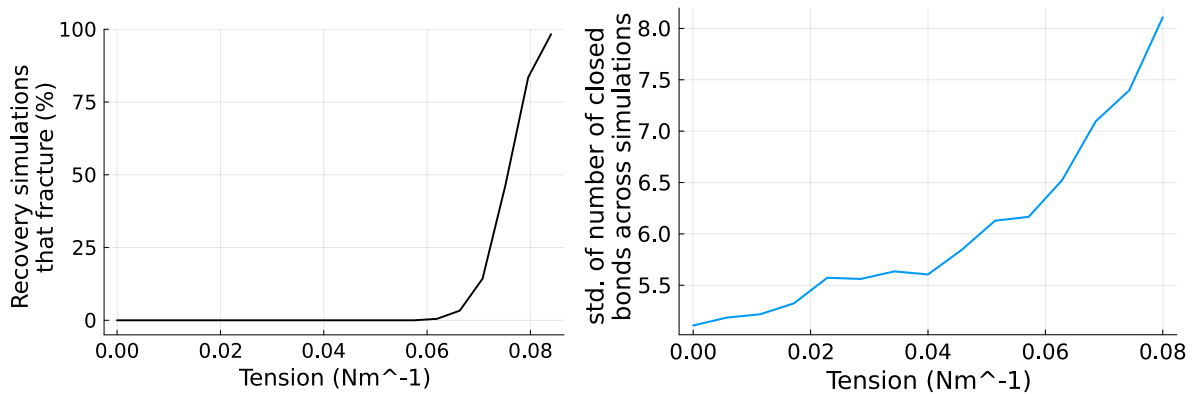


Figure SM 11: **Effect of applied tension on repair timescale response**. Left: percentage of recovery simulations resulting in rupture increases sharply close to Γ_c , indicating increased noise close to the transition. Right: standard deviation of the number of closed bonds across simulations increases close to Γ_c , also reflecting the increased variability in bond dynamics at the transition. Total simulation duration is 40,000s. Note: if the simulation duration were extended to infinity, all trajectories would eventually rupture due to stochastic fluctuation.

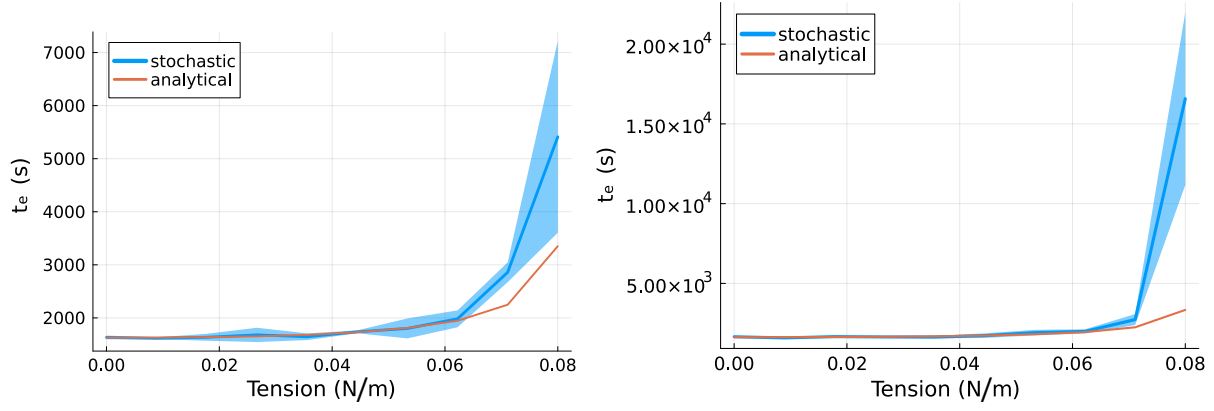


Figure SM 12: **Alternative definition of the repair timescale**, t_e , for the stochastic model, where any trajectories that ruptured as the applied tension approached Γ_c were assigned an infinite rupture time and included only up to failure to capture initial recovery behaviour. The diverging behaviour of the repair timescale close to Γ_c is now observed for both the analytical and stochastic models. Left: total duration 10,000. Right: total duration 20,000. The sharp divergence in the right plot suggests that, if simulated for sufficiently long times, a system with a finite number of bonds will always eventually rupture.

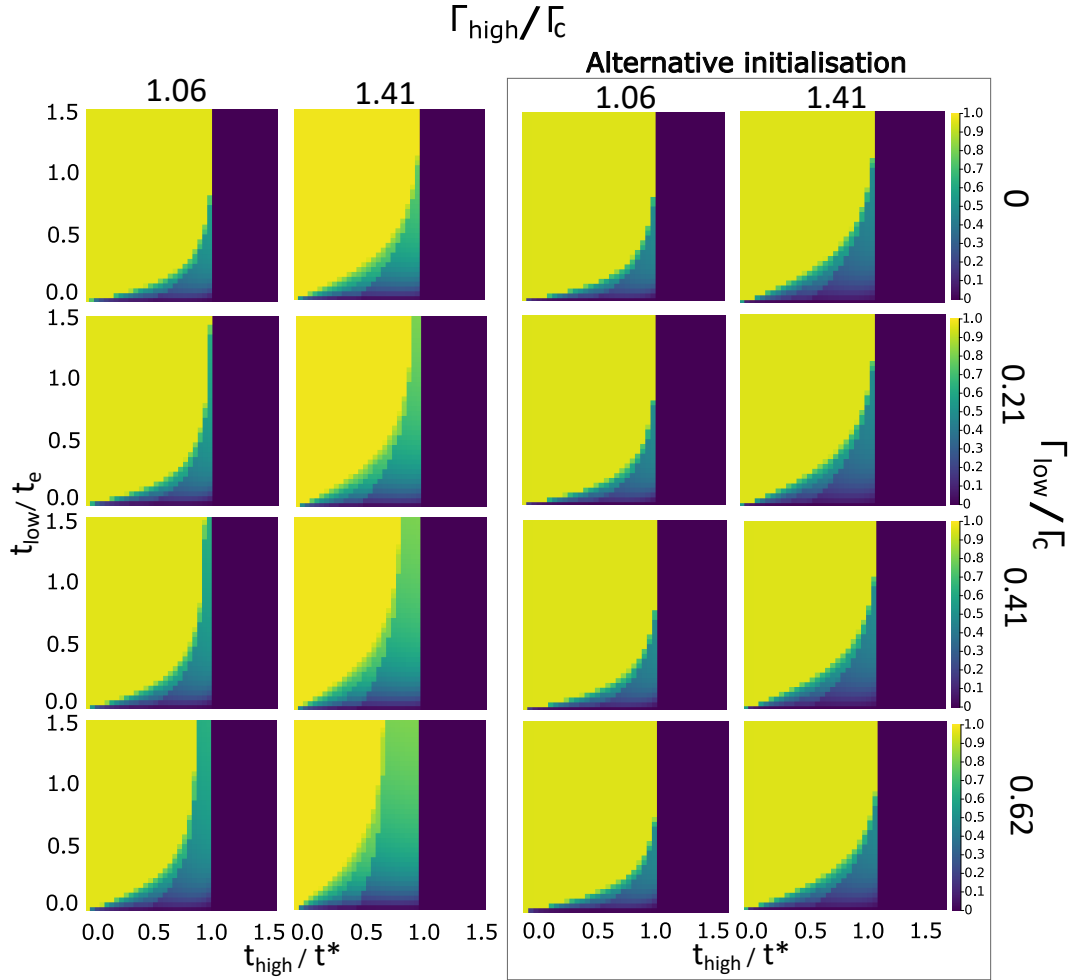


Figure SM 13: **Alternative initialisation strategy**. An alternative initialisation strategy was considered for the map generation, whereby the initial number of closed bonds was set to the stable ratio of the specific low tension value (stable solution branch (black) on figure 3c). This removed the fourth regime of continuous diverging at both Γ_{high} and Γ_{low} , as shown in the analytical model plots within the gray border.

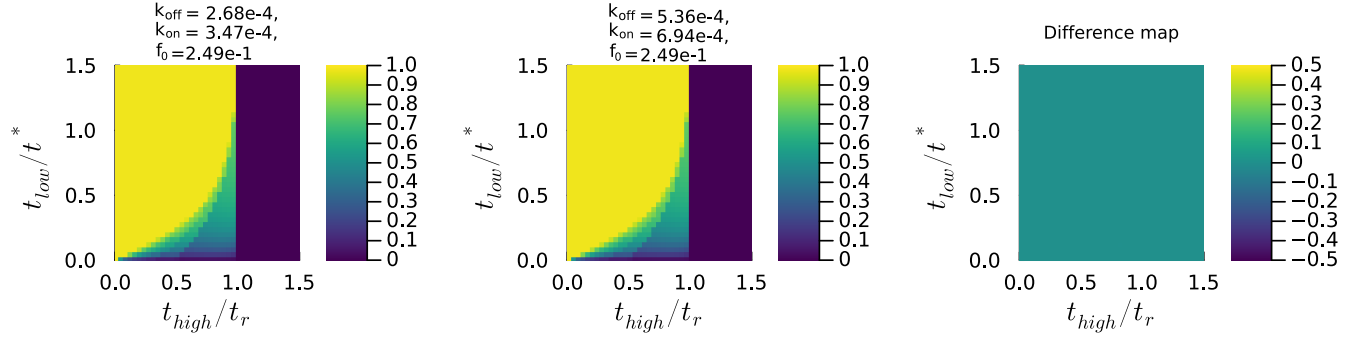


Figure SM 14: **Effect of doubling kinetic parameters while maintaining the k_{on}/k_{off} ratio.** Heat maps show the normalised rupture probability as a function of the loading and relaxation durations (t_{low}/t_e and t_{high}/t^*) for the original parameter set (left) and for parameters doubled in magnitude while preserving the k_{on}/k_{off} ratio (middle). The difference map (right) indicates negligible change across the parameter space, demonstrating that the model behaviour depends primarily on the relative balance between association and dissociation rates, rather than their absolute values.

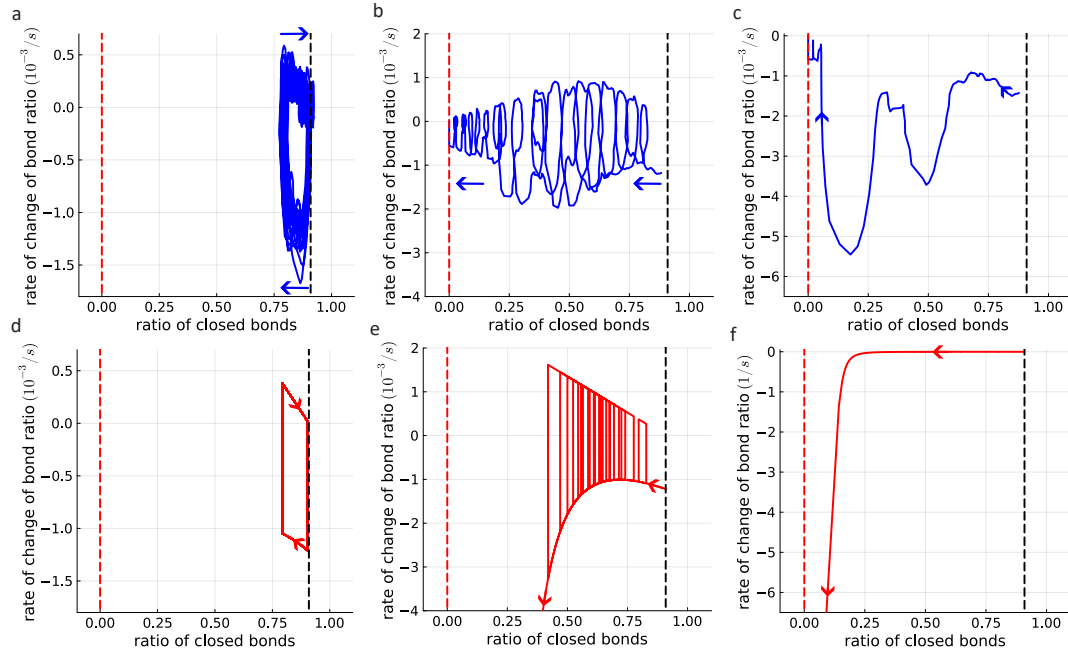


Figure SM 15: **Phase-space trajectories of junctional bond dynamics under cyclic loading.** Phase diagrams showing the rate of change of the fraction of closed bonds as a function of the instantaneous fraction of closed bonds for different combinations of high- and low-tension phases. Panels a–c) show stochastic simulations of a single junction (blue), and panels d–f) show corresponding analytical results (red). The black dashed lines mark stable fixed points, while red dashed lines denote unstable fixed points separating rupture and recovery regimes. Arrows indicate the direction of trajectory evolution during cyclic loading. Oscillatory trajectories a,b) correspond to partial bond recovery within the slow-damage regime, whereas monotonic collapse to the left c,f) reflects irreversible rupture. Stable limit cycles near the right boundary a,d) indicate sustained recovery and junctional stability.

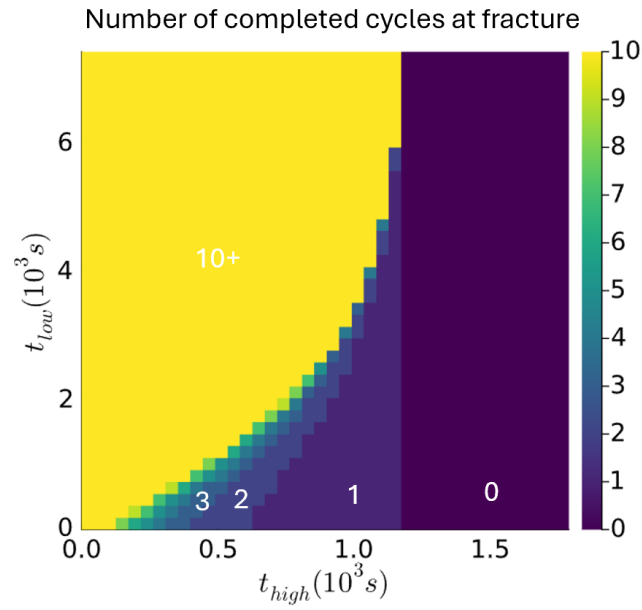


Figure SM 16: **Maps scaled by rupture cycles (number of cycles-to-rupture)**. Number of completed load cycles at rupture as a function of high- and low-load durations defined between $\Gamma_{high} = 0.15\text{Nm}^{-1}$ and $\Gamma_{low} = 0.015\text{Nm}^{-1}$. Colours indicate the number of cycles, with yellow representing 10 or more cycles (maximum duration) and purple indicating zero cycles (immediate rupture). In the slow damage accumulation regime, the fringes illustrate transitions to larger cycle numbers, in a direction towards smaller t_{high} and larger t_{low} indicating that slower damage accumulation and increased recovery extend the junction's lifetime to a higher number of cycles.

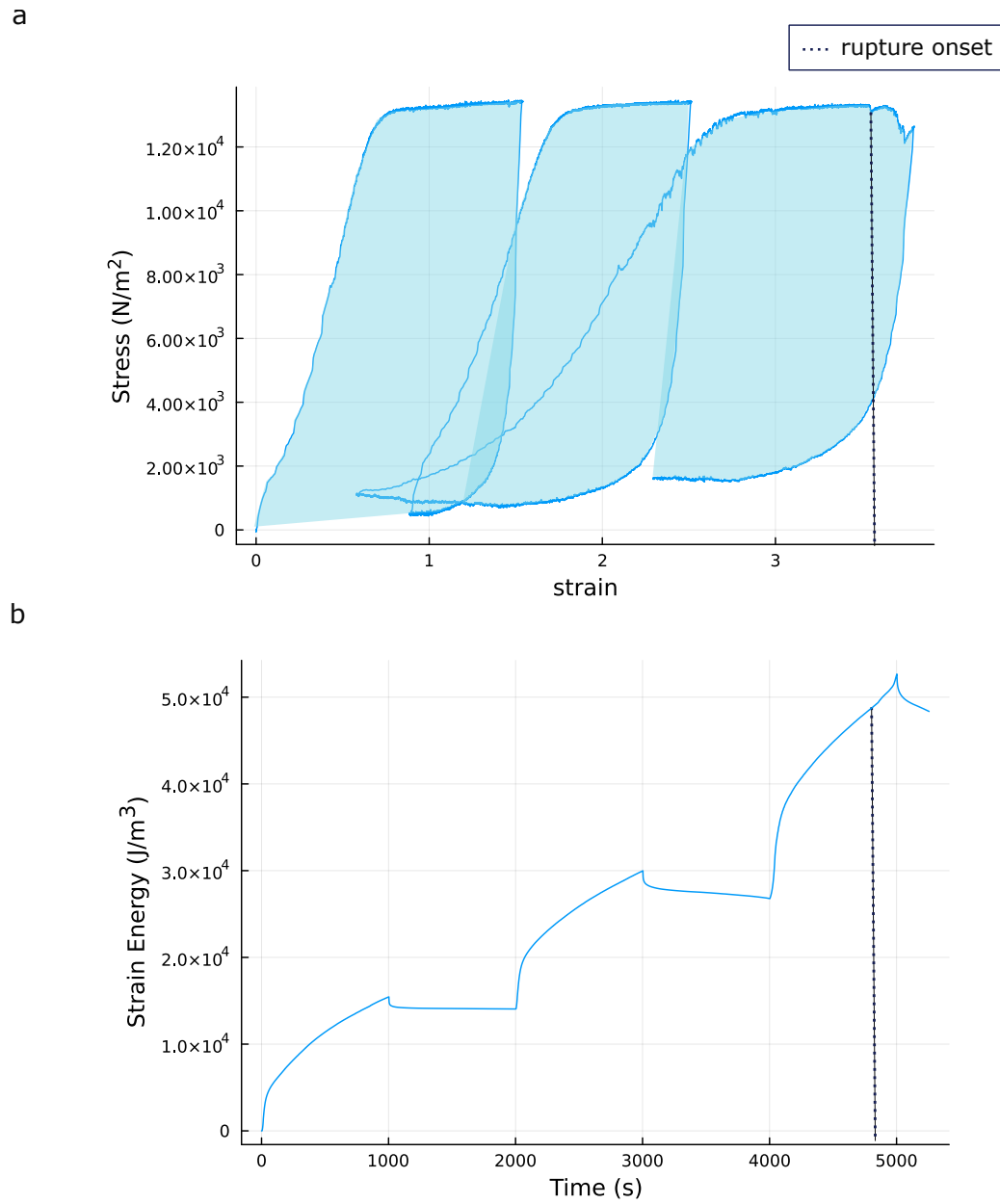


Figure SM 17: Energy dissipation up to rupture onset under cyclic loading. a) Stress–strain curves for successive loading cycles. The shaded region represents the integrated stress–strain response, reflecting turnover prior to failure. b) Temporal evolution of the accumulated strain energy, showing the progressive increase in mechanical work ($4.84 \times 10^4 \text{ J/m}^3$) dissipated over successive cycles until rupture.