Chapter 8
Regulation of Immune Responses by Lectins



Shamna Naseemashahul and Femi John Fawole

Abstract Lectins are carbohydrate-binding proteins having receptors for recognizing different carbohydrate moieties such as mannose, galactose, rhamnose, fucose, lactose, N-acetyl glucosamine, and N-acetyl galactosamine with specificity. They perform this function with or without the support of divalent cations. Moreover, some lectins can bind erythrocytes and result in agglutination, and, hence called "phytohaemagglutinin." Lectins are structurally diverse and are present in both plant and animal kingdoms. Lectins participate mainly in innate immune defense in animals by identifying carbohydrate and glycoconjugate moieties in cells and biological fluids. Based on the carbohydrate recognition domain (CRD), lectins can be classified into various groups. The plant lectins are very well studied, while among the animal lectins many groups including fish and shellfish lectins are less studied compared to plant lectins, especially the regulation of lectins.

Keywords Fish lectin · Shellfish lectin · Lectin mediated immunity · Carbohydrate recognition domain · Types of lectin

Abbreviations

AGPs	Arabinogalactan proteins
Artin M	Artocarpin mannose binding
CFL	*Cratylia argentea* lectin
CLCP	Charcot Leyden Crystal Proteins
CLP	Collectin like protein

S. Naseemashahul
Fish Nutrition Biochemistry and Physiology Division, ICAR-Central Institute of Fisheries Education, Mumbai, India

F. J. Fawole (✉)
Fish Nutrition and Biochemistry Unit, Department of Aquaculture and Fisheries, University of Ilorin, Ilorin, Nigeria
e-mail: fawole.fj@unilorin.edu.ng

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CLRs	C-type lectin receptors
Con A	Concanavalin A
ConBr	*Canavalia brasiliensis* lectin
CR1	Complement receptor 1
CRD	Carbohydrate recognition domain
CTLs	C-type lectins
DORN1	Does not Respond to Nucleotides 1
FTLs	F-type lectins
GalNAc	N-acetylgalactosamine
GluNAc	N-acetyl glucosamine
IFNγ	interferon-gamma
IL	interleukin
LecRLKs/RLPs	lectin receptor-like kinases/proteins
LOX-1	Lectin like oxidized low-density lipoprotein receptor 1
LysM	Lysin motif domain
MBPs	Mannose-binding proteins
Nictaba	Nicotiana tabacum agglutinin
OniL	*Oreochromis niloticus* Lectin
PAMPs	Pathogen associated molecular patterns
proPO	Prophenoloxidase system
PRRs	Pathogen recognition receptors
RBLs	Rhamnose-binding lectins
RcaL	*Rachycentron canadum* Lectin
Siglecs	Sialic acid-binding immunoglobulin-like lectins
Th1	T1 helper cell
TLR	Toll-like receptors
UAE1	Ulex europaeus agglutinin 1

8.1 Introduction

Lectins are glycoproteins that play important role in non-specific immune system activation and function as receptors for recognizing microbial pathogens via their carbohydrate surface (Ewart et al. 2001a, b; Wang and Wang 2013; Elumalai et al. 2019). One of the most exciting and significant functions of lectins is in the area of immune system activation occasioned by their ability to recognize sugars on the surface of a potential pathogen, and this often incites the lectins-protein to engulf invading microbial pathogens thereby resulting in their destruction by the phagocytic cells (Lu 1997; Ewart et al. 2001a, b; Vasta et al. 2011). Lectins are of different types and differ in their molecular weight, amino acid sequences, and sugar-binding specificity. They are widely found in microbes, yeast, plants, and animals, and play an important biological role in teleost, animal, and plants such as antitumor, immunoregulatory, pathogen recognition and cell adhesion, antimicrobial, cells

agglutination, and cell signalling (McGreal et al. 2004; Wang and Wang 2013; Ng et al. 2015; Brinchmann et al. 2018). A recombinant mannose-binding lectin homologue was reported to agglutinate *Streptococcus agalactiae* and *Aeromonas hydrophila* in vitro (Mu et al. 2017). Furthermore, in vivo challenge with bacterial pathogen showed higher expression of mannose-binding lectin gene in the liver, spleen, and kidney of Nile Tilapia (*Oreochromis niloticus*) (Mu et al. 2017). Several other studies have also documented the role of lectins in host protection against bacterial infection (Arasu et al. 2013; Rajan et al. 2013; Wang et al. 2014; Zhou and Sun 2015; Zhou et al. 2016; Liu et al. 2016; Wang et al. 2017; Huang et al. 2020). This points to the agglutination capability and immune response activation of lectins against bacterial pathogens in fish. Aside from immune system function, some lectin types have been reported to bind endogenous ligands and are involved in different physiological functions such as preventing polyspermia or acting as antifreeze protein (Vasta et al. 2011). Nevertheless, these molecules (i.e. lectins) with potency to fight microbial pathogen in host cells are involved in many intra- and extracellular processes. Hence, this chapter presents information on different types of lectin, role on immune responses in animal and plant, mode of actions, regulation of lectins, and the prospect of lectins in fish health management.

8.2 Type of Lectins

Lectins are produced by a wide variety of living organisms and they can be grouped according to their species of origin, such as animal lectins, fish lectins, fungal lectins, bacterial lectins, and plant lectins (Mishra et al. 2019). Several types of lectins families have been identified and characterized especially in fish based on their specificity for carbohydrate recognition domain, structure, and calcium dependency (Taylor and Drickamer 2003; Elumalai et al. 2019). Among them include C-type lectins, F-type lectins, ricin type, lily type, galectins, rhamnose-binding lectin, fish-egg lectin, interlectin, and tectonin-type lectins (Ogawa et al. 2011; Ng et al. 2015; Brinchmann et al. 2018; Elumalai et al. 2019). In addition to C-type and galectins, other animal-specific families of lectins have also been reported such as I-type lectins, L-type, pentraxins, R-type, F-box lectins, ficolins, P-type lectins, and tachylectins (Ogawa et al. 2011; Wang and Wang 2013). Comprehensive reviews on different lectins family in animals, fish, and plants are available (Weis et al. 1998; Kilpatrick 2002; Zelensky and Gready 2005; Ogawa et al. 2011; Elumalai et al. 2019; Tsaneva and Van Damme 2020), and some important groups are mentioned in Table 8.1.

Table 8.1 Classification, localization, and functions of important groups of lectins

Sources	Lectins family/ domain	Localization	Function	Reference
Animal	C-type lectins, Rhamnose-binding Lectins, Pentraxins, I-type lectins, L-type, R-type, F-box lectins, ficolins, P-type lectins, and tachylectins	Gill, liver, intestine, spleen, muscle, heart, kidney, and mucus	Immune response, opsonization, phagocytosis, and activation of complement pathway	Ogawa et al. (2011), Wang and Wang (2013), Gabius (1997), Lis and Sharon (1986), Huang et al. (2020)
Plant	Agaricus bisporus lectin,	Nucleus, cytosol	Recognize glycoconjugate on cell surface, host–pathogen interaction, cell signalling and cell–cell communication, protection against harmful phytopathogenic microorganisms, insect, and predatory animals	Lannoo and Van Damme (2014), Tsaneva and Van Damme (2020)
	Cyanovirin, Chitinase related agglutinin, Jacalin,	Nucleus Vacuole, membrane bound Nucleus, cytosol, vacuole		
	Legume lectin, LysM,	Vacuole, nucleus, Cytosol or membrane-bound		
	Ricin-B,	Vacuole, nucleus, cytosol		
	Amaranthin	nucleus, cytosol		
Algal	N-glycan specific, High mannose type N-glycan specific		Anti-inflammatory, Anti-tumor, Antiviral	Mishra et al. (2019)
Fungal			Growth, development, morphogenesis, molecular recognition, early-stage infection	Mishra et al. (2019)

8.3 Immune Functions

Glycosylation of proteins can be considered as one of the major posttranslational modifications and many proteins have specificity in recognition to these glycosylated sites. The carbohydrate-binding proteins can recognise and bind different forms of carbohydrate in glycoproteins, proteoglycans, free sugars etc. This carbohydrate-binding property of proteins can be a basis for the initiation of immunity in all kingdoms of life. Generally, there are three receptor groups, galactins, siglecs (Sialic acid-binding immunoglobulin-like lectins) and C-type lectins (CTLs).

In multicellular organisms, various physiological functions like cell migration, homeostasis balancing, and innate immune signalling etc are depending on the capability of cells to distinguish glycoconjugates (glycoproteins and glycolipids) via glycan-binding proteins or lectins. Despite the differences between the immune system of animals and plants, there are some striking similarities like lectin-mediated immune response. Lectins can act as pattern recognition receptors or pathogen recognisers and initiate the immune response. However, lectins are involved in damage associated patterns as well as immune pathology. Various immunological processes such as phagocytosis, opsonisation, cell adhesion, cell activation, migration and differentiation and cell death or apoptosis involve lectin-carbohydrate interactions. Enhanced respiratory burst activity and bactericidal activity were also stimulated by lectins. In cells, the lectin receptors are present on the cell surface or insoluble form (Ni and Tizzard 1996).

8.3.1 Lectin Mediated Immunity in Plants

Plant lectins are well known for their immunomodulatory functions and studies revealed that there are 12 families of lectins having cytokine and other immune mediator production functions (Da Silva and Correia 2014; Coelho et al. 2017). Plant lectins can bind with the bacterial cell as well as with the glycan moiety of immune cells. Hence, their action can be on both microbial cells and immune cells and enhance the signalling and further action of activator mechanism (Van Damme et al. 2008; Souza et al. 2013). Con A, ConBr, CFL, UEA1, Cramoll, Artin M, and Jacalin lectins are widely observed in plants and studied on the immunomodulatory activity in response to various pathogens. Con A when treated in murine macrophages stimulated the toll-like receptors (TLR) to enhance the secretion of cytokines and nitric oxides and promoted various signalling pathways (Sodhi et al. 2007). Moreover, in mice, the ConA enhanced the survival of Klebsiella infection and reduced liver necrosis (Kuo et al. 2007).

8.3.2 Lectin Mediated Immunity in Animals

Similar to plant lectins, lectins obtained from animal tissues are found to be potent immune boosters. Though not identified as lectin, it seems that the first reported animal lectin is Charcot Leyden Crystal Proteins (CLCP). However, the lectin activity was first observed in snakes. But, the biochemical analysis of the eel agglutinin revealed the non-immunoglobulin nature for the first time. The animal lectin has both immune and non-immune functions. The action of lectin is carried out by agglutination, structure-activity divergence, tertiary structure convergence, non-carbohydrate binding, etc. Similar to plant lectins, animal lectins also recognize molecules in the immune system. This function is possible by the direct defense,

suppression or enhancement of the immune system, blocking autoimmunity, and recognition and molecule trafficking. Animal lectins showed an extraordinary ability to bind protein, lipid, and nucleic acid, i.e., the animal lectin binds with non-carbohydrate moieties also. Hence, the animal lectins can be called "bifunctional molecules" (Kilpatrick 2002). The collectin like protein (CLP) cloned from serum and hepatic tissues of humans can promote phagocytosis (Nakamura et al. 2001; Ohtani et al. 2001).

8.3.3 Lectin Mediated Immunity in Fish

Several reports are published on the immune-stimulating effect of lectin and it is depicted that several fish lectins mediate pathogen recognition (Yano 1996; Vasta et al. 2011; Lino 2013; Elumalai et al. 2019). Galectins, C-type lectins (CTLs), F-type lectins (FTLs), and pentraxins are the predominant lectin families observed in cartilaginous and bony fishes (Cammarata et al. 2016). The presence of humoral and membrane-associated lectins which helps in establishing positive interactions with colonizing microbes are also observed in fishes (Vasta et al. 2011). Most of these lectins are C-type and several other functions such as agglutination, immobilization with complement-mediated neutralization are also performed by fish lectins (Table 8.2). Different types of CTLs were isolated from species such as zebrafish, *Silurus asotus*, fugu, Japanese eel, rainbow trout, common carp, etc. (Tasumi et al. 2002; Singha et al. 2008; Zhang et al. 2011). Mucus lectin from the skin surface of *Genypterus blacodes* was the first isolated fish lectin (Oda et al. 1984).

Isoforms are also observed in fish lectins, for example, the F-type lectins from Japanese eel had various forms (Honda et al. 2000). Serum of *Sparus aurata* contains fucose binding lectin (Sau FBP32) and mucus extract of loach is also reported to have lectins having agglutination property (Sun et al. 2019). The smelt lectin classified in type II antifreeze protein belongs to the C-type lectin group. The same category of lectins is isolated from herrings (Ewart and Fletcher 1993). Lectin isolated from the ovaries and roe of grass carp showed mitogenic activities on splenocytes of murine and phagocytic activity in seabreams (Ng et al. 2003). Similarly, a lectin (at 14 μM) with a mannose-binding property, isolated from the ovary of *Rachycentron canadum* also exhibited a mitogenic activity on a mouse (Ngai and Ng 2007). Serum lectin from *Rachycentron canadum* or RcaL is also reported to be an immunomodulator. Furthermore, lectin isolated from grass carp ovaries having specificity towards rhamnose enhanced the expression of interferon-gamma (IFNγ) and interleukin 2 (IL-2) expressions in splenocytes (Lam and Ng 2002). Rhamnose-binding lectins are one of the unique fish lectins present in serum, embryos, and eggs of fish (Watanabe et al. 2009; Cammarata et al. 2014). In vitro T1 helper cell (Th1) was inducted by a mannose-specific serum lectin (OniL) in *Oreochromis niloticus.*

Table 8.2 Immunomodulatory and biological roles of fish lectins

Lectin family	Lectin	Source	Mode of action	Target	References
F-type lectins	GANL (220 kDa)	Bighead carp	Growth inhibition by agglutination	*Vibrio harveyi*	Pan et al. (2010)
Galectins	AJL-1 (30 kDa) PfGAL9 (35.12 kDa) Congerin, LcGal9 Pufflectin	Japanese eel Yellow catfish Conger eel Yellow croaker Pufferfish	Growth inhibition by agglutination Cellular encapsulation Binds parasite	*Streptococcus difficile, E. coli, Aeromonas hydrophila, B. Subtilis*, *B. Megaterium,* and *S. aureus* Bacteria Parasitic nematodes *V. Alginolyticus* and *A. hydrophila Heterobothrium Okamotoi*	Tasumi et al. (2004) Wang et al. (2016) Nakamura et al. (2012) Zhang et al. (2016) Suzuki et al. (2003)
Intelectins	SaIntL (35 kDa	Catfish	Growth inhibition by agglutination	*Aeromonas salmonicida*	Tsutsui et al. (2011)
Fish-egg lectins	RbFEL (28.2 kDa) Coho egg lectin zFEL KPL (140 kDa) RBL (30 kDa)	Rock bream Coho salmon Zebrafish Skipjack tuna Chinook salmon	Enhanced lectin activity on challenged with pathogens Binds bacteria Phagocytosis of microbes by macrophages Hemagglutination Anti-proliferative activity	*Edwardsiella tarda*, *Streptococcus iniae* and red sea bream iridovirus (RSIV) *Aeromonas salmonicida* *Aeromonas hydrophila* Blood type A erythrocytes Human breast cancer MCF-7 cells and hepatoma Hep G2 cells	Kim et al. (2011) Yousif et al. (1995) Wang et al. (2016) Jung et al. (2003) Bah et al. (2011)
C-type lectins	HjCL BGL (80 kDa) SauFBP32 LycCTLR OniL (17 kDa)	Japanese bullhead Shark Potcha fish Atlantic salmon Yellow croaker Tilapia fish	Blood coagulation Binds bacteria Activates immune response Exhibit strong cytotoxic effects Binds to bacteria	Erythrocytes (Shark blood) *Aeromonas hydrophila*	Tsutsui et al.(2015) Fock et al. (2001) Ewart et al. (1999) Ao et al. (2015) da Silva et al. (2012) Coriolano et al. (2012) Ngai and Ng (2007)
	RcaL MBL	Cobia fish Cobia fish	Induced higher IFN-γ Production Lower IL-10 as well as Nitrite release Induced production of IL-2 and IL-6 and proliferative responses, Mitogenic activity	Mice *Vibrio anguillarum* and *A. salmonicida* *E. coli* and *B. cereus* Mouse splenocytes	

Adopted with permission from Elumalai. et al. (2019): The Role of Lectins in Finfish: A Review. Reviews in Fisheries Science & Aquaculture, 27(2), 152–169

8.3.4 *Lectin Mediated Immunity in Shellfish*

The lectins present in the invertebrate hemolymph have a role in neutralizing pathogens and support the phagocytosis in hemocytes. The lectins of C-type are observed in crustaceans and arthropods, while pentraxins are quite common in shellfish. Beta, 1–3 glucan and tachylectins identified from yeasts and bacteria has the capacity to activate the prophenoloxidase system (proPO) in shrimp. The activity of lectins on clotting proteins in crustacean immunity is also well studied (Hall et al. 1999). However, compared to other arthropod groups, the studies on crustacean lectins are less. A lectin isolated from *Limulus polyphemus* (Horseshoe crab) showed a property of broad-spectrum recognition to various monosaccharides (Vasta 1992). Similarly, lectin obtained from horseshoe crab also showed a binding property to sialic acid.

The structurally different lectins obtained from invertebrates act as humoral innate immunity and enhance phagocytosis, melanization, respiratory burst activity, and encapsulation (Zhang et al. 2011; Wang and Wang 2013) (Fig. 8.1). The globular region of lectin having less than 200 amino acids is considered as the sugar-binding site which promotes phagocytosis even in invertebrates. Most of these lectins belong to the C-type family while other groups also show similar action. Furthermore, the pathogen recognition receptor (PRRs) group having CTLs with C-type lectin domain (CTLD) act as the major group in crustaceans which shows the predominant role of C-type lectins in the innate immune response of crustaceans (Zhang et al. 2020a, b). Most of these groups showed agglutination of human erythrocyte or bacteria or yeast and sugar-binding nature (Elumalai et al. 2019; Zhang et al. 2019). Antiviral action was observed in a lectin (PcLec) isolated from crayfish and its action against bacterial pathogens need further investigation (Zhang et al. 2020a, b). In shrimps, lectins with multiple roles and different structure and expression patterns are observed and are of seven groups, viz., galactins, L-types, M-types, C-type, p-type, fibrinogen like, and calnexins (Wang and Wang 2013). The

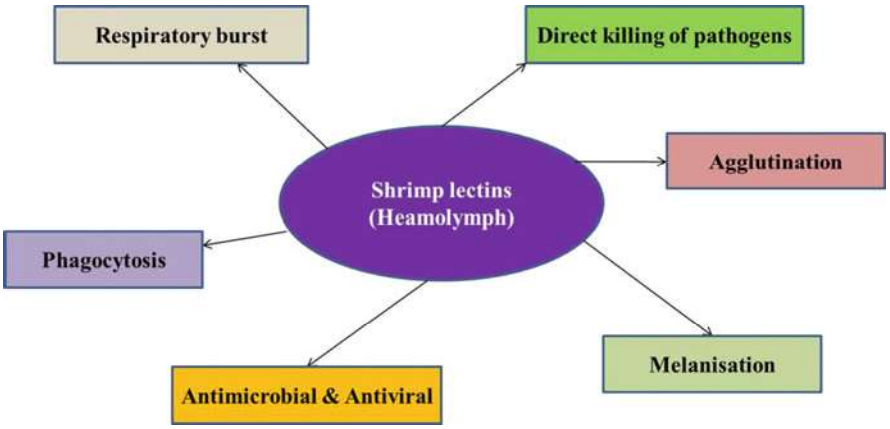


Fig. 8.1 Mode of action of shrimp lectins. Adapted and modified from Wang and Wang (2013)

rhamnose-binding lectins (RBLs) showed specificity towards L- rhamnose is found mainly in fishes and shellfishes (Tateno 2010).

8.4 Regulation in Plants

Recent researches using molecular techniques and omics studies paved the way towards the action and interaction of lectins in plants at the molecular level. Studies on the expression of lectin genes and the pathways triggering immunity by lectins showed the importance of the molecule in innate immunity and cell signalling. The surface localized pathogen recognition receptors (PRRs) that recognize apoplastic elicitors and the intracellular receptors regulate the immune action in cells (Jones and Dangl 2006; Bos et al. 2010). During the entry of pathogen through the cell wall, the cell wall degrading enzymes get activated and AGPs and degradation products of pectin (oligogalacturonides) can be recognized by lectin receptor-like kinases/proteins: LecRLKs/RLPs (Benedetti et al. 2015; Davidsson et al. 2017). In addition to the vacuolar and surface lectins, the cytoplasmic and nuclear lectins also play a major role in lectin mediated immunity in plants.

Cytoplasmic lectins such as Nictaba (Nicotiana tabacum agglutinin) are involved in cell signalling (Van Damme et al. 2004). Nictaba is specific to GlcNAc or GalNAc residues (Lannoo et al. 2006). ConBR from *Canavalia brasiliensis* enhanced the production of Interleukin 2, 6 (IL-2, IL-6) and interferon-gamma (IFN-γ) and decreased the production of IL-10 in the splenocyte murine (de Oliveira Silva et al. 2011). ArtinM interacts with intracellular matrix Mannose by a process called haptotaxis and supports the migration of neutrophils (Souza et al. 2013). In mice, another lectin called Cramoll regulate the level of reactive oxygen species (ROS) and different interleukins such as IL-6, 10, 17, and IFN-γ. Arabidopsis have a lectin designated as DORN1 have recognition to peptides and nucleotides. The universal LysM lectins and LecRLKs have both positive and negative roles in plants as microorganisms having these lectins will suppress plant immunity (Rovenich et al. 2016). Effectors have the capacity to downstream signalling events. Plants have both constitutive and inducible lectins. Constitutive lectins are highly abundant in cells while in response to stress exposure the inducible lectins are expressed at a low base level. Stress inducible lectins are mostly nucleoplasmic in nature (Van Damme et al. 2004). However, the mechanism of action and regulation of nucleocytoplasmic lectins are studied less.

8.5 Regulation in Animals

Lectins are associated with pathogen-associated molecular patterns (PAMP) and they recognize pathogen recognition receptors (PRRs) in a reversible binding and activate the lectin pathway (Vasta et al. 2011). The binding of carbohydrate moieties

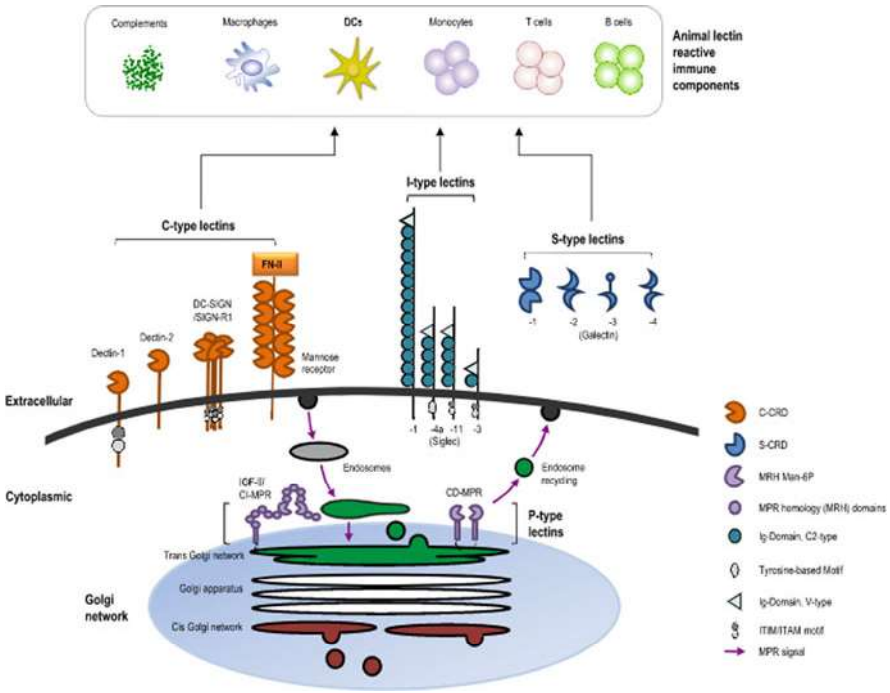


Fig. 8.2 Domain structure models, the location of several representative animal lectins, and the immune cells reacting with animal lectins
Carbohydrate recognition domains (CRDs) of each lectin are depicted in the respective whole structures. S-type lectins are secreted into the extracellular matrix of body fluids. C-type and I-type lectins are localized to the plasma membrane, and P-type lectins are located in luminal compartments of the secretory pathway. CD-MPR, cation-dependent mannose-6-phosphate receptor; CI-MPR, cation-independent mannose-6-phosphate receptor; CRD, carbohydrate recognition domain; DC, dendritic cell; DC-SIGN, DC-Specific Intercellular adhesion molecule-3-Grabbing Non-integrin; IGF, insulin-like growth factor; MRH Man-6P, mannose 6-phosphate receptor homology; ITIM/ITAM motif, immunoreceptor tyrosine-based inhibition/immunoreceptor tyrosine-based activation motif. (modify)
Adapted from Loh et al. (2017): Animal lectins: potential receptors for ginseng polysaccharides. Journal of gingseng 41: 1–9

activates various immune responses such as complement pathway, agglutination, immobilization, and opsonization (Sharon and Lis 2004) (Fig. 8.2). The animal lectin galectin has a carbohydrate recognition domain with two β-sheets folded antiparallel producing a ligand-binding site that is concave in shape. This helps in binding β-galactoside glycans specifically (Diehl et al. 2010). Other important lectins in animals such as Calnexin and Calreticulin have both globular and proline-rich (P) regions and are made of two β-sheets, and hence, look like a β-sandwich (Chouquet et al. 2011). Divalent cations have a role in carbohydrate recognition in some animal lectins, especially in the C-type lectin family. C-type

lectin receptors play a critical role in immune responses and inflammation and recognition of molecular pattern of pathogens. The micro-RNA regulated C-type lectin receptors can distinguish molecular patterns of endogenous and exogenous pathogens and initiate downstream signalling cascade which induce the production of effector immune cells and inflammatory responses, furthermore, this maintain homeostasis in cell (Ganguly et al. 2020).

Lectins with N and O-glycan specificity control regulatory signals of cell homeostasis. They amplify or silence autoimmunity responses or neoplastic disease responses (Rabinovich and Croci 2012). Conformational and allosteric changes on a specific protein as a result of post translational modifications can leads to generation of new epitopes (binding site). This can lead to the formation of signalling complexes in cell. Some cell surface glycans acting as a ligand for lectins that facilitate receptor dynamics (Dennis and Brewer 2013). Hence, density of glycan epitopes has an important role in lectin mediated receptor and transporter regulation in animals (Dam and Brewer 2010). This might be due to the higher regulation of glycan binding sites on cell surface and hence we can derive a conclusion that interaction between surface glycan and lectin and golgi glycosylate transferase helps the regulation of cell surface receptors and transporters to respond to cellular signalling and environmental changes (Dennis and Brewer 2013). In addition, in autoimmune diseases, C-type lectin receptor controls the expression of BIC/MicroRNA 155 and therefore a feedback regulation of T cell development through thymus control is indirectly mediated by CD69, a C-type lectin receptor (Sánchez-Díaz et al. 2017). In humans, lectin like oxidized LDL receptor (LOX-1) plays an important role in cardiovascular diseases and is regulated by several mechanisms like upregulation of receptor expressions, signalling modulation through internalization or proteolysis, and adjusting the activity of surface receptors (Mentrup et al. 2021).

In human serum, five different pathways of lectin are observed and only two pathways (ficolin and collectin 11) are found to be effective on activation of complement pathway during pneumococci infection (Ali et al. 2012). However, the mannan binding lectin which (a group of collectin proteins) pathway activate complement pathway in human serum is upregulated by *Neisseria gonorrhoeae* through activation of complement receptor 1 (CR1) (Gulati et al. 2002).

Several studies are reported in recent years explaining the mode of actions of lectin in fishes (Elumalai et al. 2019). In fishes, the presence of lectins is observed in the intestine, plasma, liver, gills, eggs, embryos, serum, and surface mucus (Mistry et al. 2001a, b; Ng et al. 2015). In response to stress or infection, the concentration of the F-type lectin is observed to be increased in the plasma of teleosts fish (Parisi et al. 2015). The F-type lectins prove to be effective against *Vibrio harveyi* as they agglutinate the bacterial cells (Pan et al. 2010). Similarly, they are found to be effective against heavy metal accumulation in fishes (Guardiola et al. 2015). Functionally analogous lectins in fish, including mannose-binding lectin, have been reported to effectively agglutinate *Streptococcus agalactiae* and *Aeromonas hydrophila* in vitro. Also, in vivo challenge with bacterial pathogen showed higher expression of mannose-binding lectin gene in the liver, spleen, and kidney of Nile

Tilapia *Oreochromis niloticus* (Mu et al. 2017). In Obscure pufferfish *Takifugu obscurus*, the gene expression of two novels CTLs (ToCTL1 and ToCTL2) were found up-regulated after being challenged with both the gram-positive and gram-negative bacteria (Huang et al. 2020). The authors opined that the CRDs inhibited the growth of the bacteria by binding to the sugar on the bacteria surface, thereby establishing the vital role of CTLs in clearing invading foreign agents. In addition to agglutinate cells as well as precipitate carbohydrate moieties, fish lectin can initiate an innate response against pathogens and helps in the establishment of positive interaction with colonizing microbial species (Vasta et al. 2011).

Mucus lectin in the skin of fish is the first line of defense against pathogens and it was confirmed when intelectins got activated in response to *Aeromonas salmonicida* infection. However, in the study, the gene expression of mucus lectin was not inducted in vivo (Tsutsui et al. 2011). Moreover, an egg lectin isolated from coho salmon could not prevent the growth of *Aeromonas salmonicida* indicating multiple roles of lectin other than disease prevention (Yousif et al. 1995). The C-type galactose binding lectins such as eCL-1 and eCL-2 were found to be downregulated in Japanese eel when it was shifted to the marine environment from freshwater (Mistry et al. 2001a, b). Lectin isolated from yellow croaker (galectin) also showed an agglutination property against bacteria (Zhang et al. 2016). Fully developed immune tissues of rock bream had lily type lectin (OfLTL-2 and 3), which play an important role in immunity.

8.6 Prospects of Lectins in Fish Health Management

One of the most exciting and significant functions of lectins is in the area of immune system activation occasioned by their ability to recognize sugars on the surface of a potential pathogen, and this often incites the lectins-protein to engulf invading microbial pathogens thereby resulting in their destruction by the phagocytic cells (Lu 1997; Ewart et al. 2001a, b; Vasta et al. 2011). In the innate part of the immune system, lectins are one of the important agglutinins which help in the agglutination of a foreign body (i.e. non-self). Since fish depends majorly on the innate immune system, and antibiotic resistance is one of the problems confronting the global aquaculture industry, lectins could be useful in fish health management as different lectins have been reported to show cytotoxic effects against the bacterial pathogen (Iordache et al. 2015). Furthermore, the opsonizing lectins present in fish serum could be useful in fish health, both in terms of gaining a better understanding of the immune system and the development of applications for disease prevention. Functionally analogous lectins in fish, including mannose-binding lectin, have been reported to effectively agglutinate bacterial cells. This points to the agglutination capability and immune response activation of lectins against bacterial pathogens in fish. Ottinger et al. (1999) showed that salmon lectins could opsonize *A. salmonicida* and make the bacterial more susceptible to the action of phagocytes. He further reiterated that the binding of the salmon serum lectin to the bacteria prior to

phagocytosis improved the bactericidal activity of macrophages and increased the degradation of internalized bacteria (Ottinger et al. 1999). Other documented literature have also clearly indicated that many of these lectins have the capability to enhance the phagocytic activity of immune cells, however, further research should be directed at understanding the molecular mechanism of the functional aspect of different types of lectins in fish immunity and the regulatory aspects. This will help in the development of fish lectins-based immunotherapeutic agent or nutraceuticals to fight infectious diseases in aquaculture.

8.7 Conclusion

Lectins are a versatile protein distributed widely in various kingdoms with innate immune modulation. Reports on the role and function of lectins in plants, animals especially in aquatic organisms are continuously increasing in recent years. There are several in vitro as well as in vivo studies to understand the mode of action of lectins in various plants and animals. They have biomedical, clinical, antimicrobial, and immune modulating applications. Different types of lectins have different mode of actions and regulations in different tissues. Researches showed that more than pattern recognition, lectins play a major role in immunity by regulating the glycan receptor binding through density levels. Different lectin types will have different mode of actions and regulations. In animals especially humans, the regulation part has been studied in a better way. However, very few studies have reported on the regulation of lectins in fishes and shellfishes. More researches have to be carried out in this regard and various genomic and feedback regulation through signalling has to be studied in detail.

Conflict of Interest The authors have no conflicts of interest to declare.

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