How do primitive metazoans survive without an acquired immune system?

How important is innate immunity for higher metazoans?

### Mechanisms of Generating Immunological Diversity in Primitive Metazoans

<table>
<thead>
<tr>
<th>Taxonomic group</th>
<th>Genetic variability (polymorphisms, etc.)</th>
<th>Somatic variability</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nematodes</td>
<td>Lectins</td>
<td>Variable lymphocyte receptor</td>
</tr>
<tr>
<td>Molusks</td>
<td>Fibronectin-related proteins (immunglobulin SF), lectins</td>
<td>Variable lymphocyte receptor</td>
</tr>
<tr>
<td>Arthropods</td>
<td>Fibrinogen-related proteins (immunglobulin SF), lectins</td>
<td>Variable lymphocyte receptor</td>
</tr>
<tr>
<td>Echinoderms</td>
<td>Surrogate receptor system rich (b), (c)</td>
<td>Variable lymphocyte receptor</td>
</tr>
<tr>
<td>Prochordates</td>
<td>Variable region-containing chain binding protein (immunglobulin SF), lectins</td>
<td>Variable lymphocyte receptor</td>
</tr>
<tr>
<td>Agnathans</td>
<td>Variable region-containing chain binding protein (immunglobulin SF), lectins</td>
<td>Variable lymphocyte receptor</td>
</tr>
<tr>
<td>Gnathostomes</td>
<td>Many immunoglobulin SF multigene families (b), Toll-like receptor, lectins</td>
<td>Variable lymphocyte receptor</td>
</tr>
</tbody>
</table>

Red, membrane-associated; Black, soluble

From: Du Pasquier Science 309:1826, 2005
Dscam, a Secreted Opsonic Protein with Extensive Alternative Splicing in *Drosophila*

From: Watson et al., *Science* 309:1874, 2005

The Innate Immune Response to Bacterial and Fungal Infections
The Innate Immune Response is Conserved Throughout Evolution and is Triggered by Pattern Recognition

Lipopolysaccharide is Composed of Lipid and Polysaccharide

Collectins and Innate Immune Recognition

Some Functions of Collectins

The Scavenger Receptor Superfamily

KEY TO DOMAINS
- Complement control protein (CCP)
- Somatomedin B
- C-type Lectin
- Epidermal growth factor (EGF)
- Partial Epidermal growth factor (EGF)
- Potential N-linked glycosylation
- Potential O-linked glycosylation

Receptors Important in The Systemic Response to Infection

Local infection with gram-negative bacteria
Macrophages activated to secrete TNF-α in the tissue
Increased release of plasma proteins, hypoproteinemia, and lymphocyte migration into tissue
Increased platelet adhesion to blood vessel wall

Systemic infection with gram-negative bacteria (sepsis)
Macrophages activated in the liver and spleen secrete TNF-α into the bloodstream
Systemic edema causing decreased blood volume, hypoproteinemia, and neutropenia, followed by neutrophilia
Decreased blood volume causes collapse of vessels
History of Endotoxin Research

The post-microbial era began with the discoveries of Koch and Pasteur (1865). Four phases of discovery are depicted: the recognition that infection is "poisonous" (red); search for poisons culminating in the identification of endotoxin (green); the chemical and biological characterization of endotoxin (orange); the identification of the endotoxin receptor and its role in promoting the immune response (purple).

A human homologue of the Drosophila Toll protein signals activation of adaptive immunity

Section 1

Endotoxin = LPS

C3H/HeJ mutation reported

TNF mediates LPS-induced shock

Discovery of CD14

LPS = TLR4

Pasteur, Koch: the germ theory of disease

Pfeiffer describes "endotoxin"

Primary structure of LPS reported

The post-microbial era began with the discovery of Koch and Pasteur (1865). Four phases of discovery are depicted: the recognition that infection is "poisonous" (red); search for poisons culminating in the identification of endotoxin (green); the chemical and biological characterisation of endotoxin (orange); the identification of the endotoxin receptor and its role in promoting the immune response (purple).

Use of adjuvant to stimulate the immune response

"Infectious-non-self" model of immunity described by Janeway

A Re-interpretation of the Endotoxin Research Timeline

Dryad

TLR Signaling: Not So Simple

Cartoon of major signal transduction pathways following engagement of TLRs. TLR4 is the major sensor of LPS. TLR3 recognizes dsRNA and is important in the anti-viral response. The IRF pathway leading to production of Type I IFNs (i.e., IFN-α/β) is particularly prominent in a minor subset of dendritic cells (called "plasmacytoid DCs") that are the major source of these IFNs in response to viral infections. Do not memorize this cascade but rather appreciate that it consists of two parallel pathways, one that activates NF-κB, leading to production of pro-inflammatory proteins, and one that activates the IRF pathway, leading to production of Type I IFNs.

Selected Pro-inflammatory TLR Signaling Components

Vertebrates

Drosophila

TIR = Toll/IL-1 receptor
DD = Death domain
IKK = IκB kinase

From: Moynagh, Trends Immunol. 20:469, 2005
Positive Feedback Loops in Innate Immunity

The existence of multiple, overlapping positive feedback loops helps explain why targeting any one pro-inflammatory mediator is typically ineffective in the treatment of severe, systemic inflammation (e.g., during Gram-negative septicemia).

From Werner et al., Science 309:1826, 2005

Primitive Specificity in Target Recognition by the Innate Immune System
Recruitment of TLR2 to Yeast Phagosomes


Specificity of TLR Transcriptional Programs

Newer Innate Immune Proteins

NOD Proteins: Intracellular Peptidoglycan Sensors

Polymorphisms in Nod-2 are associated with up to 30-40% of cases of Crohn’s disease (an inflammatory bowel disease)

CARD, caspase-recruitment domain; LRR, leucine-rich repeat; RICK, a CARD-containing protein kinase
Mutations in Pyrin, Another CARD-containing Innate Immune-like Protein, is Responsible for Familial Mediterranean Fever

Contrast-enhanced abdominal CT from a 31 year-old patient with Familial Mediterranean Fever suffering an acute attack of abdominal pain, nausea, vomiting, and arthritis. Note mesenteric vessel with thickened mesenteric fold (white arrow). Histopathology demonstrated neutrophilic infiltrate and associated vasculitis. Treatment with an IL-1 receptor antagonist (Anakinra) resulted in prompt cessation of symptoms.

The Dendritic Cell and Development of The Primary Immune Response
Dendritic Cell Maturation


The Innate Immune Response Orchestrates DC Trafficking to Secondary Lymphoid Organs

Functional Differences Between Immature and Mature DCs

<table>
<thead>
<tr>
<th>Principal Function</th>
<th>Immature DCs</th>
<th>Mature DCs</th>
</tr>
</thead>
<tbody>
<tr>
<td>Antigen capture</td>
<td>++</td>
<td>--</td>
</tr>
<tr>
<td>Antigen presentation</td>
<td>+</td>
<td>++</td>
</tr>
<tr>
<td>Expression of Peyer patch receptors</td>
<td>++</td>
<td>+</td>
</tr>
<tr>
<td>Expression of molecule triggering T cell activation</td>
<td>++</td>
<td>--</td>
</tr>
<tr>
<td>Class II MHC molecule</td>
<td>++</td>
<td>++</td>
</tr>
<tr>
<td>Half-life on surface</td>
<td>12 hr</td>
<td>&gt;48 hr</td>
</tr>
<tr>
<td>Percentage of surface molecules</td>
<td>&gt;9%</td>
<td>&gt;95%</td>
</tr>
</tbody>
</table>

The (Primary) Acquired Immune Response is Initiated by Innate Immune Recognition
Chemokines Direct Trafficking of Immune Cells


The Early Antiviral Response:
Cytokines of the Innate Immune System

Fig 2.41 © 2001 Garland Science
The Antiviral Response: a Cascade of Transcriptional Events

Multiphasic induction of murine type I IFN genes can be divided into three phases. (a) The immediate early phase. Virus infection stimulates a phosphorylation cascade, leading to the activation of at least three families of transcription factors, including NF-κB, AP-1 and IRF3. Activation of the IFN-α promoter requires all three transcription factors. (b) IRF7 induction phase. Secretion of early IFN produces an autocrine response through stimulation of the JAK-STAT pathway. Among the pathway’s target genes is IRF7, itself. (c) Delayed early (amplification) phase. Many members of the IFN-α gene family possess promoter binding sites for activated IRF7 and become transcriptionally active.

Some targets of IRFs

<table>
<thead>
<tr>
<th>Gene</th>
<th>Function</th>
</tr>
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<tbody>
<tr>
<td>p21</td>
<td>Cell cycle arrest</td>
</tr>
<tr>
<td>IL-15</td>
<td>NK cell maturation</td>
</tr>
<tr>
<td>FasL</td>
<td>Cell death</td>
</tr>
<tr>
<td>IL-12</td>
<td>Th1 immune response</td>
</tr>
</tbody>
</table>

NK Cells are an Important Early Source of IFN-γ

Summary

1. Innate immunity is conserved throughout evolution and is triggered by recognition of repetitive molecular patterns (e.g., LPS) by “pattern recognition receptors.”

2. Collectins (e.g., SP-A, C1q, MBP) recognize carbohydrates on pathogen surfaces and perform multiple anti-microbial functions (e.g., opsonization). Collectins are essential for innate immunity, but also help clear apoptotic debris.

3. Members of the Scavenger Receptor superfamily recognize bacteria as well as glucose-modified proteins (AGEs) and oxidized lipoproteins. They are implicated in the response to infection as well as atherosclerosis and other degenerative diseases.

4. TLR4 is the major LPS receptor in mammalian cells. Via engagement of a series of adaptor proteins and kinases, it triggers activation of NF-κB (leading to production of TNF-α, for example) and the IRF pathway (and production of IFN-α/β).

5. Dendritic cells undergo a maturation program: immature DCs, which traffic to the periphery, capture antigen, and mature DCs, which traffic to the lymph node, present antigen.

6. NK cells, a component of innate immunity, especially to viruses, represent an early source of IFN-γ which serves to stimulate macrophages in inflammatory sites.