Towards an Understanding of the Role of Aragonite in the Mechanical Properties of Nacre

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ABSTRACT

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Nacre, also known as mother-of-pearl, is a biocomposite material that exhibits higher strength and fracture toughness than its component materials. It derives its strength from the brick-and-mortar layering of aragonite (CaCO_3) platelets and organic binder. It is believed that the protein binder helps redistribute the stress throughout all tablets for optimal mechanical performance. In this study, we attempt to measure the mechanical properties of aragonite within nacre and compare them to bulk aragonite and bulk nacre and understand the redistribution of stresses. Here we show that x-ray diffraction techniques are useful for isolating and measuring strain of crystallites within a composite material. Our results show that the apparent stiffness of aragonite varies with crystallographic directions and is higher than the stiffness of bulk nacre in all cases, meaning that aragonite tablets are exposed to less than the average bulk stress. The average force applied to the bulk sample is partitioned between the aragonite and the binder, so that the protein layer bears as much as 27.2% of the total applied force. Different crystallographic directions exhibit behaviors different than bulk aragonite or bulk nacre. These are related to texture of aragonite platelets (i.e. preferred orientations within nacre). By examining nacre, we can obtain a better understanding of the mechanical relationship between the ceramic and polymer in composite materials. We expect that x-ray diffraction will become the standard method for probing the mechanical properties of composite materials.

INTRODUCTION

Nacre, commonly known as mother-of-pearl, is a biocomposite material found lining the shells of many mollusks. It has a non-isotropic microstructure composed of aragonite (CaCO_3) platelets bound together by organic polymers. Depending on the species, these platelets can be arranged in columns or in a staggered formation. The platelets are polygonal with diameter $10\text{ }\mu\text{m}$ and thickness of $0.3\text{ }\mu\text{m}$ [1]. Aragonite, which constitutes approximately 95% of nacre by volume [2], has an elastic modulus between 80 and 205 GPa, depending on the crystallographic direction [3, 4, 5, 6].

Bulk aragonite has a strength of 30 MPa. Nacre, however, has a strength of 170 MPa or more and exhibits brittle fracture upon failure [1, 2, 7]. Nacre has an extremely high interlaminar fracture toughness, believed to be created by the protein matrix [8, 9]. The elastic modulus has been measured to be 72.6 GPa [10]. Currey showed that nacre is extremely good at stopping cracks caused by external sources. The work of fracture has been measured to be 1650 J/m^2 - much higher than aragonite itself [1]. Fleischli et al. demonstrated that nacre has a fracture toughness of $2.9\text{ MPa m}^{1/2}$ [10], which is 3000 times higher than that of aragonite. The mechanical properties of nacre are much more desirable than those of aragonite alone, due to its composite structure.

To understand the emergent behavior in this composite material, the interactions between the individual components must be understood. To do so requires a characterization technique that allows the isolation of the properties of one material within the composite. X-ray diffraction is such a technique and therefore ideal for investigating the origin of nacre's emergent mechanical properties.

The aragonite platelets (each a single crystal) diffract an incident x-ray beam. The organic material is not crystalline in nature and makes only 5% by volume of nacre; therefore its diffraction pattern is very weak and not easy to separate from the diffraction pattern for

aragonite. The diffraction pattern, collected with a large area detector, can be analyzed with reciprocal space techniques to determine the strain present in the aragonite tablets [11].

In this work, we are investigating why nacre has superior properties when it is composed of weak materials. To do this, we will characterize the partitioning of forces between aragonite and organic polymer in nacre. We will restrict this study to uniaxial load as it presents the simplest loading geometry. As uniaxial loads are applied, the apparent elastic modulus for aragonite in nacre can be determined and can be used to characterize force partitioning. Elastic modulus can therefore easily be extracted without making many assumptions. This will establish methods for examining crystalline components of composite materials while explaining the relationship between the structure and strength of nacre.

MATERIALS AND METHODS

Epoxy Selection

Previous studies in this lab to examine properties of nacre under tension were limited by the method to attach samples to the test rig. Samples tested during July, 2008 used A+B Epoxy Paste, experienced failure at the epoxy-sample boundary. As a result, the tensile region of the stress-strain curve could not be determined. In order to collect tensile data, a stronger epoxy was required.

Epoxies to test were selected based on shear strength published in technical data sheets. 3M 1838 B/A Green epoxy has a reported shear strength of 3000 psi [12] and Armstrong A-661 [13] has a shear strength of 3300 psi. These two epoxies were selected for testing along with epoxies used previously in this lab. Of these epoxies, A+B Fast Epoxy Paste and Loctite QuickSet 5 Minute Epoxy did not provide technical data sheets.

Epoxies were tested using aluminum holders designed to simulate the conditions present in tensile testing of nacre. The holders have slots with width and depth of 1.9 mm and a

length of 5.7 mm. This creates an aspect ratio for the slot of 3.00, which is close to the that of the nacre grips which have an aspect ratio of 2.85. Steel wires with diameter 1.4 mm were prepared by sanding the surface to be bonded in order to roughen it. This surface was coated in epoxy and placed in the holders to cure overnight. A tensile load was applied to the samples with a MTS test rig with a 1 kN load cell at Stanford University. Samples were elongated at $10 \mu\text{m s}^{-1}$ until failure occurred. The load at failure was compared to evaluate the relative strengths of these epoxies.

Internal Standard Selection

A powder x-ray diffraction standard was desired for calibration of x-ray diffraction images. A wrongly calibrated image can show strain in the diffraction rings that is not present in the sample. The calibration standard would be a powder applied directly to the sample. Then each image can be calibrated to the standard powder rings before strain analysis is performed. The ideal standard would have well-defined diffraction rings that don't interfere with any of the diffraction rings of nacre. Such a standard could not be found - every standard we examined overlapped with some of the nacre diffraction rings.

Diffraction data was calibrated instead with external LaB_6 powder placed next to the sample. LaB_6 is widely used as a diffraction standard. Silver powder applied to one edge of the gage section was selected as a secondary internal standard despite overlap with nacre rings.

Sample Preparation

Rectangular prismatic samples of red abalone were prepared by Karen Magid and coworkers in Zurich in a materials science laboratory. Sample cross-sections ranged from $88,900 \mu\text{m}^2$ to $185,760 \mu\text{m}^2$. The corresponding maximum stresses for these samples ranges from 237 to 495 MPa at a force of 44.5 N (the upper limit of the load cell). Red Abalone was selected

for its widespread use in scientific research.

Experimental Setup

The tension/compression test rig is comprised of the load cell, grips, and a strain gage. The load cell, shown in Figure 1, has an upper limit of 44.5 N. Epoxy was applied to samples and test grips and allowed to cure overnight at room temperature. Silver paint was applied as a secondary calibration standard to one edge of the gage section. Black spray paint was applied for use with a Digital Image Correlation (DIC) camera. This camera acts as an optical strain gage to find bulk strain of nacre, while diffraction techniques find microscopic strain of aragonite only. Figure 2 shows the entire experimental setup including the position of the DIC camera, large-area detector, sample, and x-ray beam.

Data Analysis

Diffraction data was analyzed using Area Diffraction Machine (ADM) [14] and Matlab. ADM was used to calibrate the data, convert circular diffraction patterns (Figure 3) to rectilinear coordinates (Figure 4), and export peak lists showing the position of the diffraction rings. These rings were then analyzed with Matlab scripts to extract 3-dimensional strain for each load point, diffraction ring, and position along the length of the sample. Diffraction patterns were recalibrated using LaB_6 standard at each load point to account for possible beam and sample movement. Stress-strain curves and elastic moduli were extracted from this data. Gaps in the diffraction rings represent a preferred orientation of the tablets. This direction preferences is called texture and is seen in many biological materials. Due to limitations on time, texture was not included in this analysis.

RESULTS

The results from the epoxy experiment are detailed in Table 1. The epoxy adhered to two components with different adhesion areas and the tested strength was a combination of tensile and shear strength. For this reason, the data was left in units of force instead of stress. But under identical conditions, 3M and Armstrong epoxies performed significantly better than previously used epoxies.

Figure 5 shows the fracture surface of an epoxy sample. The epoxy separated from the steel sample along half of the curved surface. The flat sections along the bottom of the fracture surface show where the epoxy separated from the aluminum holder. Between those two regions is a jagged edge where the epoxy separated from itself. Steel-epoxy, epoxy-epoxy, and aluminum-epoxy failures were exhibited in this fracture surface. This fracture surface indicates that the three different bonds between materials have roughly the same strength in this failure mode.

In uniaxial loading, red abalone exhibited strain in x-ray diffraction rings as well as DIC. A sample diffraction pattern is shown in Figure 3. The best rings for tension and compression were selected based on the intensity, lack of near overlap with adjacent rings, and completeness of the ring (many rings were incomplete due to texture). Compression and tension data were collected on unloading, as recommended by ASTM standards. Unloading compression and tension data were analyzed separately, then combined to form one full data set encompassing both. Figure 4 shows diffraction rings after being “unwrapped” by ADM. Strain is seen as wiggles in the unwrapped peak lines.

Table 2 shows the apparent elastic modulus values for different hkl crystallographic directions for tension and compression. Units are in GPa. Lattice parameters are as follows: $a = 4.96 \text{ \AA}$, $b = 7.97 \text{ \AA}$, and $c = 5.74 \text{ \AA}$. Values range from 79.3 GPa ([232] in tension) to 119.3 GPa ([200] in compression). The value of elastic modulus as measured by DIC was

72.4 GPa, lower than any value of aragonite elastic modulus. This result is preliminary — further DIC data analysis will be published by Magid et al. in the near future.

Figure 6 shows stress-strain plots for 3 normal directions and 3 shear directions. The applied displacement is in the yy -direction. The x-ray beam is in the xx -direction where there are the fewest data points, so error on xx -strain is much larger and data is often skewed by small variations in the x-ray beam. The zz - and yy -data is unaffected by small variations and the error is much smaller.

By comparing the elastic modulus in all three normal strain directions, we can determine Poisson’s ratio for aragonite in nacre. This value was measured to be 0.33. There was no correlation between measured strain and shear strain in any diffraction ring.

The force felt by each platelet can be estimated by examining the elongation in each crystallographic direction. By comparing the apparent elastic modulus measured by x-ray diffraction to the theoretical modulus based on the compliance tensor, we can find the stress intensity factor. This factor describes the force felt by tablets aligned in a particular direction.

$$E_{meas} \frac{SI_A}{1 - x} = E_{theo} \quad (1)$$

where E_{meas} and E_{theo} are the measured and theoretical elastic moduli, SI_A is the stress intensity factor for the protein, and x is the volume fraction of protein (taken to be 5%). This model is derived from a force balance and assumes that forces are distributed between aragonite platelets and the protein.

The stress intensity factor is used to compare the force felt by each individual component to the average applied force. A stress intensity factor less than 1 indicates that a component is exposed to less than its volume fraction of force. Data from Table 3 shows stress intensity values for aragonite. The stress intensity factor for aragonite is exactly the same as the ratio

of the theoretical modulus of aragonite to the apparent (measured) modulus.

DISCUSSION AND CONCLUSIONS

The epoxy selected for use on the final samples was 3M 1838. The tested strength was 3% lower than the Armstrong epoxy. However, the 3M epoxy was much easier to measure, mix, and apply than the Armstrong epoxy. 3M 1838 was selected for use with the nacre samples due to its greatly increased strength over previously used epoxies.

X-ray diffraction is a valid technique for measuring strain in composite materials. Microstrain information, unavailable from conventional methods to measure strain, is easily found by examining the diffraction pattern of aragonite in nacre. This technique can be applied to a wide range of composite materials of natural and synthetic origin.

The measured value for Poisson's ratio of 0.33 fits perfectly with that of many other materials (a general rule of thumb is Poisson's ratio ≈ 0.3). This falls within the range for aragonite measured by Zhang et al. [15]. More data is required to measure the Poisson's ratio in different crystallographic directions. Poisson's ratio can be observed in the left plots of Figure 6 by comparing the slopes (elastic moduli) in the xx -, yy -, and zz -directions. The three shear plots in the right plots of Figure 6 show that the applied force is uniaxial, as expected, and there is no relationship between stress and shear strain.

For all hkl directions, the value for combined tension/compression data was closer to the tension-alone data. The data suggests that compression is consistently stiffer than tension, but the difference between the two is comparable to the error. Tension and compression data were grouped together to form one data set for further analysis. The different crystallographic directions exhibit elastic moduli anisotropically (see Table 2). This is the first indication that aragonite in nacre has an altered mechanical response.

Data tabulated in Table 3 shows that different crystallographic directions bear different

amounts of total applied force. The first column is the calculated value for stress intensity factor of aragonite based on Equation 1. The second column is the ratio of theoretical to apparent elastic moduli. This should tell us the actual force experienced by each crystallographic direction. A value higher than 1 indicates that this direction experiences more stress than expected. The third column gives the estimated percentage of the load borne by protein based on the stress intensity factor for aragonite.

It appears that the estimated stress intensity factor is significantly different along certain crystallographic directions. For example, the [031] direction is 0.76 whereas the [202] direction is 1.29. Measurement of the stress intensity factor for aragonite in different crystallographic directions helps to better understand this effect. Future studies on the role of texture in nacre would elucidate the relationship between crystallographic direction and force partitioning. A detailed texture analysis could be used to find the crystallographic direction-averaged strain. Nacre samples taken in different directions could also be used to determine the role of texture in the mechanical properties of nacre.

Nature harnesses the anisotropic behavior of aragonite to create nacre, which exhibits anisotropic properties distinct from aragonite's. This additional anisotropic behavior is created by biological processes to create a superior composite material. From measured strains, we can deduce that the polymer layer bears different amounts of the applied force in different directions. The protein layer alone is not the sole source of nacre's strength and toughness — texture plays a crucial role as well.

Protein constitutes only 5% of nacre by volume, but it can absorb up to 27.2% of the applied force (seen in the [031] direction). The protein layer works to shield weak properties, such as strength of aragonite, from excessive force. The texture present in nacre helps ensure that the strongest crystallographic directions of aragonite absorb the most stress. Arrangement of aragonite platelets and interactions with the biopolymer helps partition forces among strong and weak directions ultimately resulting in superior material properties.

However, to fully quantify this effect, texture studies are required.

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FIGURES

Figure 1: The test rig. The load cell is located on the left. The green epoxy and silver calibration paint are visible on the sample.

Figure 2: The x-ray diffraction setup. The x-ray beam enters from the right and diffracts off the sample. Diffraction patterns were collected using a large area detector (white box with black circle on left). The DIC camera is located above the sample.

Figure 3: A circular diffraction pattern collected with a large area detector.

Figure 4: Diffraction rings after being “unwrapped”. (a) Rings exhibiting no strain. Peaks form a straight line. (b) Rings exhibiting compression. (c) Rings exhibiting tension.

Figure 5: The fracture surface of a tensile epoxy sample.

Figure 6: Stress-strain curves for 3 normal directions (left column) and 3 shear directions (right column). Scale is the same on all plots. The applied displacement is in the yy -direction.

TABLES

Epoxy	Yield strength
Armstrong A-661	394.5 ± 24.5
3M 1838	380.5 ± 30.5
A+B Epoxy Paste	134.0 ± 50.5
Loctite	127.7 ± 69.8

Table 1: Yield strength for an epoxy bond between aluminum and steel. Units are Newtons.

Crystallographic Direction hkl	Measured Elastic Modulus			Theoretical Modulus
	Compression	Tension	Combined	
200*	119.3 ± 7.7	100.5 ± 4.1	105.6 ± 1.9	142.9
232	84.4 ± 5.1	79.3 ± 3.7	79.6 ± 1.3	86.7
321	96.1 ± 7.4	95.7 ± 6.3	97.6 ± 2.0	106.2
231	93.9 ± 4.8	89.7 ± 3.0	90.1 ± 1.2	91.3
141	85.7 ± 4.1	85.2 ± 2.1	83.9 ± 1.0	80.5
202	89.0 ± 4.3	84.0 ± 2.0	81.9 ± 1.1	67.7
031	108.7 ± 6.2	97.2 ± 3.4	98.7 ± 1.5	79.6

Table 2: Elastic modulus for different crystallographic directions in tension and compression, arranged in order of decreasing stress intensity (see Table 3). Units are GPa. *Near overlap with [102].

Crystallographic Direction hkl	SI _A	% load borne by protein
200*	1.29	-22.1%
232	1.03	1.7%
321	1.03	1.8%
231	0.96	8.5%
141	0.91	13.4%
202	0.79	25.4 %
031	0.76	27.2%

Table 3: Values for stress intensity factor and percent load borne by protein. The stress intensity factor is the same as the ratio of theoretical elastic modulus to apparent (measured) elastic modulus of aragonite. *Near overlap with [102].