

Annual Review of Physical Chemistry Biomimetic Structural Materials: Inspiration from Design and Assembly

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Abstract

Nature assembles weak organic and inorganic constituents into sophisticated hierarchical structures, forming structural composites that demonstrate impressive combinations of strength and toughness. Two such composites are the nacre structure forming the inner layer of many mollusk shells, whose brick-and-mortar architecture has been the gold standard for biomimetic composites, and the cuticle forming the arthropod exoskeleton, whose helicoidal fiber-reinforced architecture has only recently attracted interest for structural biomimetics. In this review, we detail recent biomimetic efforts for the fabrication of strong and tough composite materials possessing the brick-and-mortar and helicoidal architectures. Techniques discussed for the fabrication of nacre- and cuticle-mimetic structures include freeze casting, layer-by-layer deposition, spray deposition, magnetically assisted slip casting, fiber-reinforced composite processing, additive manufacturing, and cholesteric self-assembly. Advantages and limitations to these processes are discussed, as well as the future outlook on the biomimetic landscape for structural composite materials.

1. INTRODUCTION

Strength: a measure of the maximum amount of stress, or force per unit area, a material can withstand before fracturing

Toughness: a

measure of the amount of energy a material can absorb before fracturing The design and fabrication of materials possessing desired mechanical, optical, thermal, and electrical properties has been a critical goal in the field of materials science and engineering (1, 2). Over the past several decades, man-made techniques to process these materials have been largely successful, yet still exhibit numerous drawbacks (1, 2). Traditionally, these processes are labor-intensive, require conditions of high temperature and/or pressure (which can be energy demanding and costly), and rely upon the use of harsh or environmentally unfriendly (acidic, basic, toxic) chemicals and components (1, 2). Moreover, it is well known that the desired properties (mechanical, optical, thermal, electrical, etc.) for end applications are highly dependent on the material composition and structure, which in turn are related to the processing conditions and techniques. The ability to predict optimal structural designs for an end application and to precisely and uniformly control the nano- and microstructure and composition over large areas remains a great challenge in materials engineering (3–7). Thus, in the past decade, there has been increasing interest in developing novel processing techniques in order to address these issues and fabricate materials with a high level of control (3–11).

Nature provides an excellent inspiration for the design and fabrication of next-generation functional materials. For hundreds of millions of years, nature has evolved efficient strategies for synthesizing a diverse range of materials. These materials exhibit exceptional mechanical, thermal, and optical properties while also incorporating multifunctionality, including features such as adapting, sensing, and self-repair (11–15, 125–127). Organisms rely upon these structures for functions such as structural support, feeding, and defense against predators. Such diversity in material properties and functions is remarkable, considering the limited set of constituent elements and building-block materials nature has at its disposal.

Biological materials, which are typically found as composite structures, can be divided into nonmineralized (typically soft) and mineralized (hard) tissues. Both contain organic components, typically soft biopolymers such as proteins (e.g., collagen, keratin, and elastin) or polysaccharides including chitin, cellulose, hemicellulose, lignin, and suberin (12–15). Whereas nonmineralized tissues typically derive their strength from intermolecular bonding, such as hydrogen bonding or covalent cross-linking of polymer chains, biomineralized tissues additionally incorporate stiff inorganic crystals to achieve high strength. The most common inorganic components, which are of either amorphous or crystalline form, include calcium carbonate, calcium phosphate, silicon dioxide, and even iron oxide (13–15). Although the soft biopolymer and stiff, biologically mineralized ceramic constituents are not inherently strong or tough, respectively, the resultant biological composites are both (12–15). This can be attributed to the precise manner in which they are hierarchically assembled from the bottom up, yielding complex yet well-defined structures at the nano-, micro-, and macroscales.

Beyond learning from nature about the unique structural designs that yield exceptional mechanical, thermal, or optical properties, much can be gleaned from studying how nature fabricates its materials. Unlike conventional synthetic materials processing, nature synthesizes its materials under mild conditions: ambient temperature, ambient pressure, and near-neutral pH. Understanding the parameters that control biological self-assembly and mineralization can provide new insights into low-energy processing of materials. For example, in biomineralized tissues, biopolymers (proteins, peptides, and polysaccharides) containing specific chemical functionalities are often used to control the nucleation and growth of minerals. The result of this directed synthesis is precise control of inorganic phase, crystallinity, morphology, orientation, and gradients, contributing to the end-material properties.

Whereas nature has a fairly limited selection of constituent materials from which to choose in constructing its structures, today's scientists and engineers have a wide range of synthetic material

components that possess high strength or high toughness. Thus, by studying and understanding both the synthesis–structure and structure–property relationships of biological materials, we can gain insight useful for the efficient fabrication of next-generation high-performance multifunctional materials. These ideas are at the heart of biomimetics. Biomimetic materials engineering, which differs from bio-inspired or biomediated materials engineering, focuses on recreating or mimicking the structural design of biological materials or the process by which biological materials are synthesized (16, 17). Although it is closely related, we define bio-inspired materials engineering as taking design cues from natural fabrication processes and applying them to other synthetic systems. An example of bio-inspired processing would be the synthesis of nanostructured oxides that utilize synthetic control parameters exhibited by biological systems (i.e., pH, precursor concentration, organic ligands, etc.) to yield materials with controlled size, shape, and phase. Biomediated engineering focuses on implementing or incorporating the inherent biological elements into some other system to take advantage of the properties of that biological material. An example would be utilizing enzymes as a platform to synthesize semiconducting oxide materials (18–20).

Because the field of biomimetics and bio-inspired and biomediated materials is vast, this review focuses only on structural and mechanical aspects of materials that mimic specific biomineralized tissues. We begin this review by examining structural design elements and structure-mechanical property relationships of two biomineralized tissues that have been well documented in the recent literature: the nacreous layer of a mollusk shell and the arthropod cuticle. We discuss biomimetic efforts to replicate the structure and mechanical properties of these natural materials. In addition, we provide a brief discussion of biomimetic routes toward calcium carbonate and phosphate minerals. We conclude by discussing limitations and challenges to the field as well as an outlook on the future of biomimetic materials engineering.

2. BIOMIMETIC STRUCTURAL PROCESSING

For decades, the nacreous structure found within the inner lamellar layer of mollusk shells has provided scientists and engineers with a blueprint for fabricating tough and strong materials. Its brick-and-mortar design has been considered the gold standard for biomimetic composites (5, 6, 21, 22). Recently, another architecture, a fibrous helicoidal (Bouligand) composite that provides enhanced toughening over that from the nacreous design, was revealed in the cuticles of arthropods (23). Here we describe the structure, mechanical properties, and formation of the nacre and helicoidal structures, and we provide a review of the biomimetic processing used to replicate both.

2.1. Structure, Mechanics, and Formation of Nacre

Biomimetic structural processing begins with a concrete knowledge of the multi-length-scale structure and organization of components within the biological material. This knowledge, coupled with mechanical analysis, can reveal mechanisms contributing to the overall strength and toughness of the material. Understanding the natural assembly and crystallization routes can then provide clues as to appropriate biomimetic processing routes.

2.1.1. Structure. Many marine organisms of the Mollusca phylum rely on exoskeletal structures (i.e., shells) for support and protection against predators. Mollusk shells feature a multilayered structure, with different layers containing unique microstructures, the most common of which are prismatic, nacreous, and cross-lamellar (24). Nacre, also known as mother of pearl, is the structure forming the inner layer of many mollusk shells and has become one of the most widely studied biological materials in the literature, largely because of its impressive mechanical properties and unique underlying composite architecture (**Figure 1**) (25–36). Nacre is composed of 95%

Biomimetics:

imitating or replicating biological structures

Bio-inspired materials engineering:

applying biological principles of fabrication to synthesize new materials

Biomediated materials engineering:

implementing or incorporating an inherent biological material into another system to take advantage of the properties of that biological material

Nacre:

the brick-and-mortar composite structure that makes up the inner layer of some mollusk shells

Arthropod cuticle:

the outer exoskeletal structure that provides protection and support

Helicoid: a laminated structure featuring stacked layers of unidirectional fibers, where each fiber layer is rotated from the adjacent layer about an axis normal to the fiber direction, producing a twisted appearance



Figure 1

Ultrastructure and toughening mechanisms in nacre. (*a*) Overview of inner iridescent region of shell from the smooth Australian abalone (*Haliotis laevigata*). (*b*) Scanning electron micrograph (SEM) of fractured cross section of the nacreous layer from *H. laevigata*. (*c*) High-magnification SEM micrograph showing mineral nanoasperities on the surface of tablets and mineral bridging between tablet layers after deproteination treatment. (*d*) Schematic of the brick-and-mortar architecture (*top*) and SEM micrograph showing tablet pull-out under tensile loading (*bottom*). (*e*) Schematic showing different modes of tablet sliding and pull-out. (*f*) SEM micrograph showing tablet showing tablet waviness and dovetail-like structure providing resistance to pull-out. Panels *a* and *b* adapted from Reference 44; panel *c* adapted from Reference 31. Panels adapted with permission.

inorganic and 5% organic material by volume, yet remarkably achieves strength and fracture toughness several orders of magnitude higher than those of its constituent materials (25, 27, 28). This has been attributed to its well-ordered microstructure, featuring a brick-and-mortar-like arrangement of inorganic and organic constituents. Polygonal tablets composed of biomineralized aragonite, an orthorhombic polymorph of calcium carbonate, make up the inorganic component. These tablets, which are approximately 500 nm thick and 5–15 μ m in diameter, stack to form densely packed parallel lamellae (**Figure 1***b*). The lamellae are separated by porous sheets of organic material (20–30 nm thick) comprising the organic matrix, which is composed of a mixture of β -chitin polysaccharide, fibroin silk-like proteins, and a variety of acidic proteins (24, 37). The templating effect of the organic matrix components on the nucleation and growth of aragonite also results in the tablets being nearly single crystalline and crystallographically aligned, with their *c*-axes [i.e., (001) lattice planes] aligned nearly perpendicular to the plane of the tiles, which contributes to the anisotropic mechanical properties (38–40). At the mesoscale, mineral nano-asperities coat the surface of aragonite tablets, providing roughness, and some of these asperities cross plate boundaries, fusing them together via mineral bridges (**Figure 1***c*) (34, 38).

2.1.2. Mechanics. Much work has been done to characterize the mechanical properties of nacre and to identify the strengthening and toughening mechanisms yielding nacre's enhanced damage tolerance and fracture resistance (25, 27-32, 34). Sarikaya et al. (35) conducted three- and fourpoint bending tests on single-edge notched beam samples of the red abalone, Haliotis rufescens. Samples were loaded in the transverse direction, perpendicular to the tablet planes (35). The samples had a fracture strength of 185 ± 20 MPa and a fracture toughness, $K_{\rm IC}$, of 8 ± 3 MPa \cdot m^{1/2} (35). Other studies reported a tensile strength of 100–170 MPa when the nacre was loaded parallel to the layer plane (34, 36). Intuitively, nacre achieves its high strength because of its high volume fraction of stiff ceramic material; the origins of its toughness, however, are less obvious. Several mechanisms have been attributed to its high toughness. First, crack deflection at the hard/soft interfaces between the ceramic tablets and organic matrix leads to a tortuous crack path, providing an enhanced work of fracture (27–31, 34). Second, viscoplastic deformation of the interlamellar organic sheets allows for tablet sliding (Figure 1d), providing some ductility (27-31, 34). Finally, resistance to tablet sliding under tensile loading due to mineral nanoasperities on the surface of tablets, mineral bridging between tablet layers (Figure 1e), and wavy tablet morphology provides an interlocking and strain hardening effect (Figure 1f) (27-31, 34).

2.1.3. Formation. Aside from structure–mechanics relationships, the formation mechanisms of the nacre structure, namely the self-assembly of the organic matrix and the role of organic macro-molecules in controlling the nucleation and growth of aragonitic crystals, has attracted much interest from the biomineralization community (37, 41–43). Although organic polysaccharides and proteins make up only 5% of nacre by volume, these biopolymers play a critical role in the formation of the well-ordered and crystallographically aligned mineral phase.

Nacre growth occurs by the alternate growth and arresting of mineral tablet layers controlled by the secretion and deposition of interlamellar organic matrix material (**Figure 2**). Heinemann et al. (44) and Nudelman (24) recently provided reviews on the mechanisms of crystal nucleation and growth in nacre. Initially, an organic layer is secreted. Calcium carbonate then nucleates on the surface of this layer and grows in all directions, with the fastest growth along the *c*-axis. Thereafter, a second organic layer is secreted, which arrests mineral growth in the *c*-direction and allows growth to continue in the *a*- and *b*-directions. However, small pores within the organic layer allow the mineral to continue growing in the *c*-direction, through the membrane, and seed the growth of new tablets in the next layer. This process repeats itself, resulting in the characteristic Christmastree pattern shown in **Figure 2**. The continuation of mineral across tablet layers accounts for the observed mineral bridges that also display crystallographic continuity (**Figure 2***f*). Although the specific mechanism of nucleation and growth of mineral is still under debate, it is generally accepted that acidic proteins containing residues with certain chemical functionalities (e.g., carboxylates, sulfates) adsorbed to the chitinous scaffold are responsible for initiating nucleation and arresting growth of the aragonite tablets (24).

2.2. Nacre-Mimetic Materials

Because of the remarkable mechanical performance of nacre, significant efforts have been taken to mimic its hierarchical structure using engineering materials to create high-performance composites exhibiting exceptional stiffness, strength, and toughness. In theory, engineered composite materials could be manufactured that exceed the mechanical properties of nacre, given the diverse range and performance of synthetic building-block materials, as well as processing conditions, that engineers have at their disposal.

In this section, we examine recent biomimetic efforts to replicate the nacre structure, focusing on the materials used, fabrication methods, and the resulting structural features and mechanical

Nacre (hypostracum)

Prismatic calcite (ostracum) Periostracum



Figure 2

Nacre formation mechanism. (*a*) Schematic of cross section through growing shell of *Haliotis laevigata*. (*b*) Schematic showing development of nacreous tiles through repeated arrest and growth of CaCO₃. (*c*) Scanning electron microscopy micrograph showing a Christmas-tree pattern of growing nacre tablets. (*d*) Schematic showing various stages of tablet growth: (*i*) deposition of organic layer arresting tablet growth in the *c*-direction; (*ii*) continued lateral growth in the *a*- and *b*-directions with small amount of *c*-direction growth into next layer through pores of organic layer; (*iii*) formation of new tablet in adjacent layer with growth in all directions; and (*iv*) *c*-axis growth arrested with deposition of new organic layer. Processes *i-iv* are repeated. (*e*) Transmission electron microscopy (TEM) micrograph showing nacre tablet growth. (*f*) TEM micrograph of nacreous layers and selected area electron diffraction patterns confirming crystallographic continuity across tablet layers through mineral bridging. Panels *a* and *e* adapted from Reference 44; panels *b*-*d* adapted from Reference 14; panel *f* adapted from Reference 38. Panels adapted with permission.

performance. For in-depth reviews of nacre-inspired layered composites, the reader is referred to the works of Yao et al. (6, 21), Cheng et al. (45), and Wang et al. (5). The focus here is on more recent reports of nacre-mimetic materials. Additionally, we focus on three of the most prominent fabrication techniques reported in the literature: freeze casting, layer-by-layer (LBL) deposition, and self-assembly.

2.2.1. Freeze casting. Inspired by the process by which salts and other impurities become expelled from ice crystals growing from seawater and are trapped within the channels between them, freeze casting is a technique that allows for the formation of well-ordered, layered porous materials (46, 47). It has become a popular technique for the mimicry of nacre, whereby the controlled unidirectional freezing of ceramic suspensions can yield oriented lamellar microstructures. Deville (7) provided a review of the freeze-casting process and of recent progress in producing porous ceramic materials. As the water freezes and the growing ice front advances, ceramic particles are repelled from the ice–water interface and settle, becoming trapped between the growing dendritic ice crystals, as shown in **Figure 3***a*. The ice can then be removed by sublimation via freeze drying, leaving



Figure 3

Freeze-cast nacre-mimetic structures. (*a*) Schematic phase diagram describing the freeze-casting process. Ceramic slurry is initially prepared. Controlled freezing of the solvent traps ceramic particles between the growing crystals. The solvent is then sublimated, leaving behind an ordered lamellar structure, which can then be sintered to fuse the ceramic particles. (*b*) Design strategy for the production of nacre-mimetic alumina composites by freeze casting. (*c*) Schematic showing densification of the composites by rearrangement of silica and calcia particles and filling of void spaces between platelets. (*d*) Scanning electron microscopy (SEM) micrograph of fractured surface of alumina nacre-mimetic structure. (*e*_i*f*) SEM micrographs of single-edge notched beam test specimens showing long-range crack deflection and crack bridging. (*g*) Stress–strain curve showing results of single-edge notched beam testing comparing natural nacre, nacre-like alumina at 25°C, and nacre-like alumina after heat treatment to 600°C. The heat-treated alumina retains a high fraction of its original strength and fracture toughness. Shading indicates where test parameters are outside an acceptable range and no longer comply with standard testing procedure per the American Society for Testing and Materials (ASTM). (*b*) Flexural strength of alumina composites with varying composition of constituents. The alumina nacre-mimetic composite containing alumina nanoparticles as well as a silica-calcia liquid phase shows the best performance. Panel *a* adapted from Reference 7 with permission; panels *b*–*b* adapted from Reference 49 with permission.

behind a lamellar structure reminiscent of the aragonite bricks (without the mortar) in nacre. Subsequent sintering of the ceramic structure followed by infiltration of the void space with a second polymer can effectively replicate the organic matrix, yielding a two-phase nacre-like composite structure. By carefully controlling the freezing kinetics, specifically the speed of the solidification front, Deville et al. (46) were able to vary the thickness of ceramic (alumina and hydroxyapatite) lamellae from 1 μ m to 200 μ m without affecting the long-range ordering. In addition, a most intriguing result, stemming from the trapping of ceramic particles between ice dendrites and from the occasional dendrite spanning the channels between lamellae, was the formation of roughness on the lamellae's surface and of linkages between lamellae, reminiscent of the nanoasperities and mineral bridges found in nacre (46). After infiltration of alumina scaffolds with an aluminumsilicon alloy, the alumina/Al-Si composites (45/55 vol%) were able to achieve a strength of 400 MPa and a fracture toughness of 5.5 MPa \cdot m^{1/2} under three-point bend loading. Moreover, after doping of the alloy with 0.5 wt% titanium, the strength and fracture toughness increased to 600 MPa and $10 \text{ MPa} \cdot \text{m}^{1/2}$, respectively (46). Munch et al. (48) used a similar freeze-casting process to create aluminum oxide/polymethyl methacrylate (PMMA) composites, which exhibited yield strength, fracture toughness, and maximum strain to failure of 200 MPa, 30 MPa · m^{1/2}, and 1.4%, respectively. Ceramic bricks were 5–10 μ m wide and 20–100 μ m long. The authors were able to achieve densely packed, high-volume fraction (up to 80 vol% ceramic) composites through additional pressing (perpendicular to the lamellae) and double sintering steps, which helped reduce the average lamella thickness to 5 µm and promote the formation of ceramic bridges between lamellae (48). Moreover, the addition of sucrose to the slurry mixture assisted in the formation of ice crystals with microscopic roughness, which translated into ceramic tablets with rough surfaces and also promoted interlamellar bridging (48). Indeed, the adhesive properties at the inorganicorganic interfaces play an important role in the inelastic deformation response in nacre. Here, chemical grafting of the methacrylate group onto the alumina surfaces prior to PMMA infiltration ensured stronger covalent bonding between the ceramic and polymer phases and resulted in a noticeable improvement in the strength and crack-initiation fracture toughness, $K_{\rm IC}$ (48). Bouville et al. (49) eliminated the ductile polymeric phase and created an alumina nacre-like mimetic material using only ceramic constituents (Figure 3b-b). The aqueous colloidal suspension contained 100-nm alumina nanoparticles (98 vol%) mixed with a liquid-phase precursor containing 20-nm silica (1.3 vol%) and calcia (0.2 vol%) nanoparticles. The role of this smaller nanoparticle phase was to fill the remaining gaps between alumina lamellae, mimicking the organic matrix of nacre, and resulted in nearly fully dense (>98% relative density) samples after a pressure-assisted sintering step to 1,500°C (Figure 3b,c) (49). Alumina bridges and nanoasperities, reminiscent of nacre, were also observed within the structure. Although the structure featured no ductile components, the resultant ceramics exhibited an impressive combination of strength (470 MPa), toughness (22 MPa \cdot m^{1/2}), and stiffness (290 GPa) under single-edge notched beam testing while also exhibiting mechanical stability up to 600°C (Figure 3g) (49). Toughening mechanisms such as crack deflection and crack bridging were also observed in the single-edge notched beam-loaded specimens (Figure 3e,f) (49). Bai et al. (50) recently introduced a bidirectional freezing technique capable of imparting large-scale, long-range (centimeter-scale) ordering of aligned single-domain lamellar nacre-like structures. This was achieved using a polydimethylsiloxane wedge of varying slope to cover the cold finger, which imparts dual temperature gradients, allowing for the control of ice growth in both the vertical and horizontal directions and eliminating the formation of randomly oriented submillimeter-scale lamellar domains in the plane perpendicular to the freezing direction (50). The technique was used to create hydroxyapatite/PMMA nacre-mimetic composites as large as $4 \times 8 \times 25$ mm that displayed an excellent combination of properties: elastic modulus of \sim 20 GPa, bending strength of \sim 100 MPa, and work of fracture as high as 2,075 J/m², two orders of magnitude higher than that of monolithic hydroxyapatite (51). The composites possessed ceramic content as high as 75–85 vol% as a result of a postcasting/sintering uniaxial compression step to densify the scaffold (originally 70 vol% porous, 30 vol% ceramic). PMMA was incorporated by grafting 3-(trimethoxysilyl) propylmethacrylate onto the scaffold, followed by infiltration and in situ polymerization of methyl methacrylate (51). Although hydroxyapatite is not a structural ceramic, these nacre-mimetic hydroxyapatite/PMMA composites offer promise as bone substitute materials in orthopedic applications because of their improved inorganic volume fraction and mechanical properties compared to similar hydroxyapatite composites reported in the literature (51).

2.2.2. Layer-by-layer deposition. LBL assembly is a bottom-up processing technique that allows for the precise ordering of nanoscale building blocks. Based on the sequential adsorption of single monolayers, LBL deposition yields multilayered materials exhibiting well-defined nanostructures. As a result, the LBL technique has become a popular method to produce biomimetic nacre-like materials. Podsiadlo et al. (52) created inorganic-organic nanocomposites of montmorillonite clay with nanoplatelet morphology (sheets with thickness of ~ 1 nm and diameter of 100–1000 nm) and polyvinyl alcohol (PVA). The films were fabricated by sequential immersion of a clean glass slide (substrate) into 1 wt% PVA solution and 0.5 wt% montmorillonite, with washing and drying steps between each immersion. The procedure could be repeated until the desired number of layers had been deposited (52). Films were post-treated with glutaraldehyde to further facilitate bonding between the polymer matrix and clay tablets through covalent bonding of the -OH groups in the polymer chains to hydroxyls on the montmorillonite sheets (52). Crosssectional analysis of 200- and 330-bilayer films revealed a final thickness of $\sim 1 \ \mu m$ and $\sim 1.5 \ \mu m$, respectively, suggesting an average bilayer thickness of approximately 5 nm. Thermogravimetric analysis showed a final composition of \sim 50 vol% (\sim 70 wt%) montmorillonite. Microtensile analysis revealed a maximum ultimate tensile strength of up to 480 MPa (10 times higher than that of pure PVA) and a modulus of 125 GPa (two orders of magnitude higher than that of pure PVA), with the glutaraldehyde cross-linked films displaying increased strength, stiffness, and brittleness over the non-cross-linked composites (52). Wang et al. (53) fabricated artificial nacre from alumina microplatelets/graphene oxide nanosheets-polyvinyl alcohol (Al2O3/GO-PVA), which demonstrated remarkable strength (143 \pm 13 MPa) and toughness (9.2 \pm 2.7 MJ/m³), exceeding those of natural nacre (Figure 4). The authors attribute the enhanced performance to the hierarchical structure, which combines microplatelets (Al_2O_3) and nanosheets at different length scales. Al₂O₃/GO-PVA films exhibited a tensile strength 2.8 times higher than that of Al₂O₃/PVA films and a toughness approximately 6 times higher than that of GO-PVA films, suggesting that the three-component composite is beneficial for balancing strength and toughness (53).

A summary of the biomimetic composites outlined in Sections 2.2.1–2.2.3, as well as their mechanical properties, is provided in **Table 1**. The mechanical properties of nacre are also listed. The reader should bear in mind that the toughness values reported in **Table 1** are primarily measurements of the crack-initiation fracture toughness, K_{IC} ; however, where indicated, values are measurements of the elastic-plastic fracture toughness, K_{IC} . It is important to note this distinction. The crack-initiation fracture toughness, K_{IC} , describes only resistance to crack initiation; it does not account for contributions to fracture resistance during crack propagation. Many biological structures, including nacre, feature extrinsic toughening mechanisms that activate during crack propagation, allowing for stable crack growth and energy dissipation. These mechanisms lead to increased fracture resistance as the crack propagates, a phenomenon known as the R-curve effect. K_{JC} measurements account for this R-curve behavior, and this is why we observe higher toughness values than those obtained from K_{IC} measurements.

2.2.3. Other methods. Gravitational sedimentation may provide a means to manufacture thicker composites consisting of well-aligned platelet-shaped particles. Behr et al. (54) prepared centimeter-thickness (10–12-mm) sediments of alumina platelets (300 nm thick, 10 μ m in diameter) in solutions of polyvinyl butyral and ethanol, which displayed good alignment of the platelets.



Figure 4

Layer-by-layer assembly of alumina microplatelet/graphene oxide nanosheet–polyvinyl alcohol (Al₂O₃/GO-PVA) nacre-mimetic composites. (*a,b*) Fabrication begins with the mixing of ultrasonically dispersed GO nanosheets and aqueous PVA solution. (*c*) GO-PVA solution spin-coated onto a glass substrate. (*d*) Substrate dip-coated into solution containing monolayer of silane-modified Al₂O₃ platelets. (*e*) Al₂O₃ platelet monolayer transferred to substrate, forming one nacre-mimetic organic/tablet layer. (*f*) Process is repeated to build up nacreous lamellae layer by layer. Composite film is peeled away from the substrate, yielding the nacre-mimetic structure (*top*). The mimetic structure is flexible and foldable. (*g*) Scanning electron microscopy cross-sectional micrograph showing highly ordered layered structure of Al₂O₃/GO-PVA composites films containing the same volume fraction of inorganic platelets. Results indicate that the hybrid Al₂O₃/GO-PVA composite exhibits an adequate combination of strength and toughness derived from the GO and the PVA, respectively. Figure adapted from Reference 53 with permission.

Table 1 Mechanical properties of nacre and biomimetic nacre composites

			Fracture		
Composite	Fabrication method	Strength (MPa)	$(MPa \cdot m^{1/2})$	Reference	Year
Nacre (Haliotis rufescens)	NA	185 ± 20	8 ± 3	35	1989
Al ₂ O ₃ /Al-Si-Ti	Freeze casting	400–600	5.5-10	46	2006
Al ₂ O ₃ /PMMA	Freeze casting	120–210	3-5, 15-30 ^a	48	2008
Al ₂ O ₃ -SiO ₂ -CaO	Freeze casting	470	6.2, 22 ^a	49	2014
Hydroxyapatite/PMMA	Freeze casting	68.6–119.7	6.4 ^b	51	2015
Montmorillonite/PVA-GA	Layer by layer	$400 \pm 40^{\circ}$	NR	52	2007
Al ₂ O ₃ /GO-PVA	Layer by layer	$143\pm13^{ m d}$	$9.2 \pm 2.7 \text{ MJ/m}^3$	53	2015
Al ₂ O ₃ /polyvinyl butyral	Gravitational sedimentation	NR	NR	54	2015
Al ₂ O ₃ /epoxy	Thermal spray	250	6	55	2015
Al ₂ O ₃ /SiO ₂	MASC	650	6, 14 ^a	60	2015
Al ₂ O ₃ /SiO ₂ -PMMA	MASC	182.3 ± 13.8	2.40 ± 0.35	61	2016
Al ₂ O ₃ /SiO ₂ -(PUA-PHEMA)	MASC	168.1 ± 17.5	3.39 ± 0.28	61	2016

^aFracture toughness *K*_{JC} calculated from J-R curve measurements.

^bK_{IC} calculated on the basis of elastic modulus and work of fracture measurements.

^cUltimate tensile strength.

^dTensile yield strength.

Abbreviations: GO-PVA, graphene oxide nanosheet–polyvinyl alcohol; MASC, magnetically assisted slip casting; NA, not applicable; NR, not reported; PMMA, polymethyl methacrylate; PUA-PHEMA, polyether urethane diacrylate-copoly 2-hydroxyethyl methacrylate.

High-energy X-ray diffraction was used to quantify the orientation distributions of the single crystalline particles within the composite pellets. The average misalignment of platelets was found to be 21.5 \pm 1.4° and 14.6 \pm 0.4° for nonpressed and pressed pellets, respectively (54). Although the pellets achieved high alignment of microplatelets and ceramic fraction up to 65 vol% with pressing, the pellets displayed fairly poor mechanical properties, with microhardness and Young's modulus reaching only 5 MPa and 5 GPa, respectively (54). This could be attributed to the significant pore volume within the pellets (approximately 35 vol%), the nonuniform distribution of polymer within the sediments, and stacking defects (misalignment resulting from edge-on settling of particles) (54). Dwivedi et al. (55) recently employed a thermal spray processing technique to fabricate alumina/epoxy nacre-like structures (Figure 5). The thermal spray process allows for a high rate of manufacture, wide material feedstock capability, and thick coatings. The feedstock material (powder, rod, or wire morphology) is fed into a high-temperature, high-velocity gas jet, which forms micrometer-sized droplets that are impacted upon a surface. Upon impact, the droplets flatten out, rapidly cool, and solidify into disk-shaped structures (thickness $0.5-2 \mu m$) that form a lamellar microstructure reminiscent of nacre (Figure 5b,d) (55). Once infiltrated with a polymer (epoxy), the composites attained flexural strength and fracture toughness of approximately 250 MPa and 6 MPa · m^{1/2}, respectively, while only containing 4-5 vol% polymer, which is similar to the polymer content of nacre (Figure 5f,g) (55). Microstructural analysis of fractured specimens reveals deformation/toughening mechanisms similar to those reported in nacre, such as a tortuous crack path, localized interlayer sliding and pullout, and polymer bridging (55). Finally, the authors were able to incorporate an aspect of many damage-tolerant natural composites that has not received much attention in biomimicry literature: a stiff, hard, and dense outermost layer. This structural feature has been identified in numerous biological tissues, such as the nanoparticulated outermost layers found within the abrasion-resistant chiton (mollusk) tooth (56, 57, 128) and impact-resistant stomatopod (crustacean) dactyl club (23, 58, 59). Such layers are thought to act as a first line of defense against stress localization and penetration from external threats such as predators. Dwivedi et al. (55) coated the nacre-like mimetic structures with a 300- μ m fully dense top layer of alumina using high-velocity combustion thermal spray processing,



Figure 5

Spray-formed nacre-mimetic composites. (*a*) Schematic depicting the spray process. (*b*) Scanning electron microscopy (SEM) micrograph of polished cross section of spray-formed lamellar structure. (*c*) Comparative SEM micrograph of polished nacre from abalone shell. (*d*) SEM micrograph of fractured surface of spray-formed structure. (*e*) SEM of fractured surface of nacre from abalone shell. Note the difference in scale bars in panels *b* and *d* versus panels *c* and *e*. Feature sizes obtained in the mimetic structure are larger than those observed in natural nacre. (*f*) Flexural strength measurements of unnotched disordered and brick-and-mortar spray-formed structures. (*g*) Fracture toughness of notched disordered and brick-and-mortar spray-formed structures. (*b*) SEM micrograph of fractured composite, featuring fully dense Al_2O_3 coating atop a nacre-mimetic underlayer. Results show brittle fracture though dense outer coating and tortuous crack path through nacre-mimetic layer. (*i*) Representative SEM micrograph of natural abalone shell showing prismatic outer layer and nacreous inner layer. (*j*) Mechanical testing showing stiff initial response from dense outer layer followed by stable fracture and energy dissipation through the nacre-mimetic layer. Figure adapted from Reference 55 with permission.

which resulted in a bilayer structure that exhibited a stiff initial response and localized brittle failure followed by stable crack propagation and energy dissipation up until ultimate failure (Figure 5b*i*). More recently, Le Ferrand et al. (60) demonstrated a powerful and versatile technique, called magnetically assisted slip casting (MASC), that allows for the fine control of ceramic platelet orientation, thus allowing for the manufacture of not only biomimetic nacre-like structures, but also periodic architectures and multilayered heterogeneous composites. Slip casting is a fabrication process by which a fluid suspension of particles is deposited onto the walls of a dry porous mold. Capillary forces draw the liquid phase into the pores of the mold, thus building a layer of iammed particles on the mold wall. MASC allows for control of particle assembly and texturing through use of magnetically responsive, anisotropic ceramic particles, which are coated with superparamagnetic iron oxide nanoparticles, in combination with an applied external magnetic field (60). By carefully tuning the direction of the magnetic field as a function of time in concert with the position of the consolidation front as a function of time, one can fabricate heterogeneous structures with any desired orientation of particles throughout the thickness of the material (60). The volume fraction of inorganic platelets in centimeter-sized samples can span from 40% to 100% while maintaining a high degree of alignment through cold/hot pressing and sintering steps (60). Subsequent infiltration of polymer into the scaffold allows for the introduction of a second continuous phase (60). The authors were also able to introduce mineral bridges between platelets in their nacre-mimetic materials by introducing silica or alumina nanoparticles into the initial alumina casting suspension followed by partial sintering between adjacent platelets (60). As a result, the fracture toughness reached 14 MPa \cdot m^{1/2}, which was a three- to fourfold improvement compared to that of monolithic untextured alumina, and the fracture strength was maintained at 650 MPa (60). Finally, the versatility of the MASC process was demonstrated by fabricating a tooth-mimetic structure that contained a bilayer structure featuring a higher-density, stiffer, harder outer (enamel-like) layer with vertically aligned platelets and a lower-density, softer inner (dentin-like) layer with horizontally aligned platelets (60). This structure demonstrates the power of the MASC technique for biomimetic materials processing, enabling fine control over aspects such as bulk morphology, local density, stiffness/hardness, microstructure and texturing of inorganic components, and chemical composition (60).

Niebel et al. (61) examined the effect of the choice of polymer used for the organic matrix on the mechanical properties of nacre-like composites. Three different polymers with drastically different mechanical properties, polylauryl methacrylate, PMMA, and polyether urethane diacrylate-copoly 2-hydroxyethyl methacrylate (PUA-PHEMA), were infiltrated into Al₂O₃ scaffolds prepared by MASC, yielding composites with a ceramic fraction of 57 vol% (61). Through a combination of three-point bending and single-edge notched beam tests, fracture analysis via scanning electron microscopy (SEM), and finite-element analysis, it was determined that the inherently stiffer and tougher polymers PMMA and PUA-PHEMA significantly increase the flexural strength (to 182 MPa and 168 MPa, respectively), strain to failure (both to approximately 0.43%), and fracture toughness $K_{\rm IC}$ (to 2.4 MPa \cdot m^{1/2} and 3.39 MPa \cdot m^{1/2}, respectively) of the nacremimetic composites as compared to polylauryl methacrylate, a soft and weak elastomer (100 MPa, 0.25%, and 1.43 MPa · m^{1/2}, respectively) (61). The improvements in strength were attributed to the ability of the PMMA and PUA-PHEMA composites to generate a more homogeneous stress distribution and to avoid formation of stress concentrations near platelet interfaces, and PUA-PHEMA's improvement in toughness was attributed to its ability to promote crack propagation at the organic-inorganic interface rather than through the organic phase, which was observed for polylauryl methacrylate and PMMA composites (61). The authors conclude that the toughening and strengthening mechanisms observed in natural nacre can be best incorporated into artificial

nacre composites by choosing a polymer, representing the organic matrix, that is strong and stiff and that possesses a high modulus of toughness (61).

2.2.4. Biomimetic mineralization. Rather than assembling prefabricated inorganic platelets, such as those commercially purchased, several reports have combined the techniques of LBL or freeze casting with polymer-mediated growth of calcium carbonate tablets in an attempt to replicate the biomineralization process. Finnemore et al. (62) created artificial nacre using LBL in combination with a crystallization step that closely mimics the mineral tablet formation process in natural nacre. First, a continuous organic thin film was deposited onto a glass slide by dip coating into a solution of polyacrylic acid (PAA) and poly 4-vinyl pyridine. The PAA in the film was then dissolved (and the film thus made porous) by immersion in basic solution followed by stabilization using UV cross-linking and COO⁻ surface functionalization by PAA immersion to promote mineral nucleation. Amorphous calcium carbonate (ACC) was then grown on the film surface using an ammonium carbonate diffusion technique and a PAA solution containing 1:5 Ca²⁺:Mg²⁺, yielding polymer-induced liquid precursor droplets that coalesced on the film surface (63-65). The ACC then phase transformed into calcite through a dissolution-recrystallization process upon exposure to high humidity. This process was then repeated to produce a multilayered lamellar structure (62). This synthesis route allows for several structural features similar to those of natural nacre. The resultant tablets are 400-nm-thick pseudosingle crystalline calcite, with grain sizes varying from 5 μ m to 35 μ m. The porous (pore diameter: 99.3 \pm 40.3 nm) organic layers promote intertabular mineral growth (mineral bridges) as well as transference of crystallographic information and continuity across layers, which was confirmed by transmission electron microscopy (TEM) selected area electron diffraction (62). Nanomechanical analysis via nanoindentation revealed similar deformation behavior in the artificial nacre as compared to natural nacre. At loads that induce cracking and shear faulting in monolithic calcite and aragonite, respectively, the artificial nacre displays pile-up behavior and plastic deformation, resisting fracture (62).

More recently, Mao et al. (66) used a matrix-directed mineralization method to form millimeter-thick synthetic nacre with a 91 wt% fraction of aragonitic platelet layers. A chitosan organic matrix with a predefined laminated structure was fabricated by freeze casting and then acetylated, forming β -chitin. The scaffold was then mineralized using a direct peristaltic pump-driven circulatory system to decompose calcium bicarbonate [Ca(HCO₃)₂] in the presence of PAA and Mg²⁺, followed by silk fibroin infiltration and hot pressing (66). Complete mineralization of the organic scaffold can occur in only two weeks, yielding aragonitic tablets (100–150 nm thick) that are randomly (crystallographically) oriented mesocrystals with 10–100-nm nanograins (66). Similar to reports by Finnemore et al., the resulting nacre-mimetic composites do not display crack propagation at loads that induce long crack formation in monolithic calcite and aragonite by nanoindentation, indicating their enhanced toughness and damage-tolerant behavior.

2.2.5. Advantages and disadvantages of nacre-mimetic processing techniques. There are advantages and disadvantages to each of the biomimetic processing routes described in Sections 2.2.1–2.2.4 for producing high-strength and high-toughness nacre-like composites. Freeze casting allows for fine control over layer thicknesses, down to 1 μ m. The nature of the freeze-casting process works well for mimicking some of the nanostructural features of nacre. For example, the forming of inorganic walls from entrapped particles creates a characteristic surface roughness that mimics very well the nano-asperities that coat the surface of nacre tablets (46). In addition, ice dendrites occasionally span the channels between lamellae, effectively replicating the mineral bridges observed in nacre (46). As a result, biomimetic nacre composites fabricated by this method achieve some of the highest reported properties of strength and toughness. Freeze casting also permits

the use of a wide range of inorganic and organic components. However, although freeze casting works well for mimicking lamellar structures such as nacre because of the inherent nature of ice crystals as a structural template, it may not be suitable for replicating other complex structural geometries. In addition, this process does not provide the finely controlled size distribution of platelets, especially at a scale below 1 µm. Conversely, LBL assembly allows for high-precision ordering of nanoscale building blocks, yielding well-defined nanostructures. The constituent materials are often cast, using techniques such as spin coating, resulting in nanoscale-thick layers or even monolayers. Not only does this allow for a highly laminated and ultrathin composite material, but the intimate contact between layers usually results in high interfacial bonding and mechanical strength. However, because of its multistep nature, requiring the repetitive deposition of each layer, one at a time, the LBL technique is very time-consuming and labor-intensive. The gravitational sedimentation technique mentioned in Section 2.2.3 is highly energy-efficient and shows promising scalability, with the potential for fabricating large-volume, bulk-sized materials; however, the long processing time remains an issue (54). The technique allows for fairly good alignment of platelet particles; however, since particles are settling under the fairly weak force of gravity, the resulting composites often possess low density, high porosity, and some particle misalignment, leading to poor mechanical performance. Such issues can be partially improved with additional mechanical pressing and heat treatment steps during the fabrication process (54). Thermal spray processing allows for a high rate of manufacture and produces nacre-mimetic composites with excellent mechanical properties (55). However, because of the droplet forming process, the ability to fabricate platelet structures with consistent geometry, morphology, and packing appears to be limited (55). MASC is a highly versatile process that allows for fine control of bulk morphology, local density, microstructure and texturing of inorganic components, and chemical composition (60). The ability to fabricate multiregional composites with compositional and mechanical gradients has already been demonstrated, highlighting the enormous potential of this technique (60). As with many of the previously mentioned techniques, addressing scalability while minimizing processing time will be an ongoing challenge. Biomimetic mineralization techniques provide the opportunity to properly mimic many aspects of the mineral phase in nacre, such as crystallinity, crystallographic orientation, phase, and morphology, which are crucial for enabling high-performance mechanical properties.

2.3. Structure, Mechanics, and Formation of Arthropod Cuticle

One of the defining features of organisms in the Arthropoda phylum is the cuticle, a tough and robust exoskeletal structure whose purpose is primarily to provide support and protection from predators and other environmental pressures. Its ability to withstand impact-, crushing-, abrasion-, and puncture-type loads has inspired a new generation of multifunctional, damagetolerant composite materials. The cuticle is a composite material composed of the polysaccharide α -chitin and associated proteins, which demonstrate a helicoidal or twisted plywood architecture known as a Bouligand structure (67–69). In Crustacea, amorphous or crystalline forms of calcium carbonate and calcium phosphate mineral are added to the cuticle to provide strength, stiffness, and hardness. In contrast, the nonmineralized cuticle, such as that of hexapods (e.g., insects), relies on the cross-linking of proteins (tanning/sclerotization) or the incorporation of metal ions to stiffen and harden the exoskeleton. In this section, we focus on the mineralized cuticle of crustaceans. Paris et al. (70) and Fabritius et al. (71) recently provided overviews of the structure and mechanical properties of the mineralized crustacean cuticle. Discussions of the mineralogical aspects of the crustacean cuticle have recently been provided by Bentov et al. (72) and Luquet (73). Biomimetic efforts inspired by the crustacean cuticle have recently been reviewed by Grunenfelder et al. (74). **Pitch length:** the repeat distance within a helicoidal structure over which the fiber layers produce a complete 180° rotation **2.3.1. Structure.** The cuticle is a multilayered structure, consisting of an epicuticle, a procuticle, and underlying epidermal tissue. A schematic depicting the hierarchical structure of the cuticle is shown in Figure 6a. The epicuticle is the thin waxy outermost layer, which is composed of lipids and proteins and functions as an impermeable barrier providing microbial resistance. The procuticle makes up the bulk of the exoskeleton and provides the primary load-bearing properties. It consists of two layers: the exocuticle and the endocuticle (**Figure 6***b*). The procuticle features a hierarchical assembly of organic and inorganic constituents. At the nanoscale, proteins are often covalently associated with α -chitin molecules, forming nanofibrils, which aggregate to form nanofibers (71). These nanofibers subsequently self-assemble, forming the twisted plywood (Bouligand) structure (Figure 6a,g), which features unidirectional fiber layers that are stacked, with each layer offset by a specific rotation angle with respect to the adjacent layer. This produces a lamellar architecture of helicoidally oriented fibers, where the fiber layers are aligned parallel to the cuticle surface and rotate about an axis that is normal to the cuticle surface. The helicoidal architecture of the fibers results in a characteristic banded or nested arc appearance observed in cross section, where alternating light and dark bands correspond to the varying local fiber orientation within the helicoidal microstructure. Moreover, fibrous pore canal tubules aligned normal to the cuticle surface interpenetrate the rotating fibers and serve as channels for transport of mineral ions during resorption and deposition of minerals during periodic molting and growth (Figure 6c, d) (75). The exocuticle is typically more densely packed than the endocuticle, containing a smaller helicoidal pitch length. Here, we define the pitch length as the repeat distance over which the fiber layers make a full 180° rotation. In fact, the pitch length is often graded throughout the procuticle, typically with decreasing pitch length observed in the direction from the cuticle surface toward the interior of the exoskeleton. The observation of a smaller pitch length could be attributed to one of two possible structural changes: a larger rotation angle between fiber layers or a denser packing of fibers (decreased interlayer distance).

2.3.2. Mechanics. The Bouligand architecture and, more broadly, helicoidal structures of biological origin have been identified as advantageous designs for biomimetic structural materials. Naleway et al. (11) recently identified the helicoidal architecture as one of the eight most common structural motifs among biological materials in a variety of animal taxa and detailed its mechanical advantages. The helicoidal design has been identified not only in the exoskeletal structures of arthropods, but also other systems, such as the mineralized collagen layers in osteonal bone, in fish scale dermal armor, and in the cellulose fibrils in wood cell walls (76–79, 129). In fact, the

Figure 6

Ultrastructural features of the crustacean cuticle. (*a*) Schematic showing hierarchical structure of the cuticle. (*b*) Optical image of the claw of the American lobster (*Homarus americanus*) and scanning electron microscopy (SEM) micrographs showing cuticle cross section highlighting the helicoidal microstructure of fibers within the exocuticle and endocuticle regions. The right micrograph provides a higher-magnification view of the exocuticle region. (*c*) SEM of cuticle fractured parallel to the cuticle surface showing chitinous pore canal tubule (pct) fibers interpenetrating the helicoidally rotating fibers. (*d*) Schematic showing fiber architecture of the Bouligand structure. Helicoidal fibers rotate in the plane parallel to the cuticle surface with interpenetrating pore canal tubule fibers oriented normal to the cuticle surface. (*e*) Overview of the mantis shrimp with an arrow highlighting the raptorial appendage (dactyl club). (*f*) Energy-dispersive spectroscopy mapping and line scans showing relative concentrations and gradients of phosphorus, calcium, carbon, and magnesium through the cuticle cross section. (*g*) Schematic showing the helicoidal architecture (twisted plywood or Bouligand structure) of mineralized chitin fibers within the dactyl club. (*b*) SEM of fractured section showing nested-arc pattern characteristic of stable microcracking and crack deflection. Panels *a* and *b* adapted from Reference 67; panel *c* adapted from Reference 75; panel *d* adapted from Reference 23. Panels adapted with permission.



wrapping of the tubule structures in bone osteons and in wood cell walls with helicoidal fiber layers has been found to enhance their torsional and bending rigidity (76, 80). The helicoidal arrangement of fibers provides several toughening mechanisms, leading to enhanced damage tolerance and energy absorption. Because of the anisotropic stiffness of chitin nanofibers, which are stiffer in the *c*-direction (i.e., the long axis) (81), the helicoidal architecture provides inherent elastic modulus oscillation resulting from the periodic nature of the helicoidal microstructure. Such modulus oscillation has been identified via nanoindentation in cross sections of osteonal bone (82), lobster cuticle (71), and the mantis shrimp dactyl club (Figures 6e and 7a) (23). The gradient thus results in reduction in local driving force for crack propagation between layers. Moreover, the modulus difference between the stiff mineral and the soft organic polymer within the mineralized fibers allows for extrinisic toughening in the form of crack deflection at the fiber interface. Fratzl and coworkers (83, 84) showed that for laminated structures containing mineral-organic interfaces, when the ratio between Young's moduli of the stiff and the soft layer is sufficiently high (greater than five), the soft layers can act as crack arresters. Combined with the helicoidal architecture, the result is a crack twisting effect that forces interfibrillar crack propagation along a tortuous helicoidal path, thereby increasing the work of fracture and allowing for high energy dissipation. The crack twisting effect has been observed at the osteon boundaries in bone as well as the mantis shrimp dactyl club, where nested-arced cracks were visualized within the endocuticle region by charge-contrast SEM imaging (Figure 6i) (23, 85, 130). Helicoidal reinforcement has also been shown to provide enhanced toughness through the reorientation of fiber layers in response to external loading: Zimmermann et al. (77) used in-situ synchrotron small-angle X-ray scattering to measure the Bouligand-type arrangement of collagen fibril lamellae in the scales of Arapaima gigas, a freshwater fish (77). Their measurements showed that the collagen fibril lamellae rotate and reorient in response to tensile loading so as to deform in tension or compression, thus enhancing their ductility and toughness (Figure 7e).

An additional advantage of the Bouligand design found in arthropod cuticle is enhanced resistance to interfiber-lamellar delamination resulting from the incorporation of interpenetrating pore canal fibers. Similar to the effect of z-pinning in fiber-reinforced composites (86), pore canal fibers aligned through thickness effectively stitch the stacked rotating fiber layers together, providing not only resistance to delamination, but also improved tensile and compressive strength in the direction normal to the cuticle surface (**Figure 7***f*,*g*). Chen et al. (69) examined tensile properties of the sheep crab cuticle (*Loxorbynchus grandis*) in directions longitudinal and normal

Figure 7

Mechanical properties and toughening mechanisms of the arthropod cuticle. (*a*) Nanoindentation mapping and line scan through a transverse section of the stomatopod (*Odontodactylus scyllarus*) dactyl club, showing graded reduced Young's modulus, *E*, and hardness, *H*. Line scan reveals smooth transition from stiff and hard exocuticle surface to softer endocuticle with oscillating modulus as a result of the periodic helicoidal fiber architecture. (*b*) Nanoindentation mapping along cuticle cross section of edible crab (*Cancer pagurus*). Results show similar trends of increasing reduced modulus and hardness moving from the endocuticle to the exocuticle surface. (*c*) Cross-sectional scanning electron microscopy (SEM) micrograph through the molar tooth of the crayfish (*Cherax quadricarinatus*). (*d*) Optical micrograph and nanoindentation line scan through the cross section of the crayfish molar tooth. Results show increasing reduced modulus and hardness moving from the endocuticed apatite reinforced with amorphous mineral. (*e*) Schematic showing deformation mechanism of Bouligand-like fiber lamellae within the natural dermal armor of the freshwater fish *Arapaima gigas*. Lamellae reorient in response to loading, enhancing the scale's ductility and toughness. (*f*) Schematic showing tensile failure of crab cuticle in the z-direction. (*g*) SEM micrographs showing result of cuticle tensile loading in the z-direction. Pore canal tubules provide reinforcement and show necking, providing evidence of ductile fracture. Panel *a* adapted from Reference 23; panel *b* adapted from Reference 92; panels *c* and *d* adapted from Reference 96; panel *e* adapted from Reference 77; panels *f* and *g* adapted from Reference 69. Panels adapted with permission.



to the cuticle surface and found a high density of pore canal tubules showing necking and ductile fracture, which are thought to play an important role in enhancing toughness in the z-direction. Moreover, the tensile strength of dried specimens in the longitudinal direction (12.9 MPa) was approximately 50% lower than that in the normal direction (19.8 MPa), where pore canal fibers were directly engaged (69). Additionally, Yaraghi et al. (58) recently identified a modified Bouligand architecture within the stiff and hard impact surface region (exocuticle) of the stomatopod dactyl club, which features sinusoidally arranged helicoidal fiber layers as opposed to the traditionally flat sheets found within most arthropod cuticles. It was determined that this herringbone structure provides improved stress redistribution and strain-to-failure under compressive loading in the normal direction as compared to the Bouligand design because of the flattening of sinusoidal fiber layers. Moreover, through finite-element analysis, it was shown that by varying the aspect ratio (amplitude:wavelength) of the sinusoidal fiber layers, the relative Young's modulus in the loading direction can be increased (58). Guarín-Zapata et al. (87) also showed that periodic Bouligand structures, such as that found within the mantis shrimp dactyl club, can exhibit band gaps at frequencies related to the stress pulse generated during impact loading, thus providing a shear wave filtering effect that enhances energy absorption.

While many strengthening and toughening mechanisms associated with the helicoidal structure have been identified and discussed here, we acknowledge that, unlike in our treatment of nacremimetic materials in Section 2.2, we have provided little information on the quantification of bulk-scale mechanical properties such as strength and toughness. This is in part because of the small physical size and nonstandard geometry of many cuticle structures featuring the helicoidal design. It is difficult to obtain test specimens of adequate size and geometry that conform to standard test protocols required for measuring bulk mechanical properties. Specialized techniques, such as in situ mechanical testing at the submicron scale, are just now being developed and implemented.

2.3.3. Cuticle formation. Although the microstructure of the chitinous organic matrix provides numerous mechanical advantages, the mineral component also plays a vital role in modifying the stiffness, strength, and hardness of the crustacean cuticle. Perhaps most fascinating is nature's ability to precisely regulate the biomineralization process, demonstrating exceptional control over the local distribution, morphology, phase, crystallinity, and orientation of inorganic material. Such control is responsible for providing the diverse range of mechanical properties and functions of cuticular structures and is achieved by specific proteins and biological macromolecules colocalized on the organic scaffold, which promote/inhibit nucleation and growth of mineral. One of the most common strategies is to vary the degree of mineralization throughout the cuticle, resulting in graded composition and mechanical properties (Figure 6f). This can be achieved via the molting process. Because of the rigid exoskeletons encompassing their soft tissue, arthropods must periodically shed (molt) their exoskeletal structure in order to grow (88). The molt cycle begins with apolysis, the onset of premolt, during which the hypodermis is separated from the cuticle through the work of secreted chitinase and other enzymes that dissolve the membranous layer (88). The new epicuticle and exocuticle are next laid down beneath the endocuticle of the old cuticle; however, they are not mineralized until the old cuticle is shed (88). Concurrently, mineral from the old cuticle is resorbed. Pore canal tubules provide transport pathways for the resorption of mineral (89). Amorphous mineral is advantageous in its ability to be easily resorbed and stored in structures such as gastroliths during molting (90). The crystalline mineral, which is usually less soluble, cannot be resorbed and is thus shed with the old exoskeleton. The molt stage follows, in which the old cuticle is shed from the body. The postmolt follows, during which the exocuticle is tanned/sclerotized (cross-linked with protein), and mineralization begins within

the outermost regions of the epicuticle and the exocuticle (88). Transport of mineral ions is again achieved through the pore canal network (89). The deposition of the endocuticle follows, and the endocuticle becomes mineralized as the organic layers are being deposited. It is here that we expect formation of the endocuticle to take on a chiral nematic ordered structure by the cholesteric self-assembly of chitin-protein fibrils. The exocuticle additionally becomes further mineralized; the end of the postmolt occurs when the membranous layer is deposited and new mineralization is ceased (88).

The result of this controlled molting process is a smooth gradient in Young's modulus and hardness across the exocuticle-endocuticle interface, which allows for a stiff and hard surface to be combined with a compliant and tough interior while preventing stress concentration at the interface. Despite the sharp transition in packing density of Bouligand fiber layers from the endocuticle to the exocuticle, fracture does not appear to commonly initiate at the exocuticleendocuticle interface. Energy-dispersive spectroscopic mapping combined with nanoindentation mapping through the cuticle cross section reveals a strong correlation between increasing calcium content and increasing Young's modulus and hardness moving from the endocuticle to the exocuticle in various other systems, including the American lobster (Homarus americanus), the edible crab (*Cancer pagurus*) (Figure 7b), and the mantis shrimp dactyl club (Figure 7a) (23, 91, 92).

The exocuticle and endocuticle of crustacean cuticle most commonly feature amorphous minerals, such as ACC mineral and amorphous calcium phosphate (ACP) mineral. Because of their high solubility in water and thermodynamic instability, amorphous minerals are usually stabilized within the cuticle by proteins or cations, such as magnesium (93-95). Crystalline minerals such as calcite or apatite may also be incorporated into the cuticle for added stiffness and hardness and are most often confined to the exocuticle, closer to the outermost surface, where stresses from impact, crushing, or puncture from predators may be concentrated. A good example of the colocalization of multiple mineral phases lies within the mandibles of the freshwater crayfish, Cherax quadricarinatus, which feature ACC, ACP, calcite, and fluorapatite organized within the cuticular structure (96). Calcite is found within the incisor; the molar contains a mixture of the softer amorphous minerals (ACC and ACP) within the bulk endocuticle-like interior, which transitions to fluorinated apatite within the enamel-like outer surface (96). Nanoindentation line scans through the cross section of the molar show a gradient of increasing reduced Young's modulus and hardness moving toward the surface, as ACC becomes more substituted for ACP, with the highest values within the fluorapatite surface layer (Figure 7c,d) (96). Similar mineral composition/distribution and mechanical gradients have been identified within the mantis shrimp dactyl club (23, 58, 97). The orientation of the mineral crystals within crustacean cuticle can be controlled so that they exhibit preferential alignment (texture), yielding anisotropic mechanical properties advantageous for certain types of loading. This results from the inherent mechanical anisotropy of many crystalline minerals. This is also observed within the crayfish mandible and mantis shrimp dactyl club, where apatite crystals within the outer layer are textured with their *c*-axes preferentially coaligned normal to the molar/impact surface, parallel with the pore canal tubules and the loading direction, providing improved hardness and wear resistance (23, 59, 87, 96, 97).

As mentioned in the examples of the crayfish mandible and the stomatopod dactyl club, enhancement of the local stiffness and hardness can also be achieved by the substitution of various elements into either the crystal lattice of the mineral phases or the organic matrix. Substitution of fluorine into the apatite crystal lattice not only decreases the solubility, enhancing the resistance to mineral dissolution, but also may improve the elastic modulus and hardness (98-100). The incorporation of zinc, in the form of zinc oxide nanogranules, and chlorine into the chitin matrix of spider (Cupiennius salei) fang was shown to correlate with higher stiffness and hardness at the fang tip, enabling its function for puncture through insect cuticle (101). Zinc has been found to play an important role in hardening the jaws, teeth, claws, and stings of many other nonmineralized arthropods, such as ants and scorpions (102–105).

2.4. Helicoidal/Bouligand Mimetic Materials

Biomimetic efforts to replicate arthropod cuticle have only recently gained popularity, with the majority of reports published within the past few years. Research efforts have focused primarily on taking advantage of one of two kinds of properties of the cuticle, mechanical or optical. Here we focus on structural applications and examine biomimetic composite materials that feature the laminated helicoidal architecture of fibers characteristic of the arthropod cuticle. We examine three popular processing techniques for their fabrication, which include fiber-reinforced composite processing, additive manufacturing, and self-assembly.

2.4.1. Fiber-reinforced composite processing. Fiber-reinforced composites (FRCs) are popular structural materials because of their combination of high strength and toughness and are currently found in a wide range of commercial applications, such as aerospace, automotives, defense, and high-performance sporting equipment. FRCs consist of fibers (reinforcement), typically woven fabrics or unidirectional sheets that are impregnated and embedded in a continuous matrix phase. Because of the wide range of mechanical properties (e.g., tensile strength, Young's modulus, ductility) of reinforcement and matrix materials, FRCs can find great versatility in application. One could imagine FRCs as candidates for cuticle-mimetic materials because of the reinforcement and matrix components being analogous to chitin-protein fibers and biomineral, respectively. Cheng et al. (106) fabricated biomimetic helicoidal laminated composites using glass fiber preimpregnated with epoxy resin (prepreg). Panels measuring $0.3 \text{ m} \times 0.3 \text{ m}$ containing 24 stacked fiber layers (plies) were produced. Single helicoid as well as double helicoid panels were produced, featuring continuous interply rotation angles of 7.8° and 16.4°, respectively. An industry-standard quasiisotropic design, which features a stacking sequence of $[0/-45^{\circ}/45^{\circ}/90^{\circ}]_{3s}$, was used as a baseline control for comparison of mechanical performance (106). The mechanical properties were assessed under flexure and short-beam shear, and it was found that the biomimetic helicoidal structures displayed higher flexural stiffness as well considerably increased residual strength (damage tolerance) compared to the baseline (83% for single helicoid, 56% for single helicoid with midplane symmetry, and 23% for double helicoid) (106). Grunenfelder and colleagues (107, 131; see also 132) more recently fabricated 48-ply carbon fiber/epoxy biomimetic helicoidal composites by a similar out-of-autoclave prepreg processing route and examined their mechanical response to impact loading (Figure 8). Biomimetic helicoidal panels featuring three different rotation angles, 7.8° (small), 16.3° (medium), and 25.7° (large), were fabricated and compared to unidirectional as well as quasiisotropic controls (Figure 8a,d) (107). The purpose of testing various interply rotation angles of

Figure 8

Helicoidal fiber-reinforced composite processing. (*a*) Optical micrographs showing postimpact 48-ply carbon fiber/epoxy composite laminates: (*i*) unidirectional, (*ii*) quasi-isotropic, (*iii*) small-angle helicoid, (*iv*) medium-angle helicoid, and (*v*) large-angle helicoid. (*b*) Measured postimpact dent depths for the quasi-isotropic and three helicoidal panels. All biomimetic helicoidal panels show reduction in dent depth in comparison to the quasi-isotropic control, with the medium-angle panel showing the greatest reduction in dent depth. (*c*) Ultrasonic C-scans showing extent of internal damage of impacted composite panels. Results indicate that helicoidal fiber architecture deflects cracks laterally over a larger area. (*d*) Scanning electron microscopy micrographs of polished cross sections of the composite panels. Contrast variation corresponds to changing local fiber orientation. (*e*) Schematic showing multiscale toughening routes of fiber-reinforced composites using carbon nanofibers and *z*-pins. Panels *a*–*d* adapted from Reference 107 with permission; panel *e* adapted from Reference 108 with permission.

the helicoid was to determine if there is an optimal helicoidal geometry for impact resistance and to understand the mechanical implications of having a graded pitch length throughout the cuticle cross section (107). Results of the impact testing, using a drop tower test with 100 J of impact energy, showed that each of the helicoidal mimetic composites outperformed the unidirectional and



quasi-isotropic controls in terms of dent depth, a measure of surface damage, with the panel with medium rotation angle showing a 49% reduction in dent depth compared to the quasi-isotropic control (**Figure 8b**) (107). Moreover, the medium- and large-angle panels showed 16% and 18% improvement, respectively, in residual compressive strength compared to the quasi-isotropic control (107). Postimpact ultrasonic imaging revealed internal damage within the composite panels and showed that the biomimetic helicoidal fiber architecture promotes lateral crack propagation as opposed to through-thickness damage that may lead to catastrophic failure (**Figure 8***c*) (107).

One aspect of the arthropod cuticle that remains difficult to mimic, yet plays a vital mechanical role, is the network of aligned out-of-plane pore canal tubule fibers. Because of the difficulty in incorporating this network of fibers into the helicoidally stacked laminate, they are often disregarded and the mimetic structure is falsely termed a Bouligand/cuticle mimic. A potential route to incorporate this structural feature into helicoidal mimetic composites is through *z*-pinning (**Figure 8***e*). *z*-Pinning (e.g., 3D weaving, stitching, braiding) is a common technique already used in industry for reinforcing FRC laminates in the through-thickness direction, improving delamination resistance and impact tolerance (86). The challenge is that *z*-pinning does not work for unidirectional prepregs, which are most commonly used to mimic cuticle, because they result in excessive fiber damage that degrades the in-plane mechanical properties (86). Ladani et al. (108) recently incorporated macroscale carbon *z*-pins (~280 μ m in diameter) and through-thickness carbon nanofibers (~100 nm in diameter) into carbon-epoxy composite laminates. When *z*-pins were implemented, the interlaminar fracture energy, under quasistatic loading, was increased by approximately 200%. When *z*-pins and nanofibers were used concurrently, the fracture energy was increased by 400% (108).

2.4.2. Additive manufacturing/3D printing. Additive manufacturing or 3D printing provides a facile route to preparing biomimetic helicoidal structures with the help of computer-aided design software. There are now a wide range of additive manufacturing techniques (e.g., direct ink write, stereolithography, inkjet, laser sintering, electrophoretic deposition) and concurrently a diverse range of feedstock materials that are capable of being printed (e.g., ceramic and metal slurries, polymers, and living cells) into functional materials (109). Some techniques, such as the extrusionbased direct ink write process, allow for open-source ink development, providing the opportunity to print unique materials such as biopolymers (e.g., chitosan) or hydroxyapatite mineral suspensions (110–113). 3D printing also allows for the manufacture of parts with complex geometry, which is advantageous for the production of arthropod cuticle-mimetic structures. Ribbans et al. (114) 3D printed helicoidal fiber structures that were additionally supported with peripheral ring-like and helix reinforcement; this was proposed by Milliron (115; see also 23) to mimic the circumferential fiber reinforcement along the lateral sides of the mantis shrimp dactyl club, which was hypothesized to provide resistance to lateral expansion during impact (133) (Figure 9d) Monotonic torsional testing combined with modeling was performed to study interlamellar shear resistance and to examine failure mechanisms. The results showed that the fiber-matrix modulus ratio and the pitch angles of the helix reinforcement play an important role in the interlamellar shear resistance; however, shear resistance was not substantially affected by interlayer rotation angle (114). Yaraghi et al. (58) used a multimaterial 3D printer (Connex350, Objet, USA) to produce Bouligand and herringbone modified Bouligand structures in order to compare their mechanical responses under compressive loading and validate them with finite-element analysis (Figure 9a-c). The parts were cylindrical disks 60 mm in diameter and 37 mm in height, and featured a stiff acrylic polymer for the fibers and a soft elastomeric polymer for the matrix (58).

2.4.3. Self-assembly/liquid crystalline chiral nematic ordering. The biomimetic route that perhaps most closely resembles the natural process by which the arthropod cuticle forms is the



Figure 9

3D printed helicoidal mimetic structures. (*a*) 3D model comparing the traditional helicoidal architecture within the arthropod cuticle (*left*) and the sinusoidally architected herringbone helicoidal structure found within the exocuticle of the mantis shrimp (*Odontodactylus scyllarus*) dactyl club (*right*). (*b*) Optical images of the multimaterial 3D printed cylindrical parts containing the helicoidal (*left*) and herringbone (*right*) structures shown in panel *a*. The parts feature stiff acrylic-based fibers in an elastomeric matrix. (*c*) Stress–strain plot showing results of uniaxial compression testing in the *z*-direction of the 3D printed mimetic structures in panel *b*. Mechanical strain is denoted by ε and mechanical stress by σ . Subscripts 1 and 2 denote the maximum strain values before failure of the Bouligand and herringbone mimetic structures, respectively. Results show that the herringbone structure allows for greater energy absorption compared to the traditional helicoidal design. (*d*) 3D models and printed helicoidal structures featuring no reinforcement (*left*), ring reinforcement (*center*), and helix reinforcement (*right*). Panels *a*–*c* adapted from Reference 58 with permission; panel *d* adapted from Reference 114 with permission.

cholesteric self-assembly of liquid crystals forming the chiral nematic phase (**Figure 10**). This technique takes advantage of the chiral nature of constituent nanocrystalline building block materials like cellulose/chitin nanocrystals or nanowhiskers and fine-tunes their environmental conditions (e.g., pH, concentration, hydration, etc.) to drive self-assembly toward the formation of the chiral nematic phase. Several very recent studies have successfully demonstrated the cholesteric selfassembly of biomaterials, such as chitin and cellulose, forming well-ordered helicoidal structures mimicking the arthropod cuticle, with subsequent assessment of their mechanical and optical properties.

Shopsowitz et al. (116) published one of the first reports of the synthesis of mesoporous chiral nematic structures with long-range ordering that displayed photonic properties. The focus of this work was to develop a photonic mesoporous inorganic solid by forming a chiral nematic structure from cellulose nanocrystals (CNCs) and using it as a cast for silica. CNCs were prepared by sulfuric acid hydrolysis of bleached softwood kraft pulp (116). Aqueous CNC suspensions (3 wt%) adjusted to pH = 2.4 were loaded with silica precursor (tetra-ethyl orthosilicate), homogenized, and allowed to slowly evaporate at room temperature (116). After drying, free-standing composite

films were obtained, and the formation of a chiral nematic phase was confirmed by polarized light microscopy, which showed the characteristic fingerprint pattern indicative of the ordered nematic structure (116). The films could then be calcined at 540°C under air to remove the cellulose, resulting in a pure silica film. SEM analysis confirmed that the chiral nematic structure still persisted. Although the successful production of the chiral nematic phase was only possible

over a narrow pH range, the process was relatively simple. Moreover, the authors found that the peak wavelength of reflected light (useful for the design of photonic structures), which is directly related to the helicoidal pitch of the chiral nematic phase, could be increased by increasing the silica content (116). It was speculated that increase in helicoidal pitch could be caused by a greater silica wall thickness and higher repulsive interactions between the negatively charged silica and cellulose nanocrystals during condensation (116). Majoinen et al. (117) also examined the formation of chiral nematic phases using CNCs (approximately 7.5 nm thick \times 150 nm long) obtained from the acid hydrolysis of cellulose filter paper with sulfuric acid. It was determined that charged sulfate half-ester groups on the surface of the CNCs, formed during hydrolysis, resulted in electrostatic repulsive forces that helped keep the CNCs well dispersed in water. The cast films (70 µm thick) were formed by slow evaporation of water from 1–2 wt% aqueous suspensions of CNCs. SEM analysis of the fractured film cross section revealed a helicoidal pitch distance of 1–3 µm (117).

Wang & Walther (118) recently fabricated helicoidal crustacean-mimetic CNC-derived nanocomposites and demonstrated the tailored periodicity of their structures. Using PVA as a matrix component to the system, chiral nematic phases could be formed in a simple single-step process. Here, hypercolloidal dispersions were prepared by the slow addition of dilute CNC suspensions into well-stirred PVA solution and left to evaporate, forming the solid films (118). An interesting finding was that the helicoidal pitch and photonic bandgap could be tuned by varying the CNC/PVA ratio. The cholesteric nematic phase was formed at CNC fractions above 55 wt%, and the helicoidal pitch was found to decrease with increasing CNC fraction. The pitch could be varied between 486 \pm 50 nm (CNC/PVA: 60/40 w/w) and 355 \pm 30 nm (CNC/PVA: 100/0 w/w) (118). Additionally, the authors demonstrated helicoidal structures with stacked periodicity (multiple periodicities within the same structure). This was achieved by sequential casting of dispersions of different CNC/PVA ratio on top of one another (118). The interfacial properties were determined to be mechanically robust, demonstrating no splitting or delamination. Such architectural control is important for the mimicry of crustacean cuticle, which demonstrates not only graded helicoidal pitch, but also sharp transitions in periodicity (e.g., between endocuticle and exocuticle regions). Finally, the tensile properties of the resultant biomimetic films were tested (at 55% relative humidity). Nanocomposites with ratios of CNC/PVA from 90/10 to 70/30 show stiff and strong character; however, little to no plastic deformation was observed (118). Yielding and plastic deformation was observed in composites with higher PVA content; however, their strength also decreased. Plastic deformation appears to coincide with a structural transition from the cholesteric phase to a disordered phase (118). SEM analysis of the fractured specimens reveals some evidence of toughening mechanisms, such as ligament bridging and constrained microcracking (118). These studies not only provide new insight into the biological process of helicoidal self-assembly, but also show promise for the development of scalable processing of hierarchical nanocomposite materials.

Figure 10

Cholesteric self-assembly of helicoidal chitin/CaCO₃ composites. (*a*) Schematic showing strategy for development of helicoidal chitin/CaCO₃ composites. (*b*) Atomic force microscopy image of synthesized chitin nanowhiskers. (*c*) Polarized optical image of the chitin whisker suspension indicating formation of the chiral nematic phase. (*d*) Optical image of the chitin/PAA template after polymerization. (*e*) Polarized optical image confirming that the chiral nematic phase is maintained. (*f*–*b*) Scanning electron microscopy cross-sectional micrograph of synthesized helicoidal chitin/PAA template (*f*) before crystallization, (*g*) after 1 day of crystallization with CaCO₃, and (*b*) after 7 days of crystallization. (*i*) X-ray diffraction patterns of the chitin/PAA template (1) before crystallization, (2) after 1 day of crystallization, and (3) after 7 days of crystallization. Labeled peaks indicate Miller indices for calcite. Figure adapted from Reference 119 with permission. Abbreviations: ACC, amorphous calcium carbonate; PAA, polyacrylic acid.

Matsumura et al. (119) added to this work by fabricating helicoidal chitinous nematic phases and subsequently mineralizing them with calcium carbonate, forming a hybrid composite structure (Figure 10). Chitin nanowhiskers (Figure 10b) (165 nm long \times 6 nm wide), prepared by hydrolysis in HCl solution, were used to form the chiral nematic phase, which was confirmed by polarized light microscopy. As in previous examples, the cholesteric self-assembly occurred only in certain ranges of pH and concentration. To avoid disturbing the chiral nematic phase during mineralization, the helicoidal structure was fixed by a polymer network forming a chitin/PAA template (119). The polymer network, bearing carboxylate groups, was believed to facilitate the binding of Ca²⁺ ions and induce CaCO₃ mineralization. The chitin/PAA helicoidal scaffold was mineralized by immersion in an ACC colloidal suspension for 1-7 days. Cross-sectional SEM analysis confirmed that the helicoidal structure was preserved after mineralization (Figure 10f-b) (119). Moreover, the pitch length was measured as a function of time after the onset of mineralization. The pitch length of the nonmineralized scaffold before and after drying was 7–8 μ m and 1.4 µm, respectively. The decrease was associated with water leaving the chitin/PAA scaffold. After 1 day of mineralization, the pitch length increased to 2.4 μ m, where it remained after 7 days. The increase was attributed to the infiltration of inorganic material into the scaffold (119). X-ray diffraction showed the presence of calcite after 7 days (Figure 10i); TEM selected area electron diffraction confirmed the presence of ACC occluded within the chitin/PAA template after 1 day of mineralization and of nanocrystalline calcite after 7 days. Additionally, thermogravimetric analysis revealed 12 wt% and 23 wt% CaCO₃ loading after 1 day and 7 days, respectively. The incorporation of PAA is important from a biomimetic standpoint. Attempted crystallization of non-PAA-modified chitin templates with CaCO₃ and thermogravimetric analysis results suggest that mineralization was not induced, which is a potential indication of PAA's role in triggering the infiltration of ACC into the chitin scaffold and inducing crystallization (119).

Multifunctional aspects of these biomimetic composites have also been explored. Nan et al. (120) created helicoidal thermal reduced graphene (TRG)/cellulose nanocomposites through a vacuum-assisted self-assembly technique that demonstrate electrical conductivity as well as water-responsive color changing properties. Nguyen et al. (121) used discarded crustacean endocuticles and shells to obtain chitosan nanofibrils and fabricate helicoidal photonic hydrogels.

Oh et al. (122) reproduced chiral plywood structures from chitin nanowires extracted from crustacean shell and assessed the resultant mechanical properties. The authors employed a new technique, using Ca-saturated methanol to dissolve the chitin at the molecular level under mild conditions, which does not require the use of a strong acid or base for hydrolysis. Calcium ions could also stabilize the nanowhiskers in solution by screening interfibrillar hydrogen bonding (122). Once the chitin nanowhiskers were dispersed in solution, Ca^{2+} ions were removed through solvent exchange using three different solvents: isopropanol, methanol, and deionized water. Because of the different binding affinities of the solvents for Ca^{2+} , different liquid crystal (LC) phases were formed: In methanol and isopropanol, a nematic LC formed (parallel alignment) (122). Because water has the highest binding affinity to Ca^{2+} among the three solvents, a chiral nematic LC phase was readily formed in water. The microstructural features of these composites were visualized by polarized light microscopy and TEM. By varying the water content of the hydrogel (i.e., through evaporation), the helicoidal pitch could be varied through hydrodynamic pressure placed on the chitin scaffold (122). To assess the mechanical performance of the helicoidal structures, composites were prepared, by in-filling the chiral nematic structures with epoxy, and then tested to failure in tension. As expected, both Young's modulus and toughness increased with higher-order structuring (disordered < nematic LC < chiral nematic LC) (122). The nematic and chiral nematic LC phases actually had similar Young's moduli; however, the chiral nematic LC had significantly higher toughness (122).

Zhu et al. (123) demonstrated a similar process for producing polymer/CNC cholesteric composites, instead using copolymers containing ureidopyrimidinone motifs to improve the mechanical performance. The CNC-based composites can span a wide range of mechanical properties depending on the polymer/CNC ratio, with inelastic deformation strains reaching up to 13% and stiffness reaching 15 GPa, independently, with combinations in between. This is due to the ureidopyrimidinone motifs, which promote supermolecular hydrogen bonding and can be modified to vary the molecular interactions with the CNCs (123).

3. CONCLUSIONS AND OUTLOOK

The field of biomimetic engineering has been fruitful over the past decade. In the context of the systems covered in this review, we discuss examples of mimetic nacre structures that achieve microstructural, nanostructural, and crystallographic control with features that are nearly indistinguishable from the natural systems. By designing these materials with well-defined micro- and nanostructures resembling nacre, as well as incorporating engineering materials such as alumina ceramic tablets and extensible polymers as constituent building blocks, the resultant composite materials have the potential to exceed the mechanical properties of the native biological composite. More progress, however, needs to be made on incorporating other aspects of the nacre microstructure into biomimetic designs. For instance, nanoscale features such as tablet waviness and mineral bridges have not often been recorded in the biomimetic nacre literature. Such features have been found to enhance the strength of nacre through strain hardening and resistance to tablet sliding, respectively. Faster and less labor-intensive processing techniques will also need to be developed, and processing techniques must produce materials with more consistent microstructures (e.g., uniform density, tablet geometry, orientation, and interlamellar spacing) while also being scalable.

Beyond the nacre structure, the arthropod cuticle has recently become a biological material of biomimetic interest for structural applications. Great progress has been made in mimicking its helicoidal structure and achieving the fracture-tolerant properties that this structure affords. Biomimetic helicoidal carbon fiber/epoxy composites fabricated using modern prepreg materials and industrial processing techniques have already demonstrated the ability to surpass the performance of current aerospace and automotive design standards under impact loading.

Fiber-reinforced composite processing is advantageous in that it allows for the production of large-scale bulk materials that exhibit an adequate combination of strength and toughness; however, the process of mimicking the helicoidal architecture within the arthropod cuticle requires labor- and time-intensive processing. With the help of current technology, the cutting, alignment, and stacking of fiber layers (plies) could be automated so as to reduce processing time and the possibility of human-induced error, such as ply misalignment. This suggests the need for new assembly methods. The cuticle-mimetic structures produced by the cholesteric self-assembly of nanocrystalline materials achieve highly controlled micro- and nanostructure reminiscent of natural cuticle and could provide means to scalable production.

In addition, most commercial prepreg materials are expensive and only feature reinforcements with fiber diameters on the microscale, therefore demonstrating a need for alternative materials and for a reduction in size, which will reduce weight. Chitin fibers within most arthropod cuticle have diameters on the order of tens of nanometers. Thus, in order to achieve a mimetic structure that is more representative of the natural system, there is a current demand for a 100–1,000-fold reduction in the diameter of commercially available fiber prepregs. Electrospinning is a popular current technique for the production of nanofibers; however, achieving a high degree of fiber alignment, orientation, and packing density while incorporating a laminated helicoidal

architecture remains a challenge. In addition, the tensile strength of fibers that are synthesized using electrospinning falls short of the strength of fibers synthesized using current high-temperature processing. Addressing this by looking at methods by which nature produces its fibers may yield new insights into processing strong fibers.

Another challenge in mimicking arthropod cuticle is incorporating pore canal tubule fibers. It has been well established in natural systems that interpenetrating pore canal tubule fibers provide enhanced out-of-plane properties such as tensile and compressive strength. Few biomimetic reports have successfully incorporated these tubule structures into helicoidal architected materials. As mentioned above, *z*-pinning is one potentially successful option for incorporating these structural features.

There are many widespread challenges across the field of biomimetics, including lack of control of micro- and nanostructural features and interfaces across large areas and lack of manufacturing speed and scalability. For example, in order to scale up, well-defined micro- and nano-architectures need to be maintained and uniform across the entire volume of the part in order to avoid localized defects and to provide predictable function. Rapid processing methods for these materials need to be addressed. Many of the current methods, such as LBL techniques, are time-consuming and labor-intensive. 3D printing methods continue to push this boundary, but length scale is still an issue. Moreover, control over the interfacial properties is a major challenge. Many biological materials control the strength of hard–soft interfaces through smooth compositional and mechanical gradients, which reduce stress concentration and delamination. Incorporating mechanical gradients into biomimetic materials will be beneficial for many structural applications.

Finally, more attention needs to be paid to taking advantage of the multifunctional nature of many biological systems, which not only are mechanically robust, but also incorporate aspects such as self-healing, nutrient transport, and actuation/shape-change. If these aspects can be appropriately addressed, the next generation of high-performance multifunctional materials will certainly be inspired by biology.

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LITERATURE CITED

- Roco MC. 2011. The long view of nanotechnology development: the National Nanotechnology Initiative at 10 years. J. Nanopart. Res. 13:427–45
- 2. Holdren JP. 2011. *Materials genome initiative for global competitiveness*. Rep., Nat. Sci. Technol. Counc., Off. Sci. Technol. Policy, Washington, DC
- Meng J, Zhang P, Wang S. 2015. Recent progress of abrasion-resistant materials: learning from nature. Chem. Soc. Rev. 45:237–51
- De Yoreo JJ. 2014. Mimicking biomineral systems: What have we achieved and where do we go from here? In *Bio-Inspired Nanotechnology*, ed. MR Knecht, TR Walsh, pp. 291–314. New York: Springer
- 5. Wang J, Cheng Q, Tang Z. 2012. Layered nanocomposites inspired by the structure and mechanical properties of nacre. *Chem. Soc. Rev.* 41:1111–29
- Yao H-B, Fang H-Y, Wang X-H, Yu S-H. 2011. Hierarchical assembly of micro-/nano-building blocks: bio-inspired rigid structural functional materials. *Chem. Soc. Rev.* 40:3764–85
- Deville S. 2008. Freeze-casting of porous ceramics: a review of current achievements and issues. Adv. Eng. Mater. 10:155–69
- Addadi L, Gal A, Faivre D, Scheffel A, Weiner S. 2015. Control of biogenic nanocrystal formation in biomineralization. Isr. 7. Chem. 56:227–41

5. Wang et al. survey biomimetic efforts to fabricate layered nanocomposites inspired by nacre and discuss advantages and disadvantages of such strategies.

6. Yao et al. discuss hierarchical biological structures and review assembly techniques for fabricating bio-inspired rigid functional materials.

- 9. Studart AR. 2016. Additive manufacturing of biologically-inspired materials. Chem. Soc. Rev. 45:359-76
- 10. Wegst UG, Bai H, Saiz E, Tomsia AP, Ritchie RO. 2015. Bioinspired structural materials. Nat. Mater. 14:23 - 36
- 11. Naleway SE, Porter MM, McKittrick J, Meyers MA. 2015. Structural design elements in biological materials: application to bioinspiration. Adv. Mater. 27:5455-76
- 12. Meyers MA, McKittrick J, Chen P-Y. 2013. Structural biological materials: critical mechanics-materials connections. Science 339:773-79
- 13. Chen P-Y, McKittrick J, Meyers MA. 2012. Biological materials: functional adaptations and bioinspired designs. Prog. Mater. Sci. 57:1492-704
- 14. Meyers MA, Chen P-Y, Lin AY-M, Seki Y. 2008. Biological materials: structure and mechanical properties. Prog. Mater. Sci. 53:1-206
- 15. Fratzl P, Weinkamer R. 2007. Nature's hierarchical materials. Prog. Mater. Sci. 52:1263-334
- 16. Kisailus D, Truong Q, Amemiya Y, Weaver JC, Morse DE. 2006. Self-assembled bifunctional surface mimics an enzymatic and templating protein for the synthesis of a metal oxide semiconductor. PNAS 103:5652-57
- 17. Kisailus D, Najarian M, Weaver JC, Morse DE. 2005. Functionalized gold nanoparticles mimic catalytic activity of a polysiloxane-synthesizing enzyme. Adv. Mater. 17:1234-39
- 18. Johnson JM, Kinsinger N, Sun C, Li D, Kisailus D. 2012. Urease-mediated room-temperature synthesis of nanocrystalline titanium dioxide. J. Am. Chem. Soc. 134:13974-77
- 19. Kisailus D, Choi JH, Weaver JC, Yang W, Morse DE. 2005. Enzymatic synthesis and nanostructural control of gallium oxide at low temperature. Adv. Mater. 17:314-18
- 20. Sumerel JL, Yang W, Kisailus D, Weaver JC, Choi JH, Morse DE. 2003. Biocatalytically templated synthesis of titanium dioxide. Chem. Mater. 15:4804-9
- 21. Yao H-B, Ge J, Mao L-B, Yan Y-X, Yu S-H. 2014. Artificial carbonate nanocrystals and layered structural nanocomposites inspired by nacre: synthesis, fabrication and applications. Adv. Mater. 26:163-88
- 22. Espinosa HD, Rim JE, Barthelat F, Buehler MJ. 2009. Merger of structure and material in nacre and bone-perspectives on de novo biomimetic materials. Prog. Mater. Sci. 54:1059-100
- 23. Weaver JC, Milliron GW, Miserez A, Evans-Lutterodt K, Herrera S, et al. 2012. The stomatopod dactyl club: a formidable damage-tolerant biological hammer. Science 336:1275-80
- 24. Nudelman F. 2015. Nacre biomineralisation: a review on the mechanisms of crystal nucleation. Semin. Cell Dev. Biol. 46:2-10
- 25. Jackson AP, Vincent JFV, Turner RM. 1988. The mechanical design of nacre. Proc. R. Soc. B 234:415-40
- 26. Currey JD, Taylor JD. 1974. The mechanical behaviour of some molluscan hard tissues. J. Zool. 173:395-406
- 27. Song F, Soh AK, Bai YL. 2003. Structural and mechanical properties of the organic matrix layers of nacre. Biomaterials 24:3623-31
- 28. Wang RZ, Suo Z, Evans AG, Yao N, Aksay IA. 2001. Deformation mechanisms in nacre. 7. Mater. Res. 16:2485-93
- 29. Wang RZ, Wen HB, Cui FZ, Zhang HB, Li HD. 1995. Observations of damage morphologies in nacre during deformation and fracture. 7. Mater. Sci. 30:2299-304
- 30. Meyers MA, Lin AY-M, Chen P-Y, Muyco J. 2008. Mechanical strength of abalone nacre: role of the soft organic layer. 7. Mech. Behav. Biomed. Mater. 1:76-85
- 31. Barthelat F, Espinosa HD. 2007. An experimental investigation of deformation and fracture of nacremother of pearl. Exp. Mech. 47:311-24
- 32. Wang R, Gupta HS. 2011. Deformation and fracture mechanisms of bone and nacre. Annu. Rev. Mater. Res. 41:41-73
- 33. Espinosa HD, Juster AL, Latourte FJ, Loh OY, Gregoire D, Zavattieri PD. 2011. Tablet-level origin of toughening in abalone shells and translation to synthetic composite materials. Nat. Commun. 2:173
- 34. Barthelat F, Li C-M, Comi C, Espinosa HD. 2006. Mechanical properties of nacre constituents and their impact on mechanical performance. J. Mater. Res. 21:1977–86
- 35. Sarikaya M, Gunnison K, Yasrebi M, Aksay I. 1989. Mechanical property-microstructural relationships in abalone shell. MRS Proc. 174:109

21. Yao et al. discuss routes to fabricate and applications of artificial carbonate nanocrystals and layered structural nanocomposites inspired by nacre.

45. Cheng et al. review

fabricate nacre-inspired

layered composites with

superior mechanical

performance, with a

focus on nanoscale

carbon materials.

recent efforts to

- 36. Jackson AP, Vincent JFV, Turner RM. 1988. The mechanical design of nacre. Proc. R. Soc. B 234:415-40
- Nudelman F, Gotliv BA, Addadi L, Weiner S. 2006. Mollusk shell formation: mapping the distribution of organic matrix components underlying a single aragonitic tablet in nacre. J. Struct. Biol. 153:176–87
- 38. Checa AG, Cartwright JH, Willinger M-G. 2011. Mineral bridges in nacre. J. Struct. Biol. 176:330-39
- 39. Olson IC, Gilbert PUPA. 2012. Aragonite crystal orientation in mollusk shell nacre may depend on temperature. The angle spread of crystalline aragonite tablets records the water temperature at which nacre was deposited by *Pinctada margaritifera*. *Faraday Discuss*. 159:421–32
- Gilbert PUPA, Metzler RA, Zhou D, Scholl A, Doran A, et al. 2008. Gradual ordering in red abalone nacre. *7. Am. Chem. Soc.* 130:17519–27
- Lin AY-M, Chen P-Y, Meyers MA. 2008. The growth of nacre in the abalone shell. Acta Biomater. 4:131–38
- 42. Cartwright JH, Checa AG. 2007. The dynamics of nacre self-assembly. J. R. Soc. Interface 4:491-504
- Addadi L, Joester D, Nudelman F, Weiner S. 2006. Mollusk shell formation: a source of new concepts for understanding biomineralization processes. *Chem. Eur.* 7, 12:980–87
- Heinemann F, Launspach M, Gries K, Fritz M. 2011. Gastropod nacre: structure, properties and growth—biological, chemical and physical basics. *Biophys. Chem.* 153:126–53
- Cheng Q, Jiang L, Tang Z. 2014. Bioinspired layered materials with superior mechanical performance. Acc. Chem. Res. 47:1256–66
- Deville S, Saiz E, Nalla RK, Tomsia AP. 2006. Freezing as a path to build complex composites. *Science* 311:515–18
- Worster M, Wettlaufer J. 1997. Natural convection, solute trapping, and channel formation during solidification of saltwater. J. Phys. Chem. B 101:6132–36
- Munch E, Launey ME, Alsem DH, Saiz E, Tomsia AP, Ritchie RO. 2008. Tough, bio-inspired hybrid materials. *Science* 322:1516–20
- Bouville F, Maire E, Meille S, Van de Moortèle B, Stevenson AJ, Deville S. 2014. Strong, tough and stiff bioinspired ceramics from brittle constituents. *Nat. Mater.* 13:508–14
- Bai H, Chen Y, Delattre B, Tomsia AP, Ritchie RO. 2015. Bioinspired large-scale aligned porous materials assembled with dual temperature gradients. *Sci. Adv.* 1:e1500849
- Bai H, Walsh F, Gludovatz B, Delattre B, Huang C, et al. 2016. Bioinspired hydroxyapatite/poly(methyl methacrylate) composite with a nacre-mimetic architecture by a bidirectional freezing method. *Adv. Mater.* 28:50–56
- Podsiadlo P, Kaushik AK, Arruda EM, Waas AM, Shim BS, et al. 2007. Ultrastrong and stiff layered polymer nanocomposites. *Science* 318:80–83
- Wang J, Qiao J, Wang J, Zhu Y, Jiang L. 2015. Bioinspired hierarchical alumina–graphene oxide– poly(vinyl alcohol) artificial nacre with optimized strength and toughness. ACS Appl. Mater. Interfaces 7:9281–86
- Behr S, Vainio U, Müller M, Schreyer A, Schneider GA. 2015. Large-scale parallel alignment of plateletshaped particles through gravitational sedimentation. Sci. Rep. 5:9984
- Dwivedi G, Flynn K, Resnick M, Sampath S, Gouldstone A. 2015. Bioinspired hybrid materials from spray-formed ceramic templates. *Adv. Mater.* 27:3073–78
- Grunenfelder LK, de Obaldia EE, Wang Q, Li D, Weden B, et al. 2014. Stress and damage mitigation from oriented nanostructures within the radular teeth of *Cryptochiton stelleri*. *Adv. Funct. Mater.* 24:6093– 104
- Wang Q, Nemoto M, Li D, Weaver JC, Weden B, et al. 2013. Phase transformations and structural developments in the radular teeth of *Cryptochiton stelleri*. Adv. Funct. Mater. 23:2908–17
- Yaraghi NA, Guarín-Zapata N, Grunenfelder LK, Hintsala E, Bhowmick S, et al. 2016. A sinusoidally architected helicoidal biocomposite. *Adv. Mater.* 28:6835–44
- Amini S, Tadayon M, Idapalapati S, Miserez A. 2015. The role of quasi-plasticity in the extreme contact damage tolerance of the stomatopod dactyl club. *Nat. Mater.* 14:943–50
- Le Ferrand H, Bouville F, Niebel TP, Studart AR. 2015. Magnetically assisted slip casting of bioinspired heterogeneous composites. *Nat. Mater.* 14:1172–79
- Niebel TP, Bouville F, Kokkinis D, Studart AR. 2016. Role of the polymer phase in the mechanics of nacre-like composites. *J. Mech. Phys. Solids* 96:133–46

54 Yaraghi • Kisailus

- Finnemore A, Cunha P, Shean T, Vignolini S, Guldin S, et al. 2012. Biomimetic layer-by-layer assembly of artificial nacre. *Nat. Commun.* 3:966
- Gower LB, Odom DJ. 2000. Deposition of calcium carbonate films by a polymer-induced liquidprecursor (PILP) process. *J. Cryst. Growth* 210:719–34
- Gower LB. 2008. Biomimetic model systems for investigating the amorphous precursor pathway and its role in biomineralization. *Chem. Rev.* 108:4551–627
- Olszta MJ, Odom DJ, Douglas EP, Gower LB. 2009. A new paradigm for biomineral formation: mineralization via an amorphous liquid-phase precursor. *Connect. Tissue Res.* 44(Suppl. 1):326–34
- Mao LB, Gao HL, Yao HB, Liu L, Colfen H, et al. 2016. Synthetic nacre by predesigned matrix-directed mineralization. Science 354:107–10
- Raabe D, Sachs C, Romano P. 2005. The crustacean exoskeleton as an example of a structurally and mechanically graded biological nanocomposite material. *Acta Mater.* 53:4281–92
- Bouligand Y. 1972. Twisted fibrous arrangements in biological materials and cholesteric mesophases. *Tissue Cell* 4:189–217
- Chen P-Y, Lin AY-M, McKittrick J, Meyers MA. 2008. Structure and mechanical properties of crab exoskeletons. *Acta Biomater*. 4:587–96
- Paris O, Hartmann MA, Fritz-Popovski G. 2013. The mineralized crustacean cuticle: hierarchical structure and mechanical properties. In *Materials Design Inspired by Nature: Function Through Inner Architecture*, ed. P Fratzl, JWC Dunlop, R Weinkamer, pp. 180–96. Cambridge, UK: R. Soc. Chem.
- Fabritius H, Sachs C, Raabe D, Nikolov S, Friák M, Neugebauer J. 2011. Chitin in the exoskeletons of arthropoda: from ancient design to novel materials science. In *Chitin: Formation and Diagenesis*, ed. NS Gupta, pp. 35–60. Dordrecht, Neth.: Springer
- Bentov S, Abehsera S, Sagi A. 2016. The mineralized exoskeletons of crustaceans. In Extracellular Composite Matrices in Arthropods, ed. E Cohen, B Moussian, pp. 137–63. Cham, Switz.: Springer
- 73. Luquet G. 2012. Biomineralizations: insights and prospects from crustaceans. ZooKeys 176:103-21
- Grunenfelder LK, Herrera S, Kisailus D. 2014. Crustacean-derived biomimetic components and nanostructured composites. *Small* 10:3207–32
- Sachs C, Fabritius H, Raabe D. 2008. Influence of microstructure on deformation anisotropy of mineralized cuticle from the lobster *Homarus americanus*. J. Struct. Biol. 161:120–32
- 76. Zimmermann EA, Ritchie RO. 2015. Bone as a structural material. Adv. Healthc. Mater. 4:1286
- Zimmermann EA, Gludovatz B, Schaible E, Dave NK, Yang W, et al. 2013. Mechanical adaptability of the Bouligand-type structure in natural dermal armour. *Nat. Commun.* 4:2634
- Lichtenegger H, Müller M, Paris O, Riekel C, Fratzl P. 1999. Imaging of the helical arrangement of cellulose fibrils in wood by synchrotron X-ray microdiffraction. *J. Appl. Crystallogr.* 32:1127–33
- Wagermaier W, Gupta HS, Gourrier A, Burghammer M, Roschger P, Fratzl P. 2006. Spiral twisting of fiber orientation inside bone lamellae. *Biointerphases* 1:1–5
- Weinkamer R, Fratzl P. 2011. Mechanical adaptation of biological materials—the examples of bone and wood. *Mater. Sci. Eng. C* 31:1164–73
- Nikolov S, Petrov M, Lymperakis L, Friák M, Sachs C, et al. 2010. Revealing the design principles of high-performance biological composites using ab initio and multiscale simulations: the example of lobster cuticle. *Adv. Mater.* 22:519–26
- Gupta H, Stachewicz U, Wagermaier W, Roschger P, Wagner H, Fratzl P. 2006. Mechanical modulation at the lamellar level in osteonal bone. J. Mater. Res. 21:1913–21
- Fratzl P, Gupta HS, Fischer FD, Kolednik O. 2007. Hindered crack propagation in materials with periodically varying Young's modulus—lessons from biological materials. *Adv. Mater.* 19:2657–61
- Kolednik O, Predan J, Fischer FD, Fratzl P. 2011. Bioinspired design criteria for damage-resistant materials with periodically varying microstructure. *Adv. Funct. Mater.* 21:3634–41
- Launey ME, Buehler MJ, Ritchie RO. 2010. On the mechanistic origins of toughness in bone. Annu. Rev. Mater. Res. 40:25–53
- 86. Mouritz AP. 2007. Review of Z-pinned composite laminates. Composites A 38:2383-97
- Guarín-Zapata N, Gomez J, Yaraghi N, Kisailus D, Zavattieri PD. 2015. Shear wave filtering in naturallyoccurring Bouligand structures. *Acta Biomater*. 23:11–20

- 88. Roer R, Dillaman R. 1984. The structure and calcification of the crustacean cuticle. Am. Zool. 24:893-909
- Chockalingam S. 1971. Studies on enzymes associated with calcification of the cuticle of the hermit crab Clibanarius olivaceous. Mar. Biol. 10:169–82
- Luquet G, Dauphin Y, Percot A, Salomé M, Ziegler A, et al. 2016. Calcium deposits in the crayfish, *Cherax quadricarinatus*: microstructure versus elemental distribution. *Microsc. Microanal.* 22:22–38
- Sachs C, Fabritius H, Raabe D. 2006. Hardness and elastic properties of dehydrated cuticle from the lobster *Homarus americanus* obtained by nanoindentation. *J. Mater. Res.* 21:1987–95
- Fabritius H, Karsten ES, Balasundaram K, Hild S, Huemer K, Raabe D. 2012. Correlation of structure, composition and local mechanical properties in the dorsal carapace of the edible crab *Cancer pagurus. Z. Kristall.* 227:766–76
- Aizenberg J, Addadi L, Weiner S, Lambert G. 1996. Stabilization of amorphous calcium carbonate by specialized macromolecules in biological and synthetic precipitates. *Adv. Mater.* 8:222–26
- Loste E, Wilson RM, Seshadri R, Meldrum FC. 2003. The role of magnesium in stabilising amorphous calcium carbonate and controlling calcite morphologies. *J. Cryst. Growth* 254:206–18
- Politi Y, Batchelor DR, Zaslansky P, Chmelka BF, Weaver JC, et al. 2009. Role of magnesium ion in the stabilization of biogenic amorphous calcium carbonate: a structure–function investigation. *Chem. Mater.* 22:161–66
- Bentov S, Zaslansky P, Al-Sawalmih A, Masic A, Fratzl P, et al. 2012. Enamel-like apatite crown covering amorphous mineral in a crayfish mandible. *Nat. Commun.* 3:839
- Amini S, Masic A, Bertinetti L, Teguh JS, Herrin JS, et al. 2014. Textured fluorapatite bonded to calcium sulphate strengthen stomatopod raptorial appendages. *Nat. Commun.* 5:3187
- Moreno EC, Kresak M, Zahradnik RT. 1974. Fluoridated hydroxyapatite solubility and caries formation. Nature 247:64–65
- Okazaki M, Tohda H, Yanagisawa T, Taira M, Takahashi J. 1998. Differences in solubility of two types of heterogeneous fluoridated hydroxyapatites. *Biomaterials* 19:611–16
- Menéndez-Proupin E, Cervantes-Rodríguez S, Osorio-Pulgar R, Franco-Cisterna M, Camacho-Montes H, Fuentes ME. 2011. Computer simulation of elastic constants of hydroxyapatite and fluorapatite. *J. Mech. Behav. Biomed. Mater.* 4:1011–20
- 101. Politi Y, Priewasser M, Pippel E, Zaslansky P, Hartmann J, et al. 2012. A spider's fang: how to design an injection needle using chitin-based composite material. Adv. Funct. Mater. 22:2519–28
- 102. Schofield RMS, Nesson MH, Richardson KA, Wyeth P. 2003. Zinc is incorporated into cuticular "tools" after ecdysis: the time course of the zinc distribution in "tools" and whole bodies of an ant and a scorpion. *J. Insect Physiol.* 49:31–44
- Schofield RMS, Nesson MH, Richardson KA. 2002. Tooth hardness increases with zinc-content in mandibles of young adult leaf-cutter ants. *Naturwissenschaften* 89:579–83
- Cribb BW, Stewart A, Huang H, Truss R, Noller B, et al. 2008. Insect mandibles—comparative mechanical properties and links with metal incorporation. *Naturwissenschaften* 95:17–23
- Schofield RM, Lefevre HW. 1992. PIXE-STIM microtomography: zinc and manganese concentrations in a scorpion stinger. Nucl. Instrum. Methods Phys. Res. 72:104–10
- Cheng L, Thomas A, Glancey JL, Karlsson AM. 2011. Mechanical behavior of bio-inspired laminated composites. *Composites A* 42:211–20
- Grunenfelder LK, Suksangpanya N, Salinas C, Milliron G, Yaraghi N, et al. 2014. Bio-inspired impactresistant composites. *Acta Biomater*. 10:3997–4008
- Ladani RB, Ravindran AR, Wu S, Pingkarawat K, Kinloch AJ, et al. 2016. Multi-scale toughening of fibre composites using carbon nanofibres and Z-pins. *Compos. Sci. Technol.* 131:98–109
- 109. Gao W, Zhang Y, Ramanujan D, Ramani K, Chen Y, et al. 2015. The status, challenges, and future of additive manufacturing in engineering. *Comput.-Aided Des.* 69:65–89
- Ang TH, Sultana FSA, Hutmacher DW, Wong YS, Fuh JYH, et al. 2002. Fabrication of 3D chitosanhydroxyapatite scaffolds using a robotic dispensing system. *Mater. Sci. Eng. C* 20:35–42
- Geng L, Feng W, Hutmacher DW, Wong YS, Loh HT, Fuh JY. 2005. Direct writing of chitosan scaffolds using a robotic system. *Rapid Prototyp. J.* 11:90–97

- 112. Dellinger JG, Cesarano J, Jamison RD. 2007. Robotic deposition of model hydroxyapatite scaffolds with multiple architectures and multiscale porosity for bone tissue engineering. *J. Biomed. Mater. Res. A* 82A:383–94
- 113. Almeida CR, Serra T, Oliveira MI, Planell JA, Barbosa MA, Navarro M. 2014. Impact of 3-D printed PLA-and chitosan-based scaffolds on human monocyte/macrophage responses: unraveling the effect of 3-D structures on inflammation. *Acta Biomater*. 10:613–22
- Ribbans B, Li Y, Tan T. 2016. A bioinspired study on the interlaminar shear resistance of helicoidal fiber structures. J. Mech. Behav. Biomed. Mater. 56:57–67
- 115. Milliron GW. 2012. Lightweight impact-resistant composite materials: lessons from mantis shrimp. PhD Thesis, Univ. Calif., Riverside
- Shopsowitz KE, Qi H, Hamad WY, MacLachlan MJ. 2010. Free-standing mesoporous silica films with tunable chiral nematic structures. *Nature* 468:422–25
- 117. Majoinen J, Kontturi E, Ikkala O, Gray DG. 2012. SEM imaging of chiral nematic films cast from cellulose nanocrystal suspensions. *Cellulose* 19:1599–605
- Wang B, Walther A. 2015. Self-assembled, iridescent, crustacean-mimetic nanocomposites with tailored periodicity and layered cuticular structure. ACS Nano 9:10637–46
- Matsumura S, Kajiyama S, Nishimura T, Kato T. 2015. Formation of helically structured chitin/CaCO₃ hybrids through an approach inspired by the biomineralization processes of crustacean cuticles. *Small* 11:5127–33
- 120. Nan F, Chen Q, Liu P, Nagarajan S, Duan Y, Zhang J. 2016. Iridescent graphene/cellulose nanocrystal film with water response and highly electrical conductivity. *RSC Adv.* 6:93673–79
- Nguyen T-D, Peres BU, Carvalho RM, MacLachlan MJ. 2016. Photonic hydrogels from chiral nematic mesoporous chitosan nanofibril assemblies. *Adv. Funct. Mater.* 26:2875–81
- 122. Oh DX, Cha YJ, Nguyen H-L, Je HH, Jho YS, et al. 2016. Chiral nematic self-assembly of minimally surface damaged chitin nanofibrils and its load bearing functions. *Sci. Rep.* 6:23245
- 123. Zhu B, Merindol R, Benitez AJ, Wang B, Walther A. 2016. Supramolecular engineering of hierarchically self-assembled, bioinspired, cholesteric nanocomposites formed by cellulose nanocrystals and polymers. ACS Appl. Mater. Interfaces 8:11031–40
- 124. Al-Sawalmih A, Li C, Siegel S, Fabritius H, Yi S, et al. 2008. Microtexture and chitin/calcite orientation relationship in the mineralized exoskeleton of the American lobster. *Adv. Funct. Mater.* 18:3307–14
- 125. Salinas CL, de Obaldia EE, Jeong C, Hernandez J, Zavattieri P, Kisailus D. 2017. Enhanced toughening of the crossed lamellar structure revealed by nanoindentation. *J. Mech. Behav. Biomed. Mater.* 76:58–68
- 126. Sullivan TN, Pissarenko A, Herrera SA, Kisailus D, Lubarda VA, Meyers MA. 2016. A lightweight, biological structure with tailored stiffness: the feather vane. Acta Biomater. 41:27–39
- 127. Su FY, Bushong EA, Deerinck TJ, Seo K, Herrera S, et al. 2017. Spines of the porcupine fish: structure, composition, and mechanical properties. *J. Mech. Behav. Biomed. Mater.* 73:38–49
- 128. de Obaldia EE, Herrera S, Grunenfelder LK, Kisailus D, Zavattieri P. 2016. Competing mechanisms in the wear resistance behavior of biomineralized rod-like microstructures. *J. Mecb. Phys. Solids* 96:511–34
- 129. Sherman VR, Yaraghi NA, Kisailus D, Meyers MA. 2016. Microstructural and geometric influences in the protective scales of *Atractosteus spatula*. J. R. Soc. Interface 13:20160595
- Suksangpanya N, Yaraghi NA, Kisailus D, Zavattieri P. 2017. Twisting cracks in Bouligand structures. J. Mecb. Bebav. Biomed. Mater. 76:38–57
- 131. Yaraghi NA, Guarín-Zapata N, Grunenfelder LK, Wuhrer R, Zavattieri PD, Kisailus D. 2016. Biomimetic composites derived from an impact resistant crustacean. In *Insights and Innovations in Structural Engineering, Mechanics and Computation*, ed. A Zingoni, pp. 890–96. London: CRC Press
- 132. Kisailus D, Milliron G. 2016. Shock and impact resistant materials. US Patent No. 9452978
- 133. Grunenfelder LK, Milliron G, Herrera S, Gallana I, Yaraghi NA, et al. 2017. Ecologically driven ultrastructural and hydrodynamic designs in stomatopod cuticles. *Adv. Mater.* Accepted

v

Annual Review of Physical Chemistry Volume 69, 2018

Contents

Addressing the Challenge of Molecular Change: An Interim Report Raphael D. Levine
Biomimetic Structural Materials: Inspiration from Design and Assembly Nicholas A. Yaraghi and David Kisailus
An Active Approach to Colloidal Self-Assembly Stewart A. Mallory, Chantal Valeriani, and Angelo Cacciuto
Excitons in Single-Walled Carbon Nanotubes and Their Dynamics Amanda R. Amori, Zhentao Hou, and Todd D. Krauss
Slow Photoelectron Velocity-Map Imaging of Cryogenically Cooled Anions Marissa L. Weichman and Daniel M. Neumark
Graph Theory and Ion and Molecular Aggregation in Aqueous Solutions Jun-Ho Choi, Hochan Lee, Hyung Ran Choi, and Minhaeng Cho
Permutationally Invariant Potential Energy Surfaces Chen Qu, Qi Yu, and Joel M. Bowman
Straightening the Hierarchical Staircase for Basis Set Extrapolations: A Low-Cost Approach to High-Accuracy Computational Chemistry <i>António J.C. Varandas</i>
Connections Between Theory and Experiment for Gold and Silver Nanoclusters K.L. Dimuthu M. Weerawardene, Hannu Häkkinen, and Christine M. Aikens 205
Characterization of Intermediate Oxidation States in CO ₂ Activation Leah G. Dodson, Michael C. Thompson, and J. Mathias Weber
Measuring Electric Fields in Biological Matter Using the Vibrational Stark Effect of Nitrile Probes Joshua D. Slocum and Lauren J. Webb

Indexes

Cumulative Index of Contributing Authors, Volumes 65–69	. 541
Cumulative Index of Article Titles, Volumes 65–69	. 545

Errata

An online log of corrections to *Annual Review of Physical Chemistry* articles may be found at http://www.annualreviews.org/errata/physchem