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Tethering of soluble immune effectors to mucin and chitin reflects a convergent and dynamic role in gut immunity

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The immune system employs soluble effectors to shape luminal spaces. Antibodies are soluble molecules that effect immunological responses, including neutralization, opsonization, antibody-dependent cytotoxicity and complement activation. These molecules are comprised of immunoglobulin (Ig) domains. The N-terminal Ig domains recognize antigen, and the C-terminal domains facilitate their elimination through phagocytosis (opsonization). A less-recognized function mediated by the C-terminal Ig domains of the IgG class of antibodies (Fc region) involves the formation of multiple low-affinity bonds with the mucus matrix. This association anchors the antibody molecule to the matrix to entrap potential pathogens. Even though invertebrates are not known to have antibodies, protochordates have a class of secreted molecules containing Ig domains that can bind bacteria and potentially serve a similar purpose. The VCBPs (V region-containing chitin-binding proteins) possess a C-terminal chitin-binding domain that helps tether them to chitin-rich mucus gels, mimicking the IgG-mediated Fc trapping of microbes in mucus. The broad functional similarity of these structurally divergent, Ig-containing, secreted effectors makes a case for a unique form of convergent evolution within chordates. This opinion essay highlights emerging evidence that divergent secreted immune effectors with Ig-like domains evolved to manage immune recognition at mucosal surfaces in strikingly similar ways.

This article is part of the theme issue 'Sculpting the microbiome: how host factors determine and respond to microbial colonization'.

1. Introduction

The vertebrate immune system protects against pathogens using somatically derived, highly diverse T-cell receptors (TCRs) on the surface of cells or antibodies, which are first displayed as cell-surface receptors, and once selected, are secreted. These two receptor classes provide effective immune protection in blood and secretions and include evolved adaptations that mediate important interactions with commensal communities. Recent studies have shown that both IgG, the most abundant class of antibody in mammals, along with secretory IgA, can combine with mucin to trap and immobilize pathogens and/or facilitate the retention and colonization of commensal microbes [1–13]. These antibody-mediated actions can serve as a primary defence mechanism at the mucosal surfaces, e.g. immune exclusion, and significantly shape the composition and ecology of commensal communities that colonize the mucus gels lining epithelial surfaces [14–16]. This mucus-tethered interaction is reminiscent of what has been shown to occur in the gut of invertebrate chordates, in which V region-containing chitin-binding proteins (VCBPs) are bound to chitin-rich mucus of the gut, by way of their C-terminal chitin-binding domain (CBD) [17–20]. In aquatic organisms, chitin is thought to

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stabilize mucus, i.e. it reduces the solubility of epithelial gel mucins. Thus, the tethering of Ig domain effectors to the mucus matrix represents a fundamental immune effector mechanism that shapes or 'sculps' the ecology of host–microbe interactions, both pathogenic and commensal, in mucosal environments. This process arose at least twice in chordate evolution, suggesting that strong selective pressures have resulted in a novel functional convergence.

2. The mucosal surface paradox

Throughout metazoan evolution, animals have developed various defence mechanisms to protect themselves from a growing number of diverse pathogens [21]. One of the most significant barriers to infection is the epithelium, which lines animal tissues both on the outside, such as the skin, and on the inside, like the gastrointestinal tract or lungs [22,23]. Mucosal environments, which are layered with mucin gels that are shed on a continuous basis, are particularly effective at assembling physical barriers that protect the internal confines of animals [24]. Trillions of microbes generally colonize animals, many on mucosal surfaces where they are attracted to and graze on the carbon-rich mucin gels [25,26]. The 'restaurant hypothesis' posits that nutrient-mediated selection helps drive the initial colonization events of the gut, mostly by commensal organisms, making access more difficult for potential pathogens [27,28]. This process, especially early in the developmental maturation of the gut, serves principal roles in shaping downstream immune responses [29–31]. Most of the microbes colonizing mucosal surface niches [32] are harmless or beneficial in mediating protection from other microbes or supporting animal health via metabolic exchanges with their host.

A paradox presents for the local immune system, which must ignore most colonizing microbes while remaining vigilant toward potential pathogens. This challenge has existed since the origin of metazoans [14]. Diverse strategies that include the secretion of various immune effector molecules into the mucosal surface gels have evolved to defend animals while shaping and maintaining the outcome of symbiotic interactions with microbes that can improve fitness. Thus, it has been proposed that the evolution of the adaptive immune system is intimately associated with a role in managing complex communities of beneficial microbes [33]. In addition to cellular responses that mediate adaptive immunity, it is likely that diverse molecules evolved to negotiate the crosstalk between host and beneficial microbes to promote host survival.

Two effector approaches evolved to protect the mucosal surface. Membrane-bound effectors, like the TCR or the Toll-like receptors, bind to antigens and transduce signals that can trigger a range of effector responses; whereas soluble effectors can bind and label their targets for phagocytosis, i.e. opsonization. Soluble effectors also can bind to and neutralize their targets, e.g. immune exclusion, and are sometimes directly cytotoxic, e.g. defensins or other antimicrobial peptides (AMPs). Soluble effectors that are secreted at the epithelial surface are most helpful in mediating a variety of defensive mechanisms that include preventing potential pathogens from penetrating mucin gels and accessing the host epithelium. By binding diverse microbial targets in the lumen spaces or within the mucin-rich gels, soluble effectors can shape the ecology of the gut, protect the host from pathogen overgrowth, adherence, penetration or even influence the outcome of polymicrobial interactions and enhance settlement or retention of commensal populations. The production of microbial proteases and glycosylases is necessary for breaking down or using mucin gels, but their presence, along with free glycans, also can signal potential danger that is monitored closely by the underlying epithelium [34].

3. Mucin-rich gels are an ancient defence strategy and help define mucosal environments

It is widely accepted that the mucus gels layering epithelial surfaces function as a protective barrier, acting as the first line of defence of the innate immune system against harmful agents and pathogens [35–37]. However, the mucus layer is much more than a simple physical barrier. Because of the extensive glycosylation of mucin glycoproteins, it serves a primary selective role and factors in homeostasis with the microbiota by providing a rich source of nutrients that are targeted by specific microorganisms [24,38,39]. Because glycosylation patterns vary across mucosal sites, microbial populations become spatially dispersed, resulting in the formation of a three-dimensional scaffold for colonization and providing a source of signals that can directly impact microbial gene expression and behaviour [39].

Mucus composition in mammals is well characterized and has been described as a complex, viscoelastic and adherent secretion. Mucin is synthesized and secreted by specialized goblet or mucous-producing cells in organs and glands that interface and communicate with the external environment. Mucus contains water, gel-forming mucin glycoproteins, lipids, DNA, inorganic salts and different proteins, including AMPs and other secreted immune factors [40]. The composition and physicochemical properties of mucus vary depending on their anatomical location, such as eyes, respiratory tract, gastrointestinal tract, cervix and vagina. Within these tissues, mucus performs distinct physiological functions [41,42]. The biology of mucus is dynamic, as the processes of degradation and biosynthesis are integrated to maintain continuous mucosal protection against external stressors [43].

Mucins are classified as both membrane-associated and secreted molecules and form the glycocalyx at the apical surface of the epithelium and the overlying gel layer, respectively [40,41,44]. Mucin glycoproteins are encoded by genes referred to as 'MUC' in mammals. Twenty-two MUC genes have been identified in humans, and their biosynthetic, biochemical and anatomical properties have been described [40,41]. Both threonine and serine residues of mucins are heavily glycosylated via *N*-acetylgalactosamine (GalNAc) O-linkages, making the domains resistant to proteolysis and often resulting in a rigid matrix that can mimic a 'dense forest' [36,40,41,43]. The domains often have repeated sequences and vary in length depending on the allele, species and individual mucins [45].

Phylogenetic probing of sequenced genomes and transcriptomes reveals that secreted or gel-forming mucins were present in the early metazoans and are well developed in Ctenophores and Cnidarians. Diversification of membrane-anchored mucins appeared later in the chordates [45,46]. Glycosylation patterns among some mucins can include the polymerization of *N*-acetylglucosamine (GlcNAc) to form

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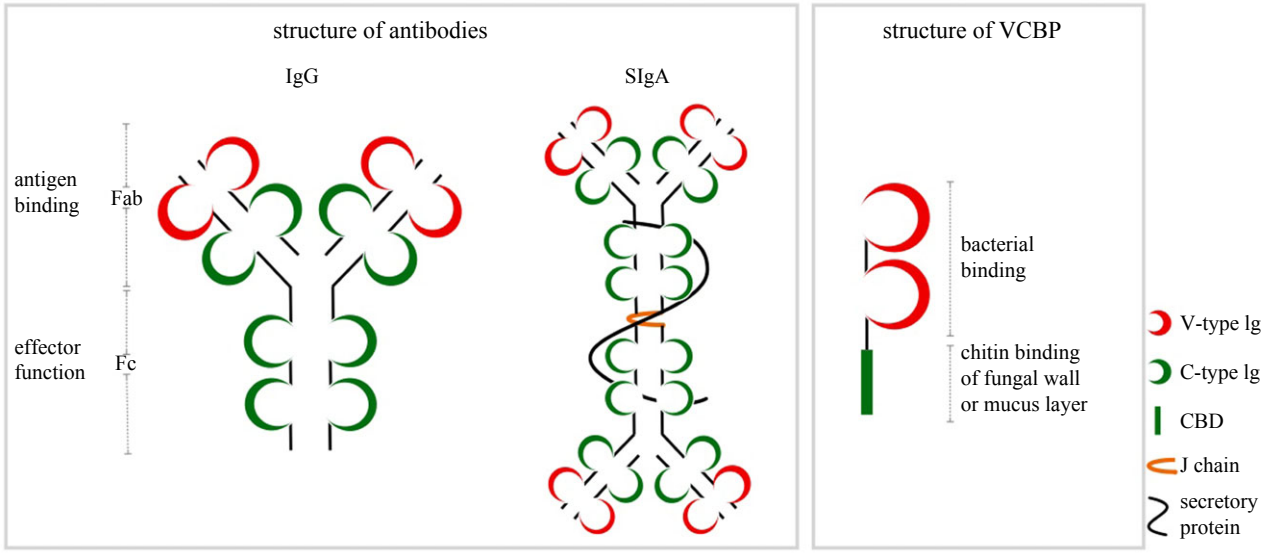

Figure 1. Schematic of immunoglobulin (Ig) domain structure in antibodies and the Variable Region Containing Chitin-Binding Protein (VCBP). V-type Ig domains, involved in antigen or microbial binding, are present in vertebrate antibodies, including IgG and secretory IgA, as well as in protochordate VCBP-C. Secondary effector functions of these molecules are mediated by the C (constant)-type Ig domain in antibodies and the chitin-binding domain in VCBP-C. In IgG and VCBPs, interactions with the Fc and CBD domains are the basis for immune effector functions at mucosal surfaces.

interwoven chitin fibres [47,48]. The peritrophic matrix (PM) is the glycocalyx barrier found in the midgut of several arthropods and consists of chitin fibres attached to peritrophins [49–51], a specialized form of glycoproteins. The peritrophins possess CBDs that facilitate the anchoring of chitin fibres and may also contain MUC domains [51]. Peritrophins may have originated in MUC genes by the acquisition of CBDs; e.g. insects also make purely MUC-based mucus gels. The PM may complement mucus gels in some compartments in order to provide additional protection against physical damage, microbial infection and toxins [52] while enhancing digestion by compartmentalizing digestive enzymes in different areas of the midgut [51,53].

Although chitin fibres were initially observed in arthropods, recent studies have revealed their presence in invertebrate chordates like the ascidian *Ciona robusta* [18], as well as in non-mammalian vertebrates such as *Danio rerio* and *Salmo salar* fishes and *Ambystoma mexicanum* amphibians [54,55]. This discovery suggests that chitin fibres within mucus barriers are more widespread in different species than previously believed. Chitin-rich mucus gels play a significant role in decreasing the solubility of mucins in water, thus enhancing the barriers to infection by adding strength and stability to the mucus [56–58]. *Ciona* produces chitin-rich mucus in its pharynx and throughout the gut [18,55]. Supplementing this mucus with exogenous chitin microparticles enhances the mucus barriers and increases protection against chemical-induced colitis, such as through ingestion of dextran sulfate sodium [19]. Chitin is abundantly found in nature because it not only adds structural rigidity but also helps to stabilize mucin gels in aquatic environments. Chitin in the midgut of insects may therefore be a vestige of an aquatic existence among arthropods [54].

It is tempting to speculate that the loss of chitin-rich mucus barriers in some vertebrates, like mammals, increased susceptibilities and/or reduced barrier protection against some pathogens. The requirement to enhance barriers may have resulted in increased selective pressures to include additional secreted immune effectors in mucus gels and drive the expansion of membrane-anchored mucin gene families. Subsequently, the intestinal epithelium of most animals produces a membrane proximal, tightly interwoven glycocalyx that remains relatively sterile while the outer layers of mucus are loose and microbe-rich [37]. Colonization of the outer layers by non-pathogens helps to enhance barriers against settlement and invasion by potential pathogens. Whereas intestinal mucus is rich in secreted immune effectors, it is likely that the specific types found in the inner versus outer layers are distinct. In this way, the effector molecules can influence colonization dynamics by encouraging the settlement of non-pathogens on the outer portions of the mucus barrier [10,11,18,59].

4. The Ig domain is a common feature of immune effectors

Ig domain-containing proteins are a superfamily of proteins with diverse functions in metazoans [21,60–62]. Because of their ability to undergo somatic diversification in antibodies and TCRs, these receptors recognize an extraordinarily large number of antigenic determinants. The ability of Ig domains to form homo- and heterodimers is a shared feature of antibodies that mediate defence capabilities, as well as many adhesion molecules that help stabilize many cell-to-cell interactions [60]. Variable, or V-type, domains are derived somatically in vertebrates and exhibit highly complex polymorphisms that define the recognition potential of the antigen-binding region. C-type, or constant, Ig domains are minimally polymorphic, provide a range of additional effector functions for antibodies, and are a common feature of adhesion molecules.

Vertebrate antibodies typically consist of two chains, as shown in figure 1, with V-type and C-type Ig domains. Soluble antibodies perform various effector functions, including antigen neutralization, agglutination and opsonization. The Fab fragment comprised the entire light chain and a portion of the heavy chain, consists of both V-type and C-type Ig domains acting like flexible arms, and is responsible for recognizing antigens. The Fc region is located at the C-terminal portion of

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the heavy chain and is made up of C-type Ig domains. When antibodies act as B cell receptors, they are bound to the cell membrane through an association involving the C-terminal transmembrane portion of the Fc region. Differential RNA splicing [63,64] replaces the transmembrane portion to create a secreted form. The Fc region of an antibody molecule can be changed through class switch recombination [65]. The secreted antibody molecule is recognized by Fc receptors and affords specificity to mononuclear phagocytes that, in turn, mediate essential effector functions such as opsonization, cytokine secretion and complement activation.

Several isotypes of vertebrate antibodies produce distinct and overlapping effector functions. For example, IgG is the most abundant form of antibody in circulation but also is present in some mucosal secretions, e.g. it is the most abundant isotype in cervicovaginal secretions [3]. Copious amounts of secretory IgA are part of normal mucosal secretions and provide diverse effector functions that include defence against pathogens. Antibodies of vertebrates are synthesized in secondary lymphoid tissues of laminar spaces; distribution into the lumen requires active transport. Polymeric Ig receptors (pIg) are expressed on the basolateral surface of the gut epithelium where they bind polymeric antibodies, such as IgA or IgM [66], the former being the most abundant antibody of mucosal secretions. The bound antibody is then transcytosed onto the apical surface where the ectodomain of the pIg receptor is cleaved and remains attached as a secretory component to help stabilize the effector molecules in the physiologically challenging environment of the gut lumen [67]. The resulting macromolecular complex helps to trap and physically clump microbes; unbound secretory component is also able to directly bind bacteria [68–72]. The secretory component of mucosal antibodies helps bridge innate and adaptive responses and serves important roles in commensal recognition responses, e.g. actively transporting some antigen back across microfold or M-cells into Peyer's patches [68,73,74]. Recent efforts have demonstrated an active role of secretory antibodies, primarily SIgA, in commensal attachment and colonization of mucosal environments [8–12].

Antibody effector function can be modified via glycosylation [75–77], e.g. distinct glycosylation patterns of Fc regions of IgG [4,15,78–81] have been shown to influence low-affinity interactions with mucins that help to immobilize microorganisms and limit their movement through mucin gels [1,2,4,13]. This newly recognized function has transformed our understanding of the functional repertoire of antibodies as essential mediators of diverse symbiotic interactions at mucosal surfaces [12,59].

5. The surprising and not-so-surprising antiquity of Ig-like domains

Ig-like domains have been identified in all kingdoms [82,83], suggesting a remarkable selective advantage that likely relates to their exceptional structural adaptability and intrinsic capacity to form homo- and heterodimers. In some microbes, Ig-like domain-containing proteins facilitate associations with components of animal membranes. Certain animal viruses and bacteriophages, viruses that infect bacteria, have been shown to interact with mucins through capsid-associated proteins that possess Ig-like domains [84]. This interaction facilitates viral access to epithelial surfaces and, in the case of phages, can provide increased access to diverse bacterial flora colonizing mucosal surfaces. Interestingly, a variety of distinct viruses may have independently acquired some of these domains from their hosts [83,85]. Some bacteria also have evolved diverse mucin-utilization programmes and can interact with host surfaces via Ig-like domains. Certain bacterial adhesins, such as IgI3, possess Ig-like domains and can specifically associate with Ig-rich CEACAM proteins on host epithelial cells [86]. The conjugation machinery in some bacteria possesses Ig-like domains that facilitate interactions and associations with other bacteria via recognition of their flagella and/or pili proteins [87]. Thus, diverse molecular structures that include Ig-like domains have evolved or have been recruited to facilitate interactions via homo- and heterodimer associations. Some of these associations help microbes access host epithelia or assist in the utilization of mucus. It is tempting to speculate that animal immunity via Ig-like domain recognition of microbial surface structures is itself a borrowed and ancient molecular feature of microbial recognition of mucosal surface structures.

In a directed search for related Ig-like domain proteins [88] that have V-type Ig domains, a small family of VCBPs was discovered in protochordates, first in amphioxus *Branchiostoma floridae* (namely, VCBP-1 to −5) [89,90] and then in the ascidian or tunicate *Ciona intestinalis* type A, or *C. robusta* (namely, VCBP-A to -D), where the function of these molecules has been investigated most thoroughly [17,18,90,91]. However, further studies using genomic resources from diverse organisms will be important to clarify if related gene families exist across diverse ascidian species, and protochordates in general.

The VCBPs possess two V-type Ig domains at the *N*-terminus, which mediate recognition of bacteria [17,92] and a CBD at the *C*-terminus (figure 1), which recognizes chitin polymers that are present in mucin gels of *Ciona*, as well as fungi whose cell membranes are chitin-rich [18,20]. Unlike mucosal antibodies of vertebrates, VCBPs are directly synthesized by epithelial cells of the gut that resemble goblet cells. The VCBPs are directly co-associated with mucus-rich vacuoles that are actively exuded [18,19].

Haplotype variation and polymorphisms are prominent in the gene segment encoding the *N*-terminal V-type Ig domain of the VCBP-2 and VCBP-5 genes in *B. floridae* [89,90,93], suggesting a co-evolving pathogen response. This feature has not been observed within *Ciona* VCBPs [17]; however, *Ciona* VCBPs A–D are more homologous to amphioxus VCBP-1, −3 and −4, which also lack the polymorphism of VCBP-2 and −5, paralogous genes that are found clustered in the genome [93].

In vitro binding experiments using polymorphic variants of VCBP-2 and −5 have yet to be performed on native gut bacteria; however, it appears that polymorphism of the *N*-terminal V-type Ig domain is driven by a variety of factors that include pathogen selection pressures and genomic instabilities of the clustered haplotypes [93,94]. VCBPs in *Ciona*, along with VCBP-1, −3 and −4 from amphioxus, are likely recognizing conserved molecular patterns on the surface of microbes [95,96].

6. Ig-domain proteins are essential in shaping microbiome settlement in the gut of chordates

Although the membrane-proximal mucus gel is often dense and forms a glycocalyx, the outer mucus is impacted by

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digestive enzymes and forms a loose layer that attracts microbial grazing and settlement [97,98]; it also represents the site where SIgA interacts with a rich assortment of microbes [59]. Dense polymicrobial biofilm communities, in specific regions of the gut, can form due to the glycan structure of mucins and other biophysical factors, with microbial nutrition preferences serving as a primary driver of settlement along with niche preference distributions of strain variants. A parallel distribution of unique immune recognition responses and secreted effectors influence these communities [16,99–101], and remains a guiding principle of the restaurant hypothesis for microbial settlement in the gut. Variation in the local distribution of antibody responses in vertebrates [5,59], and by inference, the VCBPs in protochordates [18,102], could strongly impact the dynamics of microbial settlement. This ancient selective process likely plays a significant role in shaping microbial communities of mucosal environments, with a significant role in 'sculping the microbiome', as per the focus of many companion articles in this special issue and as proposed in the evolutionary history of adaptive immune components [33].

Colonizing the animal gut is challenging due to its unique biophysical properties, distribution of cell types and immune effectors, and constant mucus shedding and replacement. Some microbes have developed strategies to overcome this barrier or work collaboratively with components of the barrier. One such strategy is the development of secreted antibody-like effectors with Ig domains, which can bind to either commensal or pathogenic microbes within the lumen, favouring settlement of beneficial microbes while preventing penetration of mucus layers by pathogens [1,3–5,8,10,59]. Microbes that are bound by antibodies may remain active and affect the microbiome, often forming biofilms that permit their interactions as complex polymicrobial communities. It is worth noting that since antibodies do not directly kill cells, certain interactions with microbes may involve structural support through their entrapment in the outer layer of mucus that may facilitate their survival [3,14]. Further investigations are required to improve our understanding of the mechanisms by which IgG and SIgA impact settlement preferences among commensal communities [10–12,14].

Animal mucin gels have an impact on microbial colonization due to their biochemical and physical makeup. The MUCs contain many O-glycan arrays, which serve as a carbon source for commensals while still maintaining an essential role for mucus, such as lubrication [25,103]. Pathogens have a tendency to overgraze and secrete proteases that target the barriers and expose the epithelium [25]. The mucin fibres, which are heavily glycosylated and cross-linked, produce a distinctive mesh-like structure, creating a porous network with different pore sizes on various mucosal surfaces that is linked to a number of different physiological processes [39]. This structure can directly obstruct or impede the diffusion of microbes or serve an adhesive role in facilitating interactions between microbes and mucins fibres, establishing specific limitations on pore size accessibility for different members of the microbiome. With a distinctive ability to entrap within the mucus mesh or gels, Ig-domain proteins can dramatically influence settlement behaviours, polymicrobial interactions and other ecological dynamics of the gut microbiome [3,8,11]. Future studies should help define if accessory interactions among Ig-containing effectors and mucus components (i.e. mucin or chitin) impact settlement dynamics, as does the secretory component for SIgA.

Thus, mucosal antibodies can operate as third-party cross-linking molecules that reinforce the barrier properties of mucus, limiting interaction between pathogens and epithelial cells or facilitating settlement dynamics among commensals without altering other functions of the mucus gels. Investigations into the mucus-trapping properties of antibodies have focused significantly on IgG, which is relatively easy to isolate and store, as opposed to IgA, which is unstable, prone to aggregate and more challenging to purify [3]. Apart from the critical roles of the secretory component, as defined above, studies focused on modifications to the Fc region have revealed that low-affinity, hydrogen bond interactions with mucin glycans help impose significant physical limitations on how pathobionts diffuse through mucus gels [1–5]. These interactions also may increase the likelihood of collisions with other microbes, reducing the amount of IgG initially required, i.e. a complex repertoire of specificity can be maintained. Secreted effectors, such as antibodies, can have a distinctive impact on downstream immune responses within the mucosal environment, depending on how their Ig domain(s), such as the Fc region, interact with and transmit signals to other types of membrane-bound effector molecules [101,104–106]. This combination of microbes, antibodies and mucins plays a crucial role in enabling interactions with innate immune receptors and regulating inflammation.

The fundamental process of mucus trapping that is mediated by convergently evolved secreted effectors [14] is pivotal in controlling microbial colonization of mucosal surfaces. The Ig domain serves an ancient and fundamental role in cell-to-cell and microbial recognition [83,107], and its diverse functionality is well-documented among diverse invertebrate lineages [96,107]. While various protein domains likely participate in mucus trapping, the composition of mucins and the associated glycans undoubtedly serve pivotal selective roles mediating recognition specificity. For example, chitin fibres serve as essential constituents of mucus gels in invertebrates and aquatic vertebrates. The identification of the VCBP genes and their expression in the digestive tract during organogenesis and in *Ciona* adults [17,89,102] led us to examine the role of these molecules within the gut. Essential microbe influencing functions of the VCBPs, as secreted immune effectors at the mucosal sites, have been documented (reviewed in [96,108]). Follow-up *in vitro* studies have been made possible with VCBP-Cs due to their ease of purification on chitin columns and feasibility for synthesizing both full-length and partial-sized recombinant versions [17].

Briefly, *in vivo* and *in vitro* experiments have shown that VCBP-C can be tethered to chitin-rich mucus of the gut and/or branchial basket via the CBD domain [18,55]. It also exits as a soluble form in the lumen [18] and can bind bacteria and tag them for opsonization via the *N*-terminal Ig domain [17]. VCBP-C also can impact biofilm formation among bacteria [18,109] and influence prophage induction among lysogenized bacteria [109]. The CBD can bind to diverse fungi, labelling bud scars in yeast, and hyphae in filamentous fungi, identifying sporangia and their spores [20]. Transcriptome data and quantitative RT-PCR reveal surprisingly specific VCBP-C responses during exposure experiments and colonization of naive juvenile animals (Natarajan *et al.*, in preparation). VCBP-C plays crucial roles in shaping host responses to diverse microbial communities that include both commensal organisms as well as potential pathogens. The integration of a CBD in VCBPs and its interaction with chitin-rich mucus support a role in

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the mucus trapping of microorganisms, as has been demonstrated for the secretory component of pIgs, and the Fc region-mediated binding of IgG to mucins. However, clear evidence that VCBPs mediate specific roles in pathogen exclusion or in the selection or retention of commensals remains to be shown. VCBP-D, which also is produced in the gut, is related to VCBP-C but lacks a CBD [17], suggesting that mucus trapping with and without tethering to chitin fibres of the mucin gels may factor in gut homeostasis in *Ciona*.

Further investigation is warranted into how glycan structures influence the selective pressures driving domain acquisition among the secreted effectors that immobilize both beneficial and harmful microbes in mucosal environments. Binding to polysaccharide-rich mucins, either directly via glycosylated Fc domains of antibodies, the secretory component, or CBDs of the VCBPs, represents an integral element in creating the chordate barriers that both trap microbes and provide the Ig-domain-dependent effector feedback for innate immune functionality. The tethering of antibodies and other secreted effectors, like VCBP to glycans and subsequent interactions with microbes, may reflect a common functionality that influences settlement dynamics by permitting commensals and excluding pathogens at the mucosal surface

7. Concluding remarks

In this essay, we have explored the critical role that mucus gels serve in conjunction with secreted immune effectors to shape microbial ecology of mucosal environments and mitigate the effects of pathogenic microbes. It is becoming increasingly more apparent that major evolutionary differences exist in the composition and structure of the mucus gels, notably the addition of diverse glycans onto mucins and in some cases the coupling of chitin fibres that is common in aquatic organisms and some terrestrial invertebrates (such as insects). We suggest that in addition to other physiological effects, diversity in mucus composition acts as a selective force driving the evolution and selection of not just the commensal flora but secretory immune effectors serving essential functions. Despite their considerable difference in the number and complexity of Ig domain arrangements, antibodies and VCBPs, as secreted immune effectors, share an essential functional convergence in regulating microbial colonization at mucosal surfaces. Future research on host–microbial interactions should consider the mucus composition to identify and investigate additional secreted immune effectors that may recognize glycans of the mucin gels, revealing them as essential players in the overall management of mucosal ecosystems.

Data accessibility. This article has no additional data.

Declaration of AI use. We have not used AI-assisted technologies in creating this article.

Authors' contributions. L.J.D.: conceptualization, investigation, supervision, writing—original draft, writing—review and editing; G.W.L.: conceptualization, investigation, writing—original draft, writing—review and editing; A.L.: conceptualization, investigation, writing—original draft, writing—review and editing.

All authors gave final approval for publication and agreed to be held accountable for the work performed therein.

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