

Abstract of “Insights into the Mechanical Functions of Glass Sponge Spicules Through a Characterization of Their Strength and Toughness Properties.” by Michael A. Monn, Ph.D., Brown University, May 2018.

Despite being composed of weak and brittle constituents, some structural biological materials, such as shell and bone, have relatively high strength and toughness. These materials are often heterogeneous and consist of a ceramic and an organic phase arranged in intricate patterns. One goal of bio-inspired engineering is to understand how the arrangement of these phases, known as the material’s architecture, can impart such remarkable strength and toughness enhancements. Establishing connections between architecture and mechanical properties can provide both a deeper understanding of a material’s bio-mechanical function(s) and help to uncover new mechanical design principles that can be used to improve engineering composites. The first step in this type of investigation is quantifying the mechanical property enhancements provided by a material’s specific architecture. The skeletal fibers of the marine sponge *Euplectella aspergillum* are an example of a biological material for which the toughness and strength enhancements provided by the architecture have not yet been quantified. These fibers—known as spicules—have an architecture that consists of a solid silica cylinder surrounded by concentric cylindrical silica layers that look like tree rings. I measured the spicule’s bending failure strains, fracture initiation toughness, and average crack growth resistance via three-point bending tests that I performed using a custom-built mechanical testing device. I then compared the properties of the *E. aspergillum* spicules to those of spicules from a related sponge—*Tethya aurantia*—that have a similar chemical composition but lack the lamellar architecture. Through this comparison I found that the toughness enhancements provided by the spicule’s architecture pale in comparison to those observed in other biological materials with similar lamellar architectures, like shell and bone. On the other hand the spicule’s architecture enhances its bending failure strain by a factor of 2.4. This work suggests that flexibility or strain tolerance may be more beneficial to the mechanical function of *E. aspergillum* spicules than toughness.

**Insights into the Mechanical Functions of Glass Sponge Spicules
Through a Characterization of Their Strength and Toughness
Properties.**

by
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This dissertation by Michael A. Monn is accepted in its present form by
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Refereed Journal Publications

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DEDICATED TO ALL OF MY TEACHERS AND MENTORS

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Chapter 1

Introduction

1.1 Architectures in biological materials serve as templates for bio-inspiration

In order to build higher, travel further, and live more sustainably, we must develop new materials that are lighter, stronger, and tougher. However, in most engineering materials, from steels to ceramics, strength and toughness are mutually exclusive [1]. This is a critical bottleneck for aerospace, transportation, and energy production technologies. The growing field of bio-inspired engineering offers a unique opportunity to overcome this apparent material properties hurdle.

Some structural biological materials (SBMs), such as bone and shell, simultaneously have both high strength and high toughness relative to the weak and brittle ceramics (e.g., calcium carbonate and silica) from which they are primarily composed [2–8]. For example, it has been shown that the iridescent portion of mollusk shells, known as nacre, can sustain much larger strains [4] and requires much more energy to fracture than its ceramic constituent, aragonite [9, 10]. These SBMs are often heterogeneous and consist of ceramic and organic phases combined in intricate patterns at the micrometer scale (see Figure 1.1) [6]. The arrangement of these phases, which is known as the SBM's architecture, is believed to be the key to their remarkable combination of strength and toughness [4, 5, 11–17].

Structural biological materials do not outperform engineering materials, like advanced ceramics,

in terms of their absolute toughness or strength. However, combining the architectures found in SBMs with modern chemistry and digital manufacturing techniques could lead to a new generation of structural materials whose mechanical properties far exceed those of today's state-of-the-art [18, 19]. Furthermore, by tuning the architectural parameters of a bio-inspired material, the resulting enhancement of mechanical properties can exceed that which is achieved by simply copying the SBM's architecture "pixel-by-pixel" [20]. The first step toward understanding why the architectures seen in SBMs enhance certain mechanical properties is quantifying the effect that the architecture has on the mechanical properties that are relevant to the SBM's primary mechanical function(s) [21].

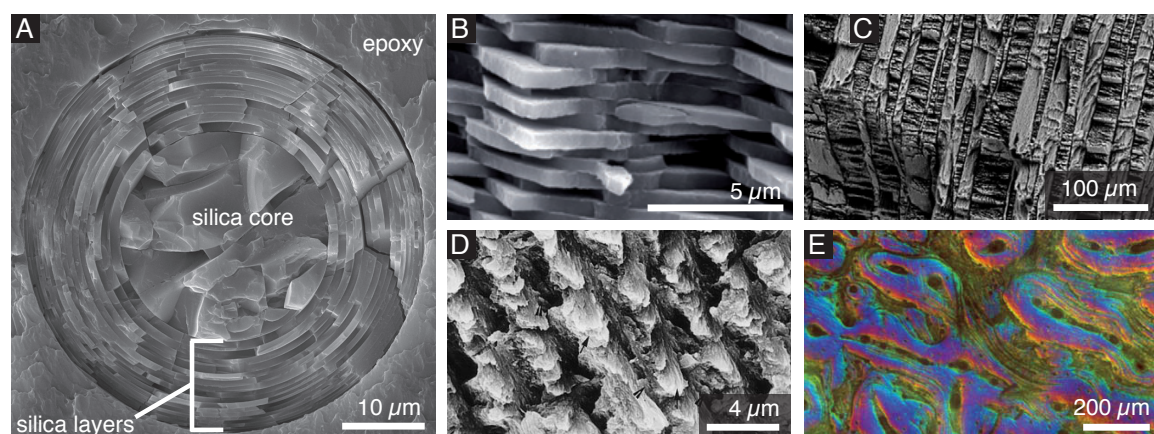


Figure 1.1: Examples of the architectures seen in SBMs. (A) Concentric silica layers make up the tree ring-like lamellar architecture of anchor spicules from the *Euplectella aspergillum* sponge (modified from [22]). Approximately 25 silica layers, each between 200 and 1000 nm thick surround a monolithic silica core. Adjacent layers are separated by ≈ 5 –10 nm thick proteinaceous interlayers. (B) The brick and mortar lamellar architecture of nacre consisting of staggered aragonite tablets separated by ≈ 30 nm thick organic interlayers (modified with permission from [1]). (C) The crossed-lamellar architecture of the shell of *Strombus gigas* (modified from [23]). Like nacre, the lamellae are composed of a calcite mineral and separated by thin organic interlayers. (D) The crossed-lamellar architecture in a rat tibia composed of mineralized collagen fibrils. The arrangement of the fibrils and the extent of hydroxyapatite mineralization define the lamellar structure (modified from [24]). (E) Differential interference contrast micrograph of compact elk antler bone showing lamellar architecture consisting of an assembly of oblong osteons. Antler is a special type of bone and is therefore composed of hydroxyapatite mineralized collagen fibrils (modified from [25]).

1.2 Spicules are a model system for exploring structure-function connections

The anchor spicules of the marine sponge *Euplectella aspergillum* (*E. aspergillum*) are a quintessential example of an architected SBM for which the functional implications of the architecture are not fully understood. Furthermore, the effect of the spicule's architecture on their strength and

toughness properties has not been adequately quantified. *Euplectella aspergillum* is a species of “glass sponge” (class Hexactinellida) that lives on the ocean floor at depths of approximately 1–2 km [15,28,29]. It is endemic to the south pacific and has been found primarily near the Philippines. Like most sponges, it is sessile and feeds by pumping seawater through its body and filtering out plankton and other micro-organisms.

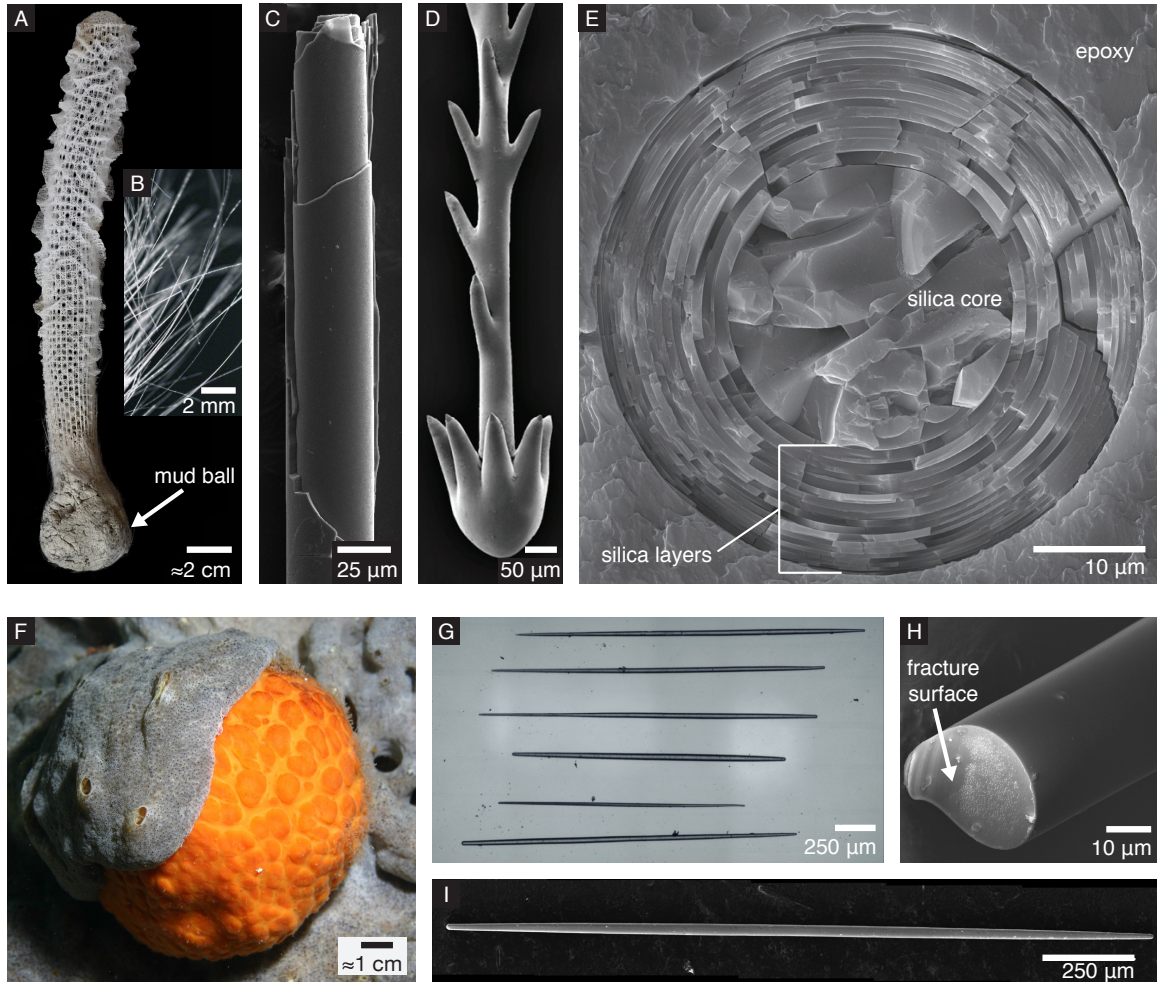


Figure 1.2: Skeleton and spicules of *E. aspergillum* and *T. aurantia* sponges. (A) An *E. aspergillum* skeleton. The mud ball at the bottom of the skeleton contains the anchor spicules (courtesy of Swee Cheng Lim). (B) The anchor spicules from an *E. aspergillum* skeleton separated from the mud ball. (C) A broken *E. aspergillum* anchor spicule exposing its architecture. (D) The distal end of a *E. aspergillum* anchor spicule is covered in barbs that help the spicule anchor to the sea floor. Reproduced from [26], Copyright 2004 National Academy of Sciences. (E) A cross-sectioned anchor spicule. Modified from [22]. (F) A live *T. aurantia* sponge (courtesy of Steve Lonhart / NOAA MBNMS). (G) Spicules from a *T. aurantia* sponge. (H) A fractured *T. aurantia* spicule. (I) The *T. aurantia* spicules are tapered along their length. Reproduced from [27] under the Creative Commons 4.0 BY license.

The sponge derives its form and the stiffness required to actively pump water through its body

from a mineralized skeleton (see Figure 1.2 (A)). In *E. aspergillum* this skeleton consists of a cylindrical, cage-like assembly of filaments called spicules (see Figure 1.2 (A)) [28, 29]. The spicules are approximately $50\ \mu\text{m}$ in diameter and up to 10 cm in length. Most of the spicules are cemented together by a ceramic matrix. However, the sponge fastens itself to the soft substrate of the seafloor using thousands of hair-like anchor spicules located at its basal end (see Figure 1.2 (B)) [29]. The proximal regions of the anchor spicules are bundled together and cemented to the main vertical struts of the skeletal lattice. The distal end of each anchor spicule is covered with a series of recurved barbs that secure the sponge into the soft sediments of the seafloor (see Figure 1.2 (D)).

Spicules in Hexactinellid sponges are composed primarily of amorphous, hydrated silica, which is thought to contain either a collagenous [30, 31] or chitinous [32–34] scaffold. This proteinaceous scaffold plays an important role in the processes of silica biomineralization [31, 33]. In *E. aspergillum* anchor spicules, this scaffold is not only distributed within the bulk of the silica but it also partitions the silica into concentric, cylindrical layers (see Figure 1.2 (C), (E)) [22]. Specifically, when viewed in cross-section, an anchor spicule consists of a $\approx 10\ \mu\text{m}$ silica core that is surrounded by a coaxial assembly of ≈ 25 cylindrical, silica layers (see Figure 1.2 (E)) [22, 28, 29]. The thicknesses of the silica layers appear to decrease from the core to the periphery [22]. It has been shown that the thicknesses of the core and layers are remarkably consistent both within and across individual organisms [22]. A thin ($\approx 5\text{--}10\ \text{nm}$ [29]) proteinaceous interlayer separates adjacent silica layers. Spicules with similar cylindrical lamellar architectures have also been found in a number of related sponge species (see Figure 2.1 (B)) [15, 35–37].

The *E. aspergillum* anchor spicules have been the subject of several previous structure-function investigations [16, 22, 29, 38–41], most of which focus on the potential connection between the cylindrical lamellar architecture and toughness enhancement [16, 38–40]. However, none of these studies directly measure the *E. aspergillum* spicule's toughness properties, such as the fracture initiation toughness or crack growth resistance. Furthermore, it is not clear how enhanced toughness at the individual spicule level would benefit the primary mechanical function of the anchor spicules—i.e., providing a robust attachment to the seafloor. Enhanced toughness allows some SBMs, like nacre and bone, to contain damage, which is a prerequisite to them to be repaired or remodeled. In contrast to these SBMs, the *E. aspergillum* spicules have not been observed to heal if they are

damaged. Therefore damage would likely result in a permanent reduction of a spicule's anchoring ability. Since the sponge is anchored by thousands of spicules, however, if a single spicule were damaged it would have little effect on the overall strength of the anchorage. Thus, I believe that the redundancy of having thousands of anchor spicules would allow the sponge's anchorage to be tough and damage tolerant even if the individual spicules were not. In the case of a single *E. aspergillum* spicule, other mechanical properties, like strength—i.e., the largest force it can withstand before the nucleation of damage—may be more important than toughness.

1.3 In spite of architecture, *E. aspergillum* spicules lack toughness

The overarching purpose of this work is to provide a more balanced perspective on the possible functional significance of the cylindrically layered architecture seen in the *E. aspergillum* spicules. To this end, I first measure the *E. aspergillum* spicule's fracture toughness properties (see Chapter 2). Specifically, I cut micrometer-size notches in the *E. aspergillum* spicules using a focused ion beam (see Figure 2.2) and performed flexural tests on them using a custom built mechanical testing device. Using data from these tests I compute their fracture initiation toughness and average crack growth resistance. Then, to isolate the toughness enhancement provided by the *E. aspergillum* spicule's architecture, I compare its toughness to the toughness of spicules from a related sponge, *Tethya aurantia* (*T. aurantia*) (see Figure 1.2 (F)). The *T. aurantia* spicules are needle-shaped fibers that are ≈ 2 mm long, ≈ 35 μm in diameter and are gradually tapered along their length (see Figure 1.2 (G), (I)). They have a similar chemical composition and volume-averaged bonding structure to the *E. aspergillum* spicules [15], however, they do not possess a lamellar architecture (see Figure 1.2 (H)) [27]. I use the *T. aurantia* spicules as a control material for quantifying the toughness enhancements provided by the *E. aspergillum* spicule's lamellar architecture¹. By comparing the *E. aspergillum* to the *T. aurantia* spicules I found that the cylindrically layered architecture provides a much smaller toughness enhancement than the lamellar architectures found in prototypically tough biological materials like nacre and bone. Thus, while the *E. aspergillum* spicule's architecture might appear similar to architectures seen in many tough SBMs, my measurements suggest

¹I believe that the best choice for the control material would be a section of the solid silica core of the *E. aspergillum* spicules. However, it is extremely difficult to remove all of the silica layers without damaging or fracturing the core. Therefore, I chose the *T. aurantia* spicules as what I believe to be a next best alternative.

that these seemingly similar architectural motifs do not provide the same magnitude of toughness enhancement.

1.4 Architecture imparts flexibility, or bending tolerance, to *E. aspergillum* spicules

The lack of toughness enhancements I observed suggests that the *E. aspergillum* spicule's architecture may be related to a different mechanical property that is more critical to their anchoring function. Motivated by the nature of the spicule's mechanical function and the fact that damaged spicules are not repaired, I hypothesize that rather than contributing to its toughness, the *E. aspergillum* spicule's architecture enhances its ability to withstand bending strains (see Chapter 3). I test this hypothesis by performing flexural tests on *E. aspergillum* spicules, measuring their bending failure strains, and comparing them to the bending failure strains of *T. aurantia* spicules. Since the *T. aurantia* spicules have a similar chemical composition to *E. aspergillum* spicules but have no architecture, I attribute any difference between their bending failure strains to the cylindrically layered architecture of the *E. aspergillum* spicules. I found that the bending failure strains of the *E. aspergillum* spicules were roughly 2.4 times larger than those of the *T. aurantia* spicules.

While the mechanical response of the *E. aspergillum* spicules in bending has been previously measured [40,42], I show that analyzing this data using standard methods (e.g., elementary beam theory) can lead to a significant underestimation of the spicule's strength properties. Since the *E. aspergillum* spicules undergo large deflections before failing, linear beam theory (i.e., Euler-Bernoulli beam theory [43]) will underestimate the spicule's bending failure strains. Having accurate estimates of the spicule's strength properties—like its bending failure strain—will provide a stronger foundation for developing models that capture the mechanisms through which their strength is enhanced. By understanding these mechanisms, it may be possible to design stronger and more flexible engineering composites.

1.5 Tapered shapes give *T. aurantia* spicules buckling resistance

During the investigation of the strength and toughness properties of the *E. aspergillum* spicules, I also identified a new structure-property connection in the *T. aurantia* spicules, which is related to their tapered shape (see Chapter 4). In contrast to most other structure-property investigations, I found that the *T. aurantia* spicule's shape is related to its resistance to structural failure through buckling rather than its strength or toughness.

The *T. aurantia* sponge's body consists of a dense, spherical core (choanosome) surrounded by a thick, fibrous shell (cortex) [44]. Both the choanosome and cortex are made of spongin—a type of collagenous tissue. These soft tissues are critical for metabolic processes but do little to provide mechanical integrity. The *T. aurantia* sponge also produces several types of spicules that are used to stiffen the choanosome and cortex. However, the spicules are not arranged in a skeletal lattice like in *E. aspergillum*. Rather, they are distributed throughout the choanosome and cortex, embedded in the spongin as a sort of biological fiber reinforced composite.

The needle-like spicules that I focus on are known as strongyloxea spicules. The strongyloxea are ≈ 2 mm long and $35\ \mu\text{m}$ in diameter. They are grouped together in bundles so that each strongyloxea's axis is oriented approximately parallel to the axis of the bundle. The bundles radiate from the center of the sponge through both the choanosome and the cortex. Along with the cortex, these bundles are thought to function like an umbrella, providing a stiff and resilient epidermal layer to the sponge [44].

I measured the shape of the strongyloxea from scanning electron micrographs, and found that their tapered shape is remarkably uniform across different strongyloxea. Motivated by this observation, I hypothesize that the tapered shape enhances a strongyloxea's ability to provide stiffness to the sponge. From the strongyloxea's slenderness and arrangement within the sponge I infer that their stiffening ability is limited by their buckling strength, which is the maximum axial force that they can transmit along their length before buckling. By measuring the mechanical response of strongyloxea using a flexural test, I found that their deformation behavior can be modeled exceptionally well using elementary beam theory [43]. I therefore modeled an individual strongyloxea spicule as an Euler column. The buckling strength of a column in this model, however, depends on

the distribution of force along the column's length as well as constraints on the column's motion. Since the strongyloxea are embedded within the soft tissue of the sponge, they likely experience a variety of force distributions along their length. I used finite element-based calculations to show that due to the strongyloxea's taper and aspect ratio, the forces are localized at the strongyloxea's ends. Finally, I estimated the load they can transmit before buckling using this structural mechanics model along with the measurements of the spicules shape. Compared to a cylinder with the same length and volume, this model predicts that the spicules shape enhances their critical buckling load by up to 30%.

Chapter 2

Quantification of toughness enhancements provided by the architecture of *E. aspergillum* spicules

Note: A version of this chapter is being prepared for submission to Nature Communications. Data and figures have been used with all co-authors' consent.

Monn MA, Mok J and Kesari H. Architecture in biological materials: a template for toughness enhancement, or a siren song?

2.1 Introduction

Despite being composed primarily of brittle ceramic materials, some SBMs have remarkably high toughness [1–3, 19, 45]. For example, nacre is composed of >95% aragonite (a brittle, calcium carbonate mineral) by volume yet it has a specific fracture initiation toughness on par with nylon and some iron alloys [45]. The toughness enhancement, compared to aragonite, has been attributed to nacre's brick-and-mortar architecture, which consists of aragonite tablets arranged in staggered layers and joined together by a proteinaceous matrix (see Figure 1.1 (B)) [4, 7, 9, 46].

Both the brick-and-mortar architecture of nacre and the cylindrically layered architecture of

the *E. aspergillum* spicules are specific examples of a broader class of architectures prevalent in SBMs, which I refer to as lamellar architectures (see Figure 1.1). The defining feature of the lamellar architecture is alternating layers of a relatively thick ceramic phase and a thin organic phase. Different forms of lamellar architectures can be seen in many SBMs that are known for their high toughness, such as bone [2, 24, 47–49], antler [25], and the shell of the queen conch (*Strombus gigas*) [23, 50] (see Figure 1.1). As a consequence of the correlation between the lamellar architecture and toughness enhancement in these materials, it has been suggested in a more general sense that SBMs containing lamellar architectures have high toughness [6, 16, 38, 39, 46]. Specifically, it is thought that the cylindrically layered architecture of the *E. aspergillum* spicules enhances their toughness [16, 38–40].

However, the toughness enhancement imparted by a lamellar architecture is highly dependent on parameters such as the thicknesses and stiffnesses of the different phases [51], the interfacial properties (i.e., bonding between the two phases), and the geometry of the layers (e.g., 2D layers versus cylindrical layers; see Figure 1.1 (A), (B)) [16, 52–54]. It is therefore important to measure a SBM’s toughness properties to determine whether its specific architecture does in fact enhance toughness.

The speculated connection between the architecture and toughness enhancement in *E. aspergillum* spicules is primarily based on the architectural motifs that the spicules share with nacre and other tough SBMs [39] and is supported by the interpretation of micrographs and mechanical responses of spicules that were fractured in flexural tests [38–40]. None of these studies, however, provide direct measurements of the *E. aspergillum* spicule’s toughness properties or quantify the toughness enhancements provided by the spicule’s architecture alone.

Spicules that have been tested in bending display jagged fracture surfaces in which individual silica layers are clearly visible [39–41]. This has been interpreted as evidence that cracks propagating across the spicule’s cross-section are arrested and deflected at the interfaces between layers [39, 40]. The process of crack arrest and renucleation (often accompanied by crack deflection) constitutes one of the dominant toughening mechanisms in nacre [55, 56] and nacre-like engineering composites [57]. Furthermore, force-displacement responses obtained from bending tests performed on *E. aspergillum* anchor spicules [42] as well as spicules from related species containing

the cylindrical lamellar architecture (see Figure 2.1 (D)) [15, 35, 36] display sawtooth-like patterns. It has been suggested that this mechanical response is also indicative of crack arrest and renucleation, resulting in increased fracture toughness. However, it was later shown that in the case of the *E. aspergillum* spicules the sawtooth pattern is not caused by fracture processes and therefore are not evidence of toughness enhancement [58].

Other studies have attempted to quantify the *E. aspergillum* spicule's toughness by measuring its work of fracture [38, 40]. However, the techniques employed in these works violate some of the basic assumptions of the work of fracture method (see Section 2.4.2) and therefore the data they present cannot be used to accurately determine the spicule's toughness properties [59, 60].

Finally, many of these studies have compared the mechanical behaviors of architected spicules to synthetic glass (see Figure 2.1 (C)) [15, 36, 38, 40, 42], despite the spicules having a lower elastic modulus and a different chemical composition [15, 61, 62]. From this comparison, it would be impossible to isolate the effect of the spicule's lamellar architecture on its toughness properties. Ideally, the *E. aspergillum* spicules should instead be compared to a specimen composed of the same biogenic silica but which is monolithic.

In this Chapter I provide the first measurement of the *E. aspergillum* spicule's crack growth resistance. I compare the *E. aspergillum* spicule's crack growth resistance to the crack growth resistance of spicules from *T. aurantia* in order to isolate the toughness enhancement provided by the architecture. The *T. aurantia* spicules have a similar chemical composition but lack the lamellar architecture. The crack growth resistance, R , is a measure of the energy that is consumed by a crack as it propagates through a material. The value of R can depend on the amount of crack growth that has occurred (see Figure 2.1 (A), (B)). Its initial value, $R(0)$ —known as the fracture initiation toughness—determines when a crack can begin to grow. If the value of R increases with crack growth then the material is said to have a “rising R curve” (see the pink curve in Figure 2.1 (B)). In this case, as the crack grows the material becomes more resistant to subsequent or continued growth. Rising R curves are a hallmark of tough materials and have been observed in some SBMs like nacre and bone [25, 46, 49, 63].

I investigate the effect of the *E. aspergillum* spicule's architecture on $R(0)$ and the average value of R , which provides a measure of how much R increases during crack growth. I measure $R(0)$ of

the *E. aspergillum* and *T. aurantia* spicules and the average crack growth resistance, $\langle R \rangle$, of the *E. aspergillum* spicules using force and displacement data obtained from flexural tests that I performed on the spicules (see Section 2.3). Before performing the bending tests I cut notches in the spicules using a focused ion beam (see Section 2.3.3). I introduce two metrics to quantify the effect of architecture on toughness (see Section 2.2). The first metric, $R(0)^{(\text{arch})}/R(0)^{(\text{con})}$ provides a measure of how the architecture enhances the material's ability to guard against the growth of crack from preexisting flaws (see Figure 2.1 (B)). The second metric, $\langle R \rangle^{(\text{arch})}/R(0)^{(\text{con})}$ provides a measure of the overall “toughness” imparted by the *E. aspergillum* spicules architecture (see Figure 2.1 (B)). I use $R(0)^{(\text{arch})}/R(0)^{(\text{con})}$ and $\langle R \rangle^{(\text{arch})}/R(0)^{(\text{con})}$ to compare the toughness enhancement provided by the *E. aspergillum* spicule's lamellar architecture with the toughness enhancements provided by the lamellar architectures of other SBMs, like nacre, antler, and bone (see Section 2.5). I find that the toughness enhancement in the *E. aspergillum* spicules is very small compared to the enhancements in these other SBMs. My measurements show that despite having a similar architecture, the cylindrical layers in *E. aspergillum* spicules do not provide the extraordinary toughness enhancements observed in many other SBMs with lamellar architectures.

2.2 Fracture mechanics background and toughness enhancement metrics

As per the fracture mechanics approach to the mechanics of materials, a material's “failure stress/strength” that is measured from a mechanical test is not a material property. Rather it is determined by the size of a preexisting crack or flaw, a_0 , in the material and the material's fracture initiation toughness, $R(0)$ —which is taken to be a material property. The relationship between these three quantities is given by [65]

$$\sigma_f \propto \sqrt{\frac{ER(0)}{a_0}}, \quad (2.1)$$

where E is the Young's modulus and σ_f is the applied stress at failure. Equation (2.1) is colloquially known as Griffith's criterion and essentially tells us that a larger crack or flaw will result in a lower failure stress. The fracture initiation toughness $R(0)$ provides a measure of how resistant a material is to the growth of a crack from this preexisting flaw. It is determined by the energy required to

create new free surfaces and feed other inelastic, dissipative processes that act in the vicinity of the crack tip.

As per Irwin's energy-based approach [66] to Griffith's theory of fracture [65], the necessary condition for the extension of a crack is

$$-\frac{d\Pi}{d\Delta A} \geq R(0), \quad (2.2)$$

where Π is the potential energy of the system and ΔA is the area of the crack growth (see Figure 2.1 (A)). The derivative $-d\Pi/d\Delta A$ is known as the energy release rate or crack driving force, G . In brittle materials—such as many monolithic ceramics—the energetic cost of crack growth remains constant as a crack grows. That is, the single value $R(0)$ is sufficient to characterize the material's resistance to crack growth [67].

However, in architected materials, the material's crack growth resistance, R , can depend on the amount of crack growth that has already occurred (see Figure 2.1 (B)). In this case the necessary condition for crack growth is

$$G(\Delta A) \geq R(\Delta A). \quad (2.3)$$

The value of R at the onset of crack growth, i.e., $R(0)$, is the fracture initiation toughness. If R increases monotonically with ΔA , then the material is said to have a “rising R curve.” I consider a material to be “tough” if it has both a large value of $R(0)$ and a rising R curve. In a tough material, the large value of $R(0)$ help to prevent cracks from growing from preexisting flaws while the rising R curve helps guard against catastrophic failure. Consider an applied stress that results in a constant value of $G = R(0)$. In this case, Eqn. (2.3) will be satisfied at $\Delta A = 0$. However, once a crack has begun growing, R will increase but G will remain constant. Therefore, after a small increment of growth, Eqn. (2.3) will no longer be satisfied and crack growth will cease in a material with a rising R curve.

Based on Equation (2.3) there are two primary ways to increase toughness. The first is to increase R . This can be accomplished both by increasing $R(0)$ and by changing its shape to impart a rising R curve behavior. The second way to increase toughness is to decrease G . As I will discuss shortly, many common toughening mechanisms in architected materials do not actually change

a material's R , but rather act to decrease G . However, it has become conventional to represent the actual decrease in G as an apparent increase in R instead.

From my definition of toughness, let us first consider ways to prevent the initial growth of cracks. That is, consider Equation (2.3) as $\Delta A \rightarrow 0$. Since $R(0)$ is determined by the strength of the molecular bonds within the material, the ways in which it can be enhanced are quite limited. Therefore, the most realistic way to prevent the initial growth of cracks is to decrease the energy release rate. This can be accomplished by adding elastic heterogeneity or architecture to a material at the micrometer scale. Doing so causes the stress/strain to be redistributed and become less concentrated at the tip of a crack [53]. This type of strain redistribution mechanism is typically invariant to the crack growth history (i.e. not dependent on ΔA) and therefore results in a negative DC offset in the G curve (or, equivalently, an apparent positive DC offset in the R curve). This type of toughening has been referred to as “intrinsic” toughening [68].

In order to satisfy my second requirement for toughness—i.e., not being susceptible to catastrophic failure—the material should have a rising R curve. There are various mechanisms that can precipitate this rising R curve behavior [68]. In SBMs with lamellar architectures, like nacre, crack bridging, and crack arrest and renucleation are thought to be the dominant extrinsic toughening mechanisms [5, 39]. For example, as a crack propagates perpendicular to lamellae or fibers it often will not sever all of the lamellae. The intact lamellae form “bridges” behind the crack tip that effectively act like sutures, holding the crack faces together. This in turn decreases the stress concentration at the crack tip, and makes it more difficult for the crack to continue propagating (i.e., decreasing G or increasing apparent R). The reason that this results in a rising R curve is that as the crack propagates, the number of bridges spanning the crack wake increases with ΔA and consequently results in a rising R curve. This type of toughening has been referred to as “extrinsic” toughening.

I propose two simple metrics for quantifying the toughness enhancement provided by an SBM's lamellar architecture. To quantify the increase in resistance to initial crack growth alone I compare the fracture initiation toughness of the architected material, $R(0)^{(\text{arch})}$, to the fracture initiation toughness of a monolithic material with the same bulk chemical composition and elastic properties, $R(0)^{(\text{con})}$ (see Figure 2.1 (B)). In the case of the *E. aspergillum* spicules, I use the *T. aurantia*

spicules to compute $R(0)^{(\text{con})}$. My first toughness metric is given by $R(0)^{(\text{arch})}/R(0)^{(\text{con})}$. A value of $R(0)^{(\text{arch})}/R(0)^{(\text{con})} = 1$ would suggest that the architecture has no effect on its fracture initiation toughness and values of $R(0)^{(\text{arch})}/R(0)^{(\text{con})} > 1$ indicate that the architecture increases the fracture initiation toughness. To quantify the overall toughness enhancement provided by the lamellar ar-

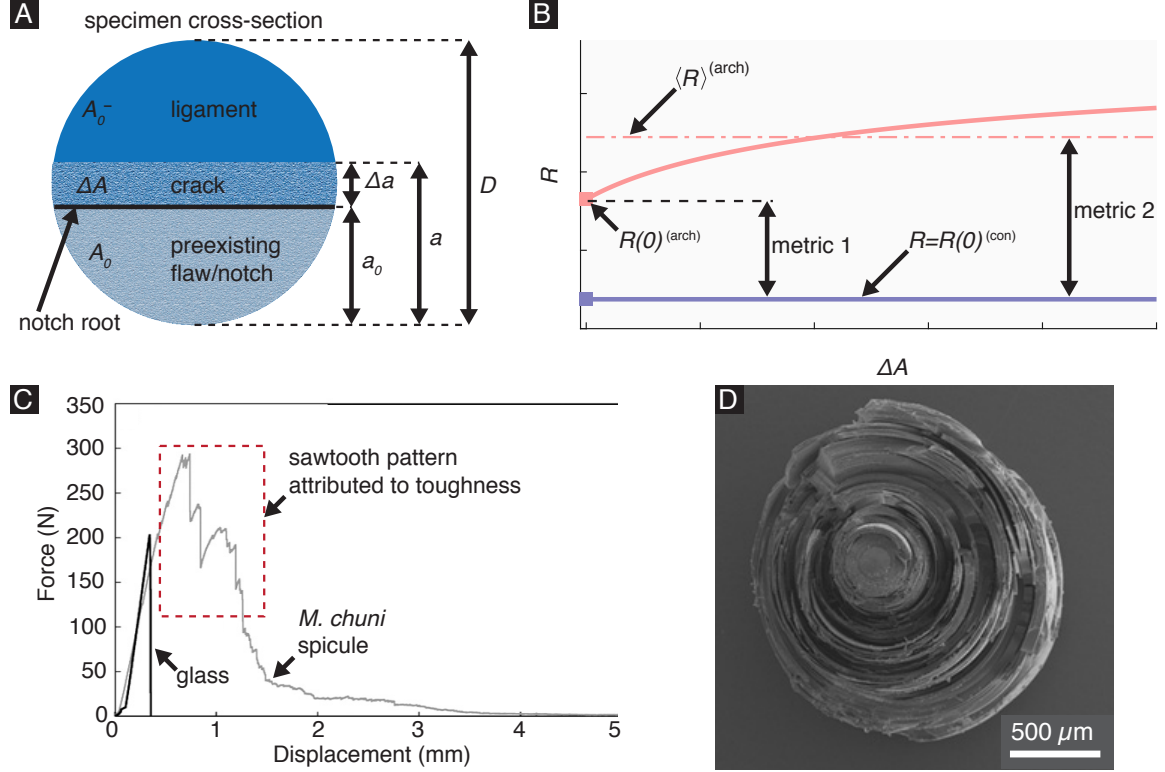


Figure 2.1: Toughness enhancement in SBMs with lamellar architecture. (A) Schematic showing a crack growing from a preexisting flaw, in this case a notch. The notch length, a_0 , crack length, Δa , are shown. (B) Schematic of a characteristic “rising” (pink) and “flat” (blue) R curve. The crack growth resistance, R , is plotted as a function of the crack area, ΔA . The toughness metrics are indicated along with the fracture initiation toughnesses and average crack growth resistance of the rising R curve. (C) The force-displacement response of a spicule from the sponge *Monorhaphis chuni* compared to that of a synthetic glass rod (modified with permission from [15]). Both specimens were tested in a three-point bending configuration and are not notched. The force at the onset of failure and the integrated area of the force-displacement response are both higher for the *M. chuni* spicule. It has been suggested that this is indicative of toughness enhancements provided by the spicule’s lamellar architecture [15]. (D) The lamellar architecture of the *M. chuni* spicule (modified with permission from [15]). The spicule is several millimeters in diameter and contains hundreds of silica layers.

chitecture, I compare the average crack growth resistance, $\langle R \rangle^{(\text{arch})}$, of the architected material to $R(0)^{(\text{con})}$ (see Figure 2.1 (B)). My second toughness metric is then given by $\langle R \rangle^{(\text{arch})}/R(0)^{(\text{con})}$. I interpret a value of $\langle R \rangle^{(\text{arch})}/R(0)^{(\text{con})} = 1$ to indicate that the *E. aspergillum* spicules have a constant R whose value is equal to the initiation toughness of the control material¹. This metric includes the

¹It is possible for a material to have $\langle R \rangle^{(\text{arch})}/R(0)^{(\text{con})} = 1$ and a non-constant R . Specifically, if the shape of the R curve were exotic [69] it would be possible for $\langle R \rangle = R(0)$. However, the R curves of other tough SBMs appear to monotonically increase [25,63]. I assume that the *E. aspergillum* spicules do not have an R curve with a complex, periodic

combined effects of toughening mechanisms precipitated by the lamellar architecture that increase the resistance to initial crack growth (i.e., the initiation toughness) and those that cause a rising R curve.

2.3 Materials and methods

2.3.1 Fracture specimen preparation

Euplectella aspergillum skeletons were received dried with the organic tissue removed (see Figure 1.2 (A)). I removed spicules from the basal portion of the skeleton using tweezers and cut ≈ 5 mm sections from roughly the midpoint along their length using a razor blade. *Tethya aurantia* spicules were received dried and separated from the organic tissue. Each *E. aspergillum* spicule section and *T. aurantia* spicule was inspected using a polarized light microscope. Sections of *E. aspergillum* spicules containing barbs (e.g. see Figure 1.2 (D)) were discarded. Due to the fragility of the outermost silica layers of the *E. aspergillum* spicules, surface cracks were commonly observed. Since pristine spicules were virtually nonexistent, only sections with missing pieces of layers were discarded. *Tethya aurantia* spicules that were not completely intact were also discarded.

All specimens were stored in dry conditions prior to testing. The mechanical properties of some SBMs change substantially if they are soaked in water before testing [7]. For example, the work of fracture of nacre that has been soaked in artificial seawater is 137% higher than that of the same nacre stored in dry conditions [7]. The soaking procedure is thought to restore the organic phases within them to their native, hydrated state. In Appendix B I compare the Young's modulus and bending failure strain of *E. aspergillum* spicule specimens stored in wet and dry conditions and find no significant difference between the two [58].

2.3.2 Fracture test overview

I performed bending tests on 27 *E. aspergillum* spicules and 17 *T. aurantia* spicules using a configuration similar to that described in [70–72]. Briefly, I placed a spicule across a trench that was

shape and therefore $\langle R \rangle^{(\text{arch})} / R(0)^{(\text{con})} = 1$ is indicative of a constant R .

Table 2.1: Summary of *E. aspergillum* and *T. aurantia* spicule fracture specimen dimensions

	<i>E. aspergillum</i> ($N = 27$)		<i>T. aurantia</i> ($N = 17$)	
	mean	s.e.	mean	s.e.
D (μm)	43.91	2.77	34.55	1.17
L (μm)	798.04	8.03	612.63	11.15
a_0 (μm)	16.37	1.19	12.14	0.72
n_r (nm)	93.3	13.4	91.2	14.0
α_0	0.38	0.02	0.35	0.02
δ	0.055	0.004	0.057	0.002

cut in a steel plate and ensured that its longitudinal axis was perpendicular to the trench edges. I used trenches whose spans were nominally 600 to 800 μm and measured the span of each trench, L , from optical micrographs (see Table 2.1). I then glued the ends of the spicule to the steel plate using conductive carbon adhesive so that only the section suspended over the trench remained exposed (see Figure 2.2 (A)). I cut a notch through part of the spicule's cross-section located mid way across the trench using a focused gallium ion beam (see Figure 2.2 (A)–(C)). This notch serves as the preexisting flaw from which a crack will grow as discussed in Section 2.2. After notching, I attached the steel plate to a motorized translation stage that is part of a custom-built mechanical testing device. I used the stage to push the spicule against a steel wedge that was positioned mid way across the trench's span until it fails (see Section 2.3.4).

The primary difference between my test configuration and that described in [70–72] is the shape of the specimen's cross-section. Specifically, the spicules have circular cross-sections instead of rectangular cross-sections. My test configuration can also be thought of as a single edge crack round bar bending (SEC-RBB) test [73, 74] modified so that the specimen's ends are encastered rather than supported by rollers.

2.3.3 Notching procedure

I cut a notch in the spicule using a focused gallium ion beam (FEI Helios). The notch's geometry is shown in Figure 2.2 (B) and (D). Briefly, the notch is nominally coincident with the spicule's

cross-section located mid way across the trench and has a length a_0 (see Figure 2.2 (B)).

Focused ion beams (FIBs) have previously been used to cut notches in micrometer-scale fracture specimens [72, 75–77]. Since the spicules are not conductive, after mounting, I coated them in 10 nm of amorphous carbon to prevent charge accumulation during the cutting procedure. In order to reduce the amount of cutting time, I cut the notch in two steps. First, I used a relatively large accelerating current of 6.5 nA to make a coarse cut (marked schematically in green in Figure 2.2 (B)). The width of this cut, B_1 , was approximately $1.5\ \mu\text{m}$ (see Figure 2.2 (B)). In the second step I used a lower accelerating current of 460 pA to make a narrower cut whose width B_2 was approximately 250 nm (shown schematically in orange in Figure 2.2 (B)). This two step cutting process and the resulting notch geometry is similar to the procedure for preparing standard edge-notch bending samples in which a notch is first cut using a diamond saw and then subsequently scored using a razor blade [78–80]. After cutting the notch, I imaged the spicule using the FIB and measured the spicule’s diameter, D , midway across the trench and notch length, a_0 from the micrographs (see Figure 2.2). A summary of these measurements for the *E. aspergillum* and *T. aurantia* spicules is given in Table 2.1 (details of each specimen’s geometry are provided in Tables 2.3 and 2.4). A representative micrograph of a notched spicule is shown in Figure 2.2 (C).

I describe the notch’s geometry using the orthonormal set of Cartesian basis vectors $\{\hat{e}_1, \hat{e}_2, \hat{e}_3\}$ and corresponding Cartesian coordinates $\{x_1, x_2, x_3\}$ shown in Figure 2.2 (A). The origin of this coordinate system is located at the point on the specimen’s longitudinal axis directly above the left trench edge (see Figure 2.2 (A)). In this undeformed configuration, the specimen’s cross-sections are normal to $\hat{e}_1$. Figure 2.2 (D) shows a schematic representation of the spicule’s cross-section at $x_1=L/2$. In this cross-section, the notched region is shown in light blue and the remaining ligament is shown in dark blue. The notch’s root (shown as a black line segment) is designed to be straight and parallel to $\hat{e}_3$. The cross-sectional area of the notch projected onto a plane whose normal is $\hat{e}_1$ is A_0 . The cross-sectional area of the remaining ligament (i.e., $\pi D^2/4 - A_0$) is A_0^- .

2.3.4 Summary of fracture test data

I used a custom-built mechanical testing device to perform the bending tests on the spicules. A detailed description of the construction and operation of this device is given in Chapter 3 Section

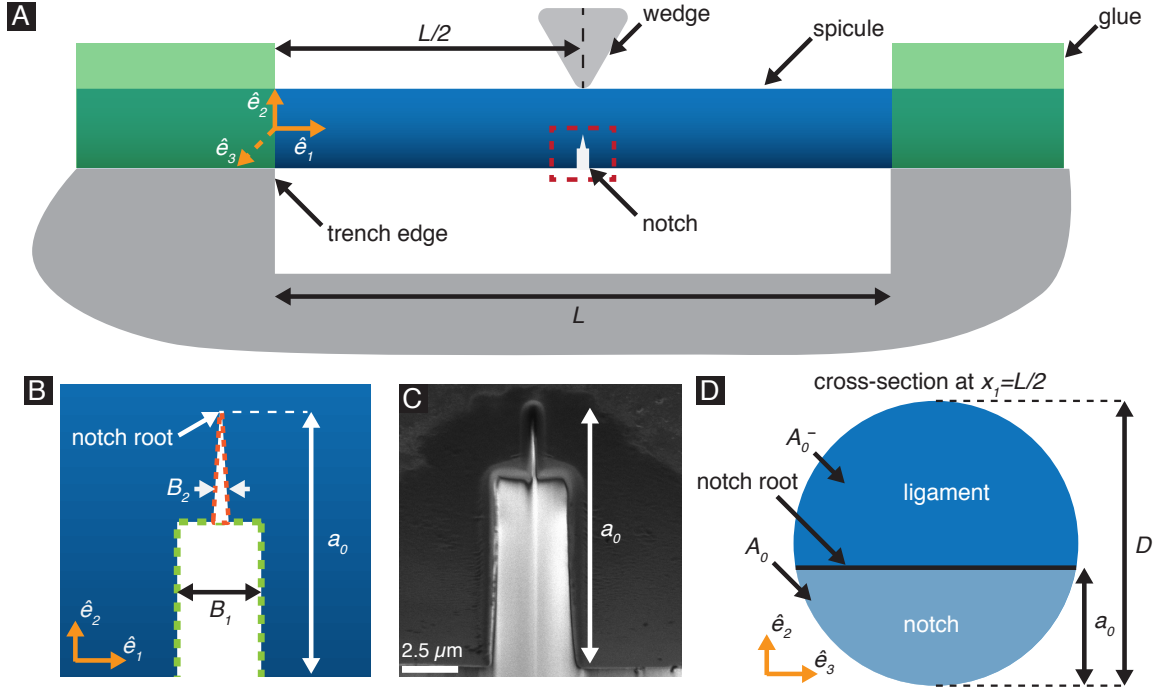


Figure 2.2: Spicule mounting and FIB notching procedure. (A) A schematic of the test configuration. (B) The region shown in the red rectangle in (A) showing the notch geometry. The portion of the notch cut using the high (resp. low) accelerating current is marked in green (resp. orange). (C) A scanning electron micrograph of the notch cut in a representative *E. aspergillum* spicule. (D) A schematic of the specimen's cross-section at $x_1=L/2$ after notching. The notch root is straight and parallel to $\hat{e}_3$. The notch length is a_0 . The remaining ligament has a cross sectional area of A_0^- and the notch has a cross-sectional area of A_0 .

3.2.2. Briefly, after notching, the steel plate to which the spicule is glued is attached to a motorized translation stage. I position the stage underneath an steel wedge so that the wedge is located mid way across the trench's span on the opposite side of the spicule from the notch (see Figure 2.2 (A)). I use the motorized translation stage to push the spicule into the wedge and measure the $\hat{e}_2$ component of the force applied by the wedge, F , and the displacement of the spicule's cross-section beneath the wedge, w_0 (see Figure 2.3 (A)).

I observed that F first increases with w_0 up to a value F_c , at which point there is an abrupt decrease in the force (see Figure 2.3 (D)). This abrupt decrease in force is commonly referred to as “pop-in”, and is a consequence of a crack beginning to grow from the notch root. The displacement corresponding to F_c is w_c . The amount of crack growth that occurs during the pop-in event determines the subsequent mechanical behavior. If the crack propagation during the pop-in event cleaves the entire cross-section, then my specimen resembles two cantilevers. If the crack propagation during the pop-in event does not completely cleave the specimen, then as F again increases, the

crack will continue to propagate across the cross-section. This case will result in a nonlinear F - w_0 response until the crack completely cleaves the specimen (see Figure 2.3 (D)).

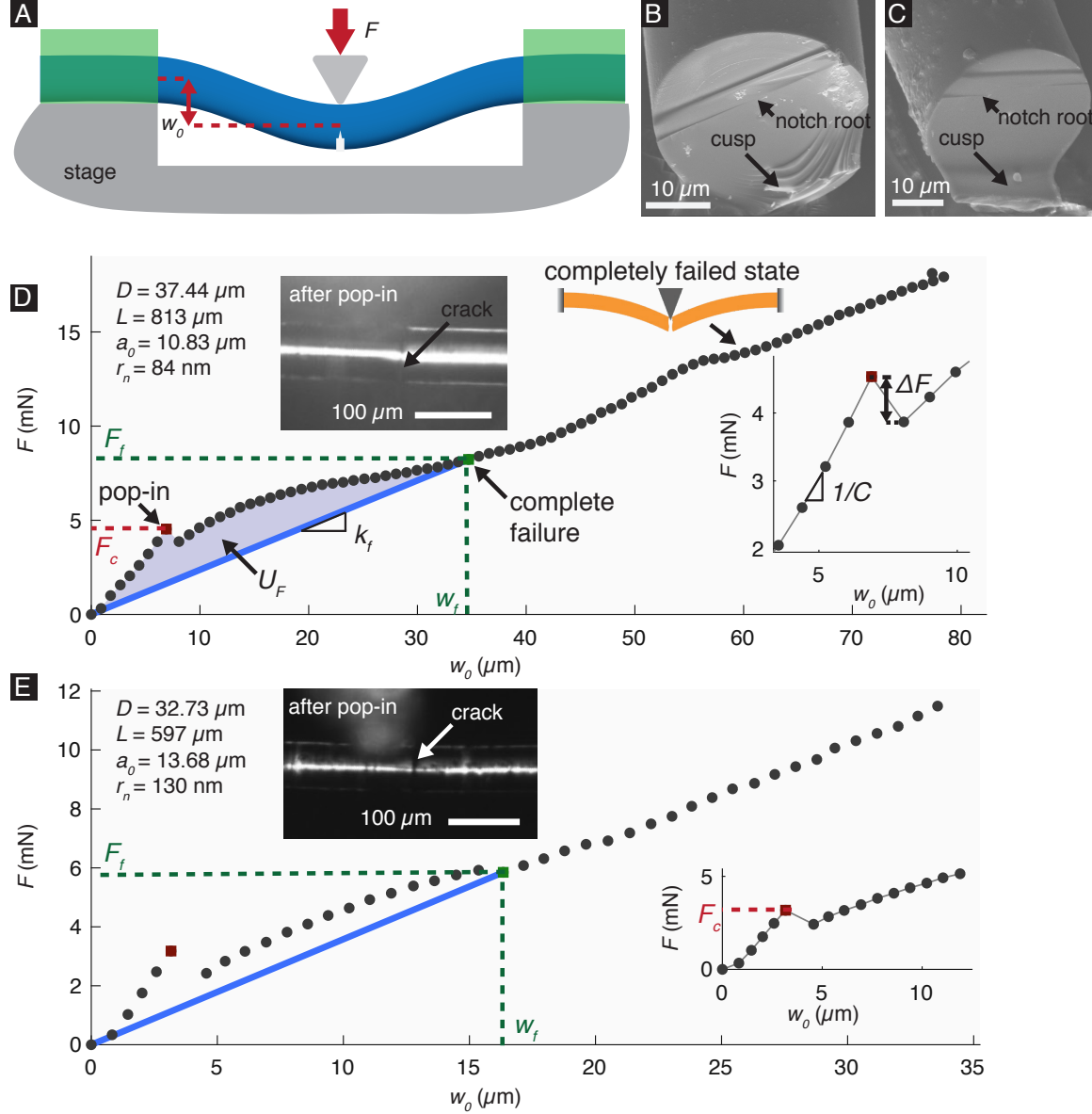


Figure 2.3: (A) The wedge shown in Figure 2.2 (A) is used to apply a force of magnitude F to the spicule. The corresponding displacement of the spicule's cross-section at $x_1 = L/2$ is w_0 . The glue (shown in green) prevents the ends of the spicule from rotating relative to the stage. (B) (resp. (C)) A micrograph of a fractured *E. aspergillum* (resp. *T. aurantia*) spicule. (D) The F - w_0 response (gray points) of a representative *E. aspergillum* spicule. The pop-in event (i.e., crack initiation) is marked as a red square. The nonlinear relationship between F and w_0 after pop-in is indicative of continued crack growth. The point of complete failure is marked with a green square. The blue line indicates the linear F - w_0 response of the completely failed spicule. The inset shows a magnified view of the F - w_0 response leading up to crack initiation. The accompanying micrograph was taken during the test immediately after the pop-in event. The crack does not appear to have cleaved the entire cross-section. (E) The F - w_0 response of a representative *T. aurantia* spicule. After the pop-in event the F - w_0 response appears nearly linear and corresponds to the completely failed state. In this case, the accompanying micrograph shows a crack that does appear to have cleaved the entire cross-section.

After the specimen has completely failed, it consists of two cantilevers each subjected to an end load (see Figure 2.3 (D)). As per Euler-Bernoulli beam theory [43] I would expect that, at least initially, the F - w_0 response of this two cantilever system will be linear. In both the *E. aspergillum* and *T. aurantia* spicules I observe that the F - w_0 response for w_0 much larger than w_c appears linear (see Figure 2.3 (D), (E)).

I could not determine the moment at which the spicule completely failed by examining micrographs taken during the test. Instead, I fit a line to this linear F - w_0 region, which occurs for $w_0 \gg w_c$. The location at which this line intersects the spicule's F - w_0 response should coincide with the moment at which the specimen has completely failed (see Figure 2.3 (D)). To find this intersection I computed the difference between the measured force, F , and the force predicted by the linear fit, $k_f w_0$, where k_f is the slope of the fitted line (see Figure 2.3 (D)). I define F_f and w_f to be the first F and w_0 for which $|F - k_f w_0|$ is less than the average change in F between two stage displacement increments; typically 200–400 μN . I also use k_f (see Figure 2.3 (D)), in Section 2.4.2 to compute the strain energy stored in the specimen after complete failure.

2.4 Results

2.4.1 Measurement of *E. aspergillum* and *T. aurantia* spicule fracture initiation toughness

The toughness metrics $R(0)^{(\text{arch})}/R(0)^{(\text{con})}$ and $\langle R \rangle^{(\text{arch})}/R(0)^{(\text{con})}$ require us to measure the fracture initiation toughnesses, $R(0)$, of the *T. aurantia* and *E. aspergillum* spicules. I use a compliance method to compute $R(0)$ since it does not place limitations on the geometry and configuration of the test specimen [81].

I assume that, at least initially, a crack propagates in the $\hat{e}_2$ direction from the notch root and that the crack front remains straight (i.e., the crack front is parallel to $\hat{e}_3$). In this case, the length of the crack with area ΔA is Δa (see Figure 2.1 (A)). I can rewrite the energy release rate $G = -d\Pi/d\Delta A$ in Eqn. (2.3) as

$$G(\Delta A) = \frac{F^2}{2} \frac{dC}{dA} \Big|_{A=A_0+\Delta A}, \quad (2.4)$$

where the compliance C is the reciprocal of the slope of the F - w_0 response prior to crack propagation (see Figure 2.3 (D) inset) and $A = A_0 + \Delta A$ is the combined cross-sectional area of the notch and the crack (see Figure 2.1 (A)) [81]. The derivative dC/dA in Eqn. (2.4) can be rewritten as $\frac{dC}{da}/\sqrt{4a(1-a)}$, where $a = a_0 + \Delta a$ is the combined length of the notch and the crack (see Figure 2.1 (A)) [73]. The energy release rate is given by

$$G(\Delta a) = \frac{F^2}{4} \frac{1}{\sqrt{a(1-a)}} \frac{dC}{da} \Big|_{a=a_0+\Delta a}. \quad (2.5)$$

Finally, I nondimensionalize the notch length and the crack length as $\alpha_0 = a_0/D$ and $\Delta\alpha = \Delta a/D$, respectively to get

$$G(\Delta a) = \frac{F^2}{4D^2} \frac{1}{\sqrt{\alpha(1-\alpha)}} \frac{dC}{d\alpha} \Big|_{\alpha=\alpha_0+\Delta\alpha}, \quad (2.6)$$

where $\alpha = \alpha_0 + \Delta\alpha$. At the initiation of crack growth (i.e. the pop-in event), $\Delta\alpha = 0$ and I take $G(0) = R(0)$ per the necessary criterion for crack growth given by Eqn. (2.3) to get

$$R(0) = \frac{F_c^2}{4D^2} \frac{1}{\sqrt{\alpha(1-\alpha)}} \frac{dC}{d\alpha} \Big|_{\alpha=\alpha_0}. \quad (2.7)$$

Notice that for $\alpha_0=0$ the encastered SEC-RBB specimen is an unnotched beam with encastered supports subjected to a concentrated load applied mid way across the span. For this case I can compute $C = L^3/192EI$ using the Euler-Bernoulli beam theory [43], where E is the Young's modulus of the specimen and $I = \pi D^4/64$ is the second moment of area of its cross-section. For $\alpha_0=1$ the notch completely cleaves the specimen into two identical cantilevers, each with a length of $L/2$. Again, I can compute C for this two beam system using the Euler-Bernoulli beam theory to be $C = L^3/48EI$. For values of α_0 between zero and unity it is natural to expect that C will lie between these two bounds. I define a dimensionless compliance $\bar{C} = 192CEI/L^3$ so that $\bar{C} = 1$ for $\alpha_0 = 0$ and $\bar{C}=4$ for $\alpha_0 = 1$. Using this nondimensionalization I can rewrite Eqn. (2.7) as

$$R(0) = \frac{F_c^2 L^3}{768D^2 EI} \frac{1}{\sqrt{\alpha(1-\alpha)}} \frac{d\bar{C}}{d\alpha} \Big|_{\alpha=\alpha_0}. \quad (2.8)$$

I can measure D and α_0 from scanning electron micrographs taken during the notching procedure (see Figure 2.2 (C)), L from polarized light micrographs of the trench, and F_c from the F - w_0

data obtained during the bending test (see Figure 2.3 (D)). By assuming that the specimen in the completely failed state consists of two cantilevers each with a length of $L/2$ subjected to an end load of magnitude $F/2$, I use the Euler-Bernoulli beam theory to compute $E = k_f L^3 / 48I$ (see Section 2.3.4) [43]. However, $\bar{C}$, and therefore its derivative $d\bar{C}/d\alpha$, depend on the specimen geometry and loading conditions and are not known *a priori* for the encastered SEC-RBB specimen that I used.

This derivative is typically estimated for a given test configuration by measuring the compliance of multiple specimens having the same diameter but different notch lengths (and therefore a variety of α values) and constructing a set of C and α values known as compliance calibration data [73, 74]. A function is fitted to this C - α data and differentiated to obtain the derivative $dC/d\alpha$ for any value of α . It is also possible to use a series of computational mechanics models (i.e. virtual experiments) instead of physical experiments to obtain the compliance calibration data [82, 83].

There are two primary limitations of this method. The first is that the choice of the function used to fit the compliance calibration curve can have a substantial effect on the computed derivative and therefore on $R(0)$. Previous studies have used a fourth or fifth order polynomial as the fitting function [73, 74]. While these functions will provide a close fit to the data, since they are not motivated by the underlying physics/mechanics they do not necessarily respect some innate properties of the compliance function. For example, the compliance for $\alpha=0$ and $\alpha=1$ are known since these cases correspond to the compliances of an uncracked beam with encastered supports subject to a concentrated load applied midway along its length and of two cantilever beams with concentrated end loads, respectively (see Section A.1). These boundary values could be used to place restrictions on the form of the fitted polynomial.

The second major shortcoming is that this method often requires a large amount of data. It has been suggested that the number of data points needed in a nonlinear regression ² should exceed the number of fitting parameters by a factor of 5–10 in order to prevent overfitting [84]. This would mean that ≈ 25 –50 experiments are required for the fifth order polynomial typically used for fitting. Furthermore, it has been shown that the compliance of a notched beam in a three-point bending

²The suggested polynomial fitting requires a multilinear, rather than a nonlinear regression. However, I later require that the fitted polynomial must be monotonically increasing (see A.5) and impose this requirement by introducing a nonlinear constraint.

configuration depends on the slenderness of the beam, $\delta = D/L$ [73, 85]. This means that a set of compliance calibration data would only be valid for a single value of δ . In the case of the spicules, I cannot control the diameter and have limited flexibility in my choice of L (i.e., it is impractical to cut a new trench in the sample stage tailored for each individual spicule specimen). Thus, each spicule specimen will have a different value of δ and require a different set of compliance calibration data. I tested a total of 27 *E. aspergillum* spicules and 17 spicules from the control species *T. aurantia*. Consequently, using this method I would need to perform ≈ 2000 calibration experiments, which would be impractical.

I explored whether I could refine the aforementioned compliance method to

- i. Account for the dependence on δ so that a single set of compliance calibration data could be used for specimens with various values of δ
- ii. Use a mechanics motivated fitting function in order to reduce the number of fitting parameters needed.

To this end, in A.1 I show using dimensional analysis that $\bar{C} \rightarrow \bar{C}(\alpha, \delta)$. Then in A.2 I show that a first order estimate of $\bar{C}$'s dependence on δ is given by $\bar{C}(\alpha, \delta) = 1 + g(\alpha) + \delta h(\alpha)$, where g and h are unknown functions of α only and therefore should be independent of the value of δ for each specimen. Using a computational mechanics model of the encastered SEC-RBB specimen, I obtained $\bar{C}$ for 10 different values of α and a fixed value of δ (see A.3). I approximate g and h as polynomials and fit them to the $\bar{C}$ - α data from the computational mechanics model to obtain an approximation for $\bar{C}$ that I can differentiate to get $d\bar{C}/d\alpha|_{\alpha=\alpha_0}$ (see A.4). Finally, I find that the fracture initiation toughness is given by

$$R(0) = \frac{F_c^2}{16D^2k_f} \frac{1}{\sqrt{\alpha_0(1-\alpha_0)}} Y(\alpha_0, \delta)$$

$$Y(\alpha_0, \delta) = 5a_5^* \alpha_0^4 + 4(a_4^* + \delta b_4^*) \alpha_0^3 - 3(6 + 2a_4^* + 3a_5^* + 2\delta b_4^*) \alpha_0^2 + 2(9 + a_4^* + 2a_5^* + \delta b_4^*) \alpha_0, \quad (2.9)$$

where $a_4^* = 69.82$, $a_5^* = -35.38$ and $b_4^* = 4.39$ are the best fit coefficients of the polynomial approximations of g and h and are independent of δ (see A.4 for details).

The accurate estimation of $R(0)$ using Eqn. (2.9) is predicated on the assumption that the cut

notch behaves like a sharp crack (i.e., one whose root radius is vanishingly small). It has been shown that the FIB cutting technique produces notch root radii that are small enough to act like sharp cracks [77]. This is supported by additional work [79,86] that has shown that if the notch root radius is less than twice the smallest microstructural length scale, then the measured value of $R(0)$ becomes insensitive to the notch root geometry. The *E. aspergillum* spicules' layers are composed of silica nanoparticles that are approximately 100 nm in diameter [28]. I take the scale of these nanoparticles to be the smallest microstructural length scale present in the *E. aspergillum* spicules. I assume that the *T. aurantia* spicules have a similar smallest microstructural length scale. Therefore, in order for the value of $R(0)$ to be insensitive to the notch geometry, the notch root radius r_n (see Figure 2.2) should be less than 200 nm. I measured r_n for each specimen from scanning electron micrographs by manually selecting three points along the profile of the notch root and fitting a circle to these points. The mean value of r_n for the 27 *E. aspergillum* spicules and 17 *T. aurantia* spicules was 93.3 nm and 91.2 nm, respectively (see Table 2.1). I identified 3 *E. aspergillum* spicule specimens and 2 *T. aurantia* spicule specimens for which r_n exceeded 200 nm, and consequently I did not compute $R(0)$ for these specimens. Additionally, there was 1 *E. aspergillum* spicule specimen for which I was unable to reliably identify the pop-in event by inspecting the F - w_0 response.

I computed $R(0)$ for 15 *T. aurantia* spicules to be 2.30 ± 0.41 J/m² (mean $\pm$ standard error). The values of $R(0)$ are similar to those expected from glass or other brittle ceramic materials (i.e., ≈ 1 -10 J/m²) [87]. By plotting $R(0)$ as a function of α_0 (see Figure 2.4 (A)) I see that $R(0)$ increases with α_0 but appears to be relatively constant for α_0 less than ≈ 0.4 . A similar phenomenon has been observed in previous studies on specimens with circular cross-sections [88]. I speculate that this increase is partly due to local compressive stresses near where the wedge is in contact with the spicule reducing G , and thus making the measured $R(0)$ artificially high [89]. This effect becomes more pronounced for larger α_0 since the notch root is closer to the wedge. In my tests, I also observed that $R(0)$ values have a greater scatter at larger α_0 . I believe that this is a consequence of how I compute the term $d\bar{C}/d\alpha|_{\alpha=\alpha_0}$ in Eqn. (2.8). An error in the measurement of α_0 from the scanning electron micrographs will result in an error in the calculation of $d\bar{C}/d\alpha|_{\alpha=\alpha_0}$, and the magnitude of this resulting error is determined by the rate at which $d\bar{C}/d\alpha$ changes—i.e., the magnitude of $d^2\bar{C}/d\alpha^2|_{\alpha=\alpha_0}$. I find that for the range of α_0 that I used, $d^2\bar{C}/d\alpha^2|_{\alpha=\alpha_0}$ is greatest

for larger values of α_0 (see Figure A.1 (E)). Consequently, for a fixed error in the measurement of α_0 , the error in $d\bar{C}/d\alpha|_{\alpha=\alpha_0}$ will increase with α_0 . As a consequence of these two effects, I believe that the mean value of $R(0)=1.81$ for the 11 specimens in which $\alpha_0 \leq 0.4$ is a better representation of the actual fracture initiation toughness of the *T. aurantia* spicules.

I measured $R(0)$ for 23 *E. aspergillum* spicules to be 5.12 ± 1.18 J/m² (mean $\pm$ standard error). I also observe an increase in $R(0)$ with α_0 for the *E. aspergillum* spicules (see Figure 2.4 (A)). I attribute this increase to the same causes that I proposed for the *T. aurantia* spicules. The range of α_0 values was larger for the *E. aspergillum* spicules than for the *T. aurantia* spicules—0.18 to 0.64 versus 0.24 to 0.44 (see Figure 2.4 (A)). However, by using larger values of α_0 , the mean value of $R(0)$ becomes larger as well. The mean value for the 15 specimens in which $\alpha_0 \leq 0.4$ is $R(0)=2.09$. I believe that this is a better representation of the actual fracture initiation toughness of the *E. aspergillum* spicules. Therefore, when computing the toughness metrics $R(0)^{(\text{arch})}/R(0)^{(\text{con})}$ and $\langle R \rangle^{(\text{arch})}/R(0)^{(\text{con})}$, I use the mean values of $R(0)$ for the *E. aspergillum* and *T. aurantia* spicules in which $\alpha_0 \leq 0.4$.

2.4.2 Measurement of *E. aspergillum* spicule average crack growth resistance

The toughness metric $\langle R \rangle^{(\text{arch})}/R(0)^{(\text{con})}$ depends on the average crack growth resistance of the architected material, $\langle R \rangle^{(\text{arch})}$. This can be computed in a straightforward manner if the complete R curve of the specimen is known. The R curve is typically measured by using Eqn. (2.6) to compute the energy release rate at every load step after crack growth initiation [90]. This requires us to measure the crack length, Δa at every loading increment. One option is to directly measure the crack length from SEM micrographs taken during the fracture test [91]. This is straightforward so long as the propagation of the crack proceeds in a relatively simple manner so that

- i. the crack follows a straight path (i.e., the new surfaces created are flat and parallel to the $\hat{e}_1$ plane),
- ii. the shape of the crack front does not change (i.e., it remains a line segment parallel to $\hat{e}_3$), and
- iii. the topology of the crack does not change (i.e., it does not branch and no new cracks nucleate).

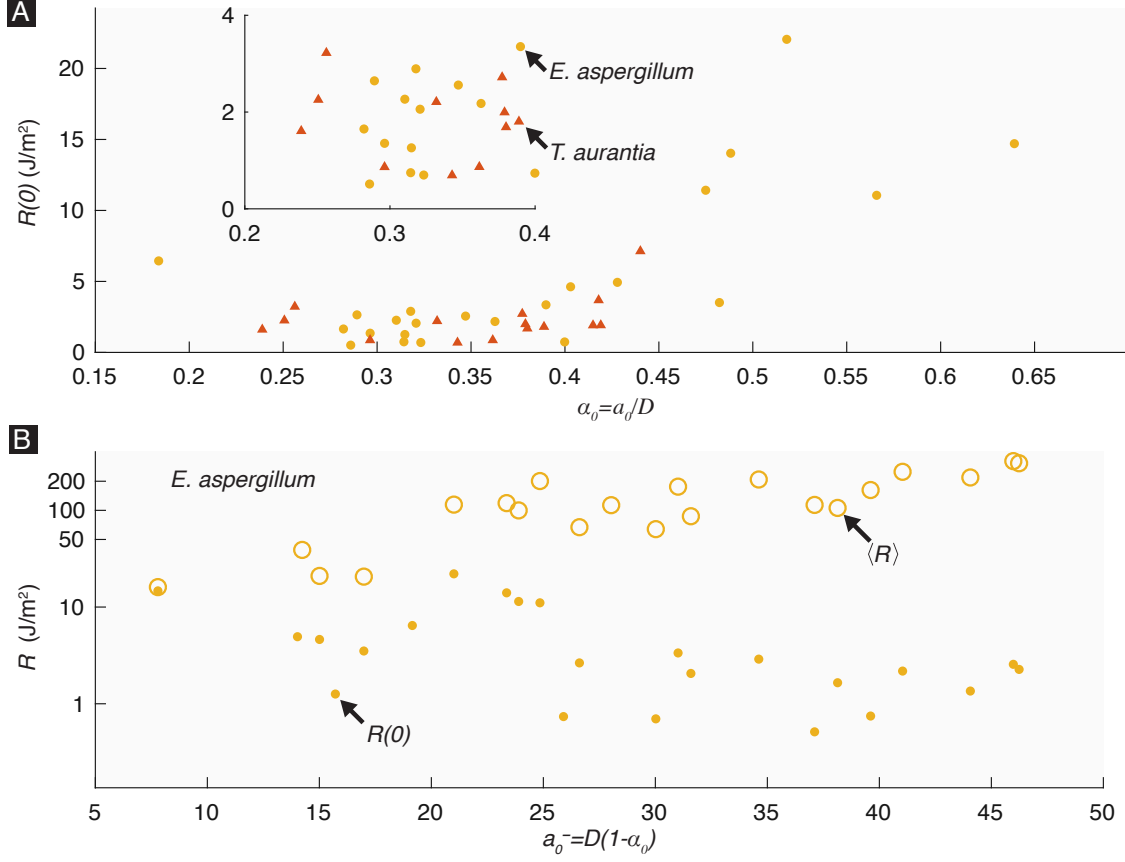


Figure 2.4: Crack growth resistance of *E. aspergillum* and *T. aurantia* spicules. (A) Fracture initiation toughness, $R(0)$ of 23 *E. aspergillum* (yellow circles) and 15 *T. aurantia* (red triangles) spicules. The fracture initiation toughness increases with the dimensionless notch length $\alpha_0 = a_0/D$. This increase becomes more pronounced for $\alpha_0 \geq 0.4$. The inset shows a zoomed view of the data for which $\alpha_0 \leq 0.4$. (C) Comparison of $R(0)$ (solid circles) and $\langle R \rangle$ (hollow circles) for the *E. aspergillum* spicules. As the ligament length $\bar{a}_0 = D(1 - \alpha_0)$ decreases, the values of $R(0)$ increase and approach the values of $\langle R \rangle$.

If any of these conditions are not met, then it becomes difficult to define the crack length at each loading increment. Some toughening mechanisms like crack wake bridging, crack arrest and renucleation (accompanied by deflection) and the Cook-Gordon mechanism [92] make it difficult to both define and measure the crack length directly. Thus, the direct measurement method is typically only useful for homogeneous materials that lack these toughening mechanisms and it becomes problematic for SBMs or other architected materials.

The crack length can also be estimated indirectly. The indirect method involves partially unloading the specimen after every few loading increments. The elastic compliance of the specimen, C , can then be measured from the slope of the unloading branch of the F - w_0 data. This can be used in conjunction with the compliance calibration data presented in A.3 in order to estimate the crack

length Δa . The main problem with the indirect method is that some toughening mechanisms, like crack bridging, depend on the loading history of the specimen and therefore will be affected by this series of loading and unloading steps. For example, in the case of crack bridging it has been shown that unloading the specimen can cause lamellae that bridge the crack wake to buckle and fail. The deterioration of the crack bridges will reduce the influence of crack bridging on the crack growth resistance. Thus, this measurement method will have an effect on the measured value of R . Other methods exist for computing the R curve, such as the crack tip opening displacement and crack tip opening angle methods [90, 93], but all of these methods depend on the accurate measurement of the crack geometry at each loading increment.

It has been shown that the average crack growth resistance can be computed using the work of fracture method [94]. This method was originally developed by the material science community for measuring the total energy that is consumed by the fracture process, U_F , of ceramic materials [59, 60, 95]. Specifically, the work of fracture is given by

$$\gamma_{WOF} = U_F / 2A_0^-, \quad (2.10)$$

where $A_0^- = \pi D^2/4 - A_0$ is the cross-sectional area of the ligament (see Figure 2.2 (D)). For a specimen with a circular cross-section,

$$A_0^- = D^2(\pi + 2(1 - 2\alpha_0)\sqrt{\alpha_0(1 - \alpha_0)} - \cos^{-1}(1 - 2\alpha_0))/4. \quad (2.11)$$

If a crack propagates in a stable, quasi-static manner then the change in kinetic energy of the system as the crack extends is negligible. An energy balance at each increment of stable crack extension then gives us

$$d\Pi = dU_e - dW = -dU_F, \quad (2.12)$$

where $d\Pi$ is a decrement in the potential energy which corresponds to an increment in the energy dissipated by the fracture process, dU_F . The reduction of the potential energy is given in terms of the change in the elastic strain energy, dU_e , and the change in the work done by applied forces, dW .

Integrating both sides of Eqn. (2.12) I get

$$\int_0^{U_F} dU_F = \int_0^{w_f} \underbrace{F dw_0}_{dW} - \int_0^{U_{ef}} dU_e, \quad (2.13)$$

where U_{ef} is the elastic energy of the specimen in the completely failed state and w_f is the value of w_0 at which the specimen has completely failed. In many test configurations, such as the SEC-RBB specimen, $U_{ef} = 0$. However, for the encastered SEC-RBB configuration that I use, U_{ef} is nonzero since the specimen has a finite stiffness even in the completely failed state. Specifically, when the encastered SEC-RBB specimen is completely failed, the elastic energy is given by $U_{ef} = k_f w_f^2/2$, where k_f is the stiffness of the completely failed specimen, which I estimate in Section 2.3.4. I can therefore simplify Eqn. (2.13) to get

$$U_F = \int_0^{w_f} F dw_0 - k_f w_f^2/2. \quad (2.14)$$

Finally, it has been shown that γ_{WOF} is related to $\langle R \rangle$ as [94]

$$\langle R \rangle = 2\gamma_{WOF} = \frac{\int_0^{w_f} F dw_0 - k_f w_f^2/2}{A_0^-}. \quad (2.15)$$

In order to compute U_F using Eqn. (2.14), the change in kinetic energy of the system should be small compared to the change in potential energy [59, 60]. This requirement is satisfied if each point of the F - w_0 response represents a quasistatic equilibrium configuration. The pop-in event—the abrupt drop in F at the initiation of crack growth—is not a stable crack extension. Consequently, some potential energy will be converted to kinetic energy rather than consumed by the fracture process, and thus $\langle R \rangle$ computed using Eqn. (2.15) will be overestimated. However, if the drop in force is small, then the pop-in event should have only a minor effect on the estimation of $\langle R \rangle$.

Several steps were taken to improve the likelihood that the pop-in event will not result in catastrophic failure and that subsequent crack propagation will be stable. First, increasing the length of the notch and decreasing its root radius both will improve test stability [96, 97]. Both longer and sharper notches will result in higher stress concentrations. By increasing the stress concentration at the notch root, less potential energy is stored in the specimen before the stress at the notch

root reaches the intrinsic strength of the material and causes a crack to initiate [96]. Consequently, less energy will be available to feed the growth of a crack (see Eqn. (2.12)). This then limits the amount of possible crack extension during pop-in. It has also been shown that the encastered edge crack beam configuration increases the stability of crack growth compared to the more conventional simply supported edge crack beam (or SENB) test [71].

By inspecting the micrographs taken during the fracture tests, I determined that the pop-in event corresponded to complete fracture of the *T. aurantia* spicule specimens. That is, the abrupt crack extension during pop-in was equal to the ligament length. In this case, the change in kinetic energy cannot be neglected and the average crack growth resistance cannot be determined using the work of fracture method for these specimens.

In the case of the *E. aspergillum* spicules, a propagating crack will interact with the spicules' architecture and I do not expect the crack to grow at the same rate through all of the silica layers. Since I do not know and cannot measure how the crack interacts with the architecture as it propagates, I cannot use micrographs taken during the fracture test to determine the amount of crack extension that occurred during pop-in. Rather, the micrographs can only be used to determine the crack extension in the outermost silica layers. The micrographs suggest that the crack propagates entirely through the outermost silica layers during pop-in. However, the nonlinearity of the F - w_0 response following crack initiation (see Figure 2.3 (D)) stands in contrast to the linear post-initiation response of the *T. aurantia* specimens (see Figure 2.3 (E)) and suggests that the crack extension during pop-in does not completely cleave the *E. aspergillum* specimens.

I used the magnitude of the drop in F at pop-in as a criterion for determining which *E. aspergillum* spicule specimens could be used to estimate $\langle R \rangle$. Specifically, I measured the abrupt drop in force ΔF_c (see Figure 2.3 (D) inset) and compared it to F_c . I considered a given test to have predominantly stable crack extension if $\Delta F_c/F_c \leq 0.15$. This amounted to 21 of the total 27 *E. aspergillum* specimens tested. It is worth noting that any nonzero ΔF_c is evidence that crack growth during the pop-in event is not quasistatic and therefore the values of $\langle R \rangle$ that I compute are upper bounds to the actual value of $\langle R \rangle$.

It is also necessary to consider whether all of the energy dissipated (i.e., the change in potential energy calculated in Eqn. (2.12)) can be attributed to the propagation of a crack and its related

fracture processes. For example, it has been shown that in SENB specimens energy dissipation due to frictional sliding of the specimen on the roller supports is significant [98]. Contributions from non-fracture dissipation mechanisms like this would appear as additional terms on the left hand side of Eqn. (2.13). Ignoring these additional terms would lead to the overestimation of U_f and subsequently $\langle R \rangle$. In a series of papers, Elices, Planas, and Guinea described and quantified the main sources of non-fracture dissipation in SENB specimens [89, 98, 99]. Yet another beneficial feature of the encastered SEC-RBB specimen that I use is that the majority of the sources of dissipation described by Elices *et al* for the SENB specimen do not exist for the encastered SEC-RBB specimen. For example, since the ends of my specimens cannot rotate or slide on the test fixture there cannot be any frictional dissipation. Due to the dimensions (micrometer-scale) of the spicules, other sources of dissipation, like hysteretic behavior of the testing equipment, are negligible.

I computed A_0^- (see Figure 2.2 (D)) for each *E. aspergillum* spicule specimen using the a_0 and D that I measured from scanning electron micrographs (see Figure 2.2 (B), (C)). I then computed $\langle R \rangle$ from Eqn. (2.15) using trapezoidal integration of the $F-w_0$ data and the w_f and F_f determined in Section 2.3.4. The mean and standard error of $\langle R \rangle$ are 134.60 J/m² and 19.66 J/m², respectively, and the measurements of $\langle R \rangle$ for each spicule are shown in Figure 2.4 (B) and Table 2.3.

2.4.3 Fractography of *E. aspergillum* and *T. aurantia* spicules

After testing the *E. aspergillum* and *T. aurantia* spicule specimens, I imaged their fracture surfaces using a scanning electron microscope (see Figure 2.3 (B), (C)). In all *E. aspergillum* and *T. aurantia* specimens, failure appears to occur via a single crack that originates at the notch root. In both the *E. aspergillum* and *T. aurantia* spicule specimens the fracture surface is relatively featureless. The lack of fracture surface roughness in the *E. aspergillum* spicules stands in contrast to fractographs of other SBMs with lamellar architectures [55], such as nacre and conch shell (see Figure 1.1 (B), (C)). In these materials crack deflection at the interfaces between layers is often associated with the toughening mechanisms of crack bridging as well as crack arrest and renucleation [55, 100, 101]. This suggests that toughening mechanisms such as crack bridging and crack arrest that are prevalent in these other SBMs may not be operating in the *E. aspergillum* spicules.

Both spicules display a cusp feature adjacent to where the load is applied (see Figure 2.3 (B),

(C)). This cusp is characteristic of the three point bending configuration [102] and is a consequence of the compressive stresses caused by the wedge inducing local mixed-mode fracture conditions, which cause the crack to change the direction of propagation [67]. That is, the cusp is not a result of the *E. aspergillum* spicule's architecture.

2.5 Discussion: comparison of toughness enhancements in *E. aspergillum* spicules and other biological materials

I computed the toughness metrics using the data presented in Sections 2.4.1 and 2.4.2. Using the mean values of $\langle R \rangle^{(\text{arch})}$, and $R(0)^{(\text{arch})}$ and $R(0)^{(\text{con})}$ for $\alpha_0 \leq 0.4$ I find that $R(0)^{(\text{arch})}/R(0)^{(\text{con})} = 1.15$ and $\langle R \rangle^{(\text{arch})}/R(0)^{(\text{con})} = 74.31$. I also compute $\langle R \rangle^{(\text{arch})}/R(0)^{(\text{arch})} = 64.6$, which indicates how much larger the average R is compared to the initial value $R(0)$. I interpret this as evidence that the *E. aspergillum* spicules do have a rising R curve since in a material with a constant R , the value of $\langle R \rangle^{(\text{arch})}/R(0)^{(\text{arch})}$ would be equal to unity.

The toughness metrics alone do not tell us whether the spicule's architecture is one that is worth replicating in bio-inspired engineering composites. However, $R(0)^{(\text{arch})}/R(0)^{(\text{con})}$ and $\langle R \rangle^{(\text{arch})}/R(0)^{(\text{con})}$ can be used to quantitatively compare the toughness enhancements provided by the *E. aspergillum* spicule's lamellar architecture to those observed in other SBMs (see Table 2.2). I computed $R(0)^{(\text{arch})}/R(0)^{(\text{con})}$ and $\langle R \rangle^{(\text{arch})}/R(0)^{(\text{con})}$ for a number of prototypically "tough" SBMs, such as nacre, bone, antler and conch, using $R(0)$ and γ_{WOF} data available from literature (see Table 2.2 and Figure 2.5) [7, 9, 25, 46–50]. In all of the other SBMs shown in Figure 2.5, the architecture provides a substantial enhancement to the fracture initiation toughness. The spicules on the other hand derive little enhancement to their fracture initiation toughness. Furthermore, by comparing $\langle R \rangle^{(\text{arch})}/R(0)^{(\text{con})}$ values I see that the overall toughness enhancement provided by the spicule's architecture is small compared to that observed in nacre, bone, antler and conch. Thus, while the *E. aspergillum* spicules share a common architectural motif with many tough SBMs, my measurements suggest that these seemingly similar architectures do not provide the same enhancements to toughness.

In order for $R(0)^{(\text{arch})}/R(0)^{(\text{con})}$ and $\langle R \rangle^{(\text{arch})}/R(0)^{(\text{con})}$ to be meaningful metrics for quantifying toughness enhancement in both the *E. aspergillum* spicules and these other SBMs, it is critical that

Table 2.2: Summary of SBM crack growth resistance data. The data shown in bold are used to compute the toughness metrics in Figure 2.5.

Material	$R(0)$ (J/m ²)	$\langle R \rangle$ (J/m ²)	Ref.
nacre (<i>Pinctada margaritifera</i>)	587	2068	[7]
nacre (<i>Pinctada margaritifera</i>)		3300	[9]
nacre (<i>Pinctada margaritifera</i>)		2880	[46]
nacre (<i>Pinna nobilis</i>)		400	[46]
nacre (<i>Trochus niloticus</i>)		1400	[46]
nacre (<i>Haliotis rufescens</i>)		1100	[46]
nacre (<i>Haliotis rufescens</i>)	300		[63]
bone (<i>Homo sapiens</i>)	50–200	11250	[48]
bone (<i>Homo sapiens</i>)	224		[49]
bone (<i>Papio anubis</i>)		15520	[47]
bone		8000	[9]
antler (<i>Cervus canadensis</i>)	100	14000	[25]
conch (<i>Strombus gigas</i>)	25	26000	[50]
aragonite	≈ 3		[103]
hydroxyapatite	≈ 10		[103]

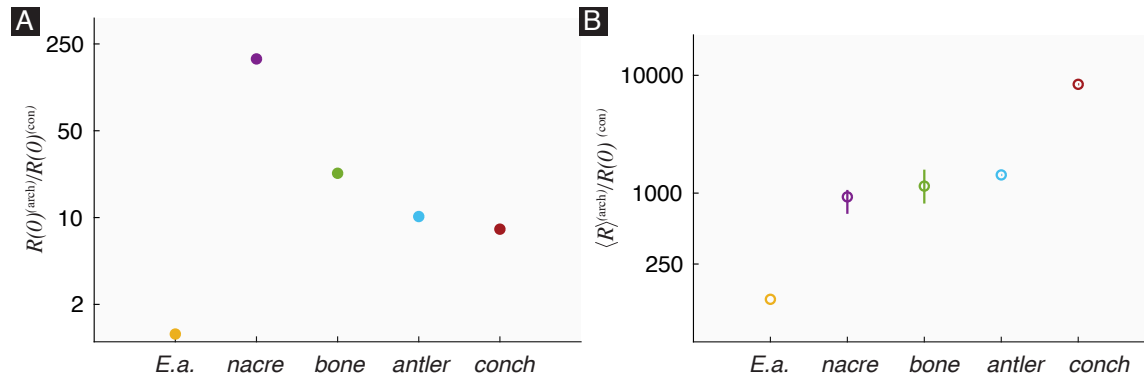


Figure 2.5: Comparison of the *E. aspergillum* spicule's toughness to the toughnesses of other SBMs with lamellar architectures. (A) The toughness metric $R(0)^{(\text{arch})}/R(0)^{(\text{con})}$ of the *E. aspergillum* spicules and other SBMs with lamellar architectures. The values of $R(0)$ for nacre [7], bone [49], antler [25], and conch (*Strombus gigas*) [50] were taken from the literature. (B) The toughness metric $\langle R \rangle^{(\text{arch})}/R(0)^{(\text{con})}$ of the *E. aspergillum* spicules and other SBMs with lamellar architectures. Again, the values of $\langle R \rangle$ that I used to compute $\langle R \rangle^{(\text{arch})}/R(0)^{(\text{con})}$ for nacre [7, 9, 46], bone [9, 47, 48], antler [25], and conch (*Strombus gigas*) [50] were taken from the literature. I used the value $R(0)^{(\text{con})} = 3 \text{ J/m}^2$ of calcite as the control material for nacre and conch [103], and $R(0)^{(\text{con})} = 10 \text{ J/m}^2$ of hydroxyapatite [103] as the control material for bone and antler when computing $R(0)^{(\text{arch})}/R(0)^{(\text{con})}$ and $\langle R \rangle^{(\text{arch})}/R(0)^{(\text{con})}$. In the case of nacre and bone, the vertical lines in (B) denote the range of $\langle R \rangle^{(\text{arch})}/R(0)^{(\text{con})}$ that results from using different values of $\langle R \rangle$ obtained from literature (see Table 2.2 for details)

the properties of the chosen control material be representative of the properties of the ceramic phase of the architected SBM. Specifically, I assume that the crack growth resistance of the control material is the same as that of the SBM's ceramic phase. The crack growth resistance is likely influenced by a material's chemical composition, molecular structure, microstructure (e.g., crystalline vs. amorphous), as well as the distribution of the protein scaffold within the ceramic phase. It is unreasonable to assume that all of these characteristics would be identical in both the *E. aspergillum* and *T. aurantia* spicules. Therefore, while I believe that the *T. aurantia* spicules are a much better choice for a control material than synthetic soda-lime glass, they are not a truly ideal control material. In order to refine the conclusions drawn from this study, I suggest using the monolithic silica core of the *E. aspergillum* spicules as the control material. Obtaining sections of the core that are large enough to perform fracture tests has proven to be very challenging. However, doing so would provide a more accurate estimate of the toughness enhancements provided by the *E. aspergillum* spicule's lamellar architecture.

By comparing $R(0)^{(\text{arch})}/R(0)^{(\text{con})}$ and $\langle R \rangle^{(\text{arch})}/R(0)^{(\text{con})}$ for the other SBMs shown in Figure 2.5 I also observed an interesting correlation between architectural complexity and toughness enhancement. Nacre, which has a relatively simple lamellar architecture consisting of a brick and mortar architecture of ceramic tablets (see Figure 1.1 (B)), has the largest value of $R(0)^{(\text{arch})}/R(0)^{(\text{con})}$, but the smallest $\langle R \rangle^{(\text{arch})}/R(0)^{(\text{con})}$ (apart from the *E. aspergillum* spicules). On the other hand, the conch shell is known for its complex and hierarchical architecture consisting of multiple layers each of which contain criss-crossed plies of much smaller lamellae (see Figure 1.1 (C)). I find that in this case, the complex, hierarchical architecture corresponds to the largest $\langle R \rangle^{(\text{arch})}/R(0)^{(\text{con})}$, but the smallest $R(0)^{(\text{arch})}/R(0)^{(\text{con})}$ (apart from the *E. aspergillum* spicules). Both bone and antler fall somewhere between nacre and conch both in terms of architectural complexity and values of $R(0)^{(\text{arch})}/R(0)^{(\text{con})}$ and $\langle R \rangle^{(\text{arch})}/R(0)^{(\text{con})}$. The primary difference between these architectures is the number of levels of architectural hierarchy (i.e., whether they contain layers within layers). The role of architectural hierarchy in enhancing toughness in SBMs has been frequently debated [104–106]. I find that hierarchy, or more generally architectural complexity, gives rise to higher overall toughness (see Figure 2.5 (B)). However, it seems that among similar lamellar architectures (e.g., nacre, bone, antler and conch) simple heterogeneity is better in terms of improving fracture initiation toughness (see Figure 2.5 (A)). Thus, both hierarchy *and* heterogeneity play two distinct roles in affecting toughness properties of SBMs.

Table 2.3: *E. aspergillum* spicule fracture toughness data

Sample ID	Sample dimensions						F and w_0 measurements				Toughness	
	L (μm)	D (μm)	a_0 (μm)	r_n (nm)	α_0	δ	F_c (mN)	w_f (μm)	F_f (mN)	k_f (N/m)	$R(0)$ (J/m ²)	$\langle R \rangle$ (J/m ²)
Ea1	604.8	50.75	16.13	25	0.32	0.084	13.37	36.5	44.95	1194.5	2.89	208.49
Ea2	813.0	37.44	10.83	84	0.29	0.046	4.54	34.7	8.23	230.8	2.65	66.96
Ea3	814.6	36.57	9.19	251	0.25	0.045	24.02	40.1	9.04	242.5	—	—
Ea4	820.5	23.46	4.31	24	0.18	0.029	1.23	34.0	1.26	35.6	6.45	—
Ea5	813.0	25.14	10.13	36	0.40	0.031	1.07	21.4	0.97	43.7	4.62	20.98
Ea6	814.6	21.60	13.81	77	0.64	0.027	0.54	16.3	0.49	29.7	14.70	16.08
Ea7	820.5	47.07	19.05	300	0.40	0.057	8.26	31.2	23.37	740.5		113.10
Ea8	808.8	46.50	14.92	120	0.32	0.057	7.60	33.3	21.07	626.2	2.06	86.89
Ea9	808.8	27.74	7.60	229	0.27	0.034	9.81	39.9	3.44	87.9	—	—
Ea10	814.6	24.51	10.49	46	0.43	0.030	0.99	12.5	0.65	48.8	4.94	—
Ea11	813.0	32.81	15.82	39	0.48	0.040	1.41	20.1	2.71	130.3	3.51	20.64
Ea12	820.5	31.33	17.09	69	0.55	0.038		21.7	2.47	110.3	—	38.97
Ea13	783.3	22.94	7.22	39	0.31	0.029	0.81	14.9	0.71	43.7	1.26	—
Ea14	814.6	45.65	22.29	45	0.49	0.056	6.51	40.1	15.12	376.4	14.03	118.77
Ea15	813.0	43.58	22.58	75	0.52	0.054	7.53	33.9	15.33	445.3	22.05	114.61
Ea16	814.6	45.51	21.62	84	0.47	0.056	6.67	31.7	14.00	435.5	11.42	99.85
Ea17	775.2	51.97	14.86	90	0.29	0.067	4.89	47.6	36.49	756.0	0.51	113.76
Ea18	783.3	57.76	18.15	178	0.31	0.074	7.33	47.1	49.30	1032.0	0.75	162.19
Ea19	790.5	62.61	18.55	84	0.30	0.079	12.01	49.8	62.13	1233.7	1.35	218.73
Ea20	820.5	64.40	23.36	89	0.36	0.078	14.19	48.3	80.08	1635.5	2.18	250.31
Ea21	813.0	67.03	20.80	81	0.31	0.082	19.64	57.0	104.29	1807.7	2.27	306.94
Ea22	814.6	70.42	24.44	44	0.35	0.086	20.28	54.9	113.74	2046.9	2.56	322.76
Ea23	779.6	43.16	17.26	52	0.40	0.055	2.54	22.9	11.93	511.2	0.74	—
Ea24	775.2	44.36	14.34	73	0.32	0.057	4.30	26.7	17.92	660.7	0.70	63.91
Ea25	813.0	50.83	19.82	118	0.39	0.063	7.58	63.2	41.44	648.7	3.35	175.80
Ea26	814.6	53.12	14.98	57	0.28	0.065	8.95	42.8	32.73	753.3	1.65	105.92
Ea27	775.2	57.24	32.40	112	0.57	0.074	10.06	34.2	44.25	1275.1	11.06	200.91

Table 2.4: *T. aurantia* spicule fracture toughness data

Sample ID	Sample dimensions						F and w_0 measurements				Toughness	
	L (μm)	D (μm)	a_0 (μm)	r_n (nm)	α_0	δ	F_c (mN)	w_f (μm)	F_f (mN)	k_f (N/m)	$R(0)$ (J/m ²)	$\langle R \rangle$ (J/m ²)
Ta1	790.5	30.34	12.71	129	0.42	0.038	0.60	19.1	0.54	27.7	1.92	—
Ta2	601.3	43.61	11.17	63	0.26	0.073	8.66	32.5	19.90	601.9	3.22	—
Ta3	597.2	32.73	13.68	130	0.42	0.055	3.18	16.3	5.85	340.3	3.68	—
Ta4	603.8	31.14	11.83	37	0.38	0.052	1.50	11.3	1.49	119.7	1.69	—
Ta5	601.3	36.17	15.92	50	0.44	0.060	4.26	25.7	8.47	325.7	7.13	—
Ta6	604.8	40.90	13.58	40	0.33	0.068	5.83	27.6	14.08	492.3	2.21	—
Ta7	597.2	34.69	8.29	35	0.24	0.058	3.46	27.3	9.35	332.7	1.61	—
Ta8	597.2	33.98	10.07	125	0.30	0.057	3.14	14.6	6.59	425.9	0.86	—
Ta9	604.8	24.05	8.69	79	0.36	0.040	0.86	13.4	1.45	102.5	0.87	—
Ta10	601.3	27.84	10.82	87	0.39	0.046	1.49	8.2	1.32	152.7	1.80	—
Ta11	603.8	34.29	14.23	65	0.41	0.057	3.05	12.9	7.29	533.7	1.91	—
Ta12	603.8	31.13	10.67	28	0.34	0.052	1.70	17.7	4.65	249.6	0.69	—
Ta13	604.8	38.37	14.47	48	0.38	0.063	5.22	16.0	9.42	578.8	2.72	—
Ta14	597.2	37.37	9.37	82	0.25	0.063	5.10	24.7	10.31	410.1	2.25	—
Ta15	603.8	38.79	18.78	211	0.48	0.064	1.06	2.4	1.07	521.6	—	—
Ta16	604.8	37.56	14.23	127	0.38	0.062	3.83	26.1	12.18	451.7	2.00	—
Ta17	597.2	34.45	7.85	216	0.23	0.058	3.94	18.5	8.01	424.0	—	—

Chapter 3

Measurement of bending failure strains of *E. aspergillum* and *T. aurantia* spicules

Note: A version of this chapter is published in the Journal of the Mechanical Behavior of Biomedical Materials. Data and figures have been used with all co-authors' consent.

Monn MA and Kesari H. Enhanced bending failure strain in biological glass fibers due to internal lamellar architecture. Journal of the Mechanical Behavior of Biomedical Materials (2017).

3.1 Introduction

I investigate the hypothesis that the *E. aspergillum* spicule's architecture enhances its bending failure strain, which allows it to provide a stronger attachment to the sea floor. My hypothesis is motivated by several observations and deductions:

- i. *The sponge obtains nutrients by filtering microorganisms from sea water. While the sponge pumps water through its body, the flow of water is facilitated by ocean currents.* I reason that in order to pump and filter water, the sponge must be robustly attached to the sea floor.

- ii. *The distal ends of the E. aspergillum anchor spicules are covered in barbs (see Figure 1.2 (D)).* I believe that the orientation of these barbs implies that the spicule's primary mechanical function is to anchor the sponge to the sea floor.
- iii. *Sponges have the ability to make spicules in a large variety of shapes [15, 107].* The presence of the barbs, therefore, suggests that there is evolutionary pressure on the spicules to enhance their anchoring ability.
- iv. *It has been shown that the force required to pull a fiber out of an elastic matrix increases with the curvature of the fiber inside the matrix [108, 109].* Therefore, I deduce that spicules will be better anchors if they are able to withstand larger bending strains (and therefore larger bending curvatures).

I test my hypothesis by performing three-point bending tests on *E. aspergillum* spicules and measuring the bending strains at which they fail. In order to quantify the effect of the architecture on the strain at which *E. aspergillum* spicules fail, I once again compare them to spicules from *T. aurantia*.

I performed three-point bending tests on 33 *E. aspergillum* and 24 *T. aurantia* spicules using a custom-built mechanical testing device (see Figure 3.2 (A), (B)). The detailed descriptions of my mechanical testing device and test procedure are given in Section 3.2.2. Briefly, sections of *E. aspergillum* spicules and *T. aurantia* spicules were suspended across a trench. A motorized translation stage was used to push each spicule against a aluminum wedge that was positioned midway across the trench—at mid span (see Figure 3.2 (B), (D)). The force applied to the spicule by the wedge and the lateral deflection of the spicule's cross-section that is in contact with the wedge were measured (see Section 3.2.2 and Figure 3.3 (E)). I also imaged the spicules during the test using a reflected light microscope and used these images to compute the strain at which each

spicule failed (see Figure 3.3 (B), (D)).

I define a spicule's effective bending strain to be the strain in the outermost material fiber of a homogeneous beam with the same curvature and cross-sectional shape as the spicule (see Section 3.3). The spicule's "bending failure strain" is the maximum effective bending strain along the spicule's length before it fails. I use the following procedure to compute each spicule's bending failure strain. I select points along each spicule's longitudinal mid-plane in the micrograph taken just before it failed (see Figure 3.4 (C), (D)). I fit a polynomial function to these points and computed the curvature of the longitudinal mid-plane using this function (see Section 3.3 and Figure 3.4 (D), (E)). After each test, I measured the radius of the cross-section at which the spicule failed from a scanning electron micrograph (see Section 3.2.3 and Figure 3.5 (A)). Finally, I computed each spicule's bending failure strain from the the maximum curvature of the polynomial function and the spicule's cross-sectional radius at the location of failure using elastica theory (see Section 3.3 and Figure 3.5 (C)) [110].

By comparing the *E. aspergillum* spicules to the *T. aurantia* spicules, I found that the *E. aspergillum* spicule's cylindrically layered architecture increases its bending failure strain by roughly 140%. This supports my hypothesis that the *E. aspergillum* spicules' architecture allows them to bend more before failing, thereby allowing them to provide a better anchorage to the sea floor.

3.2 Materials and methods

3.2.1 Three-point bending test specimen preparation

A spicule that passed the inspection procedure described in Chapter 2 Section 2.3.1 was placed across a trench cut in a stainless steel plate (see Figure 3.2 (B)). The trench has parallel, vertical walls and the trench edges support the spicules during the three-point bending tests. The trench's

span, L , was measured from optical micrographs to be $1278 \pm 3 \mu\text{m}$ (mean $\pm$ standard deviation; $N = 10$). I ensured that each spicule's longitudinal mid-plane was perpendicular to the trench edges. Since the *T. aurantia* spicules are tapered, I took efforts to also ensure that the midpoint along each *T. aurantia* spicule's length was coincident with the trench's mid span.

3.2.2 Construction and operation of the mechanical testing device

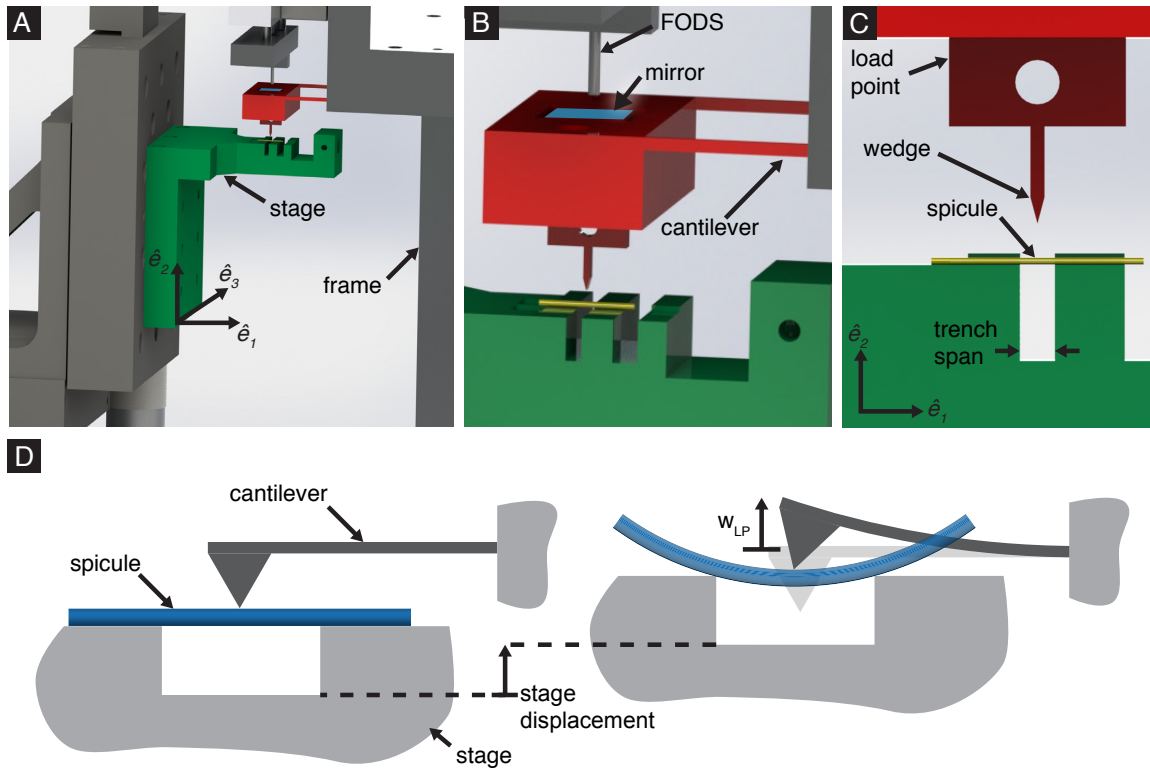


Figure 3.1: A computer aided design rendering of the mechanical testing device. (A) The stage components are highlighted in green. The force sensing subassembly (cantilever, load point) is highlighted in red. (B) A magnified view of (A). The mirror attached to the load point is shown in blue on the top surface of the load point beneath the FODS. (C) A specimen is placed across a trench cut in the stage. The wedge tip of the load point is positioned mid way across the trench span. (D) A schematic of the three-point bending configuration showing the deformation of the spicule and the cantilever to which the load point is attached.

My mechanical testing device consists of two major components: the sample stage that holds the spicules (see Figures 3.1 (A) and 3.2 (A)), and a wedge-shaped load point that applies force to the spicules (see Figures 3.1 (B), (C) and 3.2 (A)–(C)).

The sample stage consists of the steel plate containing a trench, which is attached to a three-axis

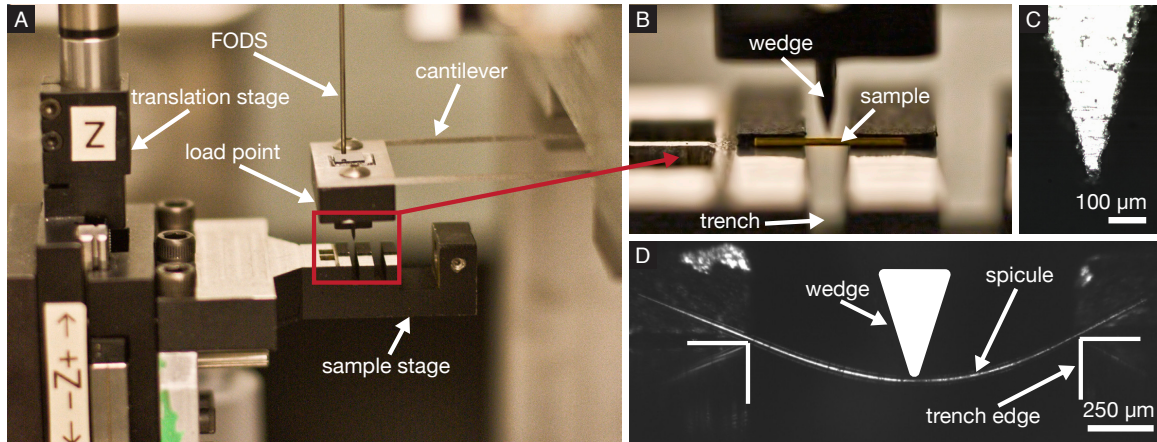


Figure 3.2: Mechanical testing device. (A) The major components of the mechanical testing device are the sample stage and the load point-cantilever assembly. (B) A closer view of the wedge-like tip of the load point and the trench in the sample stage. To aid in visualization of the experimental setup, a $\approx 125 \mu\text{m}$ diameter brass wire (sample) is shown in place of a spicule. (C) A micrograph of the load point tip. (D) A micrograph of a *T. aurantia* spicule just before failure. The trench edges and wedge are marked schematically.

translation stage that is controlled by servo motors. The motors have a minimum repeatable step size of 200 nm. Unlike the fracture tests performed in Chapter 2, the ends of the spicules are not affixed to the trench edges and are therefore free to rotate or slide on the trench edges. The load point's tip is an aluminum wedge that has an included angle of approximately 35 degrees (see Figure 3.1 (D)). The radius of curvature of the apex of this wedge is approximately $20 \mu\text{m}$ (see Figure 3.2 (C)). To perform a bending test, the load point is first centered at the trench's mid span by finding and averaging the positions of the two trench edges. After centering, the spicule is pushed into the load point in $2 \mu\text{m}$ stage displacement increments at an average rate of $1 \mu\text{m/s}$ until the spicule fails.

The load point is attached to the end of an aluminum cantilever, which is used as a force sensor (see Figure 3.1 (A), (B)). The operating principle of the load point-cantilever assembly is similar to that of an atomic force microscope [111]. As a spicule is pushed into the wedge I measure the displacement of the load point, $w_{LP}\hat{e}_2$, using a fiber optic displacement sensor (FODS), where $\hat{e}_2$ is the Cartesian basis vector shown in Figure 3.1 (A), (C). The FODS emits infrared light, which is reflected off of a mirror located on the top surface of the wedge and received by an optical fiber in the FODS (see Figure 3.1 (B)). A $\approx 5 \text{ mm}$ square piece of a polished silicon wafer is used as the

mirror and is affixed to the wedge using epoxy. The FODS measures displacements by comparing the intensities of the emitted and reflected light. After each stage displacement increment, I take 100 w_{LP} measurements and average them to reduce noise caused by mechanical vibrations. Let $w_s \hat{e}_2$ and $-w_0 \hat{e}_2$ be the displacements of the stage and the spicule's cross-section under the wedge, respectively (see Figure 3.1 (D)). Since the wedge and the spicule remain in contact until failure, w_0 is given by the difference between the stage and load point displacements (see Figure 3.1 (D)), or

$$w_0 = w_s - w_{LP}. \quad (3.1)$$

I then use the cantilever's stiffness, k_c , to compute the force applied by the wedge, $-F \hat{e}_2$. The cantilever deflections observed during the bending tests are small enough (roughly 1% of the cantilever's length) that the relationship between F and w_{LP} is linear. Therefore, the force is given by

$$F = k_c w_{LP}. \quad (3.2)$$

I hung calibration weights, whose masses I measured with ± 0.1 mg accuracy, from the end of the cantilever and measured w_{LP} . I fitted Equation (3.2) to this load–displacement data to estimate k_c to be 90.6 ± 0.3 N/m. Cantilever-based force sensors have been used in a number of micro- and macro-scale mechanical testing studies [112–116]. The specific design presented here is adapted from a mechanical testing device used for performing adhesive contact experiments [116]. A similar design has also been used in a commercially available micro-tribometer [117, 118]. My mechanical testing device has a force resolution of roughly $10 \mu\text{N}$. I verified the accuracy of the mechanical testing device by measuring the Young's modulus, E , of a tungsten wire and comparing my measurements to values cited in the literature (see Appendix C).

This mechanical testing device can measure forces ranging from 10^{-5} to 10^1 N and displacements ranging from 10^{-7} to 10^{-2} m. The primary advantage of this mechanical testing device is that the force and displacement capacities can be easily adjusted for different specimens by adjusting the stiffness of the cantilever. Commercially available, large scale, mechanical testing systems typically cannot measure forces and displacements with this resolution. While atomic force microscope-based [112, 119] or microelectromechanical systems-based [113] testing devices have adequate resolution, the maximum force (resp. displacement) they can measure is smaller than the maximum force (resp. displacement) that the spicule specimens can withstand.

I performed three-point bending tests on 33 *E. aspergillum* and 24 *T. aurantia* spicules. The load-displacement data for the *E. aspergillum* and *T. aurantia* spicules are shown in Figure 3.3 (E). Finally, after each stage displacement increment I acquire an image of the spicule's bent shape (see Figure 3.3 (A)–(D)) using a $5\times$ magnification reflected light microscope (Infinitube, AVT Manta). The image acquisition, FODS data acquisition and stage motion are all controlled using National Instruments LabView.

3.2.3 Measurement of spicule diameters

After each bending test, I collected the fragments of the broken spicule. The spicule fragments were handled exclusively using fine point brushes to avoid introducing damage to their fracture surfaces. The fragments were mounted to an aluminum stub using conductive carbon tape, sputter coated with 10 nm of carbon and imaged in a SEM. I measured the diameter of each spicule's cross-section at the location of failure from the SEM images.

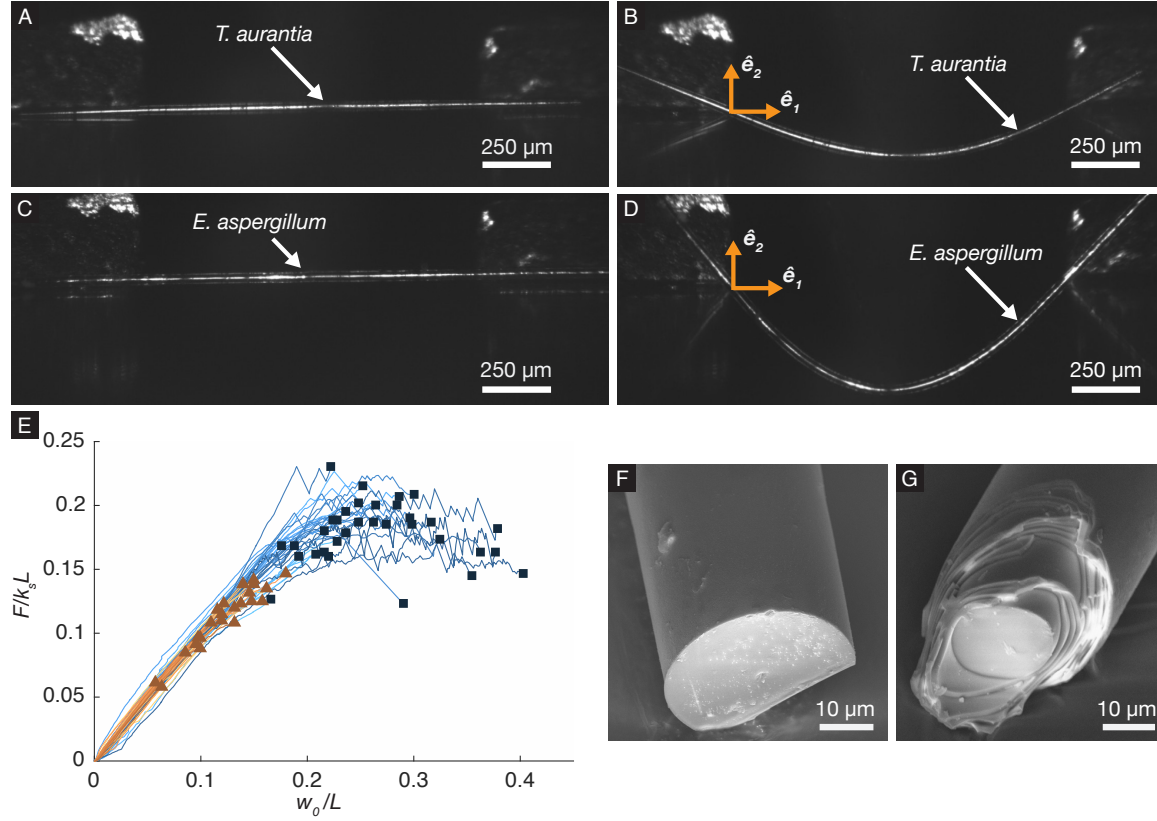


Figure 3.3: Three-point bending test data. (A) and (B) (resp. (C) and (D)) show the undeformed configuration and deformed configuration just before failure of a representative *T. aurantia* (resp. *E. aspergillum*) spicule. The coordinate system used to describe the position of points along a spicules longitudinal mid-plane is shown in (B) and (D). (E) Dimensionless load-deflection data of the 33 *E. aspergillum* (blue) and 24 *T. aurantia* (orange) spicules. The force and deflection at which each *T. aurantia* (resp. *E. aspergillum*) spicule failed is indicated by an orange triangle (resp. dark blue square). (F) (resp. (G)) A representative fractured *T. aurantia* (resp. *E. aspergillum*) spicule.

3.3 Measurement of bending failure strains of *E. aspergillum* and *T. aurantia* spicules

Figure 3.4 (A) and (B) are schematics of the undeformed and deformed configurations of a spicule, respectively, in the context of my bending experiment. The edges of the trench are shown as simple supports. In the deformed configuration, the set $\{\hat{e}_1, \hat{e}_2\}$ is an orthonormal set of Cartesian basis vectors with $\{x_1, x_2\}$ being its corresponding set of Cartesian coordinates. The origin of the coordinate system is located at the point on the spicule's longitudinal mid-plane directly above the left trench edge. I assume that the spicule deforms in the plane whose normal vector is $\hat{e}_1 \times \hat{e}_2$.

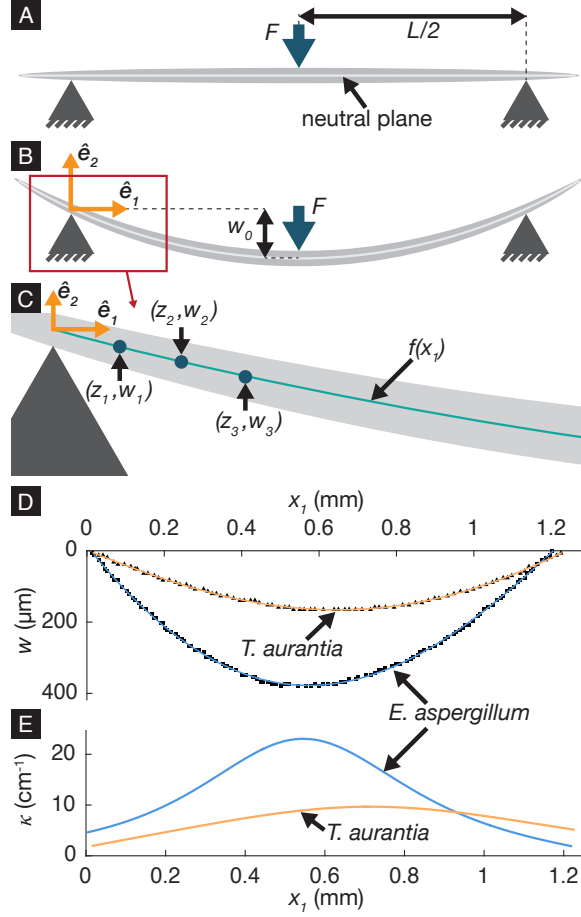


Figure 3.4: Measurement of spicule curvatures from three-point bending test images. (A) Schematic of the spicule's undeformed configuration. (B) Schematic of the spicule's deformed configuration. (C) A magnified view of (B) showing the discrete representation of the spicule's longitudinal mid-plane, $(z_i, w_i)|_{i=1...n}$, and the continuous representation of the longitudinal mid-plane, $f(x_1)$. (D) The black triangles (resp. squares) correspond to the $(z_i, w_i)|_{i=1...n}$ for the representative *T. aurantia* (resp. *E. aspergillum*) spicule shown in Figure 3.3 (B) (resp. (D)). The orange (resp. blue) curves correspond to $f(x_1)$ for the representative *T. aurantia* (resp. *E. aspergillum*) spicule. (E) the curvature, $\kappa(x_1)$, computed from the $f(x_1)$ shown in (C).

I adopted the principal strain failure hypothesis originally proposed by Saint-Venant [120]. That is, I assume that each spicule fails when the maximum principal strain within it reaches a critical value, ϵ_f , which I call its bending failure strain. The infinitesimal strain tensor is $\epsilon = \epsilon_{ij} \hat{e}_i \otimes \hat{e}_j$, where “ $\otimes$ ” denotes the dyadic product and repeated indices imply summation over the integers 1, 2, 3 (Einstein summation convention). I assume that the spicules' deformations satisfy the kinematic hypothesis of elastica theory [110, 121]. That is, I assume that cross-sections in the undeformed configuration (see Figure 3.4 (A)) remain planar in the deformed configuration (see Figure 3.4

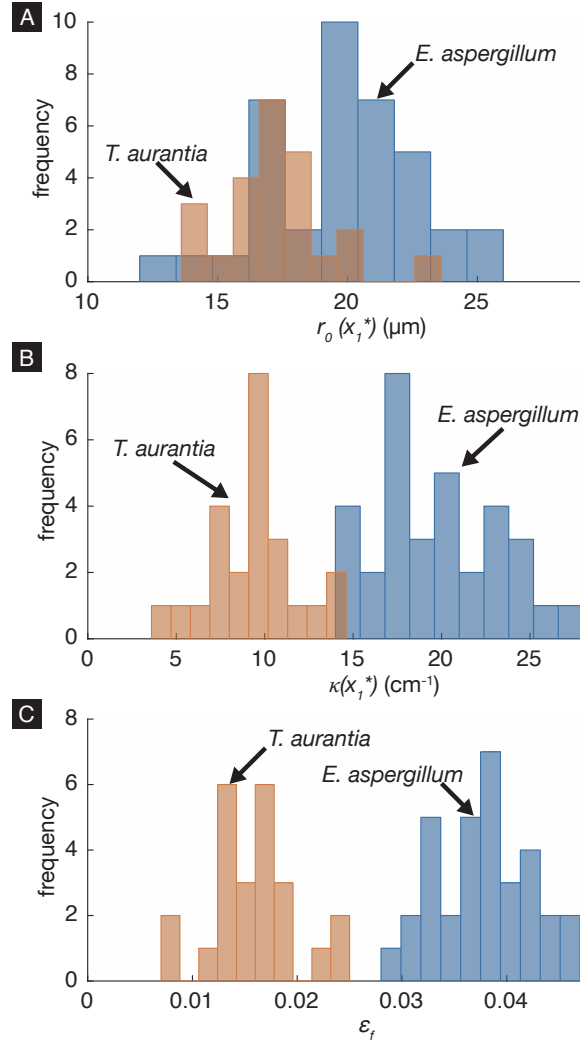


Figure 3.5: Bending failure strains of *E. aspergillum* and *T. aurantia* spicules. (A) A histogram of $r_0(x_1^*)$ for the *E. aspergillum* and *T. aurantia* spicules. (B) A histogram of $\kappa(x_1^*)$ for the *E. aspergillum* and *T. aurantia* spicules. (C) A histogram of the bending failure strains, ϵ_f , for the *E. aspergillum* and *T. aurantia* spicules.

(B)), and that there exists a neutral plane in the structure Based on micrographs of the spicules undeformed configurations (see Figure 3.3 (A), (C)), I assume that in the undeformed configuration, the spicules neutral plane has zero curvature ¹. As a result of this kinematic hypothesis, the only nonzero strain component is $\epsilon_{11} = r\kappa(x_1)$, where $r \in [0, r_0(x_1)]$ is a material point's distance from the neutral plane in the undeformed configuration, $r_0(x_1)$ is the spicule's cross-sectional radius, and

¹The neutral plane is a surface composed of material points whose shape changes as the structure deforms. In the undeformed configuration the neutral plane is normal to the $\hat{e}_2$ direction. Material fibers of infinitesimal length belonging to the neutral plane and oriented in the $\hat{e}_1$ direction in the undeformed configuration do not change length as the structure deforms.

$\kappa(x_1)$ is the curvature of the spicule's neutral plane [110, 121]. Since ε_{11} is the only nonzero strain component, it is also the maximum principal strain. This means that

$$\varepsilon_f = r_0(x_1^*)\kappa(x_1^*), \quad (3.3)$$

where $x_1^* = \operatorname{argmax}\{r_0(x_1)\kappa(x_1) : x_1 \in [0, L]\}$, and $\kappa(x_1)$ belongs to the spicule's deformed configuration just before it fails.

I assign $r_0(x_1^*)$ to be the radius of the cross-section at which the spicule fails. I measured $r_0(x_1^*)$ by collecting and imaging the broken spicule fragments after each bending test (see Section 3.2.3). A histogram of $r_0(x_1^*)$ for the *E. aspergillum* and *T. aurantia* spicules is shown in Figure 3.5 (A). The mean $\pm$ standard deviation of $r_0(x_1^*)$ are $19.7 \pm 2.8 \mu\text{m}$ and $17.2 \pm 2.0 \mu\text{m}$ for the *E. aspergillum* and *T. aurantia* spicules, respectively.

I computed $\kappa(x_1^*)$ using the following procedure. It has been shown that both *E. aspergillum* and *T. aurantia* spicules are axisymmetric [22, 27]. As a consequence of this symmetry, a spicule's neutral plane is the same as its longitudinal mid-plane in its undeformed configuration (see Figure 3.4 (A)). I built a discrete representation of the neutral plane using a set of points $(z_i, w_i)_{i=1\dots n}$, which I manually selected from the micrograph of the spicule's deformed configuration just before failure (see Figure 3.4 (C), (D)). The abscissae, z_i , were spaced in the $\hat{e}_1$ direction in increments of roughly 15-20 μm . As a result, the total number of points in the discrete representation, n , varied from 57 to 137. For each z_i , I manually chose w_i so that the point (z_i, w_i) coincided with the spicule's neutral plane. I built a continuous representation of the neutral plane by fitting a fourth order polynomial, $f(x_1) = \sum_{j=0}^4 a_j x_1^j$, to the (z_i, w_i) data, and computed the curvature of the spicule's neutral plane as $\kappa(x_1) = f''(x_1)/(1 + f'(x_1)^2)^{3/2}$ (see Figure 3.4 (D), (E) and Figure 3.5 (B)). To obtain $\kappa(x_1^*)$ I assumed that both the strain, $r_0(x_1)\kappa(x_1)$, and the curvature, $\kappa(x_1)$, attain their maximum values at

the same x_1 position. Hence, I approximate $\kappa(x_1^*) = \max\{\kappa(x_1) : x_1 \in [0, L]\}$.

Finally, I used equation (3.3) to compute each spicule's bending failure strain (see Figure 3.5 (C)). The mean $\pm$ standard deviation of ϵ_f for the *E. aspergillum* and *T. aurantia* spicules are 0.0377 ± 0.0043 and 0.0158 ± 0.0042 , respectively.

3.4 Discussion: critique of conventional data reduction and comparison procedures

Euler-Bernoulli beam theory, which assumes that both the strain and displacements are small, does not predict that the strain would redistribute even when adjacent layers can slide relative to one another. However, as can be seen in my experiments (see Figure 3.3 (D)), the *E. aspergillum* spicules undergo very large displacements before they fail. If I compute the curvature of the *E. aspergillum* spicules using Euler-Bernoulli beam theory, I find that the theory underpredicts the spicules' maximum curvature. Specifically, the maximum curvature of the *E. aspergillum* spicules predicted by Euler-Bernoulli beam theory is

$$\kappa_{EB} = \frac{12F_f}{k_s L^2}, \quad (3.4)$$

where F_f is the applied force at failure, L is the trench's span, and k_s is the slope of the linear portion of the spicule's $F-w_0$ data (see Section 3.2.2) [43]. I compare κ_{EB} to the maximum curvature measured directly from the images of the spicules' deformed configurations (see Section 3.3) for both the *E. aspergillum* and *T. aurantia* spicules in Figure 3.6. From this comparison I see that for small $\kappa(x_1^*)$, the Euler-Bernoulli theory provides a reasonable approximation for the actual curvature of the spicule. However, for most *E. aspergillum* spicules $\kappa(x_1^*)$ is large and the difference between the Euler-Bernoulli theory prediction and the measured curvature increases. Therefore, a new mechanics model of the *E. aspergillum* spicule that accounts for the *E. aspergillum* spicules' architecture

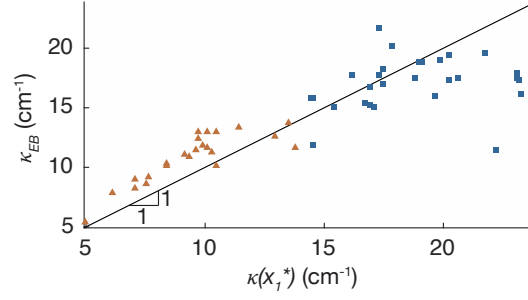


Figure 3.6: Comparison of the measured spicule failure curvatures with Euler-Bernoulli beam theory predictions. The failure curvature predicted by Euler-Bernoulli beam theory, κ_{EB} , is compared to the curvatures measured from optical micrographs using the elastica theory (see Section 3.3 for details). The curvatures, $\kappa(x_1^*)$ and κ_{EB} , of the *E. aspergillum* and *T. aurantia* spicules are shown as blue squares and orange triangles, respectively.

and also considers large displacements is needed to further investigate their bending behavior.

I also computed the Young's modulus of the *E. aspergillum* and *T. aurantia* spicules using the F - w_0 data obtained from the bending tests (see Appendix D). The Young's moduli of the 33 *E. aspergillum* spicules and 24 *T. aurantia* spicules are 28.1 ± 5.4 GPa and 34.4 ± 7.0 GPa (mean $\pm$ standard deviation), respectively. These values are both much smaller than that of synthetic glass or fused silica (SiO_2) [122]. This is not surprising since the spicules' silica is not fully condensed (i.e., it does not consist of only silicon-oxygen double bonds but also contains some functional groups, namely those containing hydrogen [15]), and also contains trace amounts of other elements [15, 123, 124]. The difference between the Young's moduli of the *E. aspergillum* and *T. aurantia* spicules (≈ 6 GPa) is much smaller than the difference between the Young's moduli of *E. aspergillum* spicules and fused silica (≈ 44 GPa) [122]. The similarity between the Young's moduli of the two types of spicules serves as additional justification for my assumption that the intrinsic mechanical properties of the *T. aurantia* spicules are the same as those of the *E. aspergillum* spicules. However, bias-corrected accelerated confidence intervals using 10^4 bootstrapped samples indicate that there is a difference in the mean Young's modulus of the two species (upper: 7.5 GPa; lower: 5.5 GPa). An unpaired t -test for independent samples ($t(41) = 3.7$, $p = 0.001$; equal variances not assumed) also suggested a significant difference between the Young's moduli of the two species. This difference

indicates that while *T. aurantia* spicules are clearly a better control material than fused silica, their intrinsic mechanical properties are not identical to those of *E. aspergillum* spicules. As a consequence of this difference I believe that one way to provide a better quantification of the toughness and bending failure strain enhancements imparted by the architecture would be to compare the *E. aspergillum* spicules to a section of their own monolithic silica core. Doing so would guarantee that the intrinsic material properties of the silica in both cases were identical.

Chapter 4

Connections between shape and buckling strength in *T. aurantia* spicules

Note: A version of this chapter is published in Scientific Reports. Data and figures have been used with all co-authors' consent.

Monn MA and Kesari H. A new structure-property connection in the skeletal elements of the marine sponge Tethya aurantia that guards against buckling instability. Scientific Reports (2017).

4.1 Introduction

Buckling is the phenomenon in which a slender, structural element that is subjected to an increasing axial compressive force abruptly starts to deform laterally when the force's magnitude reaches a critical value. This instability dramatically reduces the structure's ability to provide stiffness and structural support, and in many cases can lead to catastrophic failure.

There has always been a need for buckling-resistant designs at the large-scale, e.g., in lightweight aerospace and civil engineering structures [125, 126]. Recently, however, understanding and

controlling buckling has also become important at the small-scale as well. A number of stretchable electronics platforms being developed are based on the design of micro-scale structures whose buckling instabilities can be precisely controlled [127–129]. Bio-medical instruments, such as needles, catheter guidewires, and stents depend on buckling resistance in order to effectively penetrate tissue or be inserted through narrow ducts or capillaries [130–132]. Stents must also provide reliable and long-term mechanical support to the surrounding tissue [131, 132].

I identify a new connection between the mechanical design and buckling resistance in the spicules of the marine sponge *Tethya aurantia*. *T. aurantia* is a sessile animal that grows on rocky surfaces in the Mediterranean [133]. While *T. aurantia* produces several different types of spicules, I focus on the needle-shaped strongyloxea spicules (see Figure 4.1). The strongyloxea are monolithic, axially symmetric, silica rods (see Figure 4.4 (b)). They are roughly 35 μm thick, 2 mm long, and are tapered along their length (see Figure 4.1 (a)). I found that their tapered shape is remarkably uniform across different strongyloxea (see Section 4.3.1 and Figure 4.1 (d)). Considering that sponges have a great degree of control over the shape of their spicules, it is natural to wonder whether this tapered shape has some functional significance.

I introduce and investigate the hypothesis that the strongyloxea's taper is an adaptation aimed at enhancing their ability to provide stiffness to the sponge. My hypothesis is motivated by the following observations.

- a. *Mechanical stiffness is important for the sponge.* *T. aurantia* is primarily found in shallow, coastal environments, where it is subjected to forces exerted by underwater waves and currents [44, 133, 134]. It feeds by filtering microscopic organic particles and microorganisms from seawater. Large deformations of the sponge's body caused by ambient loads could inhibit its ability to feed. Therefore, it is critical that the sponge's body be stiff enough to limit

any such large deformations.

- b. *The sponge derives its stiffness primarily from the strongyloxea.* The strongyloxea are distributed within the sponge's spherical body and are embedded in a collagenous matrix, called spongin (see Figure 4.6 (a)) [44, 135]. Spongin is very compliant, having a Young's modulus of only 600 KPa [136]. The strongyloxea on the other hand are composed of silica, and have a Young's modulus of ≈ 34 GPa (see Appendix D). The strongyloxea also lack any internal structure (see Figure 4.4 (b)) that would imply that they perform functions other than to provide mechanical support to the sponge. Finally, a closely related sponge—*Tethya citrina*—that grows in calmer waters is more compliant and produces fewer spicules per body volume [44]. This is consistent with mechanical tests performed on spicule containing tissues, which show that the spicules drastically increase the tissue's stiffness [137].
- c. *The strongyloxea's ability to provide stiffness is limited by their resistance to buckling.*
- d. *The buckling resistance of a slender structure can be increased by tapering it.* The destabilizing bending moments arising from the eccentricity of a structure's axial compressive loads are more intense at the structure's center than at its ends. Hence, its buckling resistance can be enhanced by moving material away from its ends, towards its center. This result has been established both theoretically [138] and experimentally [139]. I elaborate on this result further in Section 4.3.5.

I test my hypothesis as follows. Based on mechanical testing (see Section 4.3.2) and sponge-anatomy informed computational mechanics calculations (see Section 4.3.3 and Appendix F), I construct a structural mechanics model for the strongyloxea (see Section 4.3.4). Using my model, I identify the shape of the structure that has the greatest resistance to buckling (see Section 4.3.5). Finally, I measure the strongyloxeas' tapers from SEM images and compare them with the shape of

this optimal structure (see Section 4.3.5). I find that the strongyloxeas' tapers are strikingly similar to the shape of the optimal structure. This similarity suggests that the strongyloxeas' tapered shape enhances their resistance to buckling.

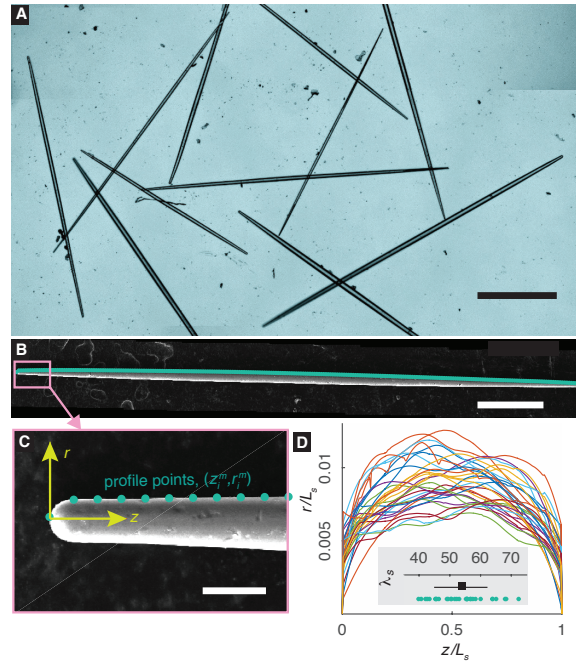


Figure 4.1: Measurement of strongyloxea profiles. (a) A micrograph of several strongyloxea. (b) An SEM image of a single strongyloxea. The strongyloxea's profile is highlighted. (c) A magnified view of (b) showing points composing the profile. (d) Dimensionless profiles of the 31 strongyloxea. The inset shows the distribution of λ_s . The square and error bar indicate the mean and standard deviation of λ_s . The scale bars in (a)–(c) are 500 μm , 250 μm and 25 μm , respectively.

The mechanical tests that I performed on the *T. aurantia* spicules in Chapter 3 are discussed in Section 4.3.2. They show that the strongyloxea behave in a linear elastic fashion until failure. They also show that the strongyloxea's deformation behavior in bending can be modeled exceptionally well using classical structural mechanics theories (see Figure 4.4 (d)). Furthermore, from the strongyloxea's arrangement within the sponge's body it is clear that the strongyloxea's primary function is to stiffen the sponge against radial compressive stresses [44, 135, 137]. I analyze a strongyloxea and a small section of its surrounding spongin matrix using computational mechanics calculations that are consistent with the sponge's skeletal anatomy (see Section 4.3.3 and Appendix F). The results from my computational mechanics calculations show that due to the

difference in the stiffnesses of the strongyloxea and spongin, the spongin matrix transmits the radial compressive stresses to the strongyloxea as highly localized surface tractions on their ends (see Appendix F). Synthesizing the knowledge gained from the mechanical tests, and the computational mechanics simulations, I model the strongyloxea as a simply supported column (see Section 4.3.4).

In the column model, the strongyloxea's stiffening ability is limited by the Euler buckling instability. Thus, the strongyloxea's stiffening ability can be quantified by what I call its buckling strength, which is the maximum axial compressive force that it can transmit without buckling. The shape that would be most consistent with my hypothesis would be the one for which the column model attains its maximum buckling strength. It has been shown using rigorous mathematical techniques that the buckling strength of a simply-supported column can be enhanced by up to 33% over that of a cylinder by tapering it so that its radius as a function of length is described by what I call the Clausen profile [140, 141]. Thus, to test my hypothesis I check how well the strongyloxea's tapered shape is described by the Clausen profile.

I imaged 31 strongyloxea using scanning electron microscopy (SEM) and measured their profiles. In order to interpret how well the measured profiles compare with the Clausen profile, I compare them to not only the Clausen profile but also to other prototypical tapered profiles (see Section 4.3.5). By fitting the profile models to the measured profiles, I find that the Clausen profile describes the strongyloxea's tapered shape the best (see Figure 4.7 (b)).

I do not directly measure the buckling strengths of the spicules. However, I use my measurements of the strongyloxeas' profiles along with my structural mechanics model to estimate the buckling strengths of the strongyloxea (see Section 4.3.6). I compare the estimated buckling strengths of the strongyloxea to the buckling strengths of equivalent cylinders—i.e., cylinders with the same length, volume, and elastic properties (see Figure 4.8). I find that the buckling strengths of the

strongyloxea predicted by my model can be as much as 30% greater than those of their equivalent cylinders. This is close to the 33% enhancement that is achieved by the Clausen profile.

The resemblance of the strongyloxea's profile to the Clausen profile is quite striking and supports my hypothesis. However, this work is only a first step in understanding the functional significance of the strongyloxea's tapered shape. It is possible that the strongyloxeas' tapered shape serves a mechanical function that is different from the one that I have presumed. Or, it is also possible that the taper is simply a consequence of the spicular growth processes, and its resemblance to the Clausen profile is only a misleading coincidence. These possibilities cannot be ruled out without having more information about the sponge's anatomy and ecology. The most direct way to reject my hypothesis would be to show that at least one of my key assumptions is incorrect. These key assumptions pertain to:

- i. the importance of stiffness to the sponge,
- ii. the primary function of the strongyloxea,
- iii. the role of the buckling instability in dictating the strongyloxea's stiffening ability, and
- iv. the effect of the spongin matrix on the strongyloxea's buckling behavior.

4.2 Materials and methods

4.2.1 Procedure for imaging the *T. aurantia* spicules

Strongyloxea from *T. aurantia* sponges were received dried and separated from the surrounding spongin. The strongyloxea were first examined using a polarized light microscope (Nikon Ci Pol). Intact, undamaged strongyloxea were mounted to aluminum stubs using conductive carbon tape. The mounted strongyloxea were sputter coated with approximately 10 nm of carbon and then imaged with a scanning electron microscope (FEI Helios, or LEO 1530 VP) at roughly 500 $\times$ magnification. At this magnification, the field of view was roughly 250 μm $\times$ 200 μm in the FEI Helios (130

$\mu\text{m} \times 90 \mu\text{m}$ in the LEO 1530 VP). Therefore, a complete image of a strongyloxea consisted of 7–14 overlapping frames. These frames were aligned and stitched together to make a single composite image using a Fourier transform-based phase correlation method implemented in ImageJ [142]. A representative composite image is shown in Figure 4.2.

4.2.2 Extraction of spicule boundary geometry from SEM images

Each composite image was first converted to a binary image in which the strongyloxea and background are made up of white and black pixels, respectively. Points on the boundary of the strongyloxea were identified using the Moore-Neighbor tracing algorithm implemented in MATLAB's Image Processing Toolbox [143] (see Figure 4.2). There were roughly 15,000 boundary points obtained for each strongyloxea. A line was fit to these points to determine the orientation of each strongyloxea's axis. I used this line as the axial— z —direction in the (z, r) coordinate system shown in Figure 4.1 (c) and Figure 4.2. The locations of the boundary points were translated so that the point with the smallest z coordinate was located at the origin. Finally, the locations of the boundary points were converted from pixels to micrometers using a scale bar taken from the first frame of each composite image.

4.2.3 Image analysis and post-processing: denoising and subsampling

I divided a strongyloxea's boundary points into 50 partitions so that the z coordinates of the points in the j^{th} partition satisfy $(j-1)L_s/50 \leq z \leq jL_s/50$, where $j = 1, \dots, 50$ and L_s is the maximum z value of the boundary points. The average z and r coordinates of the points in each partition were interpolated along with the end points, $(0, 0)$ and $(L_s, 0)$, to generate the strongyloxea's midline (see Figure 4.2). I used the midline to divide the boundary points into two halves. Boundary points whose r coordinates were greater (resp. less) than those of the midline at the same z value constitute

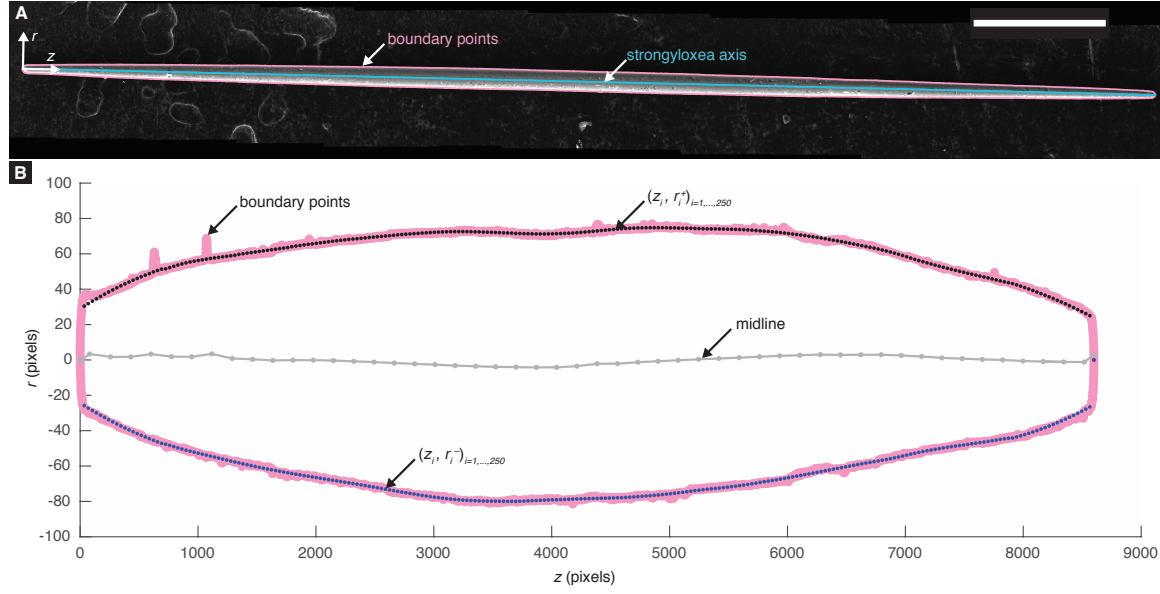


Figure 4.2: Strongyloxea profile extracted from an SEM image. (a) A SEM image of a strongyloxea. The boundary points are shown in pink, and the strongyloxea's axis is shown in blue. The scale bar measures $250 \mu\text{m}$. (b) The boundary points from (a) are shown in pink. These points are divided into two halves by the midline (gray), denoised by Savitsky-Golay filtering, and sampled to get the two sets of points $(z_i, r_i^+)_{i=1,...,250}$ and $(z_i, r_i^-)_{i=1,...,250}$ shown in black and blue, respectively.

the upper (resp. lower) half-boundary. Each half-boundary was denoised using a Savitzky-Golay filter with a kernel size of approximately $1/12^{th}$ the total number of boundary points. The two sets, $(z_i, r_i^+)_{i=1,...,250}$ and $(z_i, r_i^-)_{i=1,...,250}$, were obtained by sampling the denoised upper and lower half-boundaries, respectively. The z values of these points were selected such that $z_1 = 0$, $z_{250} = L_s$ and $z_{i+1} - z_i$, $i = 1, \dots, 249$ is a constant—i.e., the points are equally spaced in the z direction. The sets $(z_i, r_i^+)_{i=1,...,250}$ and $(z_i, r_i^-)_{i=1,...,250}$ constitute my model for a strongyloxea's boundary and are used in Section 4.2.4 for quantifying the strongyloxea's symmetries. After quantifying a strongyloxea's symmetries, the $(z_i, r_i^-)_{i=1,...,250}$ set is discarded and the $(z_i, r_i^+)_{i=1,...,250}$ set is used in the calculations and analysis in Sections 4.3.1, 4.3.5, and 4.3.6. In those sections I refer to the set $(z_i, r_i^+)_{i=1,...,250}$ as a strongyloxea's profile and denote it as $(z_i^m, r_i^m)_{i=1,...,250}$.

4.2.4 Quantification of a spicule's axial and lateral symmetries

The majority of the strongyloxea I observed were straight, radially symmetric about their axis and symmetric across their lateral plane (see Figure 4.3 (a)). For this reason I idealized the strongyloxea as straight, tapered columns with circular cross-sections in my structural mechanics model (see Section 4.3.4).

However, approximately 34% of the 47 strongyloxea I imaged did not share the highly symmetric characteristics of the majority. For example, the axes of some strongyloxea were curved (e.g., see Figure 4.3 (d)). These asymmetric strongyloxea could be accidental deviations from the strongyloxea's body plan, or could support functions different from the stiffening function that I consider in this chapter. Therefore, I chose not to compare the asymmetric strongyloxea with the profiles in Section 4.3.5. I characterized a strongyloxea as being asymmetric or not using the following procedure.

If a strongyloxea is straight and radially symmetric about its axis, then it also possesses a mirror symmetry across its transverse plane (see Figure 4.3 (a)). The metric M_B , defined in equation (4.1), gives a measure of the strongyloxea's mirror symmetry across the transverse plane.

$$M_B(r_i^+, r_i^-) := 1 - \sqrt{\frac{\sum_{i=1}^{250} [|r_i^+| - |r_i^-|]^2}{\sum_{i=1}^{250} [(r_i^+)^2 + (r_i^-)^2]}}. \quad (4.1)$$

Similarly, I define a metric M_A in equation (4.2) that provides a measure of the strongyloxea's mirror symmetry across its lateral plane.

$$M_A(r_i^+, r_i^-) := 1 - \sqrt{\frac{\sum_{i=1}^{125} [(r_i^+ - r_{251-i}^+)^2 + (r_i^- - r_{251-i}^-)^2]}{\sum_{i=1}^{250} [(r_i^+)^2 + (r_i^-)^2]}}. \quad (4.2)$$

The M_A and M_B values lie between 0 and 1. When M_A (resp. M_B) equals unity then there is

perfect mirror symmetry across the lateral (resp. transverse) plane. Several examples of shapes with different M_A and M_B values are shown in Figure 4.3 (b) and the M_A and M_B for the 47 strongyloxea are shown in Figure 4.3 (c). If a strongyloxea's M_A and M_B are both above certain values then I consider it to be symmetric, and consequently measure it and compare it to the profiles in Section 4.3.5. Otherwise, I categorize it as asymmetric and ignore it in all further analysis. I chose the cutoff values for M_A and M_B to be 0.85 and 0.84, respectively. Using this procedure I categorized 16 of the 47 strongyloxea that I imaged as being asymmetric.

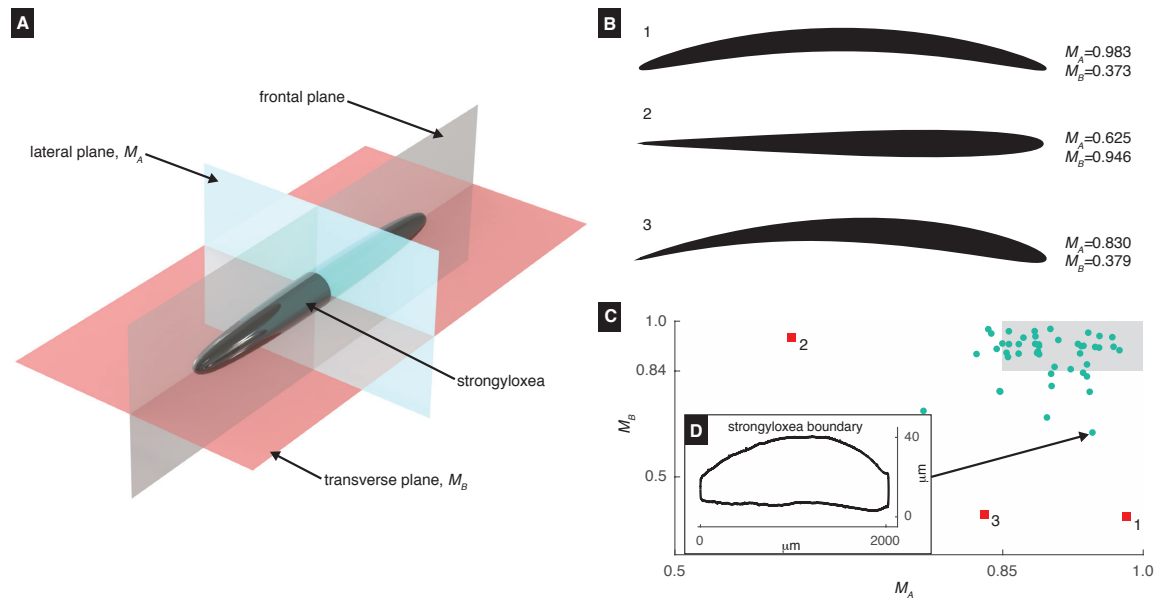


Figure 4.3: Axial and lateral symmetries of a strongyloxea. (a) Anatomical planes of a strongyloxea. Taking the frontal plane to be parallel to the imaging plane, I quantify a strongyloxea's symmetries across the transverse and lateral planes using the metrics M_B and M_A , respectively. (b) Three synthetically generated shapes with different M_A and M_B values. (c) the values of M_A and M_B for the 47 strongyloxea imaged, along with values from the three shapes in (b). The 31 strongyloxea whose (M_A, M_B) values lie inside the shaded region were used for measurement and comparison to the candidate profiles. (d) The boundary of a strongyloxea whose M_A and M_B fall outside of the cutoff values and is categorized as asymmetric.

4.3 Results

4.3.1 Measurement of *T. aurantia* spicule profile shapes

I extracted the shape of 31 strongyloxea from SEM images (see 4.2). Since the strongyloxea are axisymmetric, I describe a strongyloxea's shape using its "profile", which is a set of points (z_i^m, r_i^m) , $i = 1 \dots 250$ shown in Figure 4.1 (b)–(c). I measured the length, L_s , and maximum cross-sectional radius, R_s , of each strongyloxea from its profile. I define a strongyloxea's length $L_s = \max_i z_i^m$ and its maximum radius $R_s = \max_i r_i^m$, where $i = 1, \dots, 250$. The mean values of R_s and L_s are $18.3 \mu\text{m}$ and 1.92 mm with standard deviations of $3.0 \mu\text{m}$ and 0.24 mm , respectively.

By plotting the dimensionless profiles, $(z_i^m/L_s, r_i^m/L_s)$, for $i = 1 \dots 250$, I see that the general nature of the taper appears uniform across different strongyloxea. To make a more quantitative comparison of the strongyloxeas' tapers I compute the aspect ratio, $\lambda_s = L_s/2R_s$, for each strongyloxea. The values of λ_s are plotted in Figure 4.1 (d). The mean and standard deviation of λ_s are 53.6 and 8.7, respectively. The small scatter of λ_s further supports my viewpoint that the tapered shape is uniform across different strongyloxea.

4.3.2 Mechanical testing of *T. aurantia* spicules and comparison to Euler-Bernoulli beam theory predictions

While *T. aurantia*'s strongyloxea do not contain separate layers of protein and silica like the *E. aspergillum* spicules discussed in Chapters 2 and 3, the influence of any underlying protein scaffold on their elastic behavior is unknown. Furthermore, the composition of the silica itself varies spatially within the strongyloxea [144]. To ascertain the effect of any potential elastic inhomogeneities on a strongyloxea's deformation behavior, I performed three-point bending tests on 30 strongyloxea (see Chapter 3 for details of the test configuration).

The magnitude of the transverse force, F , and deflection of the strongyloxea's axis in the y -direction at midspan, w_0 (see Figure 4.4 (c)), were recorded until the strongyloxea failed. The w_0 – F data for the strongyloxea are shown in Figure 4.4 (a). The failure of every strongyloxea I tested was defined by a single fracture event. The fracture events are marked with red points in Figure 4.4 (a). The w_0 – F response of every strongyloxea was linear until failure. This observation indicates that the strongyloxea's mechanical behavior is linear elastic until failure.

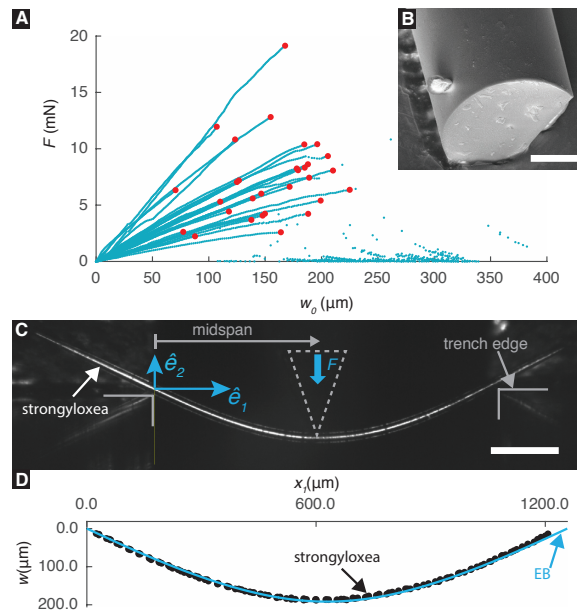


Figure 4.4: Three-point bending tests of strongyloxea. (a) Applied force, F , versus displacement at midspan, w_0 , for 30 strongyloxea. Red points indicate the load and displacement at which each strongyloxea failed. (b) A cross-section of a fractured strongyloxea. (c) micrograph of a bent strongyloxea just prior to failure. The indenter used to apply the force is outlined with dashed lines. (d) Points along the strongyloxea's axis are obtained from (e). The blue curve labeled EB is the deformed shape predicted by Euler-Bernoulli beam theory. The scale bars in (b) and (c) are $10\ \mu\text{m}$ and $250\ \mu\text{m}$, respectively.

I compared a strongyloxea's deformed shape during a bending test to that predicted by Euler-Bernoulli theory for an elastically homogeneous, tapered beam [43]. Consider a beam suspended across a trench of span L . The beam is simply supported at the trench edges and subjected a point force of magnitude F acting perpendicular to its axis at midspan—i.e., at $x_1 = L/2$ (see Figure 4.5). The deformed shape of the beam is described in terms of the transverse deflection of its axis, $w(x_1)$. I denote w at midspan as w_0 . The beam behaves in a linear elastic fashion, and its elastic modulus

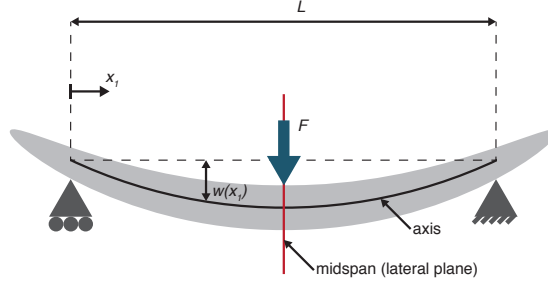


Figure 4.5: Bending of a beam with variable cross-section. A beam in a three-point bending configuration subjected to a transverse force of magnitude F at midspan. The beam's axis is indicated by a black line.

is constant along its length. Because the beam's properties are linear elastic, its w_0 – F response is linear and has a slope k_s .

The beam's second moment of area, I , can vary along its length, but I assume that the variation is symmetric across the midspan. Consequently, w will also be symmetric across the midspan. This allows us to consider only the half of the beam for which $0 \leq x_1 \leq L/2$ for calculating w . From Euler-Bernoulli beam theory, w is governed by the ordinary differential equation

$$\frac{d^2 w}{d\xi^2} = \frac{-24F}{k_s} \frac{\xi}{\eta(\xi)}, \quad (4.3)$$

where $\xi = x_1/L \in (0, 1/2)$ is the dimensionless coordinate in the $\hat{e}_1$ direction and $\eta(\xi) = I(L\xi)/I(L/2)$.

The boundary conditions for the half-beam are

$$w(\xi)|_{\xi=0} = 0, \quad (4.4)$$

$$\frac{dw(\xi)}{d\xi}|_{\xi=1/2} = 0, \quad (4.5)$$

where equation (4.5) comes as a consequence of the symmetry of w across the midspan.

To compute w , I take L to be 1.278 mm, which is the distance between trench edges in my flexural testing device (see Chapter 3 Section 3.2.1–3.2.2). For each strongyloxea, I obtained k_s by fitting a line to the w_0 – F data. Since the micrographs taken during the bending tests are low magnification,

I could not obtain detailed enough information from them to compute $\eta(\xi)$. Instead, I computed $\eta(\xi)$ using the profile of a randomly chosen strongyloxea that I measured in Section 4.3.1. I used the same $\eta(\xi)$ for each strongyloxea that I tested. For each strongyloxea I numerically computed w from equation (4.3) subject to equations (4.4)–(4.5). I found that the w I computed from equation (4.3) was relatively insensitive to which profile from Section 4.3.1 I used.

I then measured the displacement of a strongyloxea's axis in the y -direction from images taken during the bending test (see Figure 4.4 (c)). A representative comparison of these measured displacements with those predicted by Euler-Bernoulli beam theory is shown in Figure 4.4 (d). The measurements and the theoretical predictions match very well for 27 of the 30 strongyloxea. This supports that the strongyloxea's behavior is linear elastic and shows that a strongyloxea is elastically homogeneous along its length. Furthermore, it shows that a strongyloxea's deformation can be described by an Euler-Bernoulli beam theory for an elastically homogeneous, tapered, axially symmetric beam.

4.3.3 Computational mechanics model of a single *T. aurantia* spicule

Being embedded within the sponge, a strongyloxea likely experiences a complex distribution of tractions along its length. However, using computational mechanics calculations I found that due to the strongyloxeas' arrangement within the sponge and the large mismatch between the compliance of the strongyloxea and the spongin, the tractions are localized at the ends of the strongyloxea (see Appendix F). Thus, the most appropriate structural mechanics model based on Euler-Bernoulli beam theory would be a simply supported column, which is described by Equations (4.6)–(4.8).

The strongyloxea are not uniformly scattered throughout the sponge's body, rather they are grouped in bundles that extend radially from the sponge's center to its outer surface (see Figure 4.6 (a)) [44]. The strongyloxea are aligned along the bundles' lengths and are staggered with respect

to each other (see Figure 4.6 (b)). The bundles are 220–490 μm thick [44] and a bundle's cross-section contains approximately 50 strongyloxea [135]. From the average bundle thickness, number of strongyloxea per bundle, and strongyloxea diameter, I estimate the distance between the axes of neighboring strongyloxea to be $\approx 45 \mu\text{m}$ (see Appendix E). Thus, the strongyloxea within a bundle are separated from each other by a very small amount, $\approx 8 \mu\text{m}$, of spongin.

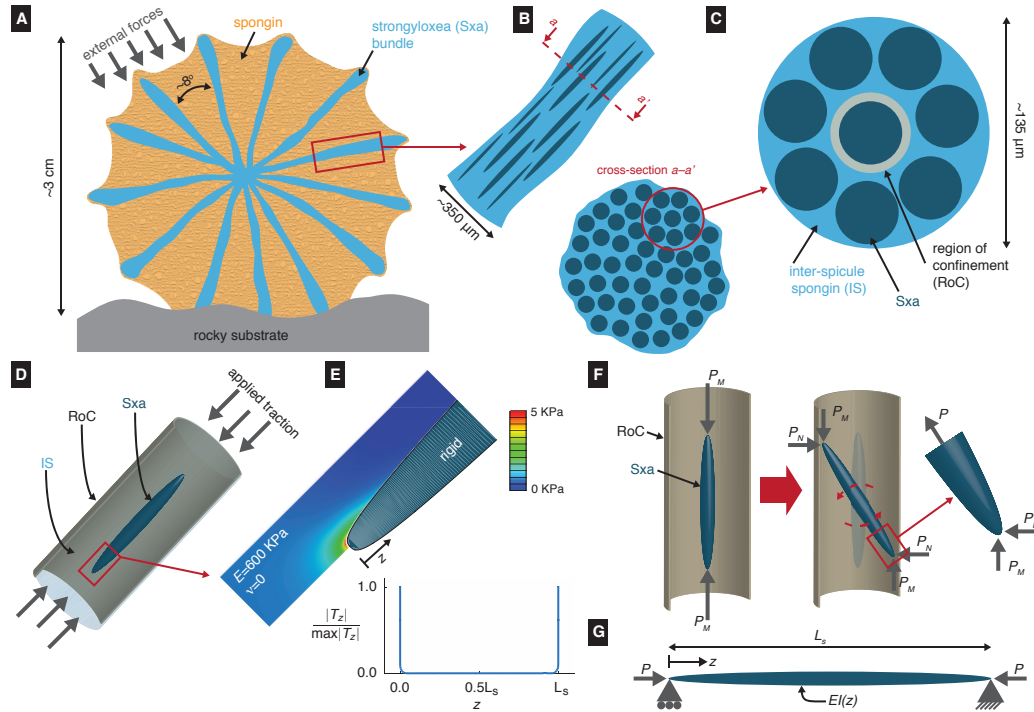


Figure 4.6: Arrangement of strongyloxea within the sponge motivates a structural mechanics model. (a) A cross-section of the sponge reveals radial bundles of strongyloxea (Sxa). (b) A bundle is composed of strongyloxea (dark) separated by spongin (light). (c) The presence of neighbors limits the deformation of a strongyloxea to a region of confinement (RoC). (d) Traction applied to the ends of the region of confinement is transferred to the strongyloxea by the inter-spicule spongin (IS). (e) Von Mises stress computed from a computational mechanics model of (d). The distribution of axial force per unit length, T_z , along the length of a strongyloxea is localized at the ends. (f) A strongyloxea within its region of confinement, subjected to opposing forces with magnitude P_M applied at its ends. A strongyloxea rotates until it is restrained by the presence of neighboring strongyloxea. The net force acting along a strongyloxea's axis has a magnitude P , which includes contributions from P_M and P_N . The strongyloxea and region of confinement in (d)–(f) are not to scale. (g) A schematic of a simply supported column.

External forces acting on the sponge are transmitted by the spongin to the strongyloxea as tractions on their surfaces. To determine the distribution of these tractions, I performed a stress analysis on a continuum mechanics model of an individual strongyloxea embedded in a cylindrical section of spongin. I refer to this cylinder as a strongyloxea's region of confinement (see

Figure 4.6 (c)–(d)). The diameter of the region of confinement is equal to the distance between neighboring strongyloxea in a bundle.

I model the spongin in the region of confinement as an isotropic, linear elastic solid with Young's modulus and Poisson's ratio of 600 KPa and 0, respectively. These values correspond to measurements of the mechanical properties of spongin in a related species [136]. Measurements of the strongyloxea's Young's modulus (see Appendix D) indicate that the spicule's silica is between four and five orders of magnitude stiffer than the spongin. Motivated by this large difference in stiffnesses, I model a strongyloxea as a rigid inclusion whose surface is bonded to the spongin in its region of confinement. I assume that external forces act normal to the sponge's surface and result in axial compressive stresses in the strongyloxea bundles. Therefore, I apply compressive tractions to the ends of the region of confinement (see Figure 4.6 (d)). Since the spongin in a region of confinement is also connected to the spongin in the region of confinements of neighboring strongyloxea, I constrain points on the lateral surface of the region of confinement from moving in the radial direction. Further details about this model can be found in Appendix F.

I computed the stress field in the spongin using finite element procedures (see Figure 4.6 (e)) [145]. I found that the axial force per unit length acting on the strongyloxea is localized on the strongyloxea's ends (see Figure 4.6 (e) and Appendix F). This localized force distribution contrasts with that predicted for an ellipsoidal inclusion embedded in a linear elastic solid subjected to far-field compressive stress. Specifically, a celebrated elasticity solution by Eshelby [146] predicts that the axial force per unit length will vary in a piecewise affine fashion along an ellipsoidal inclusion. It is not necessary, however, for this result to hold true for non-ellipsoidal inclusions. Thus, my numerical results do not contradict Eshelby's solution. In fact, they are consistent with results from computational models of short fiber reinforced composites [147, 148], full-field elasticity solutions for rigid line inclusions [149], and photoelasticity experiments on line-like inclusions [150]. This

suggests that the linear variation of the axial force per unit length predicted by the Eshelby solution is a pathology of the ellipsoidal geometry. Based on the insight gained from my computational mechanics calculations, I modeled the effect of the spongin by replacing the tractions applied to the ends of the region of confinement with opposing point forces, $\pm \mathbf{P}_M$ at the strongyloxea's ends (see Figure 4.6 (f)).

4.3.4 A structural mechanics model for the *T. aurantia* spicules

Initially a strongyloxea behaves like a column with two free ends, which is unstable when subjected to the axial forces $\pm \mathbf{P}_M$. Even if these forces are aligned with the strongyloxea's axis, small perturbations in the configuration will inevitably cause the strongyloxea to rotate about one of its transverse axes. However, after rotating by only a small amount ($\approx 1.3^\circ$), the proximity of neighboring strongyloxea in the bundle would prevent further rotation (see Figure 4.6 (f)). Due to the spongin's large compliance, it is unlikely that this small rotation will substantially change the stress state in the spongin and consequently the traction distribution on the strongyloxea's surface. However, there will be non-negligible reaction forces, $\pm \mathbf{P}_N$, at the points where a strongyloxea is restrained by its neighbors. The net force at a strongyloxea's end, $\mathbf{P}$, which includes contributions from $\mathbf{P}_M$ and $\mathbf{P}_N$, must act in the direction of the strongyloxea's axis (see Figure 4.6 (f)). This is a consequence of static equilibrium and can be deduced using a free body diagram.

Thus, a strongyloxea can be modeled using the Euler-Bernoulli beam theory in which the column's ends are subjected to compressive, axial forces and cannot move in the direction perpendicular to the column's axis. I refer to this model as a simply supported column (see Figure 4.6 (g)). In this model, the transverse deflection, w , is governed by the differential equation

$$\frac{d^2}{dz^2} \left(EI \frac{d^2 w}{dz^2} \right) + P \frac{d^2 w}{dz^2} = 0, \quad (4.6)$$

for all $z \in (0, L_s)$, and boundary conditions

$$w|_{z=0} = w|_{z=L_s} = 0, \quad (4.7)$$

$$EI(z)w_{,zz}|_{z=0} = EI(z)w_{,zz}|_{z=L_s} = 0, \quad (4.8)$$

where P , E , L_s and I are the magnitude of $\mathbf{P}$, the column's Young's modulus, length and second moment of area, respectively. Based on the results of Sections 4.3.1 and 4.3.2 I take E to be constant and $I(z) = \pi r(z)^4/4$, where $r(z)$ is the radius of the strongyloxea's cross-section—i.e. its profile.

4.3.5 Comparison of the *T. aurantia* spicule's shape with the optimal taper

The buckling strength of a simply supported column is the smallest P for which there exists a solution to equations (4.6)–(4.8) other than $w = 0$ for all $z \in [0, L_s]$. For an elastically homogeneous column, the buckling strength can be modulated by varying I , or in this case r , along the column's length [138]. My hypothesis would gain support if the profile of the simply supported column with the greatest buckling strength resembled the measured profiles of the strongyloxea.

The profile that maximizes a simply supported column's buckling strength for a given length, L_s , and volume, V , was first sought by Lagrange in the late 1700s [151]. The correct solution, however, was discovered in 1851 [152], and an accessible proof that it is in fact optimal was given in 1962 [141]. This optimal profile, which I refer to as the Clausen profile, is given by

$$\rho(\theta) = (2\lambda)^{-1} \sin(\theta), \quad (4.9)$$

$$\zeta(\theta) = \frac{1}{\pi} \left(\theta - \frac{1}{2} \sin(2\theta) \right), \quad (4.10)$$

where $\rho = r/L_s$ and $\zeta = z/L_s$ are the dimensionless radial and axial coordinates, respectively, and θ is a parameter that lies between 0 and π [140, 141]. The parameter $\lambda = (3\pi L_s/16V)^{1/2}$ is a measure

of the column's aspect ratio. I refer to a column whose taper is described by the Clausen profile as a Clausen column (see Figure 4.7 (a)).

To test my hypothesis, I compared the Clausen profile to the strongyloxea profiles. I did this by fitting equations (4.9)–(4.10) to each strongyloxea profile in the least-squares sense by varying the parameter λ . The dimensionless profile of a strongyloxea is given by the points, $(\zeta_i^m, \rho_i^m)_{i=1,\dots,250} = (z_i^m/L_s, r_i^m/L_s)_{i=1,\dots,250}$, from Section 4.3.1. I generate the Clausen profile points, $(\zeta_i, \rho_i)_{i=1,\dots,250}$, so that $\zeta_i = \zeta_i^m$ and ρ_i satisfies equations (4.9)–(4.10) for each ζ_i . For each strongyloxea, I varied λ in equation (4.9) to minimize the sum of squared residuals, $SSR = \sum_i (\rho_i^m - \rho_i)^2$, and I denote the minimum SSR as $mSSR$. To understand how well the Clausen profile describes the strongyloxea's taper, I also computed the $mSSR$ for three other profiles, which are given by,

$$\text{semiellipse: } \rho_{\ominus} = \lambda^{-1} \zeta^{1/2} (1 - \zeta)^{1/2}, \quad (4.11)$$

$$\text{triangle: } \rho_{\diamond} = \begin{cases} \lambda^{-1} \zeta, & 0 \leq \zeta \leq \frac{1}{2}, \\ \lambda^{-1} (1 - \zeta), & \frac{1}{2} < \zeta \leq 1, \end{cases} \quad (4.12)$$

$$\text{constant: } \rho_{\square} = (2\lambda)^{-1}, \quad (4.13)$$

The best fit profiles for a representative strongyloxea is shown in Figure 4.7. I found that the Clausen profile (equations (4.9)–(4.10)) has the lowest $mSSR$ for 25 of the 31 strongyloxea and the semiellipse profile (equation (4.11)) has the lowest $mSSR$ for the remaining six.

The median, mean and standard deviation (s.d.) of each profile's $mSSR$ are shown in Table 4.1, from which I see that the Clausen profile has the lowest mean and median $mSSR$. Furthermore, a two-sided Wilcoxon signed rank test indicates that the median $mSSR$ for the Clausen profile differs from that of the semiellipse profile at the 1% significance level ($p = 0.0002$). Thus, using the median $mSSR$ as a metric, I conclude that the Clausen profile describes the strongyloxeas' tapers the best

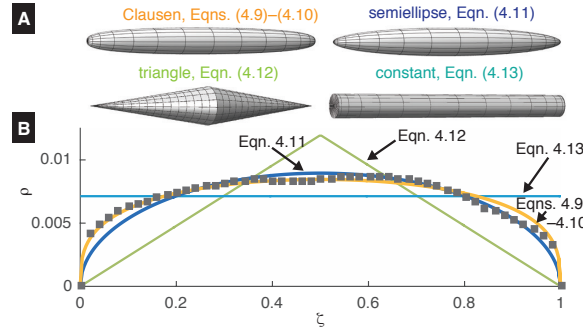


Figure 4.7: Comparison of a strongyloxea’s taper to several profiles. (a) Columns whose profiles are given by equations (4.9)–(4.10) and (4.11)–(4.13). (b) The best fit profiles for a representative strongyloxea. The dimensionless strongyloxea profile points are shown as gray squares.

Table 4.1: Comparison of the candidate profiles used to describe the *T. aurantia* spicule’s tapered shape (N=31)

	$mSSR \times 1000$		
	median	mean	s.d.
Clausen, (4.9)–(4.10)	0.157	0.156	0.077
semiellipse, (4.11)	0.247	0.281	0.165
triangle, (4.12)	2.078	2.125	0.839
constant, (4.13)	0.721	0.769	0.404

out of the different profiles that I considered.

While the mean and median $mSSR$ is lowest for the Clausen profile, the semiellipse profile did have a lower $mSSR$ for approximately 19% of the strongyloxea. I clarify how much better the Clausen profile is compared to the semiellipse profile by finding the weight of evidence that the Clausen profile is the “best” of the candidate profiles. I consider the “best” profile for a particular strongyloxea to be the one that minimizes the Kullback-Liebler (K-L) distance. The K-L distance quantifies the amount information that is lost by using a model to approximate the true function from which the data was drawn [153]. Since this function is not known, the K-L distance cannot be computed directly. However, I can obtain an estimate of it by computing the Akaike Information Criterion (AIC) [153, 154]. For each strongyloxea, the fitted candidate profile with the lowest AIC value is also expected to be the K-L best profile.

I denote the $mSSR$ and AIC of the i^{th} profile fitted to the j^{th} strongyloxea as $mSSR_i^j$ and AIC_i^j , where $i \in \{\text{Clausen, semiellipse, triangle, constant}\}$ and $j = 1 \dots 31$. The AIC_i^j is given by

$$AIC_i^j = N_j \log \left(\frac{mSSR_i^j}{N_j} \right) + 2(K_i + 2),$$

where N_j is the number of data points in the j^{th} strongyloxea profile, and K_i is the number of free parameters in the i^{th} model [153]. Here, N_j and K_i are both constants whose values are 250 and unity, respectively. From the AIC_i^j values I can compute the relative likelihood that the i^{th} profile is the the K-L best for the j^{th} strongyloxea. This likelihood is referred to as the normalized Akaike weight, ω_i^j , and is given by

$$\omega_i^j = \frac{e^{-\Delta_i^j/2}}{\sum_i e^{-\Delta_i^j/2}}, \quad (4.14)$$

where $\Delta_i^j = AIC_i^j - \min_i AIC_i^j$ [153]. The mean and standard deviation over j of ω_i^j are given in Table 4.2. The ratio $mean_j(\omega_{\text{Clausen}}^j) : mean_j(\omega_{\text{semiellipse}}^j)$ indicates that the Clausen profile is on average 4.55 times more likely to be the K-L best than the semiellipse profile.

Table 4.2: Akaike weights, ω , of the candidate profiles used to describe the *T. aurantia* spicule's tapered shape

	mean	s.d.
Clausen, (4.9)–(4.10)	$0.820 \pm$	0.374
semiellipse, (4.11)	$0.180 \pm$	0.374
triangle, (4.12)	$0.000 \pm$	0.000
constant, (4.13)	$0.000 \pm$	0.000

4.3.6 Direct estimation of the *T. aurantia* spicule's buckling strength

The fact that the Clausen profile describes the measured strongyloxea profiles the best among the prototypical tapered profiles that I considered gives strength to my hypothesis. However, it is still possible that there may exist some other profile, which corresponds to an alternate hypothesis, that

describes the strongyloxea's taper even better than the Clausen profile. If such a profile exists, would my hypothesis remain viable?

To answer this question, I numerically estimated the strongyloxeas' buckling strengths, P_s , using the measured profiles and my structural mechanics model. Briefly, I computed a strongyloxea's second moment of area $I(z) = \pi r(z)^4/4$ from its profile, $r(z)$, and used the Rayleigh-Ritz method [121] to find an approximate value for the smallest P for which there exists a solution to equations (4.6)–(4.8) other than $w = 0$. I computed P_s for each of the 31 strongyloxea whose profiles I measured in Section 4.3.1 and compared it to the buckling strength $P_c = \pi E V^2 / (4L_s^4)$ of the equivalent cylinder—i.e., the cylinder with the same length, volume, and elastic properties (see Figure 4.8). The median value of $(P_s - P_c)/P_c$ indicates that the strongyloxea's tapered enhances their buckling strength by 13.4 %. Furthermore, some strongyloxea achieve values of $(P_s - P_c)/P_c$ as large as 0.3 which is close to the enhancement of 0.33 provided by the Clausen column [140] (see Figure 4.8).

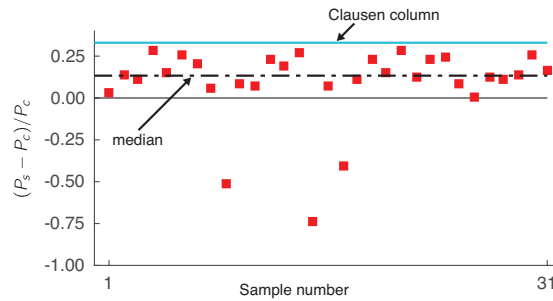


Figure 4.8: Estimated buckling strengths of strongyloxea. The relative buckling strengths, $(P_s - P_c)/P_c$, of the 31 strongyloxea whose profiles were measured in Section 4.3.1 are estimated using my structural mechanics model and are shown as red squares. The dashed, black line indicates the median of the strongyloxeas' relative buckling strengths. The solid, blue line denotes the maximum possible enhancement of buckling strength, which corresponds to the Clausen column.

So, even if there existed a profile that better resembled the strongyloxeas' tapers, the fact still remains that the strongyloxeas' tapers substantially enhance their buckling strengths. Therefore,

even if there existed a better matching profile based on an alternate hypothesis, the support for my hypothesis would still remain strong. Such a scenario would only mean that the strongyloxea serve more than one function.

Chapter 5

Conclusion

Quantifying the effect of the spicule's architecture on its toughness further develops the understanding of both the spicule's mechanical function and the effect of the architecture on its mechanical behavior. I found that their cylindrically layered architecture does not appear to provide substantial toughness enhancements compared to those observed in other SBMs. These findings are a reminder that although the lamellar architecture has been shown to dramatically affect the fracture toughness properties of SBMs like nacre and bone, similar architectures may serve other mechanical functions and enhance other properties as well. It is important to measure each SBM's toughness properties rather than categorizing it as tough solely based on it possessing a lamellar architecture. These measurements are crucial for developing truly revolutionary bio-inspired designs and avoiding the pitfalls of naive biomimicry. I hope that this study brings to light the nuanced nature of the connection between lamellar architectures and toughness properties. Assuming that the lamellar architecture of spicules evolved as a response to evolutionary pressures, it would likely benefit the property that is critical to the spicules function and overall fitness of the organism. It is therefore important to always keep in mind the ecology of the organism—i.e., the function of the SBM as it relates to the loading conditions that it experiences in its natural environment—when exploring

potential structure-property relationships [7].

I then considered the hypothesis that the *E. aspergillum* spicule's lamellar architecture allows them to sustain large bending curvatures [41]. The flexibility, or strain tolerance, that the architecture imparts could enhance the sponge's anchorage by allowing the spicules to take on tangled shapes within the sediments and consequently reducing the risk of the sponge being uprooted [41]. I showed that the bending failure strain of the *E. aspergillum* spicules is $\approx 140\%$ larger than that of the *T. aurantia* spicules. I do not believe that this result is a consequence of the *E. aspergillum* spicules' silica being intrinsically stronger than the *T. aurantia* spicules' silica. Rather, I believe that the *E. aspergillum* spicule's architecture allows it to deform differently than a monolithic beam, and reduces the maximum strain that the spicule experiences for a given $\kappa(x_1)$ compared to a monolithic beam. In Chapter 3 I compute the *E. aspergillum* spicules' bending failure strains by approximating their kinematics using elastica theory (i.e., I assume that the strain is given by $\varepsilon_{11} = r\kappa(x_1)$). However, a more refined mechanics model that accounts for the spicule's architecture is needed to understand the mechanism(s) underlying the enhancement of *E. aspergillum* spicules' bending failure strains. Specifically, while ε_{11} increases linearly with the distance, r , from the neutral plane in a monolithic beam, this may not be true for the spicules.

As a preliminary hypothesis I consider a beam model in which adjacent concentric layers slide relative to one another. Consider, for example, the 2D layered beam shown in Figure 5.1 (B). Since adjacent layers are not glued together, when the beam is bent they are able to slide relative to one another like stacked sheets of paper. This allows the layered beam to undergo larger deformations than a monolithic beam with the same cross-section (see Figure 5.1 (A)) before the strain in any layer meets the failure criterion. Consequently, the layered beam would appear to have a larger bending failure strain since it would have a larger curvature before it failed. While the mechanics of this 2D analog and the cylindrically layered *E. aspergillum* spicules are different, a similar mechanism

could be operating in the *E. aspergillum* spicules. That is, like in the 2D analog, the sliding of adjacent layers may allow strain to be redistributed across the spicule's cross-section.

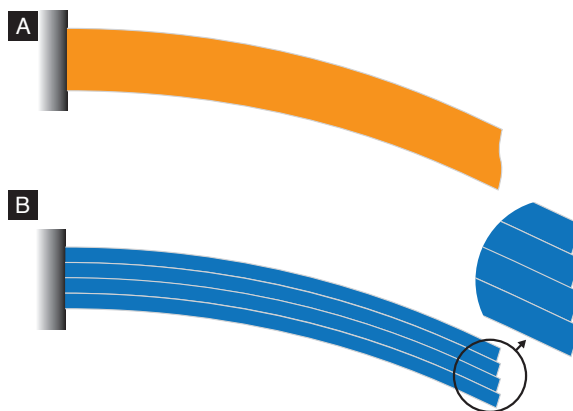


Figure 5.1: (A), (B) The deformed configurations of a monolithic and a multi-layered beam, respectively. Adjacent layers in the multi-layered beam are able to slide past one another as the beam is bent.

This layer sliding mechanism may also explain the observed enhancement in toughness properties. If the interlayers allow stress to be redistributed across the spicule's cross-section when it is bent, then this stress redistribution could effectively shield flaws (like the notches cut in Chapter 2). This would reduce the energy release rate compared to an equivalent homogeneous material, effectively increasing the measured initiation toughness. Furthermore, the sliding of adjacent silica layers would require the proteinaceous interlayers to undergo large shear strains. If the material comprising the interlayer were to experience inelastic deformation due to these large strains, that deformation would dissipate energy. This would reduce the energy that is available to propagate cracks. Unlike some toughening mechanisms like crack wake bridging, and crack arrest and renucleation this effective plastic dissipation would not necessarily manifest in micrographs of the fractured spicules. This is consistent with the smooth, featureless fracture surfaces that can be seen in Figure 2.3 (B).

Finally, the structure-property connection that I identify in the strongyloxea spicules of the *T.*

aurantia sponge represents a completely new type of entry into the growing library of structure-property connections in biological materials and structures. While the identified connection is related to the structure's stiffness, by being sharply focused on preventing buckling it is quite different from the stiffness-related structure-property connections that have been identified in biological structures, such as stems and quills [155, 156]. I hope that this work encourages the investigation of the potential buckling resistance offered by the tapered shapes of other slender biological structures, such as hedgehog quills and echinoderm spines.

I also believe that this work will increase the interest in structure-property investigations. Interest in bio-inspired engineering was originally based on the tacit assumption that evolutionary adaptation produced close-to-optimal mechanical designs [157]. However, now it is understood that for adaptations to take root they do not have to be close-to-optimal, but only “good enough” [158]. This understanding stemmed from the fact that there are very few examples of mechanical designs in biological structures and materials that have been rigorously shown to being close-to-optimal [22, 28, 159, 160]. This new understanding acts as an important bulwark against efforts that blindly imitate mechanical designs in biology without first understanding their functional significance. Unfortunately, this new understanding can also lead to excessive skepticism about the effectiveness of adaptations, and consequently, about the importance of investigating structure-property connections. Since these results show that the taper in *strongyloxea* is not just a beneficial adaptation, but is in fact a close-to-optimal adaptation, I believe that they will help alleviate such skepticism.

Appendix A

Derivation of a compliance function for an encastered SEC-RBB specimen

A.1 Motivation for the form of the dimensionless compliance, $\bar{C}$

There are five physical parameters that characterize the bending behavior of a notched beam prior to crack growth: C , E , L , D and a . These five parameters share two independent physical units (length and force), and therefore the Buckingham Π theorem [67] guarantees that there exists three dimensionless parameters. In addition to the dimensionless compliance $\bar{C}$ I take these parameters to be the dimensionless combined notch and crack length $\alpha = a/D$ and the slenderness $\delta = D/L$. As per the Buckingham Π theorem, $\bar{C} \rightarrow \bar{C}(\alpha, \delta)$.

A.2 Restrictions on the form of $\bar{C}$

By Taylor expanding $\bar{C}$ first about $\delta=0$ and then about $\alpha=0$, I get

$$\bar{C}(\alpha, \delta) = f(0, 0) + f_{,\alpha}(0, 0)\alpha + f_{,\delta}(0, 0)\delta + f_{,\alpha\delta}(0, 0)\alpha\delta + g(\alpha) + \delta h(\alpha) + o(\delta), \quad (\text{A.1})$$

where $f_{,\alpha} = \partial f / \partial \alpha$, $f_{,\delta} = \partial f / \partial \delta$, $f_{,\alpha\delta} = \partial^2 f / \partial \alpha \partial \delta$ and the functions $g \in o(\alpha)$ and $h \in o(\alpha)$.

The expression $o(\cdot)$ is the Landau symbol “little-o.” As a consequence of this, I know that

$$\begin{aligned} g(0) &= 0 \\ g'(0) &= 0 \\ h(0) &= 0 \\ h'(0) &= 0, \end{aligned} \tag{A.2}$$

where $g'(0) = dg/d\alpha|_{\alpha=0}$ and $h'(0) = dh/d\alpha|_{\alpha=0}$. By evaluating Eqn. (A.1) at $\alpha=0$ I get

$$1 = f(0,0) + f_{,\delta}(0,0)\delta + o(\delta).$$

For this to hold for all δ , this means that $f(0,0) = 1$, $f_{,\delta}(0,0) = 0$. Thus, I get

$$\bar{C}(\alpha, \delta) = 1 + f_{,\alpha}(0,0)\alpha + f_{,\alpha\delta}(0,0)\alpha\delta + g(\alpha) + \delta h(\alpha) + o(\delta). \tag{A.3}$$

The necessary condition for crack growth is $G \geq R(0)$, where $R(0) \in (0, \infty)$ is the material's fracture initiation toughness. In the limit of $\alpha \rightarrow 0$ for any fixed, finite-valued F , the energy release rate per Eqn. (2.8) will become unbounded unless $d\bar{C}/d\alpha \in O(\sqrt{\alpha})$ as $\alpha \rightarrow 0$. This means that a crack will spontaneously grow in a specimen with even the smallest imperfection or flaw. Since this would seem unphysical, I require that $d\bar{C}/d\alpha \in O(\sqrt{\alpha})$ as $\alpha \rightarrow 0$ and consequently $d\bar{C}/d\alpha(0, \delta) = 0$. By differentiating $\bar{C}$ in Eqn. (A.3) and evaluating at $\alpha = 0$ I get

$$\begin{aligned} \frac{d\bar{C}}{d\alpha} &= f_{,\alpha}(0,0) + f_{,\alpha\delta}(0,0)\delta + g'(\alpha) + \delta h'(\alpha) \\ 0 &= f_{,\alpha}(0,0) + f_{,\alpha\delta}(0,0)\delta. \end{aligned}$$

For this to hold for all δ , this means that $f_{,\alpha}(0,0) = f_{,\alpha\delta}(0,0) = 0$. Thus, I can further simplify the dimensionless compliance to

$$\bar{C}(\alpha, \delta) = 1 + g(\alpha) + \delta h(\alpha) + o(\delta). \tag{A.4}$$

I can place further restrictions on the functions g and h by evaluating Eqn. (A.4) at $\alpha=1$ and recalling that $\bar{C}(1, \delta)=4$. From this I get

$$3 = g(1) + \delta h(1).$$

For this to hold for all δ ,

$$\begin{aligned} g(1) &= 3 \\ h(1) &= 0. \end{aligned} \tag{A.5}$$

Furthermore, I also note that as $\alpha \rightarrow 1$ for any fixed F , no matter how small, the energy release rate per Eqn. (2.8) will become unbounded unless $d\bar{C}/d\alpha \in O(\sqrt{1-\alpha})$ and therefore $d\bar{C}/d\alpha(1, \delta) = 0$.

For this to hold for all δ , I require that

$$\begin{aligned} g'(1) &= 0 \\ h'(1) &= 0 \end{aligned} \tag{A.6}$$

A.3 Generation of compliance calibration data using a computational mechanics model

I generated compliance calibration data for two different values of δ using a computational mechanics model of the encastered SEC-RBB specimen. In the model I chose $D=39.466 \mu\text{m}$, which is the average diameter of *E. aspergillum* spicules measured in Chapter 3. I varied δ by using two different L ; 800 and 1600 μm . The specimen is modeled as a homogeneous linear elastic solid with $E=28 \text{ GPa}$ and Poisson's ratio $\nu=0.2$. To reduce computational cost, I modeled one quarter of the fiber specimen for which $0 \leq x_1 \leq L/2$ and $x_3 \geq 0$. The model contains an edge notch with a width of 1 μm , whose notch root has a radius of curvature of 500 nm (see Figure A.1 (A)). The length of this notch is a

I impose boundary conditions on several surfaces in my computational mechanics model that are marked in Figure A.1 (A). The surface Γ_1 is the remaining ligament of the specimen, to which I prescribe that the $\hat{e}_1$ component of the displacement is zero. I also prescribe that the $\hat{e}_3$ component of the displacement is zero on Γ_2 and set all displacement components $u_k=0$, $k = 1, 2, 3$ on Γ_3 .

Finally, I apply a displacement $u_2 = -1 \mu\text{m}$ on Γ_4 , which is a small patch whose size is $\approx 2 \mu\text{m} \times 2 \mu\text{m}$.

I solve for the stress state in the model using finite element methods. I compute the component of the traction in the $\hat{e}_2$ direction on Γ_4 from the Cauchy stress components, σ_{ij} , $i, j \in \{1, 2, 3\}$, as $t_2 = \sigma_{21}n_1 + \sigma_{22}n_2 + \sigma_{23}n_3$. The net force exerted on Γ_4 in the $\hat{e}_2$ direction then is given by $T_2 = \int_{\Gamma_4} t_2 d\Gamma$. I compute the compliance of the model by fitting a line to the u_2 - T_2 data (see Figure A.1 (B)).

For each value of δ I computed the compliance for 10 different notch lengths (i.e., values of αD) equally spaced between zero and D . From this data I constructed a plot of $\bar{C}$ versus α for the two values of δ (see Figure A.1 (C)).

It is worth noting that the compliance calibration data from my computational mechanics model corroborates the restrictions that I imposed on the form of $\bar{C}$ in Section A.2. That is, the dimensionless compliance approaches the boundary values of 1 and 4 as $\alpha \rightarrow 0$ and $\alpha \rightarrow 1$, respectively (see Eqns. (A.2) and (A.5)). Furthermore, the derivative $d\bar{C}/d\alpha$ vanishes at the boundaries as well (see Eqns. (A.2) and (A.6)).

The specimen in my computational model has a constant cross-sectional radius. However, I use the compliance calibration data obtained from this model to compute $R(0)$ for the *T. aurantia* spicules, which are tapered. I justify this decision as follows. It has been shown that the cross-sectional radius of a *T. aurantia* spicule can be described by Eqns. (D.6). Using these equations I can estimate how much the *T. aurantia* spicule's cross-sectional radius varies from the trench edge to the midspan. I assume that the spicules fail at mid-span and that therefore the radius of the cross-section at which the spicule fails is r_0 . I measure r_0 from scanning electron micrographs taken after the fracture tests. Since the *T. aurantia* spicules' lengths, L_s , are larger than the field of view of the microscope that is part of my mechanical testing device, I did not measure L_s directly. However, in Chapter 4 I show that the aspect ratio $\lambda_s = L_s/2r_0$ of the *T. aurantia* spicules is also relatively constant [27]. Therefore, in equation (D.6) I estimate L_s to be $2\lambda_s r_0$ where $\lambda_s = 53.6$ [27]. I compute the cross-sectional radius at the trench edges by setting $x_1 = 0$, $L = 600 \mu\text{m}$ and solving Eqn. (D.6) for r . I find that for the spans used in the fracture tests, the cross-sectional radius of the *T. aurantia* spicules only varies by $\approx 3\%$ from the trench edge to midspan.

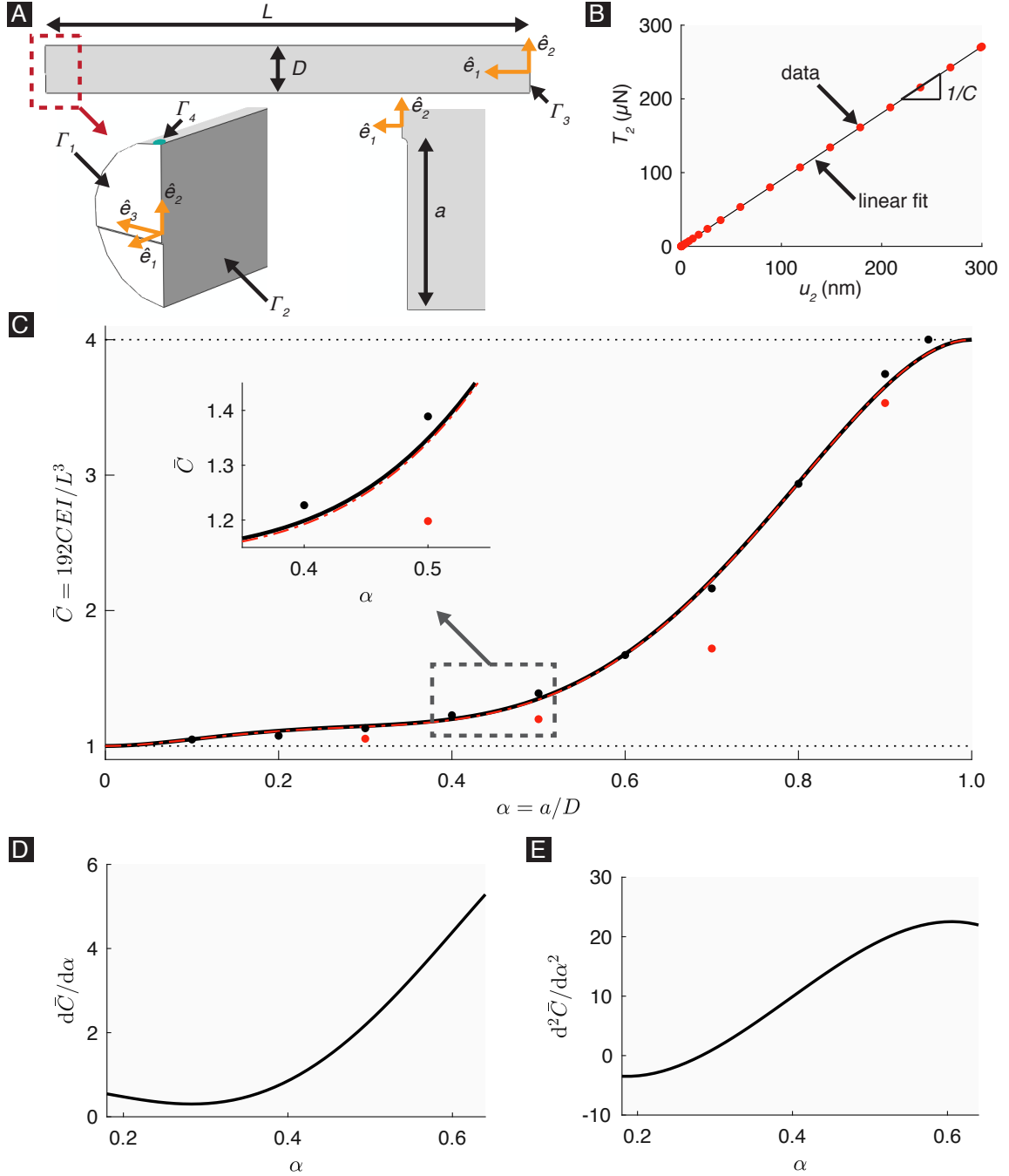


Figure A.1: Details of the computational mechanics model used to obtain the compliance calibration data. (A) geometry of the model consisting of one quarter of the encastered SEC-RBB specimen. (B) The T_2 - u_2 data obtained from the computational mechanics model for $\alpha = 0.4$. The compliance C is obtained by fitting a line to this data. (C) The compliance calibration data obtained from the computational mechanics model for two values of δ . The black and red points correspond to the data for $\delta = 0.049$ and $\delta = 0.025$, respectively. The black curve is obtained by fitting Eqn. (A.9) to the compliance calibration data for $\delta = 0.049$. The red dashed curve (see inset) is obtained by evaluating Eqn. (A.9) at $\delta = 0.025$ using the coefficients obtained for $\delta = 0.049$. (D) The derivative of the compliance calibration curve for $\delta = 0.049$ used to compute $R(0)$. (E) The second derivative of the compliance calibration curve for $\delta = 0.049$.

A.4 Approximation of g and h

I approximate g and h as polynomials given by

$$\begin{aligned}\hat{g}(\alpha) &= \sum_{i=0}^5 a_i \alpha^i \\ \hat{h}(\alpha) &= \sum_{j=0}^4 b_j \alpha^j,\end{aligned}\tag{A.7}$$

where $a_i, b_j \in \mathbb{R}$ for $i = 1 \dots 5$ and $j = 1 \dots 4$. I use the boundary conditions on g and h given in Eqns. (A.2), (A.5) and (A.6) to impose constraints on the values of a_i and b_j . Substituting Eqns. (A.7) into these equations, I find the form of $\hat{g}$ and $\hat{h}$ to be

$$\begin{aligned}\hat{g}(\alpha) &= (9 + a_4 + 2a_5)\alpha^2 + (-2a_4 - 3a_5 - 6)\alpha^3 + a_4\alpha^4 + a_5\alpha^5 \\ \hat{h}(\alpha) &= b_4\alpha^2(1 - \alpha)^2.\end{aligned}\tag{A.8}$$

The dimensionless compliance $\bar{C}$ can then be approximated as

$$\hat{C}(\alpha, \delta) = 1 + \hat{g}(\alpha) + \delta \hat{h}(\alpha).\tag{A.9}$$

I fitted Eqn. (A.9) to the compliance calibration data for $\delta=0.049$ (i.e., $L=800 \mu\text{m}$) in a least-squares sense by varying the parameters a_4 , a_5 and b_4 subject to the constraint that $\hat{C}$ must be monotonically increasing (see Section A.5 and Eqn. (A.11)). The best fit polynomial approximation to the $\delta=0.049$ compliance calibration data is shown in Figure A.1 (C).

Per the derivation presented in A.2, the values a_4^* , a_5^* and b_4^* obtained by fitting to data for $\delta=0.049$ should be independent of δ and therefore also produce a good fit for $\delta=0.025$ (i.e., $L=1600 \mu\text{m}$). In Figure A.1 (C) I see that the results obtained from the computational mechanics model predict a larger dependence on δ than what is predicted by the polynomial approximation $\hat{C}$ in Eqn. (A.9). However, I believe that this discrepancy is in part due to the large difference between the δ values of these two data sets (δ changes by a factor of 2). It is possible that the first order Taylor expansion in δ will not accurately capture the dependence on δ for such large ranges of δ . I could consider higher orders of δ in the expansion but this would increase the model's complexity,

which is exactly what I was trying to avoid to begin with.

A.5 Further constraints on the functions $\hat{g}$ and $\hat{h}$

Intuition tells us that a specimen with a longer crack will be more compliant than a specimen with a shorter crack. That is, $\bar{C}$ (and therefore $\hat{C}$) should monotonically increase with α so that

$$\frac{d\bar{C}}{d\alpha} > 0 \quad \forall \alpha \in (0, 1). \quad (\text{A.10})$$

Given that $d\bar{C}/d\alpha = 0$ at both $\alpha = 0$ and $\alpha = 1$, for Eqn. (A.10) to hold the approximation $d\hat{C}/d\alpha$ must not have any real roots in the interval $\alpha \in (0, 1)$. For arbitrary $\delta > 0$, $\hat{C}$ has four roots, two of which occur at $\alpha=0$ and $\alpha=1$ to satisfy the boundary conditions in Eqns. (A.2) and (A.6). The other two roots are conjugates of each other and depend on δ , a_4 , a_5 and b_4 . It is difficult to enforce Eqn. (A.10) directly by requiring that $d\hat{C}/d\alpha$ has no real roots in the interval $\alpha \in (0, 1)$. This is because one would need to derive constraints on δ , a_3 , b_3 and b_4 for two separate scenarios.

1. the two roots are real but lie outside the interval $\alpha \in (0, 1)$
 - (a) both roots occur at $\alpha \leq 0$, or
 - (b) one root occurs at $\alpha \leq 0$ and the other occurs at $\alpha \geq 1$, or
 - (c) both roots occur at $\alpha \geq 1$
2. the two roots are complex conjugates

Enforcing the three cases in Scenario 1 is difficult. In Scenario 2, however, the requirement is more easily satisfied. Therefore, I require that the remaining two roots are complex conjugates by requiring that the discriminant of $\hat{C}/(\alpha(1-\alpha))$ is negative. That is,

$$16a_4^2 + 105a_5^2 + 16\delta^2b_4^2 + 40a_5(9 + 2\delta b_4) + 16a_4(5a_5 + 2\delta b_4) < 0. \quad (\text{A.11})$$

Appendix B

The effect of moisture on the bending behavior of *E. aspergillum* spicules

In my experiments, I used samples from dry *E. aspergillum* skeletons. However, it has been shown that the mechanical behaviors of some biological materials (such as nacre [161, 162], antler [163, 164] and bone [164, 165]) change drastically if they are dried out prior to mechanical testing. For example, it has been shown that the work of fracture of nacre that has been soaked in water is almost triple that of nacre that is stored in dry conditions [7]. In their native state, the organic phases in these SBMs contain some amount of water, but researchers often are only able to obtain samples that have been stored in dry conditions. I believe that the ceramic phases prevent water within the organic phases of SBMs from evaporating or diffusing out of the material. However, when samples are cut from these SBMs, the organic phase is exposed and is able to dry out. By soaking nacre in water prior to testing, the thin, proteinaceous interlayers separating adjacent aragonite tablets in its brick-and-mortar architecture rehydrate and soften. This allows the proteinaceous interlayers to withstand large deformations, which then allows adjacent ceramic tablets to slide relative to each other before failing. This tablet sliding has been identified as an important extrinsic toughening mechanism in nacre.

To investigate the effect of moisture on the *E. aspergillum* spicules, I performed three-point bending tests on spicules soaked in artificial seawater. To assess the effect of this sample preparation

procedure on the spicules' mechanical behavior I computed the Young's modulus and the bending failure strain of the wet spicules and compared them to measurements of these quantities described in Chapter 3 for the dry spicules.

I soaked sections of 11 *E. aspergillum* spicules in artificial seawater for 16 days and then performed the bending tests using the same procedure described in Chapter 3. I used the slope of the linear portion of the spicule's $F-w_0$ data, k_s , to compute the Young's moduli of the 11 wet spicules using Eqn. (C.1). As described in Chapter 3 I computed the spicule's second moment of area $I = \pi r_0^4/4$ using measurements of the cross-sectional radius r_0 that I measured from scanning electron micrographs. Histograms of the Young's moduli of the wet and dry spicules (tested in Chapter 3 Section 3.4) is shown in Figure B.1 (A).

Furthermore, I computed the bending failure strain, ϵ_f , of the wet spicules using the same procedure as described in Chapter 3 Section 3.3. Briefly, I selected points along the spicule's longitudinal axis in the micrograph taken just before it failed. I fit a polynomial to these points and used it to compute the curvature of the longitudinal axis. Finally, I compute ϵ_f using the maximum value of the curvature, κ^* , and the spicule's cross-sectional radius as $\epsilon_f = r_0 \kappa^*$ [110]. I compare ϵ_f for the wet spicules to ϵ_f for the dry spicules reported in Chapter 3 in Figure B.1 (B).

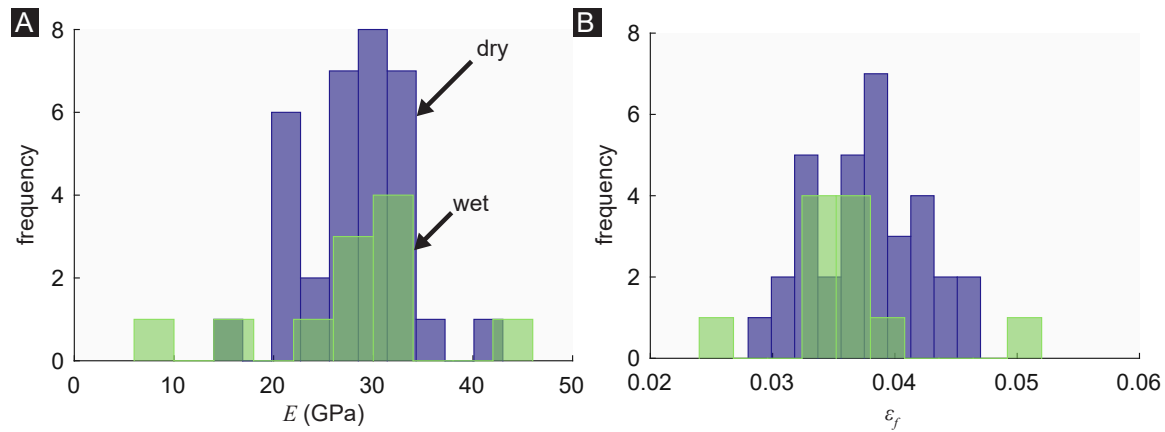


Figure B.1: Mechanical behavior of 33 dry *E. aspergillum* (light blue) and 11 wet *E. aspergillum* (green) spicules. (A) A histogram of the Young's modulus, E . (B) A histogram of the bending failure strain, ϵ_f .

Bias-corrected accelerated (BCa) confidence intervals (CI) using 10,000 bootstrapped samples indicated no reliable difference in the means of the Young's moduli of the wet and dry spicules (upper: 3.0 GPa; lower: -4.5 GPa). Furthermore, a two-sided t -test for independent samples (equal

variances not assumed) also indicated no significant difference in their means ($p=0.96$). Similar results were found when comparing the bending failure strains using both BCa CI (upper: 0.0012; lower: -0.0034) and the t-test ($p=0.35$).

Soaking the spicules in water prior to testing them does not appear to affect their mechanical properties like it does for other SBMs, such as nacre. In both spicules and nacre, the only ways for the organic phases to lose moisture are through cracks in the ceramic phases or through regions of the organic phase that are exposed to the environment, e.g., at the cross-sections of broken spicules. However, because the interlayers are very thin ($\approx 5\text{--}10$ nm [29]) I speculate that the rate of desiccation would be extremely slow. In contrast, when samples are cut from nacre, much larger cross-sections of the organic phase are exposed. This is because nacre has hundreds of organic interlayers (compared to the ≈ 25 interlayers in the *E. aspergillum* spicules) and the thickness of the interlayers can be up to an order of magnitude larger than that of the spicules' interlayers [166]. Furthermore, as a consequence of nacre's brick and mortar architecture, these interlayers are all interconnected. This interconnectivity could facilitate the transport of water within the interlayers. I believe that these two features contribute to an increased rate of desiccation in nacre compared to spicules.

Appendix C

Calibration of the mechanical testing device

I calibrated the mechanical testing device by measuring the Young's modulus, E , of a tungsten wire.

For the configuration shown in Figure 3.4 (B), the Young's modulus is given by

$$E = k_s L^3 / 48I, \tag{C.1}$$

where k_s is the slope of the linear portion of the F – w_0 data, $I = \pi r_0^4 / 4$ is the second moment of area of the cross-section, L is the trench span, and r_0 is the cross-sectional radius of the wire. Equation (C.1) comes from the Euler-Bernoulli theory for a simply-supported beam with a concentrated lateral load acting at mid span [43].

I measured the wire's diameter to be $15.15 \pm 0.03 \mu\text{m}$ (mean $\pm$ standard deviation; $N = 10$) using a scanning electron microscope (SEM). I performed 12 tests on different pieces of the tungsten wire and found the Young's modulus to be $395.2 \pm 13.4 \text{ GPa}$ (mean $\pm$ standard deviation (s.d.)). This value agrees closely with values cited in literature (see Table C.1).

Table C.1: Young's modulus of tungsten (GPa)

Measured ($N = 12$)		Reference		
mean	s.d.	[167]	[168]	[122]
395.2	13.4	404.0	409.8	410

Appendix D

Calculation of Young's modulus of *E. aspergillum* and *T. aurantia* spicules

At small deflections, F appeared to increase linearly with w_0 for both the *E. aspergillum* spicules and *T. aurantia* spicules (see Figure D.1 (C)). I describe a specimen's deformation in this region of the $F-w_0$ data using the Euler-Bernoulli beam theory for the configuration shown in Figure D.1 (A) [43]. Since the *E. aspergillum* spicules have a constant cross-sectional radius, their Young's modulus can be computed using Eqn. (C.1). The Young's modulus of the 33 *E. aspergillum* spicules is 28.1 ± 5.4 GPa (mean $\pm$ standard deviation).

Since the *T. aurantia* spicules are tapered along their length I use an Euler-Bernoulli theory that accounts for this taper to compute their Young's modulus. I derive the equation connecting E to k_s by computing the total elastic energy in the beam model of the *T. aurantia* spicule shown in Figure D.1 (A) and applying Castigliano's second theorem [169]. Euler-Bernoulli theory predicts that the only nonzero component of the Cauchy stress tensor $\boldsymbol{\sigma} = \sigma_{ij} \hat{e}_i \otimes \hat{e}_j$ is $\sigma_{11}(x_1, x_2) = M(x_1)x_2/I(x_1)$, where $M(x_1)$ is the magnitude of the net bending moment acting on the beam's cross-section located at x_1 , and $I(x_1)$ is the second moment of area of that cross-section. Specifically, $I(x_1) = \int_{\Omega(x_1)} x_2^2 d\Omega$, where $\Omega(x_1)$ denotes the beam's cross-section located at x_1 and $d\Omega$ is an infinitesimal area element on the cross-section. The dependence of I and Ω on x_1 is a consequence of the *T. aurantia* spicule being tapered. I assume that the *T. aurantia* spicule has a circular cross-section with radius r at x_1

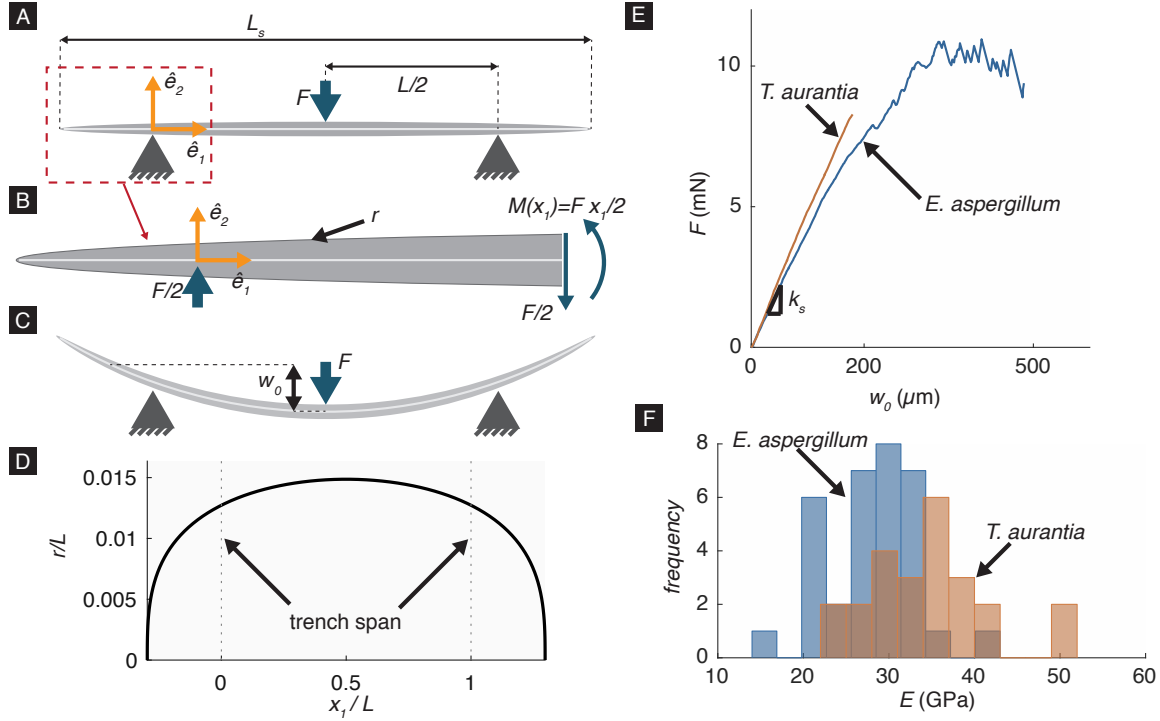


Figure D.1: Procedure for computing the Young's moduli of *E. aspergillum* and *T. aurantia* spicules. (A) A schematic of the three-point bending test configuration used to measure the Young's moduli of the spicules. (B) A free-body diagram of the bending test depicted in (A). The cross-sectional radius of the *T. aurantia* spicules r changes as a function of the axial coordinate x_1 . (C) The deformed shape of the spicule showing the rotation of the specimen at the supports. (D) The shape described by Eqn. (D.6) used to model the tapered shape of the *T. aurantia* spicules. (E) A F - w_0 response of a representative *T. aurantia* and *E. aspergillum* specimen taken from the data presented in Chapter 3. I use the slope of the initial linear portion of the F - w_0 response, k_s , to compute E . (F) A histogram of the Young's moduli of the 33 *E. aspergillum* (blue) and 24 *T. aurantia* (orange) spicules computed from the F - w_0 responses presented in Chapter 3.

(see Figure D.1 (B)) and therefore $I(x_1) = \pi r(x_1)^4 / 4$.

The total elastic energy, U_e , stored in the beam is

$$U_e = \int_{x_1=0}^L \int_{\Omega(x_1)} \frac{\sigma_{11}^2}{2E} d\Omega dx_1 = \int_{x_1=0}^L \int_{\Omega(x_1)} \frac{M(x_1)^2 x_2^2}{2EI(x_1)^2} d\Omega dx_1. \quad (\text{D.1})$$

Evaluating the inner integral in equation (D.1) I get

$$U_e = \frac{1}{2E} \int_{x_1=0}^L \frac{M(x_1)^2}{I(x_1)} dx_1. \quad (\text{D.2})$$

From a free body diagram of the beam model (see Figure D.1 (B)), $M(x_1)$ varies along the length of the spicule as $F x_1 / 2$ for $x_1 \in [0, L/2]$. In Chapter 4, I show that a *T. aurantia* spicule's taper is relatively symmetric across the midpoint along its length [27]. I assume that the midpoint along

the spicule's length is located at $x_1 = L/2$ and therefore both $M(x_1)$ and $I(x_1)$ are symmetric across $x_1 = L/2$. Thus, in terms of F , the total elastic energy of the spicule is

$$U_e = \frac{F^2}{4E} \int_{x_1=0}^{L/2} \frac{x_1^2}{I(x_1)} dx_1. \quad (\text{D.3})$$

It follows from Castigliano's second theorem [169] that

$$w_0 = \partial U_e / \partial F = \frac{F}{2E} \int_{x_1=0}^{L/2} \frac{x_1^2}{I(x_1)} dx_1. \quad (\text{D.4})$$

Solving for E and noting that the ratio F/w_0 is the slope of the linear portion of the F - w_0 response, k_s , I get

$$E = \frac{k_s}{2} \int_{x_1=0}^{L/2} \frac{x_1^2}{I(x_1)} dx_1. \quad (\text{D.5})$$

In Chapter 4 I show that the cross-sectional radius of a *T. aurantia* spicule can be described by the parametric equations

$$r(\theta) = r_0 \sin(\theta), \quad (\text{D.6a})$$

$$x_1(\theta) = \frac{L_s}{\pi} \left(\theta - \frac{1}{2} \sin(2\theta) \right) - \frac{1}{2} (L_s - L), \quad (\text{D.6b})$$

where L_s is the spicule's length, and θ is a parameter that lies between 0 and π [27]. By substituting equations (D.6) into equation (D.5) I can compute E for a given r_0 , L and L_s . I assume that the spicules fail at mid-span and that therefore the radius of the cross-section at which the spicule fails is r_0 . I measure r_0 from scanning electron micrographs taken after the test. Since the *T. aurantia* spicules' lengths, L_s , are larger than the field of view of the microscope that is part of my mechanical testing device, I did not measure L_s directly for the spicules that I tested. However, in Chapter 4 I show that the aspect ratio $\lambda_s = L_s/2r_0$ of the *T. aurantia* spicules is also relatively constant [27]. Therefore, in equation (D.6) I estimate L_s to be $2\lambda_s r_0$ where $\lambda_s = 53.6$ [27]. Using equation (D.5), the Young's modulus of the *T. aurantia* spicules is 34.4 ± 7.0 GPa (mean $\pm$ standard deviation).

Appendix E

Estimation of the distance between adjacent *T. aurantia* spicules in a bundle

The arrangement of strongyloxea within a bundle is not well-characterized and is difficult to measure. In order to estimate the distance between neighboring strongyloxea in a bundle, I assume that they are evenly distributed within the bundle's cross-section. That is, they do not clump together in some regions of the cross-section and leave large expanses of spongin in others.

I represent a bundle's cross-section as a circular region with a radius $R_b = 177.5 \mu\text{m}$, which is half the mean thickness of a strongyloxea bundle [44]. I model the strongyloxea in this cross-section as N_s smaller circles all having a radius of $R_s = 18.3 \mu\text{m}$, which is the mean R_s from Chapter 4 Section 4.3.1. A previous study found that the cross-section of a strongyloxea bundle from a closely related species (*Tethya minutia*) contains anywhere from 10 to over 100 strongyloxea [135]. From these measurements I take an approximate average value and set $N_s = 50$.

To find what constitutes an evenly distributed arrangement of strongyloxea within a bundle, I treat the N_s smaller circles as if each has a positive electrostatic charge. Consequently, each circle exerts a repulsive force on every other circle and the magnitude of this force is inversely proportional to the square of the distance between them.

I describe the positions $(x_i^s, y_i^s)_{i=1 \dots N_s}$ of the centers of the smaller circles using a cartesian coordinate system whose origin lies at the center of the circle representing the bundle's cross-section. I

write the potential energy $q(i, j)$ of the i^{th} circle due to the presence of the j^{th} circle as

$$q(i, j) = \begin{cases} C_1 [H(i, j) - 2R_s]^{-1}, & H(i, j) > 2R_s, \\ \infty, & H(i, j) \leq 2R_s, \end{cases}$$

where $H(i, j) = [(x_i^s - x_j^s)^2 + (y_i^s - y_j^s)^2]^{1/2}$ is the distance between the two circles' centers, and C_1 is a constant. I set $q(i, j) = \infty$ when the distance between the circles is less than $2R_s$ since two strongyloxea cannot occupy the same points in space. The total potential energy of the system is then given by

$$Q = \sum_{i=1}^{N_s-1} \sum_{j=i+1}^{N_s} q(i, j).$$

Without loss of generality I choose $C_1 = 1$ and vary the positions of the circles $(x_i^s, y_i^s)_{i=1 \dots N_s}$ to minimize Q , subject to the constraint that $(x_i^s)^2 + (y_i^s)^2 \leq (R_b - R_s)^2$ for $i = 1 \dots N_s$. I minimize Q numerically for 50 random initial guesses of $(x_i^s, y_i^s)_{i=1 \dots N_s}$. I take the configuration of the circles corresponding to the smallest Q and compute the distance between each circle's center and the center of its nearest neighbor. The mean nearest neighbor distance in this configuration is $45.2 \mu\text{m}$, which I use as the diameter of my region of confinement in Section 4.3.3.

Appendix F

Details of the computational mechanics model for a *T. aurantia* spicule

I model the strongyloxea in its region of confinement as a rigid inclusion embedded in an elastic cylinder (see Figure F.1 (a)). Since my goal is to determine the qualitative nature of the traction distribution on the strongyloxea at the initial stages, i.e., prior to the strongyloxea undergoing any significant motion, I assume that my computational mechanics model is axisymmetric. This assumption is expected to be valid for the strongyloxea not lying on the surface of a bundle. Similarly, during the initial stages, the applied loads and deformation are likely to be symmetric across the lateral plane (see Figure 4.3 (a)). Therefore, I only consider half of the inclusion-cylinder pair in the computational mechanics calculations (see Figure F.1 (a)). Ideally, I would like to model the exact shape of a strongyloxea. However among all models I considered, I found that the Clausen profile describes the strongyloxea's shape the best. Therefore, I represent the shape of the inclusion, $r(z)$, using a Clausen profile whose length and aspect ratio are $\text{mean}(L_m)$ and $\text{mean}(\lambda_s)$, respectively. The length and diameter of the cylinder representing the region of confinement are $1.25\text{mean}(L_m)$ and $45\text{ }\mu\text{m}$ (see Appendix E), respectively.

The different surfaces in my computational mechanics model are marked in Figure F.1 (a). The inclusion and the cylinder are rigidly bonded along the surface Γ_1 . Due to the lateral symmetry, I prescribe the displacements in the z direction on the surface Γ_4 to be zero. I also prescribe the

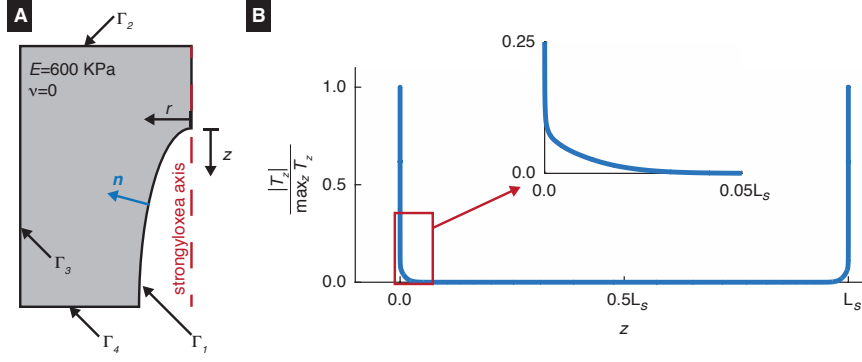


Figure F.1: Computational mechanics model of a strongyloxea embedded in an elastic matrix. (a) Model geometry. The surface between the inclusion and the elastic cylinder is denoted by Γ_1 . (b) Distribution of the axial force per unit length, T_z , along the strongyloxea's length. The inset shows a magnified view of the T_z distribution along the first 5% of the strongyloxea's length.

displacements in the r direction on the surface Γ_3 to be zero as a way of modeling the fact that the matrix surrounding a strongyloxea is also rigidly bonded to neighboring strongyloxea at distances roughly equal to the region of confinement's diameter. I apply uniform tractions on the surface Γ_2 that are parallel to the strongyloxea's axis. The magnitude of the applied traction is not important since I only wish to understand the qualitative nature of the traction distribution on the strongyloxea during the initial stages.

I computed the axial component of the traction on Γ_1 from the Cauchy stress components, σ_{ij} for $i, j \in \{r, z\}$, as

$$t_z = \sigma_{zz}n_z + \sigma_{rz}n_r, \quad (\text{F.1})$$

where n_r and n_z are the radial and axial components of the unit vector normal to Γ_1 . The components n_r and n_z can be computed from the inclusion's profile, $r(z)$, as

$$\begin{aligned} n_r &= (1 + r'^2)^{-1/2}, \\ n_z &= -r'(1 + r'^2)^{-1/2}, \end{aligned} \quad (\text{F.2})$$

where $r' = dr/dz$.

The net axial force transmitted across the strongyloxea's cross-section that is located at z' is $P_{\text{net}}(z') = \int_0^{z'} T_z(z) dz$, where

$$T_z(z) = 2\pi t_z(z)r(z)(1 + r'^2(z))^{1/2}$$

is the axial force per unit length acting on the strongyloxea. It can be seen, e.g., in Figure F.1 (b), that T_z is highly localized at the strongyloxea's end. Specifically, I find that approximately 95% of the total transmitted axial force, $P = P_{\text{net}}(L_s/2)$, is found on the first 5% of the strongyloxea's length. The idealization of a point force of magnitude P acting on the strongyloxea's end would correspond to $T_z(z) = P\delta(z)$, where $\delta(\cdot)$ is the Dirac delta distribution. As can be seen in Figure F.1 (b), the T_z distribution resembles a Dirac delta distribution.

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