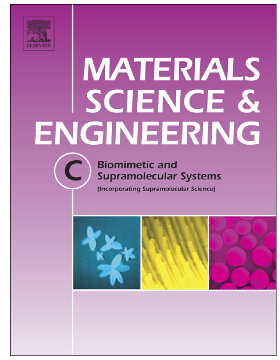
Journal Pre-proof



Marine invertebrates are a source of bioadhesives with biomimetic interest

Mariana Almeida, Rui L. Reis, Tiago H. Silva

PII: S0928-4931(19)32975-3

DOI: https://doi.org/10.1016/j.msec.2019.110467

Reference: MSC 110467

To appear in: *Materials Science & Engineering C*

Received date: 16 September 2019

Revised date: 6 November 2019

Accepted date: 18 November 2019

Please cite this article as: M. Almeida, R.L. Reis and T.H. Silva, Marine invertebrates are a source of bioadhesives with biomimetic interest, *Materials Science & Engineering C* (2019), https://doi.org/10.1016/j.msec.2019.110467

Marine invertebrates are a source of bioadhesives with biomimetic interest

Mariana Almeida[a,b], Rui L. Reis[a,b,c], Tiago H. Silva[a,b*]

[a] 3B's Research Group, I3Bs – Research Institute on Biomaterials, Biodegradables and Biomimetics, University of Minho, Headquarters of the European Institute of Excellence on Tissue Engineering and Regenerative Medicine, AvePark, Parque de Ciência e Tecnologia, Zona Industrial da Gandra, 4805-017 Barco, Guimarães, Portugal;

[b] ICVS/3B's–PT Government Associate Laboratory, Braga/Guimarães, Portugal;

[c] The Discoveries Centre for Regenerative and Precision Medicine, Headquarters at University of Minho, Avepark, 4805-017 Barco, Guimarães, Portugal

*Corresponding author: tiago.silva@i3bs.uminho.pt

Declarations of interest: none

Abstract

Protein-based bioadhesives are found in diverse marine invertebrates that developed attachment devices to adhere to various substrates. These adhesives are of interest to materials science to create bioinspired-adhesives that can perform in water or wet conditions and can be applied in a broad variety of biotechnological and industrial fields. Due to the high variety of invertebrates that inhabit the marine environment, an enormous diversity of structures and principles used in biological adhesives remains unexplored and a very limited number of model systems have inspired novel biomimetic adhesives, the most notable being the mussel byssus adhesive. In this review we give an overview of other marine invertebrates studied for their bioadhesive properties in view of their interest for the development of new biomimetic adhesives for application in the biomedical field but also for antifouling coatings. The molecular features are described, highlighting relevant structures and examples of biomimetic materials are discussed and explored, opening an avenue for a new set of medical products.

Keywords: adhesive proteins; marine invertebrates; biomimetics; marine biomaterials

1. Introduction

Biomaterials and bio-inspired designs targeted for industrial and technological applications such as pharmaceutical, biomedical and cosmetics have aroused interest in the exploration of marine biological resources (Silva et al., 2012). Approximately 97% of the animal species described to date are invertebrates belonging to more than 30 phyla, of these all except one occurs in the marine environment and 15 are exclusively marine (Bouchet, 2006; Ruggiero et al., 2015). Therefore, oceans, seas and coasts host a high diversity of invertebrates, with differing structure and physiology, in which marine materials with remarkable functional properties can be found, such as adhesives that bond surfaces underwater (Waite, 2017), ceramics with similarities with the mineral constituents of bones (Wang et al., 2012) and mutable collagenous tissues with changeable properties (Sugni et al., 2014).

In view of biomimetics, adhesives produced by marine invertebrates have received great attention because they can cure in wet environments, with potential biocompatible properties for biomedical applications such as tissue repair and wound sealants and could improve the performance of current adhesives (Cui et al., 2017). These adhesives are of strong interest to the biomedical field but also encompasses other research fields, namely the cultivation of marine species that depend on the attachment (López et al., 2010) and fouling-release coatings to prevent biofouling (Kamino, 2013), with consequences for the conservation and sustainable use of biological resources (Leal et al., 2018).

Adhesives are found in a high range of organisms that have life histories that depend upon their attachment to a substrate (Gorb, 2008). Marine invertebrates are among the organisms that produce adhesive polymers to attach permanently to a substrate (e.g. representatives among crustaceans and molluscs) or to attach and detach temporarily (e.g. most echinoderms) (Flammang and Santos, 2014). Generally, these are highly viscous or solid secretions with variable biochemical composition. Some are principally composed of proteins (Kamino, 2013; Waite, 2017) while others are a combination of proteins and carbohydrates usually also comprising alarge inorganic fraction (Hennebert et al., 2012; Li et al., 2018). Most studies have focused on the protein fraction but other polymers such as carbohydrates may have also a role in the adhesive process and little is known about the composition of other fractions. Despite the progress achieved with recent studies, their thorough physico-chemical characterization is hampered because of the insolubility of the material and the difficulty to obtain significant amounts of the adhesive.

The most well characterized marine adhesive is from marine mussels of the genus *Mytilus*, that attracted material science in the last decades and has inspired most of the biomimetic adhesives currently available. The most remarkable properties found in the mussel adhesive are proteins rich in the catecholic amino acid, 3,4-dihydroxyphenylalanine (DOPA), a residue formed by the post-translational modification (PTM) tyrosine hydroxylation, which is involved in the adsorption of the adhesive proteins to the substrate (adhesive surface bonding) and in the formation of cross-links between these different proteins (cohesive curing) (Waite, 2017). Synthetic mussel adhesive proteins and peptides were developed to produce self-healing hydrogels (e.g. Kim et al., 2014) and pH-responsive drug carriers (e.g Kim et al., 2015) while approaches using chemical groups involved in mussel adhesion, such as catechol-modified polymers, have been used to develop adhesive coatings (e.g. Carvalho et al., 2016), bioadhesives (e.g. Brubaker et al., 2010; White and Wilker, 2011) and sealants (e.g. Perrini et al., 2016) for a variety of biomedical applications. Other adhesives in marine invertebrates have gained increased interest in view of biomimetics as they have distinctive characteristics that those found in the mussel adhesive or, being similar, can fill in the understanding of adhesive processes.

This review intends to give a general overview of the molecular characteristics of the adhesives secreted by adult marine invertebrates in the context of biomimetics research, from the relatively well studied barnacles and tubeworms to the less known tunicates. It is organized by major marine invertebrate groups according to increasing organism complexity, following the organization adopted by Brusca and Brusca (2003). The adhesive of the mussel is not a focus of this review. For recent review of mussel adhesives see Waite (2017) and Balkenende et al. (2019). The marine invertebrate larvae, which may have differences in the mechanism of attachment, and small marine organisms (generally up to 1 mm) are also addressed elsewhere (Gohad et al., 2014; Lengerer et al., 2014).

The molecular characterization of the adhesives described in this review is based on histological and histochemistry procedures to characterize the secretory glands or on transcriptomic and proteomic approaches used to identify the adhesion related genes and/or the polypeptide sequences and post-translation modifications of the secreted proteins or their precursors in the cells. When available, potential applications of those marine adhesives or inspired materials in regenerative medicine and tissue engineering are also addressed. Finally, some remarks on the future use of marine invertebrate adhesives are made.

2. Protein-based adhesives in marine invertebrates

2.1 Polychaetes

Several polychaete worms with a tube dwelling lifestyle produce strong, resilient tubes that support and protect them under a variety of environmental settings, withstanding high-energy intertidal waves or providing barriers that moderate thermal and chemical extreme conditions (e.g. deep-sea hydrothermal vents) (Merz, 2015).

Some species, commonly called sandcastle or honeycomb worms, are bioengineering animals capable of forming massive reef-like mounds in coastal ecosystems by the association of thousands of individual tubes placed side by side (Fig. 1 A). Each worm builds their composite tube by collecting sand grains and calcareous shell fragments from the water and by applying a proteinaceous adhesive secreted by adhesive glands located near the mouth, to join the particles and place them at the extremity of the pre-existing tube (Stewart et al., 2004).

In particular, the tubeworm *Phragmatopoma californica* has been subject of investigation due to their remarkable adhesive properties for the development of biomimetic materials (Buffet et al., 2018; Endrizzi and Stewart, 2009; Shao et al., 2009; Stewart et al., 2004; Sun et al., 2010; Zhao et al., 2005). The adhesive is composed of several highly repetitive and oppositely charged proteins (Pcs), DOPA, sulfated polysaccharides and magnesium and calcium ions. The presence of inorganic crystals is a unique feature present in these organisms compared to the invertebrate groups described in the following sections. Pc-1, Pc-2, Pc-4 and Pc-5 proteins are rich in glycine, lysine, histidine and tyrosine from which Pc-1 and Pc-2 contain DOPA residues (Waite et al., 1992). Pc-3 protein and the variants Pc-3A and Pc-3B are characterized by serine residues largely phosphorylated and are the most unusual components of the adhesive compared to other characterized underwater adhesives (Endrizzi and Stewart, 2009; Zhao et al., 2005). The oppositely charged proteins are distributed into two types of secretory cells that produce “homogeneous” or “heterogeneous” secretory granules. Pc-2, Pc-5 and sulfated polysaccharides are located in the homogeneous granules, while Pc1, Pc-4, Pc-3A, Pc-3B and Mg^{2+} ions are located in the heterogeneous granules (Stewart et al., 2011, 2004) (Fig. 1; Table 1).

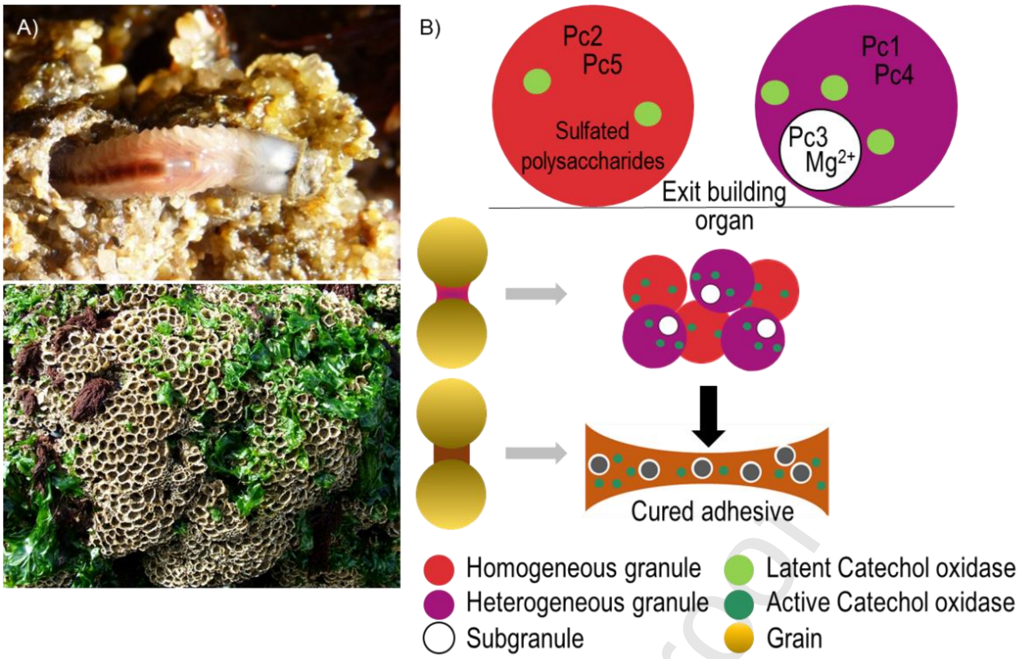


Figure 1. A) Representative species of a sandcastle worm and the reefs built by these animals; B) Model of coacervation of the tubeworm *P. californica*. Adapted with permission from (Wang and Stewart, 2013). Copyright 2013 American Chemical Society. (2-column)

A coacervation system (liquid liquid phase separation) was proposed to occur during secretory granule condensation. Briefly, within the secretory pathway of the adhesive gland cells, the entropic gain resulting from the electrostatic association of the oppositely charged proteins and divalent cations (Ca^{2+} and Mg^{2+}) condense the adhesive proteins into dehydrated secretory granules that leads to the separation of the solution into two aqueous phases (complex coacervation). After deposition onto the tube, the rehydration of the condensed granules contributes to the displacement of water from the mineral substrate to facilitate underwater adhesion and the covalent cross-linking through oxidative coupling of DOPA along with the phosphates participate in the strong adhesion of the cement (Stewart et al., 2011, 2004) (Fig. 1; Table 1).

More recently, studies on the adhesion mechanism of the tubeworms *Sabellaria alveolata and P. caudata* identified additional components of tubeworm adhesive and shed light to the role of tyrosinases and peroxidases in the chemistry of the adhesive process, which may correspond to new targets to develop biomimetic approaches (Buffet et al., 2018).

Within the polychaetes, other families build calcified, mucous or chitinous tubes (Fig. 2). The tube and adhesive of the polychaete *Hydroides dianthus* were studied in a biomineralization context and the adhesive revealed to be an inorganic-organic composite material, consisting of inorganic aragonite and Mg-calcite crystals, with an organic material associated with the crystals (Tanur et al., 2010). It would be relevant to study the functional role of inorganic crystals in these organisms and also to extend research to other tube-building polychaetes..

Figure 2. Tubes built by some polychaete species composed of calcium carbonate.
(single)

2.1.1 Biomimetic interest

Some work has been developed towards the production of adhesives based on coacervate phenomena involved in the tubeworm *P. californica* adhesion (Table 1). Shao et al. (Shao et al., 2009) designed polyacrylate adhesive proteins analogs of the adhesive secreted by *P. californica*, containing phosphate, primary amine, and catechol side chains, with molar ratios similar to their natural counterparts. The adhesive material was applied to glue wet cortical bone specimens, showing that the bond strength was around 40% of the commercial cyanoacrylate adhesives (Shao et al., 2009). More recently, an endovascular embolic agent that mimic the polyelectrolyte composition, condensed ionic strength dependent viscosity and form, proved effective for deep distal penetration and 100% de-vascularization in acute renal embolization (Jones et al., 2016).

Inspired by granule-packaged viscous adhesive secretion mechanism, a nanoparticulate formulation of a viscous adhesive based on a hydrophobic light-activating adhesive (HLAA) was developed that can be assembled into the native viscous glue state following injection and can be cured in response to on-demand external stimuli (Lee et al., 2015). Inspired by the coacervate system of *P. californica*, the multiphase adhesive created by complex coacervation of synthetic copolyelectrolytes that mimic tube worm proteins and by incorporation of polyethylene glycol diacrylate (PEG-dA) monomers in the coacervate phase also showed improved bond strength and fluid behaviour (Kaur et al., 2011). A coacervate hydrogel combining dopamine conjugated hyaluronic acid (HA-DN) and lactose modified chitosan (chitlac) and following catechol chemistry was also developed. The hydrogel showed injectable and re-moldable physical properties with potential long-term stability under water (Oh et al., 2012). A new method based on the solvent exchange concept was used to develop adhesives with rapid and robust wet adhesion performance. Zhao et al. (2016) designed a catechol-containing poly (acrylic acid) with a quaternized chitosan (QCS), ion-paired with bis(trifluoromethane-sulfonyl)imide (Tf2N) in dimethyl sulfoxide (DMSO). The water–DMSO solvent exchange whereupon electrostatic complexation, phase inversion, and rapid setting were simultaneously actuated by water–DMSO solvent exchange and a robust underwater contact adhesion was achieved in different surfaces (Zhao et al., 2016). Further characterization of the adhesive mechanisms of marine tubeworms will provide insights into the design of new and improved underwater adhesives.

2.2 Crustaceans

Barnacles are sessile crustaceans that permanently attach to various substrates. Their body is enclosed in calcified plates and the base plate is attached to a substrate by means of an adhesive layer, sometimes referred to as cement, produced by adhesive glands located just above the baseplate (Kamino et al., 2000). Barnacles are traditionally divided in acorn barnacles, those with calcareous baseplate directly attached to the substrate, and stalked barnacles which have a peduncle with a chitinous membrane attached to the substrate (Walker, 1992) (Fig. 3).

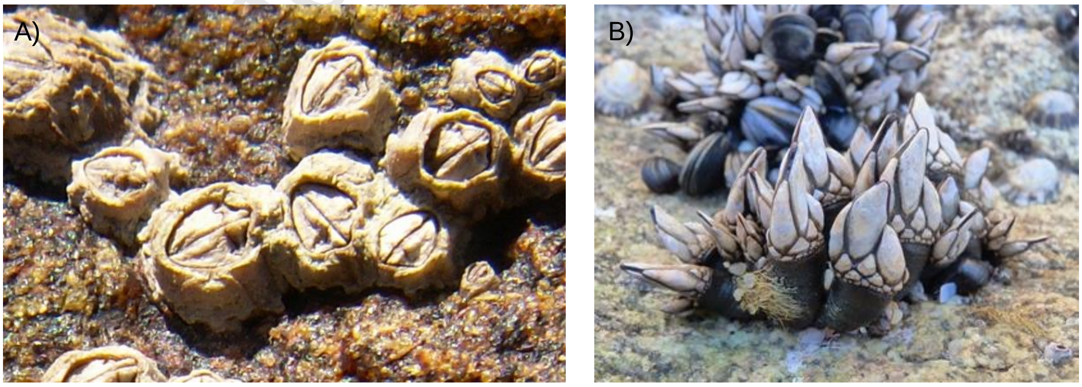


Figure 3. Representative species of A) a stalked barnacle and B) an acorn barnacle. (1.5 column)

In coastal environments, barnacles live on rocks where they face waves and tidal currents. They are also biofoulers as they adhere to a variety of man-made structures, like ships and sensors, causing economic and ecological damages (Holm, 2012). In fact, the interest in barnacle cement arise from the problem of biofouling, namely to better understand the adhesion mechanism and design strategies to hamper it, aiming to establish anti-fouling approaches, but other interests emerge as they produce a strong adhesive with potential application in the biomedical field (Kamino, 2013). Even though barnacles have been widely studied, the mechanisms allowing them to permanently attach to surfaces underwater remain unclear (Jonker et al., 2014; Kamino, 2013).

Studies performed on several acorn barnacles revealed a composition of almost entirely proteins (>90%), from which several proteins have no homologues in the available database, and apparently a molecular system of attachment different from the mussel and tubeworm models (Kamino, 2010).

Five protein sequences were found in the adhesive or adhesive gland named according to their apparent molecular weight (Cps) (mostly obtained from *Megabalanus rosa* and *Amphibalanus amphitrite*) (Fig. 4; Table 1). Two hydrophobic proteins, cp100k and cp52k, are thought to comprise the bulk of fibrillar cement. The hydrophobic nature of the proteins has been suggested to contribute to the cross-linking framework and cohesion (Kamino et al., 2000). Two hydrophilic proteins, cp19k and cp68k, rich in serine, threonine, alanine and glycine residues, participate in interfacial adhesion (Kamino, 2013). A hydrophilic protein, cp20k, rich in cysteine residues, has been suggested to be related to the adherence of the calcareous plates to the substrate (Urushida et al., 2007). Little or no occurrence of post-translational modification occurs (DOPA or phosphorylated serine) in the barnacle adhesive proteins, only cp52k has been found to have limited glycosylation (Kamino, 2013). Given the low amounts of adhesive produced by these organisms, this feature is important for the production of recombinant cp proteins for application purposes even though synthetic and recombinant adhesive materials inspired by barnacle adhesion have rarely been developed (So et al., 2018).

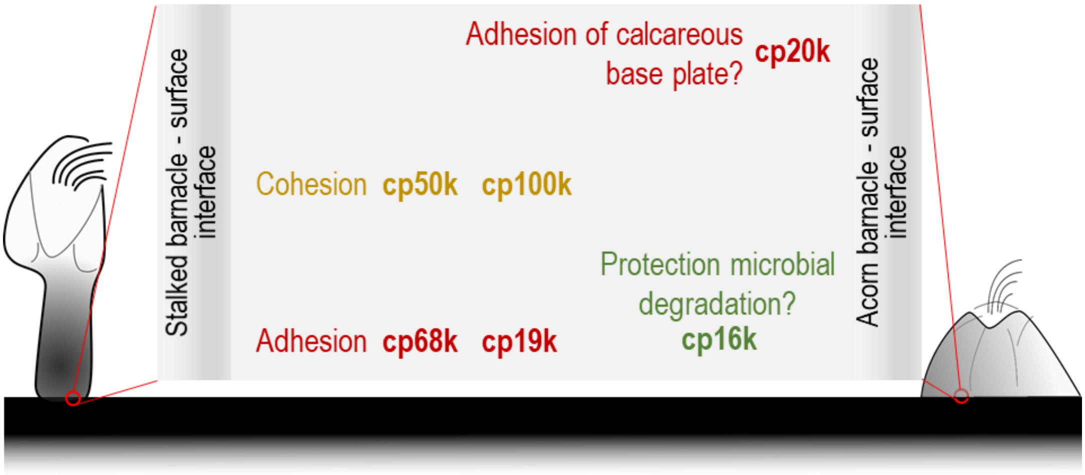


Fig. 4. Adhesive proteins involved in barnacle attachment. Note: cp20k is only described for acorn barnacles with a calcareous base plate. (2-column)

Some of these proteins (cp100k, cp68k, cp52k, cp19k) appear to be present in stalked species as well, namely in *Lepas anatifera, Dosima fascicularis* and *Pollicipes pollicipes* (Jonker et al., 2014; Rocha et al., 2019) while cp-20k may be absent, which may indicate that cp-20K is related to the adhesion of calcareous but not membranous bases to a substrate (Rocha et al., 2019) (Fig. 4).

Recently, the use of solvents to solubilize a significant unidentified portion of the cement of *A. amphitrite* revealed the existence of low complexity glycine/serine-rich cement proteins (GSrCPs) and leucine-rich cement proteins (LrCPs), as well as multiple lysyl oxidases and peroxidases. GSrCPs were found to share homology to certain silk motifs and revealed a prominent role in the construction of barnacle cement nanofibrils (So et al., 2016).

In fact, it has been suggested that structural integrity of the cured adhesive is provided by proteins that form cross-β-sheet fibres, like amyloid fibers, that provide the insolubility and stability to the complex and aggregation of components. These structures might also be important to the cohesive strength of the adhesive through the considerable number of hydrogen bonds between cross-β-sheets (Barlow et al., 2010; Liu et al., 2017; Nakano and Kamino, 2015; So et al., 2016). The molecular features of these amyloid-like proteins with silk homology found in barnacles as well as in other marine organisms (e.g. algae; Mostaert et al., 2009) are worth of further investigation. For example, other crustaceans, such as small shrimp-like tube dwelling amphipods, form tubes by collecting sand grains and organic material and by secreting an adhesive material through specialist secretory legs (Shillaker and Moore, 1978). Apparently, the secretion has a carbohydrate-protein content and is dominated by complex β-sheet structures and a high amount of charged amino acid residues with common elements of barnacle´s adhesive and spider silk (Kronenberger et al., 2012).

2.3 Molluscs

Marine mussels adhere to a variety of substrates with byssus threads secreted by the byssus gland of the foot. The adhesive properties of the mussel byssus have been under investigation in the last decades targeting the design of materials with biological origin (Balkenende et al., 2019; Waite, 2017). Despite the major focus of scientific community on mussels, the mechanisms of attachment of other molluscs are also of interest as they may provide approaches to develop new adhesives, even so their study is in its infancy (Fig. 5).

Figure 5. Molluscs with attachment devices based on adhesives. Representative species of a A) scallop; B) oyster; C) limpet and D) marine snail. (1.5 fitting image)

Scallops, for example, are bivalve molluscs that also produce byssus for attachment, however, they can detach the byssus and make movements to search for new space through the secretion of temporary byssus (Alejandrino et al., 2011) (Fig. 5A). Using a combination of transcriptomic-proteomic approaches to study the adhesion of the scallop *Chlamys farreri*, seven scallop byssus protein (SBPs) were identified from which only three showed significant aminoacid sequence homology to known proteins and only one showed homology to mussel adhesive protein, thus suggesting differences in protein composition. The analysis also suggested PTMs, namely phosphorylation and hydroxylation (Miao et al., 2015). One protein, Sbp8-1 found in byssus was annotated as an atypical metalloproteinase with two extra free cysteine residues putatively involved in the sbp8-1 polymerization and having a functional role in the cross-linking of the scallop byssus through the interaction of cysteine and DOPA (Zhang et al., 2018) (Table 1).

Another adhesive that has attracted interest in recent years is the one produced by oysters due to their economic interest and ecological role as ecosystem engineers (e.g. protection of shorelines) (Fig. 5B). A first characterization of the adhesive from *Crassostrea virginica* showed that the adhesive is a composite material consisting of proteins, polysaccharides and phospholipids possibly responsible for adhesion, together with an inorganic component largely composed of calcium carbonate and silica inclusions providing strong cohesion (Alberts et al., 2015; Metzler et al., 2016) (Table 1). The presence of cross-linked phosphorylated proteins showed to be an analogy to mussel adhesives whereas the high inorganic content is exclusive of oysters (Burkett et al., 2010). Even if the detailed mechanism of oyster's adhesion remains unclear, these animals give new insights on different type of marine adhesives from marine organisms and on the production of new types of organic-inorganic hybrid adhesives with comparable adhesion performance to mussel-inspired DOPA-based adhesives (Li et al., 2018).

Among molluscs, marine gastropods, such as marine snails and limpets, produce mucus that have not yet been studied in detail (Smith, 2016) (Fig. 5C, D). This mucus is used for a variety of functions including locomotion and protection and some are also used to adhere themselves to the substrate such as rocks and seaweeds (Davies and Hawkins, 1998). The limpet *Lottia limatula* and the periwinkle *Littoraria irrorata* for example, produce a non-adhesive gel for locomotion, use suction for strong attachment but also produce an adhesive-like material to fix very strongly to the substrate when are exposed to air in low tide conditions) (Smith, 1992). Preliminary analysis of the composition of the adhesive gel revealed a high percentage of water (95%), an organic fraction containing carbohydrates and specific proteins that confer the adhesive properties (Smith et al., 1999) which have the ability to cross link and with a gel-stiffening action (Pawlicki, 2004). Echinoderms also adhere using highly hydrated secretions (section 2.4) but mucus from gastropods apparently have a high water content. Further investigation is needed to relate the adhesive properties to the high water content and also to evaluate the gel stiffening proteins and potential useful properties.

Among cephalopods, preliminary analysis of the biochemical composition of adhesive mucus secreted by four genus (*Euprymna*, *Idiosepius, Nautilus* and *Sepia*) indicates a composition of carbohydrates and proteins. Nevertheless, so far most cephalopod-inspired tissue adhesives did not address this chemical features, having mimicked only the mechanical adhesion (von Byern and Klepal, 2006).

2.4 Echinoderms

Echinoderms have received great attention in development studies due to their proximity to vertebrates. Among several defining characteristics, a unique echinoderm feature is a water vascular system usually evident externally as muscular podia or tube feet composed by a disc at the apical extremity that contacts with the substrate, which is used for locomotion, attachment, food capture or burrowing (Brusca and Brusca, 2003). Through this disc, echinoderms produce strong but reversible adhesives to adhere to the substrate with adhesion strength in the range of values found on other marine organisms known to adhere permanently to the substrate (Flammang et al., 2016) (Fig. 6). The reversible attachment is accomplished via a duo-gland adhesion system, which produce a protein-based adhesive allowing the tube feet to attach and de-adhesive secretions, that act enzymatically on footprint proteins to enable their release from the tube feet (Flammang et al., 1998).

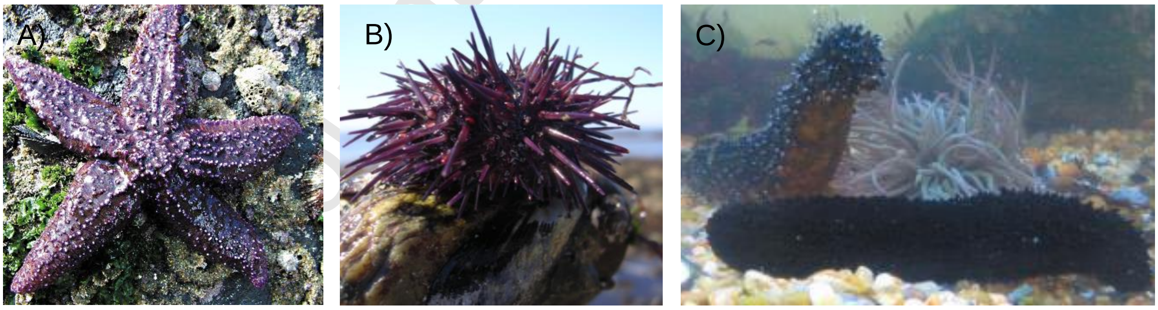


Figure 6. Echinoderms with devices based on adhesives. Representative species of a A) sea star; B) sea urchin; C) sea cucumber. (1.5 fitting image)

To the best of our knowledge, only the adhesives from the sea star *Asterias rubens* and sea urchin *Paracentrotus lividus* were analysed in detail (Fig. 6A,B). Studies conducted so far revealed an adhesive composed by an inorganic fraction and an organic fraction made up of proteins (Flammang et al., 1998; Santos et al., 2009). The protein fraction contains high amounts of charged (especially acidic) and uncharged polar amino acids, large amounts of cysteine, traits commonly observed in marine adhesives and pointed out as factors of high adhesion and cohesion and insolubility (Hennebert et al., 2012; Santos et al., 2009). Moreover, PTMs such as phosphorylation and glycosylation were also identified, in accordance with other marine adhesive proteins (Lebesgue et al., 2016; Santos et al., 2013).

Despite the recent advances in transcriptome analysis indicating putative novel adhesive proteins in echinoderms, to date only two have been characterized in echinoderm tube feet. Sea star footprint protein 1 (Sfp1) was assigned as a main constituent of the *A. rubens* adhesive. The protein forms a structural scaffold and seems to provide cohesion to the adhesive layer, rather than adhesive properties (Hennebert et al., 2014) (Table 1). Nectin, a cell adhesion protein secreted by the eggs and embryos of *P. lividus*, was also present in the adult adhesive and involved in the adhesion process (Lebesgue et al., 2016) (Table 1). As other proteins, such as actins and histone, were detected in relevant abundance in the footprints of both sea stars and sea urchins, their role in the adhesive process should be considered (Lebesgue et al., 2016).

A different adhesive mechanism is documented for some species of sea cucumbers (Fig. 6C). Adhesives in these animals are secreted by Cuvier tubules, typical instantaneous adhesive structures discharged by these animals to entangle and immobilize potential predators (Brusca and Brusca 2003). The adhesive composition of *Holothura forskali* is similar to the ones produced by other echinoderms but it differs in the carbohydrate fraction composition and by the lower inorganic content. Several proteins in the sea cucumber of the genus *Holothuria* (e.g. *Holothuria forskali, H.dofleinii*) have been identified in the adhesive, but no confirmation of their function has been provided (DeMoor et al., 2003). The identification of a C-type lectin in the *H. dofleinii* tubule raise the hypothesis of involvement of glycoproteins in sea cucumber adhesion, as well as the involvement of enzyme-like proteins in structural and/or ligand-binding properties (Peng et al., 2014) (Table 1). The elucidation of their characteristics should provide information of the underwater adhesive mechanism of holothuroid Cuvierian tubules and other animals that used this type of defense mechanism (e.g. ctenophorans).

Adhesives from other echinoderm species are currently being characterized to identify shared features of temporary adhesives in echinoderms and therefore increase the understanding the properties of temporary adhesion systems (e.g. *Asterina gibbosa*; Lengerer et al., 2018).

2.5 Tunicates

Tunicates (sea squirts), the closest living relatives of vertebrates, are sac like sessile marine organisms found attached to a variety of substrates at all ocean depths. These animals have received great attention in evolutionary and developmental studies as well as in the field of biomedicine owing to their remarkable features, such as strong adhesiveness and rapid self-regeneration (Cho et al., 2018; Pennati and Rothbacher, 2015) (Fig. 7).

Their body wall or tunic is composed of a cellulose fiber called tunicin and proteins containing DOPA and 3,4,5-trihydroxyphenylalanine (TOPA), pyrogallol amino acid, which contribute to underwater adhesion and rapid self-regeneration, as described for *Molgula manhattensis* and *Ascidia ceratodes* (Taylor et al., 1997) (Table 1). Tunicate-mimetic adhesives were developed combining gallic acids with chitin nanofibers (Oh et al., 2015) and with chitosan (Sanandiya et al., 2019) showing higher adhesion strength than mussel-mimetic adhesives and a medical adhesive, fibrin glue. Other approach combining gallic acid and metal ions has been suggested as an anesthetic solution for the treatment of dentin hypersensitivity (Prajatelistia et al., 2016). Likewise, Cho et al. (2018) highlighted the role of PG (pyrogallol) moieties in TOPA-containing compounds as bioinspired natural motifs for developing functional biomaterials. The understanding of this unique feature found in ascidians and the knowledge from regenerativestudies in these animals can advance the characterization of their adhesive properties and enhance the development of new biomimetic materials (Pennati and Rothbacher, 2015).

Figure 7. Representative species of a A) solitary and a B) colonial ascidian.(1.5 fitting image)

Table 1. Characteristics of the adhesives produced by adult marine invertebrates and examples of marine biomimetic adhesives.

Phyllum or Order	Common name	Species	Composition or characterization of the adhesive	Function	Biomimetic adhesive development phase	Potential application/speciality
Mollusca	Scallop	*Chlamys farreri*	Cystein residues in Sbp8-1 protein, atypical metalloproteinase	Cross-linking	-	
	Oyster	*Crassostrea virginica*	Organic-inorganic composite Phosphorylated proteins		ACC/PAA Hydrogel organic-inorganic adhesive (Li et al., 2018)	Wet adhesive
Polychaeta	Tubeworm	*Phragmatopoma californica*	DOPA in Pc-1 and Pc-2 Serine residues phosphorylated in Pc-3A and Pc-3B	Interfacial adhesion Cross-linking	Polyacrylate adhesive (Shao et al., 2009)	Hard mineralized tissue adhesive
					HA-DN and lactose modified chitosan polymer hydrogel (Oh et al., 2012)	Wet adhesive
					Multiphase adhesive with synthetic copolyelectrolytes and PEG-dA (Kaur et al., 2011)	Wet adhesive Drug delivery
					HLAA adhesive (Lee et al., 2015)	Glue for ophthalmic application
					In situ endovascular embolic agent (Jones et al., 2016)	Embolic application
					Polyelectrolytes by solvent exchange (Zhao et al., 2016)	Wet adhesive
		Sabellaria alveolata	Tyrosinases and peroxidase		-	
Crustacea	Barnacle	*Megabalanus rosa* *Amphibalanus amphitrite*	Cp-100k and Cp-52k Cp-68k, Cp-20k and Cp-19k	Cross-linking Interfacial adhesion	-	
		Amphibalanus amphitrite	Glycine/ serine-rich cement proteins (GSrCPs) Leucine-rich cement proteins (LrCPs) Lysyl oxidases and peroxidases	-		
Echinodermata	Sea star	*Asterias rubens*	Sfp-1	Cohesion	-	
	Sea urchin	*Paracentrotus lividus*	Nectin Actin and histones	Interfacial adhesion	-	
	Sea cucumber	*Holothuria dofleinii*	C-lectin Enzyme-like proteins	-	-	
Tunicata	Sea squirt	*Molgula manhattensis*	PG moieties in 3,4,5-trihydroxyphenylalanine (TOPA)	-	CS-GA hydrogel (Sanandiya et al.	Wet adhesive

Ascidia ceratodes		2019)	Hemostasis
		HA–PG hydrogel (Cho et al., 2018)	Wet adhesive Drug delivery
		GA/metal ion complex (Prajatelistia et al. 2016)	Coating
		Chitin nanofiber gallic acid hydrogel (Oh et al., 2015)	Wet adhesive

ACC/PAA: calcium carbonate/polyacrylic acid; HA-DN: dopamine conjugated hyaluronic acid; HLAA: hydrophobic light-activating adhesive; CS: chitosan: GA: gallic acid.

3. Conclusion

The composition and properties of adhesive secretions appears to be diverse and remains unexplored. To what the marine environment is concerned, it is therefore important to increase knowledge of biological adhesives by widening the object of study to include other marine invertebrates, namely the less familiar/studied groups.

Sponges, for example, are sessile organisms that live attached to various substrates through a root- or basal plate-like structure. These holdfasts have adhesive properties that have been very limitedly studied (Ehrlich et al., 2013). Also, cnidarians such as sea anemones can move while adhering to a substrate and by having very good adhesive properties, are biofouling animals (Floerl et al., 2016), and therefore potentially good candidates to study the adhesive properties and their biomedical potential. For example, the mechanism of adhesion of smaller organisms, such as the flatworm *Macrostomum lignano*, primarily used as a model in developmental and evolutionary studies, was recently proposed (Wunderer et al., 2019).

Marine organisms from less known marine environments characterized by different physico-chemical conditions (e.g. more acidic water, warm or colder temperatures, different current regimes, natural hard substrate of differing chemistry) may also be good candidates to study their adhesion mechanisms to know putative differences in structural, chemical and mechanical features in relation to the marine invertebrates studied so far. In this line, the study of adhesives adhering to soft substrates, including biological tissues (e.g. barnacles adhering to whale skins) could also be relevant to improve biomimicry adhesives for soft tissues, as suggested by Messersmith's research group (Balkenende et al., 2019).

It is also important to focus on other aspects of attachment, such as how organisms control the chemical environment at the interface to promote the adhesion. For example, cyprid larvae from barnacles have a bi-phasic mechanism of adhesion, with lipids having a role in the adhesion process by creating a conducive environment for the curing and crosslinking of the proteins and also by having a protective role (Gohad et al., 2014). The study of the larvae is also significant as these may have differences in composition and properties when compared to the adults. For example, the gregarious settlement in barnacle larvae is induced by a contact pheromone, SIPC (settlement-inducing protein complex), that may also have a role in the temporary larvae adhesion (Petrone et al., 2014).

The role of other biopolymers that are commonly detected in adhesive systems should also be addressed. Carbohydrates are found in temporary adhesive glands and adhesive material, but their function in the adhesive process is not yet understood. Also, the identification of novel components such as enzymes and their role in the adhesive process should be addressed with the dual use of proteomic and transcriptomic approaches (Buffet et al., 2018; So et al., 2016). The knowledge arising from these studies will certainly suggest that biomimetic adhesives could benefit from more than only protein-like molecules, namely comprehending more complex mechanisms as synergy between different classes of compounds. Biomimetic adhesives have been developed mostly based on the knowledge of the adhesive mechanism from the mussels *Mytilus* spp. and the tubeworm *P. californica*. Research opportunities should not be limited to the improvement of DOPA-bases and coacervates-enable adhesives but should also better define or uniform properties of current adhesive systems. Most studies on adhesive proteins are carried out on the adhesive organ/gland or the secreted material, the later more difficult to achieve because of the insolubility of some adhesives (e.g. barnacles). The recent use of proteomic and transcriptomic approaches overcome these problems at different levels, from genes to secreted material, and allowed the identification of a large amount of information about molecular sequences of adhesive proteins secreted by several marine organisms (Hennebert et al., 2015).

However, despite these improvements in the molecular characterization of marine adhesives, the structure-adhesive function, cohesion and interactions of these proteins remains unknown and require further investigation as suggested by Cui et al. (2017).In search for adhesives with better performance, a deep understanding of the adhesive systems from other marine invertebrates are therefore important to investigate to further develop sustainable strategies to isolate these compounds (e.g. based on supercritical fluids technology, ionic liquids and deep eutectic solvents, as being recent classes of solvents might be able to solubilize some of the targeted systems) or synthetize analogues (e.g. recombinant production; production of synthetic peptides) that mimic adhesives performance in the marine environment.

Acknowledgments

The authors acknowledge the funding from the European Union Transborder Cooperation Programme Interreg España-Portugal 2014-2020 (POCTEP) under the projects 0245_IBEROS_1_E and 0302_CVMAR_I_1_P and from European Union under the COST Action 16203 MARISTEM.

References

Alberts, E.M., Taylor, S.D., Edwards, S.L., Sherman, D.M., Huang, C.P., Kenny, P., Wilker, J.J., 2015. Structural and compositional characterization of the adhesive produced by reef building oysters. ACS Appl. Mater. Interfaces 7, 8533–8538. https://doi.org/10.1021/acsami.5b00287

Alejandrino, A., Puslednik, L., Serb, J.M., 2011. Convergent and parallel evolution in life habit of the scallops (Bivalvia: Pectinidae). BMC Evol. Biol. 11, 164. https://doi.org/10.1186/1471-2148-11-164

Balkenende, D.W.R., Winkler, S.M., Messersmith, P.B., 2019. Marine-inspired polymers in medical adhesion. Eur. Polym. J. 116, 134–143. https://doi.org/10.1016/j.eurpolymj.2019.03.059

Barlow, D.E., Dickinson, G.H., Orihuela, B., Kulp, J.L., Rittschof, D., Wahl, K.J., 2010. Characterization of the adhesive plaque of the barnacle *Balanus amphitrite*: Amyloid-like nanofibrils are a major component. Langmuir 26, 6549–6556. https://doi.org/10.1021/la9041309

Brubaker, C.E., Kissler, H., Wang, L.-J., Kaufman, D.B., Messersmith, P., 2010. Biological performance of mussel-inspired adhesive in extrahepatic islet transplantation. Biomaterials 31, 420–427. https://doi.org/10.1016/J.BIOMATERIALS.2009.09.062

Brusca, R.C., Brusca, G.J., 2003. Invertebrates, 2nd ed. Sunderland.

Buffet, J.-P., Corre, E., Duvernois-Berthet, E., Fournier, J., Lopez, J.P., 2018. Adhesive gland transcriptomics uncovers a diversity of genes involved in glue formation in marine tube-building polychaetes. Acta Biomater. 72, 316–328. https://doi.org/10.1016/j.actbio.2018.03.037

Burkett, J.R., Hight, L.M., Kenny, P., Wilker, J.J., 2010. Oysters produce an organic-Inorganic adhesive for intertidal reef construction. J. Am. Chem. Soc. 132, 12531–12533. https://doi.org/10.1021/ja104996y

Carvalho, A.L., Vale, A.C., Sousa, M.P., Torrado, E., Mano, J.F., Alves, N.M., 2016. Antibacterial bioadhesive layer-by-layer coatings for orthopedic applications. J. Mater. Chem. B 4, 5385–5393. https://doi.org/10.1039/C6TB00841K

Cho, J.H., Lee, J.S., Shin, J., Jeon, E.J., An, S., Choi, Y.S., Cho, S.-W., 2018. Ascidian-Inspired Fast-Forming Hydrogel System for Versatile Biomedical Applications : Pyrogallol Chemistry for Dual Modes of Crosslinking Mechanism. Adv. Funct. Mater. 1705244. https://doi.org/10.1002/adfm.201705244

Davies, M.S., Hawkins, S.J., 1998. Mucus from Marine Molluscs 2881, 1–71. https://doi.org/10.1016/s0065-2881(08)60210-2

DeMoor, S., Herbert Waite, J., Jangoux, M., Flammang, P., 2003. Characterization of the adhesive from Cuvierian tubules of the sea cucumber *Holothuria forskali* (Echinodermata, Holothuroidea). Mar. Biotechnol. 5, 45–57. https://doi.org/10.1007/s10126-002-0049-2

Ehrlich, H., Behm, T., Bazhenov, V. V., Ehrlich, A., Kaluzhnaya, O. V., Chernogor, L.I., Belikov, S., Tsurkan, M. V., Ereskovsky, A., Tabachnick, K.R., Ilan, M., Stelling, A., Galli, R., Petrova, O. V., Nekipelov, S. V., Sivkov, V.N., Vyalikh, D., Born, R., Janussen, D., Wörheide, G., 2013. First report on chitinous holdfast in sponges (Porifera). Proc. R. Soc. B Biol. Sci. 280, 20130339. https://doi.org/10.1098/rspb.2013.0339

Endrizzi, B.J., Stewart, R.J., 2009. Glueomics : An Expression Survey of the Adhesive Gland of the Sandcastle Worm. J. Adhes. 85, 546–559. https://doi.org/10.1080/00218460902996457

Flammang, P., Demeuldre, M., Hennebert, E., Santos, R., 2016. Adhesive Secretions in Echinoderms: A Review, in: Smith, A.M. (Ed.), Biological Adhesives. Springer, Cham, pp. 193–222.

Flammang, P., Michel, A., Cauwenberge, A. Van, Alexandre, H., Jangoux, M., 1998. A study of the temporary adhesion of the podia in the sea star *Asterias rubens* (Echinodermata, Asteroidea) through their footprints. J. Exp. Biol. 201, 2383–2395.

Flammang, P., Santos, R., 2014. Biological adhesives: from biology to biomimetics. Interface Focus 5, 1–3. https://doi.org/10.1098/rsfs.2014.0086

Floerl, O., Sunde, L.M., Bloecher, N., 2016. Potential environmental risks associated with biofouling management in salmon aquaculture 8, 407–417. https://doi.org/10.3354/aei00187

Gohad, N. V., Aldred, N., Hartshorn, C.M., Lee, Y.J., Cicerone, M.T., Orihuela, B., Clare, A.S., Rittschof, D., Mount, A.S., 2014. Synergistic roles for lipids and proteins in the permanent adhesive of barnacle larvae. Nat. Commun. 5, 4414. https://doi.org/10.1038/ncomms5414

Gorb, S.N., 2008. Biological attachment devices: Exploring nature's diversity for biomimetics. Philos. Trans. R. Soc. A Math. Phys. Eng. Sci. 366, 1557–1574. https://doi.org/10.1098/rsta.2007.2172

Hennebert, E., Wattiez, R., Demeuldre, M., Ladurner, P., Hwang, D.S., Waite, J.H., Flammang, P., 2014. Sea star tenacity mediated by a protein that fragments, then aggregates. Proc. Natl. Acad. Sci. 111, 6317–6322. https://doi.org/10.1073/pnas.1400089111

Hennebert E, Maldonado B, Ladurner P, Flammang P, Santos R. 2015. Experimental strategies for the identification and characterization of adhesive proteins in animals: a review. Interface Focus 5: 20140064. http://dx.doi.org/10.1098/rsfs.2014.0064

Holm, E.R., 2012. Barnacles and biofouling. Integr. Comp. Biol. 52, 348–55. https://doi.org/10.1093/icb/ics04

Jones, J.P., Sima, M., O´Hara, R.G., Stewart, R.J., 2016. Water-borne Endovascular Liquid Embolics Inspired By the Undersea Adhesive of Marine Sandcastle Worms. Adv. Healthc. Mater. 5, 795–801. https://doi.org/10.1002/adhm.201500825.Water-borne

Jonker, J.-L., Abram, F., Pires, E., Coelho, A.V., Grunwald, I., Power, A.M., 2014. Adhesive proteins of stalked and acorn barnacles display homology with low sequence similarities. PLoS One 9, e108902. https://doi.org/10.1371/journal.pone.0108902

Kamino, K., 2013. Mini-review: Barnacle adhesives and adhesion. Biofouling 29, 735–749. https://doi.org/10.1080/08927014.2013.800863

Kamino, K., 2010. Molecular design of barnacle cement in comparison with those of mussel and tubeworm. J. Adhes. 86, 96–110. https://doi.org/10.1080/00218460903418139

Kamino, K., Inoue, K., Maruyama, T., Takamatsu, N., Harayama, S., Shizuri, Y., 2000. Barnacle cement proteins: Importance of disulfide bonds in their insolubility. J. Biol. Chem. 275, 27360–27365. https://doi.org/10.1074/jbc.M910363199

Kaur, S., Weerasekare, G.M., Stewart, R.J., 2011. Multiphase Adhesive Coacervates Inspired by the Sandcastle Worm. ACS Appl. Mater. Interfaces 3, 941–944. https://doi.org/10.1021/am200082v

Kim, B.J., Cheong, H., Hwang, B.H., Cha, H.J., 2015. Mussel-Inspired Protein Nanoparticles Containing Iron(III)-DOPA Complexes for pH-Responsive Drug Delivery. Angew. Chemie - Int. Ed. 54, 7318–7322. https://doi.org/10.1002/anie.201501748

Kim, B.J., Oh, D.X., Kim, S., Seo, J.H., Hwang, D.S., Masic, A., Han, D.K., Cha, H.J., 2014. Mussel-mimetic protein-based adhesive hydrogel. Biomacromolecules 15, 1579–1585. https://doi.org/10.1021/bm4017308

Kronenberger, K., Dicko, C., Vollrath, F., 2012. A novel marine silk. Naturwissenschaften 99, 3–10. https://doi.org/10.1007/s00114-011-0853-5

Leal, M.C., Rocha, R.J.M., Rosa, R., Calado, R., 2018. Aquaculture of marine non-food organisms: what, why and how? Rev. Aquac. 10, 400–423. https://doi.org/10.1111/raq.12168

Lebesgue, N., da Costa, G., Ribeiro, R.M., Ribeiro-Silva, C., Martins, G.G., Matranga, V., Scholten, A., Cordeiro, C., Heck, A.J.R., Santos, R., 2016. Deciphering the molecular mechanisms underlying sea urchin reversible adhesion: A quantitative proteomics approach. J. Proteomics 138, 61–71. https://doi.org/10.1016/j.jprot.2016.02.026

Lee, Y., Xu, Chenjie, Sebastin, M., Lee, A., Holwell, N., Xu, Calvin, Miranda-Nieves, D., 2015. Bio-inspired Nanoparticulate Medical Glues for Minimally Invasive Tissue Repair. Adv. Healthc. Mater. 4, 2587–2596. https://doi.org/10.1002/adhm.201500419.Bio-inspired

Lengerer, B., Bonneel, M., Lefevre, M., Hennebert, E., Leclère, P., Gosselin, E., Ladurner, P., Flammang, P., 2018. The structural and chemical basis of temporary adhesion in the sea star *Asterina gibbosa*. Beilstein J. Nanotechnol. 9, 2071–2086. https://doi.org/10.3762/bjnano.9.196

Lengerer, B., Pfaller, K., Berezikov, E., Ladurner, P., Schärer, L., Salvenmoser, W., Rodrigues, M., Hess, M.W., Wunderer, J., Egger, B., Obwegeser, S., Pjeta, R., Arbore, R., 2014. Biological adhesion of the flatworm *Macrostomum lignano* relies on a duo-gland system and is mediated by a cell type-specific intermediate filament protein. Front. Zool. 11, 12. https://doi.org/10.1186/1742-9994-11-12

Li, A., Jia, Y., Sun, S., Xu, Y., Minsky, B.B., Stuart, M.A.C., Cölfen, H., Von Klitzing, R., Guo, X., 2018. Mineral-Enhanced Polyacrylic Acid Hydrogel as an Oyster-Inspired Organic-Inorganic Hybrid Adhesive. ACS Appl. Mater. Interfaces 10, 10471–10479. https://doi.org/10.1021/acsami.8b01082

Liu, X., Liang, C., Zhang, X., Li, J., Huang, J., Zeng, L., Ye, Z., Hu, B., Wu, W., 2017. Amyloid fibril aggregation: An insight into the underwater adhesion of barnacle cement. Biochem. Biophys. Res. Commun. 493, 654–659. https://doi.org/10.1016/j.bbrc.2017.08.136

López, D.A., López, B.A., Pham, C.K., Isidro, E.J., De Girolamo, M., 2010. Barnacle culture: Background, potential and challenges. Aquac. Res. 41, 367–375. https://doi.org/10.1111/j.1365-2109.2010.02508.x

Merz, R.A., 2015. Textures and traction: how tube-dwelling polychaetes get a leg up. Invertebr. Biol. 134, 61–77. https://doi.org/10.1111/ivb.12079

Metzler, R.A., Rist, R., Alberts, E., Kenny, P., Wilker, J.J., 2016. Composition and Structure of Oyster Adhesive Reveals Heterogeneous Materials Properties in a Biological Composite. Adv. Funct. Mater. 26, 6814–6821. https://doi.org/10.1002/adfm.201602348

Miao, Y., Zhang, L., Sun, Y., Jiao, W., Li, Y., Sun, J., Wang, Y., Wang, S., Bao, Z., Liu, W., 2015. Integration of Transcriptomic and Proteomic Approaches Provides a Core Set of Genes for Understanding of Scallop Attachment. Mar. Biotechnol. 17, 523–532. https://doi.org/10.1007/s10126-015-9635-y

Mostaert, A.S., Giordani, C., Crockett, R., Karsten, U., Schumann, R., Jarvis, S., 2009. Characterisation of Amyloid Nanostructures in the Natural Adhesive of Unicellular Subaerial Algae. J. Adhes. 85, 465–483. https://doi.org/10.1080/00218460902996366

Nakano, M., Kamino, K., 2015. Amyloid-like conformation and interaction for the self-assembly in barnacle underwater cement. Biochemistry 54, 826–835. https://doi.org/10.1021/bi500965f

Oh, D.X., Kim, S., Lee, D., Hwang, D.S., 2015. Tunicate-mimetic nanofibrous hydrogel adhesive with improved wet adhesion. Acta Biomater. 20, 104–112. https://doi.org/10.1016/j.actbio.2015.03.031

Oh, Y.J., Cho, H., Lee, Haeshin, Park, K.-J., Lee, Hyukjin, Park, S.Y., 2012. Bio-inspired catechol chemistry: a new way to develop a re-moldable and injectable coacervate hydrogel. Chem. Commun. 48, 11895–11897. https://doi.org/10.1039/c2cc36843a

Pawlicki, J.M., 2004. The effect of molluscan glue proteins on gel mechanics. J. Exp. Biol. 207, 1127–1135. https://doi.org/10.1242/jeb.00859

Peng, Y.Y., Glattauer, V., Skewes, T.D., McDevitt, A., Elvin, C.M., Werkmeister, J.A., Graham, L.D., Ramshaw, J.A.M., 2014. Identification of Proteins Associated with Adhesive Prints from *Holothuria dofleinii* Cuvierian Tubules. Mar. Biotechnol. 16, 695–706. https://doi.org/10.1007/s10126-014-9586-8

Pennati, R., Rothbacher, U., 2015. Bioadhesion in ascidians: a developmental and functional genomics perspective. Interface Focus 5, 20140061. https://doi.org/10.1098/rsfs.2014.0061

Perrini, M., Barrett, D., Ochsenbein-Koelble, N., Zimmermann, R., Messersmith, P., Ehrbar, M., 2016. A comparative investigation of mussel-mimetic sealants for fetal membrane repair. J. Mech. Behav. Biomed. Mater. 58, 57–64. https://doi.org/10.1016/j.jmbbm.2015.07.009

Petrone, L., Aldred, N., Emami, K., Enander, K., Ederth, T., Clare, A.S., 2014. Chemistry-specific surface adsorption of the barnacle settlement-inducing protein complex. Interface Focus 5, 1–11. https://doi.org/10.1098/rsfs.2014.0047

Prajatelistia, E., Ju, S.W., Sanandiya, N.D., Jun, S.H., Ahn, J.S., Hwang, D.S., 2016. Tunicate-Inspired Gallic Acid/Metal Ion Complex for Instant and Efficient Treatment of Dentin Hypersensitivity. Adv. Healthc. Mater. 5, 919–927. https://doi.org/10.1002/adhm.201500878

Rocha, M., Antas, P., Castro, L.F.C., Campos, A., Vasconcelos, V., Pereira, F., Cunha, I., 2019. Comparative Analysis of the Adhesive Proteins of the Adult Stalked Goose Barnacle *Pollicipes pollicipes* (Cirripedia: Pedunculata). Mar. Biotechnol. 21, 38–51. https://doi.org/10.1007/s10126-018-9856-y

Sanandiya, N.D., Lee, S., Rho, S., Lee, H., Kim, I.S., Hwang, D.S., 2019. Tunichrome-inspired pyrogallol functionalized chitosan for tissue adhesion and hemostasis. Carbohydr. Polym. 208, 77–85. https://doi.org/10.1016/j.carbpol.2018.12.017

Santos, R., Barreto, Â., Franco, C., Coelho, A.V., 2013. Mapping sea urchins tube feet proteome - A unique hydraulic mechano-sensory adhesive organ. J. Proteomics 79, 100–113. https://doi.org/10.1016/j.jprot.2012.12.004

Santos, R., da Costa, G., Franco, C., Gomes-Alves, P., Flammang, P., Coelho, A. V., 2009. First insights into the biochemistry of tube foot adhesive from the sea urchin *Paracentrotus lividus* (Echinoidea, Echinodermata). Mar. Biotechnol. 11, 686–698. https://doi.org/10.1007/s10126-009-9182-5

Shao, H., Bachus, K.N., Stewart, R.J., 2009. A Water-Borne Adhesive Modeled after the Sandcastle Glue of *P. californica*. Macromol. Biosci. 9, 464–471. https://doi.org/10.1002/mabi.200800252

Shillaker, R.O., Moore, P.G., 1978. Tube building by the amphipods *Lembos websteri* Bate and *Corophium bonnelli* Milne Edwards. J. Exp. Mar. Bio. Ecol. 33, 169–185.

Smith, A.M., 2016. The Biochemistry and Mechanics of Gastropod Adhesive Gels, in: Smith, A.M. (Ed.), Biological Adhesives. Springer International Publishing, Cham, pp. 177–192.

Smith, A.M., 1992. Alternation between attachment mechanisms by limpets in the field. J. Exp. Mar. Bio. Ecol. 1600, 205–220. https://doi.org/10.1016/0022-0981(92)90238-6

Smith, A.M., Quick, T.J., Peter, R.L. ST., 1999. Differences in the Composition of Adhesive and Non-Adhesive Mucus from the Limpet Lottia limatula. Biol. Bull. 196, 34–44.

So, C.R., Fears, K.P., Leary, D.H., Scancella, J.M., Wang, Z., Liu, J.L., Orihuela, B., Rittschof, D., Spillmann, C.M., Wahl, K.J., 2016. Sequence basis of Barnacle Cement Nanostructure is Defined by Proteins with Silk Homology. Sci. Rep. 6, 36219. https://doi.org/10.1038/srep36219

So, C.R., Yates, E., Estrella, L., Schenck, A., Yip, C., Wahl, K.J., 2018. Wet Adhesive Nanomaterials Inspired by the Barnacle Adhesive. Biophys. J. 114, 192a-193a. https://doi.org/10.1016/j.bpj.2017.11.1075

Stewart, R.J., Wang, C.S., Shao, H., 2011. Complex coacervates as a foundation for synthetic underwater adhesives. Adv. Colloid Interface Sci. 167, 85–93. https://doi.org/10.1016/j.cis.2010.10.009

Stewart, R.J., Weaver, J.C., Morse, D.E., Waite, J.H., 2004. The tube cement of *Phragmatopoma californica*: a solid foam. J. Exp. Biol. 207, 4727–4734. https://doi.org/10.1242/jeb.01330

Sun, C.J., Srivastava, A., Reifert, J.R., Waite, J.H., 2010. Halogenated DOPA in a Marine Adhesive Protein. J. Adhes. 85(2–3), 126. https://doi.org/10.1080/00218460902782188.Halogenated

Tanur, A.E., Gunari, N., Sullan, R.M.A., Kavanagh, C.J., Walker, G.C., 2010. Insights into the composition, morphology, and formation of the calcareous shell of the serpulid *Hydroides dianthus*. J. Struct. Biol. 169, 145–160. https://doi.org/10.1016/j.jsb.2009.09.008

Taylor, S.W., Kammerer, B., Bayer, E., 1997. New Perspectives in the Chemistry and Biochemistry of the Tunichromes and Related Compounds. Chem. Rev. 333–346. https://doi.org/10.1021/cr940467q

Urushida, Y., Nakano, M., Matsuda, S., Inoue, N., Kanai, S., Kitamura, N., Nishino, T., Kamino, K., 2007. Identification and functional characterization of a novel barnacle cement protein. FEBS J. 274, 4336–4346. https://doi.org/10.1111/j.1742-4658.2007.05965.x

von Byern, J., Klepal, W., 2006. Adhesive mechanisms in cephalopods: a review. Biofouling 22, 329–338. https://doi.org/10.1080/08927010600967840

Waite, J.H., 2017. Mussel adhesion – essential footwork. J. Exp. Biol. 220, 517–530. https://doi.org/10.1242/jeb.134056

Waite, J.H., Jensen, R.A., Morse, D.E., 1992. Cement Precursor Proteins of the Reef-Building Polychaete *Phragmatopoma californica* (Fewkes). Biochemistry 31, 5733–5738. https://doi.org/10.1021/bi00140a007

Walker, G., 1992. Cirripedia, in: Harrison, F.W., Humes, A.G. (Eds.), Microscopic Anatomy of Invertebrates, Crustacea (Volume 9). Wiley-Liss, NewYork, pp. 249–311.

Wang, C.S., Stewart, R.J., 2013. Multipart Copolyelectrolyte Adhesive of the Sandcastle Worm, *Phragmatopoma californica* (Fewkes): Catechol Oxidase Catalyzed Curing through Peptidyl-DOPA. Biomacromolecules 14(5), 1607–17. https://doi.org/doi: 10.1021/bm400251k

Wang, X., Schröder, H.C., Wiens, M., Ushijima, H., Müller, W.E.G., 2012. Bio-silica and bio-polyphosphate: Applications in biomedicine (bone formation). Curr. Opin. Biotechnol. 23, 570–578. https://doi.org/10.1016/j.copbio.2012.01.018

White, J.D., Wilker, J.J., 2011. Underwater bonding with charged polymer mimics of Marine mussel adhesive proteins. Macromolecules 44, 5085–5088. https://doi.org/10.1021/ma201044x

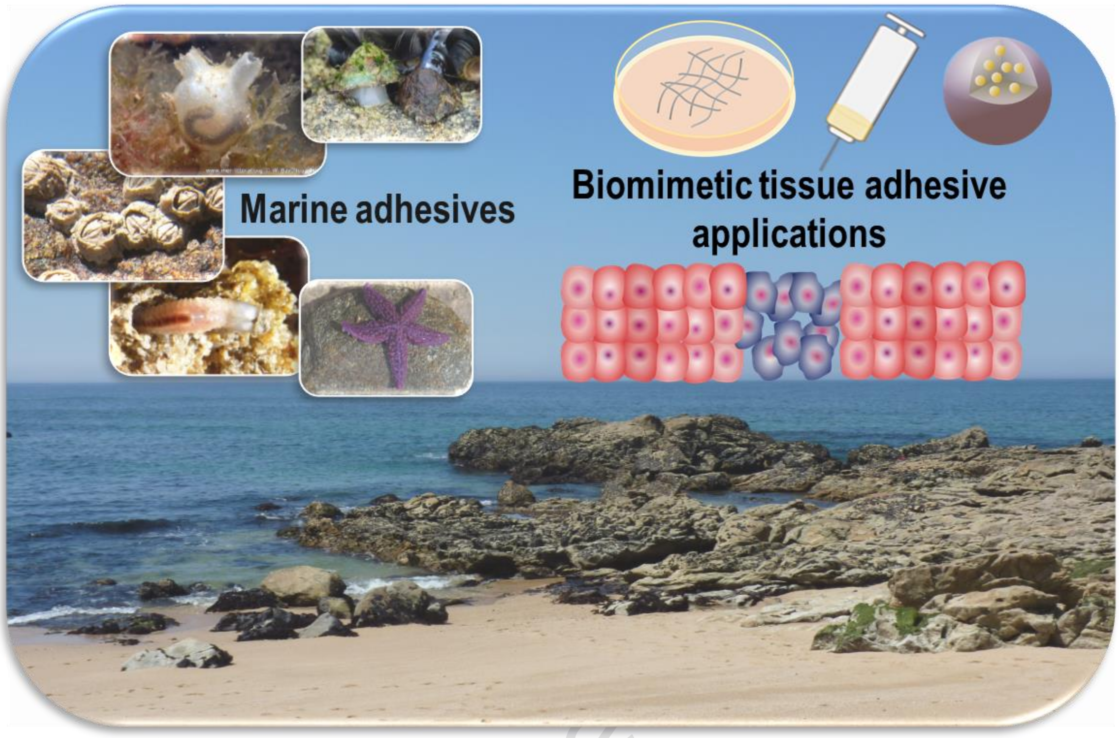
Wunderer, J., Lengerer, B., Pjeta, R., Bertemes, P., Kremser, L., Lindner, H., Ederth, T., Hess, M.W., Stock, D., Salvenmoser, W., Ladurner, P., 2019. A mechanism for temporary bioadhesion. Proc. Natl. Acad. Sci. 116, 4297–4306. https://doi.org/10.1073/pnas.1814230116

Zhang, X., Dai, X., Wang, L., Miao, Y., Xu, P., Liang, P., Dong, B., Bao, Z., Wang, S., Lyu, Q., Liu, W., 2018. Characterization of an atypical metalloproteinase inhibitors like protein (Sbp8-1) from scallop byssus. Front. Physiol. 9, 1–9. https://doi.org/10.3389/fphys.2018.00597

Zhao, H., Sun, C., Stewart, R.J., Waite, J.H., 2005. Cement Proteins of the Tube-building Polychaete *Phragmatopoma californica*. J. Biol. Chem. 280, 42938–42944. https://doi.org/10.1074/jbc.M508457200

Zhao, Q., Lee, D.W., Ahn, B.K., Seo, S., Kaufman, Y., Israelachvili, J.N., Waite, J.H., 2016. Underwater contact adhesion and microarchitecture in polyelectrolyte complexes actuated by solvent exchange. Nat. Mater. 15, 407–412. https://doi.org/10.1038/nmat4539.Underwater

Graphical Abstract



Highlights (Each highlight must be 85 characters or fewer, including spaces; 3 to 5)

- Some marine invertebrates produce protein-based bioadhesives that not contain DOPA.
- The tubes of the sandcastle worm inspired the development of adhesive coacervates.
- No DOPA is found in barnacle cement and TOPA is a feature of tunicate adhesive.
- Temporary adhesives can inspire the development of biomimetic reversible adhesives.
- The study of diverse marine animals can benefit the development of wet adhesives.