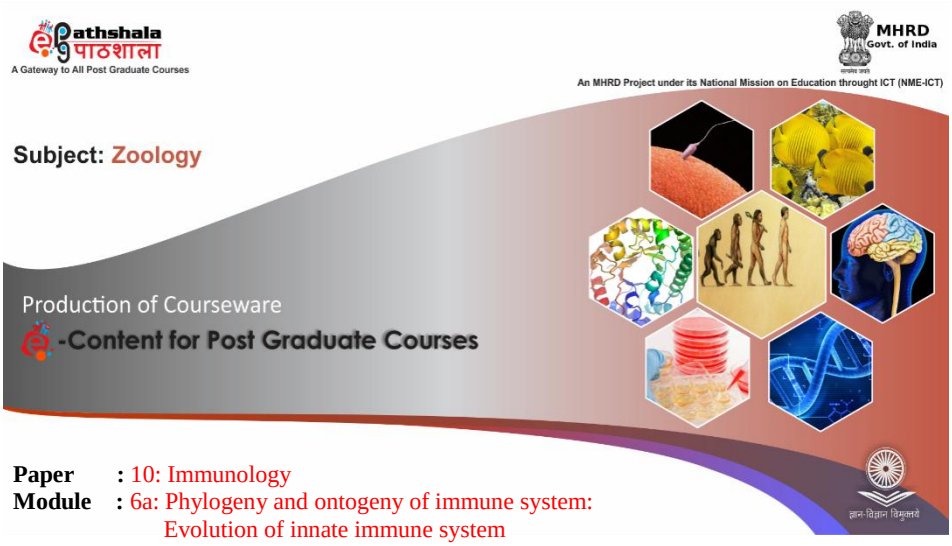

Paper : 10: Immunology
Module : 6a: Phylogeny and ontogeny of immune system: Evolution of innate immune system

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ZOOLOGY

Immunology

Phylogeny and ontogeny of immune system: Evolution of innate immune system

Description of Module	
Subject Name	ZOOLOGY
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Module ID	M6a: Evolution of innate immune system
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Contents

1. Learning Outcomes

i) All forms of life possess some kind of defense system

ii) Defense system needs to discriminate not only "self" from "non-self" but also defective, damaged and transformed "self" from normal cells.

iii) Basic components of innate immunity are shared not only by a number of invertebrates but also by all vertebrates and rely on germ-line encoded receptors which are evolutionarily conserved as they target molecular patterns which are integral part of various pathogens

iv) All cells of innate immunity express a large variety of PRRs (polyspecificity) and cells belonging to one class possess identical receptors. This is a striking difference between cells of the innate and adaptive immune branches as in the latter; each lymphocyte bears a unique receptor. As all the cells binding a given microbial pattern are able to respond without the need for clonal expansion, innate effector responses can occur very rapidly.

v) Adaptive immunity has not replaced innate immunity but continues to be present along with, and working in close association with innate branch to provide better protection to the organisms. Adaptive immune system requires signals from innate immunity for its activation.

vi) Building blocks of immune system are ancient and appeared and perfected at various stages during evolution due to selection pressure generated by dramatic environmental events on geological time scale, climatic changes and mass extinctions.

2. Introduction

All organisms are continuously exposed to pathogenic microorganisms (bacteria, viruses, fungi and parasites) but because of the powerful defense mechanisms, most of the organisms are capable of dealing with the onslaught of pathogens. Defense mechanisms have been shown to occur in all forms of life including bacteria, invertebrates and vertebrates. All cells have a definite life span after which they die. Therefore, an equally important job that the immune system must perform is to dispose of dying cells to prevent unnecessary inflammation and maintain homeostasis.

During the 20th century, pioneering discoveries by Jules Hoffmann, Bruce Beutler and Ralph Steinman among others led to the identification of the components of the immune system. Invertebrates have been shown to possess only the innate immunity, the first line of defense. That invertebrates make up more than 90% of all the earth's species, itself suggests that, even when considered primitive as compared to adaptive system, innate immunity is itself quite effective at providing adequate defense.

A clear understanding of various aspects of immune system in different organisms might help researchers discover certain mediators which may prove useful for human therapies.

3. Evolution of innate immunity

All forms of life have developed mechanisms to protect themselves from foreign intruders. Restriction enzymes and clustered regularly interspaced palindromic repeats (CRISPRs) are able to degrade invading foreign pathogens in prokaryotes. As life evolved into more complex forms from unicellular eukaryotic cells to multi-cellular organisms, additional defense mechanisms arose to maintain cellular integrity and host survival. Various components of innate immunity already exist in the organism without prior exposure to the pathogens, hence the name innate.

3.1 Cells of innate Immunity

In 1882, Elie Metchnikoff was the first to observe aggregation of cells around a thorn pushed into a starfish larva. He called these cells phagocytes. Phagocytosis, first appearing in amoebae for acquiring food, seems to have been retained in invertebrates and vertebrates where it plays a role in immune response. This property is due to cells which appear like macrophages and have been given different names in various groups such as amebocytes, hemocytes, coelomocytes, granulocytes, monocytes, macrophages. Advanced invertebrates and cephalochordates and urochordates possess both granulocyte-like cells and macrophages. Increased diversity of cell types is encountered in jawless vertebrates and gnathostomes. Resident populations of macrophages are present in most tissues.

Phagocytes, which functioned as digestive cells to begin with, retained their role to engulf the old, dying cells as well as the foreign microbial invaders. It was the observation of Metchnikoff that macrophages internalize apoptotic neutrophils at the end of inflammatory response that suggested a role for macrophages in the subsequent resolution of inflammation.

In 1975, a killer lymphocyte was discovered, which showed a rapid cytotoxic response against tumors and virus infected cells without prior sensitization and hence was named as **natural killer cell** (NK) by Eva Klein. This lymphocyte subset did not possess T- and B-cell receptors and was also referred to as null cell. Thus the natural question was: as these cells function like CD8+ T cells, should they be part of adaptive immunity or because they lack the receptors of T- and B-cells and respond to pathogen intruders almost instantaneously, should be included in innate immunity. Though the issue has not been resolved, it has been demonstrated by Weissman *et al* in 1997 that NK cells comprise a third lineage of lymphocytes, being derived from the same lymphocyte progenitor as T and B cells. More populations of "innate" lymphocytes have been reported e.g., **NKT cells, γδ T cells, CD8 αα T cells, mucosa-associated invariant T cells**, **B1 B cells** (phagocytic with bactericidal abilities) and **marginal zone B cells**, which require RAG recombinases for their development.

Since the discovery of dendritic cells in mice by Steinman and Cohn in 1973, not only a number of subsets have been described but also their presence has been reported from all vertebrates. These cells typically show gene expression profile such as TLR, MHC class II, B7 family, CD83 and CD209/DC-SIGN.

One study has reported that XL cells (with long cytoplasmic processes) from the spleen of Xenopus are capable of retaining unprocessed antigen at their plasma membrane and migrating into the white pulp following immunization. These XL cells have been proposed to be **primitive follicular DCs**. As shown by many studies, follicular DCs play an important role in humoral immune response by presenting native antigen, trapped in antigen-antibody complexes, to B cells. These cells are of non-hematopoietic lineage, different from both macrophages and conventional DCs.

XL cells of Xenopus also express high levels of MHC class II and are capable of presenting antigen to T cells, thus indicating their role in both humoral and cell mediated immune responses. DCs expressing MHC class II have also been identified in thymus and skin of Xenopus, indicating that *specialization of cells analogous to Langerhans cells occurred at the latest in amphibians*.

Other cells playing a role in innate immunity are **eosinophils, basophils and mast cells**. Leukocytes in teleost fish show morphological and functional similarity to mammalian macrophages, dendritic cells, neutrophils, monocytes, thrombocytes and natural killer-like cells. *However, presence of mast cells, basophils and eosinophils has not been unequivocally reported in teleost fish*. Eosinophils, basophils and mast cells have been implicated in cellular responses to helminth infection. Their evolution may have occurred to counter metazoan parasites (worms and parasitic arthropods) which are too large to be phagocytosed.

3.2. Receptor molecules of innate immunity

In order for the immune system to work efficiently in any organism, there is the need for discrimination between self and non-self which is achieved with the help of receptors. Receptors of innate immunity are encoded in the germ line and include a number of receptors as shown in table 1.

Even primitive organisms such as sponges show the presence of pattern recognition receptors (PRRs). These receptors have the specificity to bind pathogen associated molecular patterns (PAMPs).

The first family of PRRs to be discovered was Toll-like receptors (TLRs). In 1996 J. Hoffman and B. Lemaitre discovered that *Drosophila* fruit flies bearing mutations in toll gene were very susceptible to lethal infection with *Aspergillus fumigatus* as compared to wild type flies. This observation and other studies indicated that toll protein is required for activation of innate immune responses in invertebrates. In1997 C. Janeway and R. Medzhitov discovered a human gene for a protein similar to Toll which could activate the expression of genes of innate immunity in human cells. Therefore, this and other vertebrate Toll relatives which were discovered thereafter were named *Toll-like receptors (TLRs*). In addition to

TLRs, other PRRs were also discovered, as shown in the table, which play very important role in innate immune defense mechanisms.

Table 1. Properties of Pattern Recognition Receptors

PRR	**Localization**	**Ligands**	**Ligand Sources**
TLR Toll-like receptors	Plasma membrane	Lipoproteins, DNA, RNA, endotoxin, endogenous danger signals	Bacteria, viruses, parasites, altered self
NLR NOD-like receptors	Cytoplasm	Endogenous danger signals, muramyl dipeptides	Altered self, bacteria
CLR C-type lectin receptor	Plasma membrane	Beta-glucans	Fungi, mycobacteria, viruses, parasites and some allergens
RLR Retinoic acid-inducible gene-1-like receptors	Cytoplasm	Double-stranded RNAs	RNA viruses

Many invertebrate species contain very large families of innate receptors, for instance sea urchin genome codes for more than 200TLRs, nearly 300 Nod-like receptors and 180 scavenger receptors. Even though this diversity is less than that generated in adaptive immune responses, the possibility of further somatic variation has not yet been explored. 7 TLRs have been identified in most primitive fish, agnathans, 18 in bony fish, 14 in amphibians, 10 in birds and 13 in mammals. Decrease in number of innate receptors in gnathostomes may have occurred due to the presence of adaptive immune system.

C-type lectin receptor (**CLR**) is another family of cell surface PRRs. In humans there are at least 15 CLRs most of which recognize one or more specific sugar moieties such as mannose, fucose.

Retinoic acid-inducible gene-1-like receptors (**RLRs**) are soluble PRRs present in the cytoplasm of many cell types and function as sensors of infection by RNA viruses. These receptors are able to distinguish viral RNA from normal cellular RNA due to certain

structural features. Viral RNA shows double- stranded regions, virus- specific sequence motifs and RLR of the RIG-1 type identifies 5' triphosphate modification that arises during viral RNA synthesis and processing.

Nod-like receptors and nucleotide oligomerization domain/ leucine rich repeat-containing receptors (**NLRs**) represent another family of PRRs. They are present in the cytoplasm, detect damage and activate beneficial innate immune responses. Human genome contains approximately 23 NLR genes and mouse genome has about 34 NLR genes. Pattern recognition receptors (PRRs) may be categorized in two ways based on their function:

Signaling PRRs e.g., TLRs and NLRs;

Endocytic PRRs facilitate attachment, engulfment and destruction of microorganisms by phagocytes, without relaying an intracellular signal, e.g., mannose receptors of macrophages, glucan receptors on all phagocytes, scavenger receptors that recognize charged ligands and mediate removal of apoptotic cells.

Soluble pattern- recognition proteins

These proteins bind to conserved, repeating components on the surfaces of microbes. Once bound to microbe surfaces, these are recognized by membrane receptors on phagocytes, enhancing phagocytosis and thus are called **opsonins.** Many soluble proteins function as opsonins e.g., surfactant protein-A (SP-A), surfactant protein-D (SP-D), mannose-binding lectin (MBL), L-ficolin (binds to acetylated sugars), complement component C1q (binds bacterial cell wall component such as lipopolysaccharides), C-reactive protein (CRP) (binds to phosphorylcholine and carbohydrates on bacteria, fungi and parasites) as well as IgA antibody and some IgG subclasses.

Although recognition regions of MBL, Ficolins and C1q have different specificities, their polymeric structures are quite similar and so all are bound by CD91 opsonin receptor on phagocytes and promote phagocytosis of pathogen. CRP, IgG and IgA bind to Fc receptors on phagocytes facilitating phagocytosis of pathogen bound to them.

It is important to note that microbes of quite different biochemical composition and with entirely different life cycles are recognized by rather similar mechanisms by host PRRs. An equally important property of the innate recognition system is that each class of pathogen is identified by a number of different PRRs to ensure rapid and potent inflammatory response.

Lectins (a group of ubiquitous proteins, suggesting their ancient origin, found in bacteria, plants, invertebrates and vertebrates) bind to sugar molecules on cells, causing them to clump and get easily phagocytosed). Hemolin, binds to microbial surfaces enhancing their removal and RIG-like receptors (for binding viral RNA) have also been reported from forms more primitive than the earliest known vertebrates.

All of these represent pathogen-recognition receptors (PRR) capable of binding pathogen-associated molecular patterns (PAMP). These molecular patterns are conserved among various pathogens such as viruses, bacteria, fungi, protozoan and helminthes; they are intrinsic to these microorganisms and necessary for their function and existence. These patterns are not found in host cells.

All cells of innate immunity express a large variety of PRRs (polyspecificity) and cells belonging to one class possess identical receptors. This is a striking difference between cells of the innate and adaptive immune branches as in the latter; each lymphocyte bears a unique receptor. As all the cells binding a given microbial pattern are able to respond without the need for clonal expansion, innate effector responses can occur very rapidly.

3.3. Effector responses of innate immune system

Innate immune system works against infectious agents and injured tissues by

i) Producing antimicrobial peptides and proteins
ii) Phagocytosis
iii) Producing cytokines which play important role in inducing/controlling acute inflammation,
iv) Providing anti-viral state and,
v) Stimulating adaptive immune system in vertebrates.

Epithelial barriers like skin and mucus epithelial layers protect the body's interior from the microbial invasion not just by physical and mechanical means but also by actively producing a great variety of antimicrobial peptides and proteins (AMP). Some of the AMPs are lysozyme, lactoferrin, calprotectin, psoriasin, magainins, defensins, cathelicidins and surfactant proteins, serum amyloid-A (SAA) and serum amyloid-P (SAP).

AMPs, generally less than 100 amino acids long, represent an ancient form of innate immunity, as they are present in fungi, plants, invertebrates and vertebrates. Hultmark et al first characterized cecropin, an AMP, from Cecropia moths. In Drosophila, seven AMP families have been characterized such as Attacins, Cecropins, Diptericins, Drosomycins, Defensins, Drosocins, and Metchnikowins, in addition to having a number of immune inducible proteins in the hemolymph with antimicrobial properties.

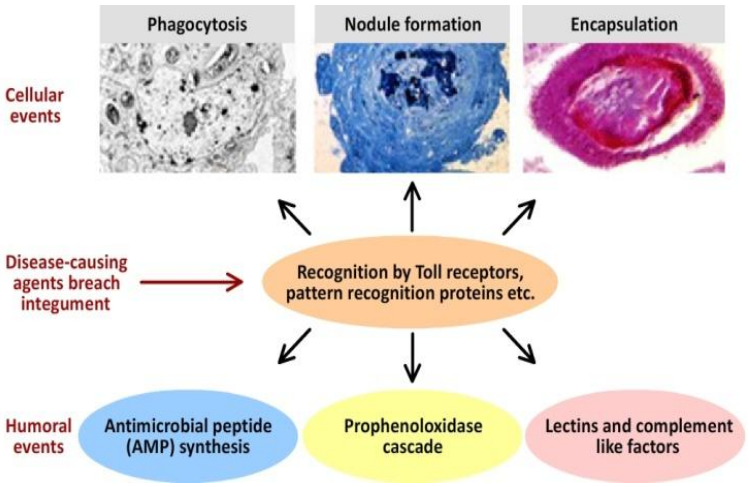

Fig.1. Arthropods show various mechanisms to deal with parasites and pathogens. PRRs recognize such organisms either free in the plasma or associated with various cell types. Phagocytes remove pathogens by phagocytosis. A sheath of cells removes large number of microorganisms from the hemocoel by forming a nodule. Larger invaders or damaged self-tissues are recognized and encapsulated by a multilayer of hemocytes. Antimicrobial peptides and phenoloxidase may kill microbes while lectins and complement-like molecules may play a role in recognition and elimination of pathogens.

A number of reports have shown that unicellular parasites such as *Plasmodium*, *Toxoplasma*, *Babesia*, and *Trypanosoma* can be taken care of by host-derived defensins.

Glands in frog skin secrete peptides called dermaseptins and magainins which have antimicrobial activity against bacteria, yeast and protozoans.

i) Phagocytosis

Even with strong defense mechanisms in place at the epithelial barriers, pathogens may gain entry into the interior due to wounds or insect bites. Next line of defense is made up by phagocytes. Phagocytosis is as old as the animal life itself. Upon insertion of a rose thorn into a larval starfish, Metchnikoff observed large phagocytes streaming towards the foreign body and eventually forming a syncytium around it. A similar sequence of events occurs upon injury or infection to mammalian tissue. Studies of Metchnikoff suggested that vertebrate leukocytes may have evolved from phagocytes of invertebrates.

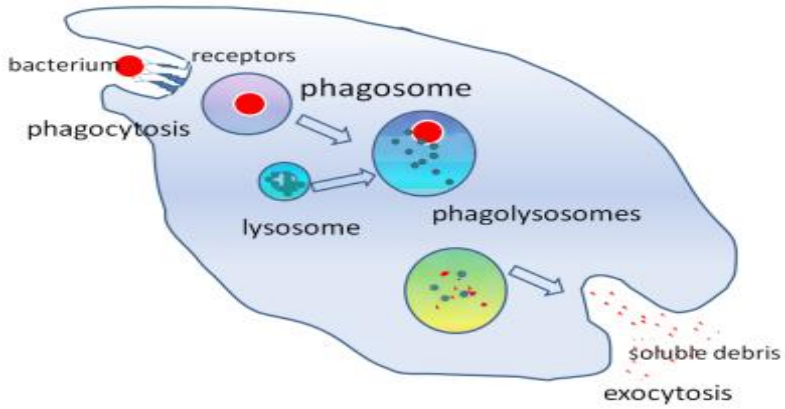

Fig. 2. Diagram showing the process of phagocytosis.
(Source: https://en.wikipedia.org/wiki/Phagosome)

When microbial organisms bind to phagocytes via PRRs or opsonins, signaling pathways get activated leading to polymerization of actin allowing membrane to extend around and internalize the microbe forming phagosome (Fig.2). This is followed by fusion of phagosome with lysosomes and in neutrophils with preformed primary and secondary granules. A number of antimicrobial agents attack and kill the pathogen. Phagolysosomes contain defensins, cathelicidins, low pH environment, a number of hydrolytic enzymes and specialized enzymes to generate **reactive oxygen species (ROS) and reactive nitrogen species (RNS**) as shown by sponges, gastropods, fish and several other organisms.

Inflammatory Responses

In response to tissue damage and resulting infection, innate immunity induces a cascade of events called inflammatory response. A parallel sequence of events has been reported in the invertebrate inflammatory response.

Invertebrate TNF-like molecule shows similarities to vertebrate TNF in size and activity. IL-6 like protein isolated from echinoderm coelomic fluids shows physicochemical properties and activity similar to vertebrate IL-6. Biochemical studies on phagocytosis mechanism in echinoderm amebocytes and vertebrate macrophages have revealed many functional homologies: microtubule inhibitor colchicine and inhibitor of microfilament organization, cytochalasin B inhibit phagocytosis by both; cycloheximide, inhibitor of protein synthesis, and fluoride, inhibitor of glycolysis, inhibit phagocytosis by both. Echinoderm coelomocytes could be activated to produce reactive nitrogen intermediate (RNI) in the presence of echinoderm IL-1, the same way as the mammalian phagocytes show increased activity of nitrogen oxide synthase upon activation.

Cytokine like molecules have been characterized from protostome and deuterostome invertebrates, tunicates and primitive vertebrates. *Thus, as we go up the phylogenetic ladder, we see evidence of a continuity of cytokines and conservation of inflammatory processes through evolution.*

Cytokine	Main cell source	Principal cellular targets and biologic effect	Animal groups
IL-1	Macrophage, endothelial cell, epithelial cell, dendritic cell, keratinocytes	Pro-inflammatory. Endothelial cell: activation (inflammation, coagulation); liver: synthesis of acute phase proteins; lymphocytes : enhances activity	Probably emerged earlier than vertebrates, and relatively conserved during evolution
IL-2	Th2	Pro-inflammatory. Stimulate T cell growth, augments NK cytolytic activity and induces differentiation of regulatory T cells	All vertebrate classes
IL-6	Macrophages, endothelial cells, T cells, epithelial cells, dendritic cells, NK cells	Pro-inflammatory. Liver: synthesis of acute phase proteins; Bone marrow : promotes hematopoiesis	All classes of vertebrates except cartilaginous fish and reptiles

Chemokines (IL-8)	Macrophages, endothelial cells, T cells, fibroblasts, platelets, dendritic cells	Leukocytes: chemotaxis, activation	All vertebrate classes
IL-12	Macrophage, dendritic cell	Pro-inflammatory. NK cells and T cells: IFN-gamma synthesis, increased cytolytic activity; T cells: Th1 differentiation	All vertebrate classes
IFN	NK cells, T cells	Pro-inflammatory. Activation of macrophages	All vertebrate classes
Type 1 IFNs	Virus infected cells. IFN-α: macrophages; IFN-β: fibroblasts	All cells: antiviral activity, increased MHC 1 expression; NK cell: activation	All vertebrate classes
TGF-β	All leukocytes including macrophages and stromal cells	Plays a crucial role in stem cell differentiation as well as T cell regulation and differentiation. Inhibits the function of inflammatory cells. Inhibits production of IL-2 required for activation of T cells, NK cells and other immune cells	All vertebrate classes (relative conservation of genes in even phylogenetically distant species because of their importance in other biological functions)
TNF-α	Macrophage, dendritic cell, mast cell, NK cell, epithelial cell	Pro-inflammatory. Endothelial cell and neutrophil: activation; hypothalamus: fever; many cell types: apoptosis; Liver: synthesis of acute phase proteins. Tumor : cytotoxic for many tumor cells	All vertebrate classes (evolutionarily conserved as it has non-immune as well as immune functions, like apoptosis, regeneration, tissue remodeling, metamorphosis.
IL-15	Macrophages, dendritic cells, fibroblasts, keratinocytes	Pro-inflammatory. NK cells and T cells : proliferation	Birds and mammals only
IL-18	Macrophages	NK cells and T cells : IFN- synthesis	Birds and mammals only
IL-4	Th2 cells	Anti-inflammatory. Inhibits proliferation of Th1, Tc and NK cells	Mammals only
IL-10	Macrophages, dendritic and mast cells, NK, T, B cells	Anti-inflammatory. Macrophages: inhibition of IL-12 production, reduced expression of co-stimulators and class II MHC molecules	Mammals only, (small no. of studies indicate their presence in birds)

Table 2. Cytokines of Innate Immunity

Thus it is clear from this table that during vertebrate evolution number of cytokines increased dramatically. Mammals show the largest number of cytokines (especially anti-inflammatory)

in addition to the expanded network of cytokines controlling killer machinery of adaptive immunity. *Selection pressures of auto-immunity and reproductive efficacy might have led to evolution of this pattern of cytokine repertoire*. Feto-placental unit in mammals represents a situation similar to a successful transplant. This is achieved by a specific cytokine network built up by NK cells, dendritic cells, lymphocytes, neutrophils, macrophages and other decidual cells in the endometrium. Th2 cells and Th2 type of cytokines (IL-4, IL-6, and IL-10) predominate in normal decidua, inhibiting proliferation of Th1 and cytotoxic cells, Tc and NK cells. If Th1 cell activation occurs, there is release of pro-inflammatory cytokines such as IL-1, IL-2, IL-6, IL-12, IL-15, IL-18, and TNF-α and IFN-γ which increase permeability of blood vessels allowing access of immune cells which can reject the placenta as an allotransplant.

Like NK cells, γδ T cells, function in innate immunity as they recognize antigen directly, independently of MHC class I and class II molecules but function in a specific fashion, exhibiting memory. It has been proposed that γδ TCR-like receptor may have appeared before the rearranging antigen-binding receptors. These cells are present in large numbers at the mucosal surfaces and mount immune response to various infective organisms entering via the oral/respiratory route.

γδ T cells have been implicated in a number of functions such as secretion of a number of cytokines (which can regulate functions of other immune cells), providing help to naive B cells to produce IgM, IgG and IgA antibodies, enabling macrophage recruitment and upon activation exhibit cytotoxic activity against many tumor cells using Fas/Fas-ligand-dependent and perforin/granzyme- or granulysin-dependent pathways.

The **complement system**, a bridge between innate and adaptive branches of immune system in vertebrates, has been postulated to have been rather very ancient. C3, a central component and Bf have been proposed to constitute the primitive complement system, having emerged more than 1300 mya. They are similar to mammalian alternative pathway and have been retained by deuterostomes. Next evolutionary step of complement system occurred when chordates appeared (900mya) with the recruitment of mannose binding lectin (MBL), MBL-associated serine protease (MASP), and ficolin genes, which established the lectin pathway.

At this stage, lectins and alternative pathways might have played a role in opsonization and induction of inflammation. Final change involved complement gene duplications in the vertebrate lineage. Multiplication of C3 gave rise to C3, C4 and C5 before the cartilaginous fish emerged since sharks have all the three genes and such as those between Bf/C2 and MASP/C1r/s about 600mya, before the appearance of cartilaginous fish and probably contributed to classical pathway of complement activation. Components C6/C7/C8/C9 (membrane attack components) might have appeared due to duplication of ancestral TCC genes before the jawed vertebrates appeared.

	Sea urchin	Ascidian	Lamprey	Shark	Carp	Frog	Snake	Chicken	Mammal
Classical pathway				+	+	+	+	+	+
C3 and factor B	+	+	+	+	+	+	+	+	+
MBL		+			+			+	+
MASP1		+				+	+	+	+
MASP2						+	+	+	+
MASP3			+	+	+	+	+	+	+
Ficolins		+				+		+	+

Table 3. Evolution of different component of complement system across various animals.C3 and C2/factor-B-like sequence have been identified in sea urchin, bearing similarity to alternative pathway of complement activation. Lectin pathway of complement activation evolves in ascidians with such molecules as GBL (homologous to MBL), ficolins, MASPs, C3 and C3 receptor. All three pathways of complement activation are seen in jawed vertebrates. GBL, glucose-binding lectin; MBL, mannose-binding lectin; MASP, MBL-associated serine protease.

Higher vertebrate host organisms show different **pathways of macrophage function known as M1 andM2**. In the M1 pathway, arginine can be converted to nitric oxide (NO) by inducible nitric oxide synthase (iNOS) enabling macrophage to fight and kill pathogenic organisms. In the M2 pathway, also known as repair pathway, arginine is converted by arginase to ornithine and urea. Cytokines secreted by M1 and M2 cells are different. In vertebrates, macrophages have acquired MHC II molecules thus enabling them to play a role in adaptive immune response by antigen presentation to T lymphocytes.

Plasmacytoid dendritic cells (DCs), mainly found circulating in blood, have been shown to play a role in innate immunity by producing immune- enhancing cytokines, interferons, which mobilize innate lymphocytes (NK, NK-T, γδ T) and help fight against viral and intracellular infections. As classical DCs can capture and process the antigen effectively, migrate to lymphoid tissues and present the antigen to T lymphocytes for their activation, DCs bridge innate branch with adaptive branch of immunity.

Mast cells, basophils and eosinophils also play important role in innate immunity. Eosinophils, mostly localized to mucosal surfaces, are able to respond rapidly to pathogens and bridge innate and adaptive immunity within secondary lymphoid tissues. They respond to helminth infections and allergens.

Exposure of Drosophila to bacteria or fungi results in transcription of a number of genes resulting in antimicrobial peptides. Fungi and Gram-positive bacteria lead to the activation of Toll pathway, very similar to Toll-like receptor pathway of mammals. Gram-negative bacteria activate IMD pathway, very similar to TNF-receptor pathway of mammals. (Hoffman, Strasbourg). These studies suggest that *there may be an evolutionary link between the regulations of anti-microbial peptide (AMP) gene expressions in flies and the mammalian innate immune response. (Hoffmann & Reichhart, 2002)*

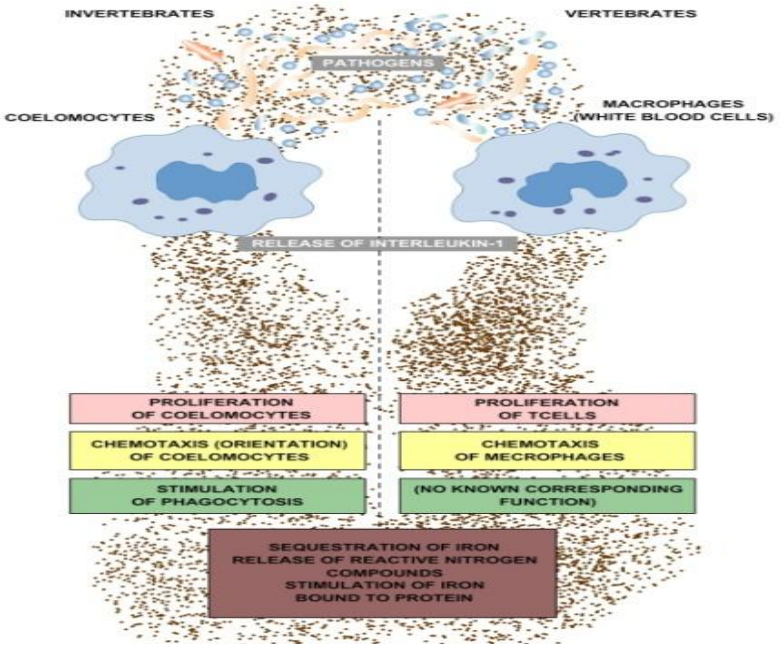

Fig 2. : Functions stimulated by cytokines are quite similar in invertebrates and vertebrates and help in host defense.

4. Applications

- Controlling disease in commercially important species. 25% of the global production of shrimp is lost due to disease. There is a need to understand the immune defenses of commercially important species. Viruses might have been responsible for the disease and attempts have been made to develop vaccines against viral pathogen is suggestive of the fact specific or primed immunity exists in insects and crustaceans and it can be transferred from parents to offspring.

- Since many effector molecules seem to be shared by a large number of organisms, they might prove to be of therapeutic value.

- Dermaseptins S (isolated from the skin of South American frog) are polycationic peptides and inhibit a number of microbial cells including *N. gonorrhea, Candida albicans, Plasmodium falciparum,* rapidly and efficiently without any toxic effect on mammalian cells.

- *Limulus* amebocyte lysate provides the most sensitive and specific method for detection of bacterial endotoxin in pharmaceuticals and drugs intended for human use.

- The complement of Alligator mississippiensis has been shown to have antiviral activity against human immunodeficiency virus type 1, West Nile virus and herpes simplex virus. Various species of Naegleria and Acanthamoeba that have been found to be resistant to lysis by human complement show lysis by alligator complement.

- If parasite structures targeted by IgE are characterized, we may be able to identify homologous molecules and potential allergens in genetically modified organisms and new food items.

- Many studies point to the possible use of δγ T cells for antitumor therapies (colon & pancreatic carcinoma, acute lymphoblastic leukemia) as well as anti-viral (influenza, HIV, HCV) and anti-bacterial infections (tuberculosis, salmonellosis).

5. Summary

The innate immunity, though non-specific and without memory, is present in both invertebrates and vertebrates. All cells of innate immunity express a large variety of PRRs (polyspecificity) and cells belonging to one class possess identical receptors. This is a striking difference between cells of the innate and adaptive immune branches as in the latter; each lymphocyte bears a unique receptor. As all the cells binding a given microbial pattern are able to respond without the need for clonal expansion, innate effector responses can occur very rapidly.

It provides not only the first line of defense in containing the infection and conferring antiviral defense but also stimulates adaptive immunity in jawed vertebrates. Research from a number of laboratories suggests the possibility of at least some of the molecules of adaptive immunity having arisen from structurally similar precursors serving related innate immune functions.

For instance, complement cascade exemplifies gradual evolution of related molecules from innate (MASP) to adaptive (C1) function and from non-immune (α2M) to immune (C3 and C4) function. MHC molecules, though play important role in active immunity by presenting antigenic fragments to T lymphocytes, also have many functions in innate immunity.

From fish to mammals, we can see the presence of cytokines playing important roles in haematopoiesis, inflammation and cytokine network seem to be conserved throughout vertebrates. During vertebrate evolution number of cytokines increased dramatically. Mammals show the largest number of cytokines (especially anti-inflammatory) in addition to the expanded network of cytokines controlling killer machinery of adaptive immunity.

Selection pressures of auto-immunity and reproductive efficacy might have led to evolution of this pattern of cytokine repertoire.

Comparative studies of innate immune system among different organisms provide us with many possible applications.