Intrinsic and innate defenses

Lecture 13
Biology 4310
Virology
Spring 2021

The trouble with facts is that there are so many of them
–Anonymous
Host defenses

Anatomical and chemical barriers  Intrinsic  Innate immunity  Acquired immunity

CONTINUOUS  IMMEDIATE  MINUTES/HOURS  HOURS/DAYS

Physical Barriers  Intrinsic  Innate  Adaptive

- Mucus
- Saliva
- Stomach acid
- Tears
- Skin
- Scabs
- Defensins
- Interferons
- Autophagy
- Apoptosis
- MicroRNAs
- CRISPRs
- Natural killer cells
- Complement
- Antigen-presenting cells
- Neutrophils
- Cytokines
- T cells
- B cells
Host defenses

- Intrinsic
  - *Always present* in the uninfected cell
  - Apoptosis, autophagy, RNA silencing, antiviral proteins
- Innate immune system: *Induced* by infection
- Adaptive immune system: *Tailored* to pathogen
RNA interference

Plant & invertebrate cells
Mammals - present or not needed?

Countermeasures!
APOBEC3 and HIV-1

(Apolipoprotein B mRNA editing catalytic polypeptide)
Epigenetic silencing

Countermeasures encoded in many viral genomes

**HCMV** pp71 causes degradation of cell Daxx, needed for histone deacetylation

**EBV Ebna5, Ad E4 Orf3** affect Pml protein localization or synthesis

Unintegrated but not integrated retroviral DNA is silenced
Apoptosis is monitored by sentinel cells
Viral regulators of apoptosis

HIV, HBV, HCV, VZV, EBV, RSV, ZIKV, HPV, Influenza virus, Reovirus

HCV

Bax
Bak

BCL-2
BCL-XL

Cytochrome c

HIV, HBV, WNV, ZIKV

HSV-1, HSV-2, HCMV, EBV, HCV, HPV, HIV-1, VACV

HCMV, MCV, EHV-2, EBV, HCV, HPV, ADV

FasL
Fas

FADD

DISC
pro-caspase-8

TNF
TNF

FADD
TRADD
RIP1
TRAF2

TRAIL
TRAIL-R

FADD

DISC
pro-caspase-8

HCV

Caspase-8/10

Caspase-9

Caspase-3/7

Apoptosis
**Ancient intrinsic defense: CRISPR**

*Clustered regularly interspaced short palindromic repeats*

- 90% of Archaea
- 50% of Bacteria
Intrinsic defenses are always present. Which of the following are included?

A. Antibodies
B. T cells
C. Epigenetic silencing
D. Skin
E. Mucus
- Activated within minutes to hours after infection
- Cytokines, sentinel cells (dendritic cells, macrophages, NK cells), complement
- Can inform adaptive response when infection reaches dangerous threshold
How does the innate system recognize microbes and not self?


- 1996: Toll found to have a role in immunity of fly to microbes

- 1997: Toll-like receptors identified in mammals
C/N/R/TLRs - Pattern recognition receptors

**c-type lectin receptors (CLRs)**
- Transmembrane proteins localized at the plasma membrane
- Recognize glycans from the wall of fungi and some bacteria

**Toll-like receptors (TLRs)**
- Transmembrane proteins localized either at the plasma membrane or in endosomes
- Broad range of specificities recognizing proteins, nucleic acids, and glycans

**Nucleotide binding oligomerization domain-like receptors (NLRs)**
- Cytoplasmic sensors
- Multiple subfamilies:
  - NLRs recognize bacterial, viral, parasitic and fungal PAMPs
  - AIM2 detects viral and bacterial DNA
  - Form multiprotein signaling complexes known as inflammasomes

**RIG-I-like receptor receptors (RLRs)**
- Cytoplasmic sensors of viral RNA
- Signal via the mitochondrial adaptor protein MAVS
- Trigger antiviral responses including the production of type 1 interferon

PAMP = Pathogen associated molecular pattern
Recognition of PAMPS

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Principles of Virology, ASM Press
Sensing DNA

DNA virus → DNA in cytoplasm → cGAS → cGAMP(2'-5') → STING → TBK1 → IκK → IRF3 → Transcription of proinflammatory genes → Nucleus

ATP → GTP

AMP → GMP

Principles of Virology, ASM Press
Viral modulators of sensing
Which of the following allow the innate immune system to distinguish microbes from self?

A. Cytoplasmic helicases and TLRs  
B. Antibodies  
C. Apoptosis  
D. Apobec  
E. All of the above
Interferons

- 1957: Issacs & Lindenmann; chicken cells exposed to non-infectious influenza virus produce substance that “interfered” with infection of other cells
- Produced by virus-infected cells and uninfected sentinel cells in response to products released from cells (e.g. viral nucleic acid)
Interferons

**Type I: IFN-αs**
- (13 subtypes)

**IFN-β**
- (1 type)

**IFN-α**
- IFNAR1

**IFNAR2**

**Type II: IFN-γ**
- IFN-γ
- IFNGR1
- IFNGR2

**Type III: IFN-λs**
- (3 subtypes)

**IFN-λ**
- IL10R2
- IFNLR1

**JAK1**
- TYK2

**JAK1**
- JAK2

**JAK1**
- TYK2

**JAK1**
- JAK2
Production of IFNα/β is rapid: within hours of infection, declines by 10 h

IFN binding to IFN receptors leads to synthesis of >1000 cell proteins (ISGs, IFN stimulated genes)

Mechanisms of most ISGs not known
Endogenous retrovirus LTRs regulate the interferon response

LTRs contain motifs that respond to innate immune signals

Highly lineage-specific and occupy 6-14% of mammalian genomes

TWiV 382: Everyone’s a little bit viral
microbe.tv/twiv/twiv-382
Tetherin, CD137

HIV-1 Nef protein is a tetherin antagonist
Interferon-induced proteins: IFIT1

IFN-induced protein with tetratricopeptide repeats 1

Ifit1 binds RNAs lacking 2'-O methylation

http://www.pnas.org/content/early/2017/02/28/1612444114.short?rss=1
Escape from IFIT1

**Cap snatching**
Influenza virus

m7GpppNm

**5’end—Independent translation**
Picornavirus

**Viral N-7 and 2′-O methylase**
- Paramyxovirus
- Rhabdovirus
- Flavivirus
- Reovirus
- Poxvirus
- m7GpppNm

**Host N-7 and 2′-O methylase**
- Polyomavirus
- Herpesvirus
- Parvovirus
- Retrovirus

**RNA structure**
Alphavirus

m7GpppN

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Principles of Virology, ASM Press
Interferon-induced proteins: IFITM3

Inhibition of fusion during virus entry
The IFN system is dangerous

- IFN induces the expression of many deleterious gene products - most of our cells have IFN receptors
- IFNs have dramatic physiological consequences: fever, chills, nausea, malaise
- *Every viral infection results in IFN production*, one reason why ‘flu-like’ symptoms are so common
How do interferons (IFNs) limit viral replication?

A. IFNs directly inhibit viral translation
B. IFNs lyse viral particles
C. IFNs induce ISGs
D. IFNs damage cells
E. None of the above
Sentinel cells

- Dendritic cells, macrophages, natural killer (NK) cells
- They patrol all our tissues looking for signs of change
Dendritic Cells

Intestine

Virus

Skin

Eye

Vagina

Epithelia

Blood DC
CD8α+ DC
CD4+ DC
CD11b-CD8α+ DC
Plasmacytoid DC

Tissue DC
Langerhans cell
Dermal/submucosal DC
CD103+CD11b+ DC
CX3CR1+ CD11b+ mononuclear phagocyte

Lymph node

Afferent lymph

B cell zone

T cell zone

CCR7↑

CCR7↑

VEIN

ARTERY
DCs

Virus binds to epithelial cells

Replication and local spread of infection

Spread to lymphatics

Adaptive immunity takes over

Cytokines

Immature dendritic cell

Mature dendritic cell

Lymph node

Also activated by viral proteins/nucleic acids released from infected cells

Circulatory system

CTL

Antibodies

Principles of Virology, ASM Press
Yes, there are viral modulators of NK cells.
Complement

Yes, there are viral modulators
Infection leads to the inflammatory response

- Infected cells produce cytokines & chemokines
- Redness; pain; heat; swelling, the four classic signs of inflammation (rubor, dolor, calor, tumor, originally recorded by the Roman medical encyclopedist Celsus in the first century AD)
- Increased blood flow, increased capillary permeability, influx of phagocytic cells, tissue damage
Initially function locally in antiviral defense
In larger quantities, enter circulation, have global effects (sleepiness, lethargy, muscle pain, no appetite, nausea)

Three classes of cytokines

<table>
<thead>
<tr>
<th>Group</th>
<th>Some members</th>
<th>Activity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Proinflammatory</td>
<td>IL-1, Tnf, IL-6, IL-12</td>
<td>Promote leukocyte activation</td>
</tr>
<tr>
<td>Antiinflammatory</td>
<td>IL-10, IL-4, Tgf-β</td>
<td>Suppress PICs</td>
</tr>
<tr>
<td>Chemokines</td>
<td>IL-8</td>
<td>Recruit immune cells</td>
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</table>

A localized viral infection produces global effects
## Viral Cytokine Countermeasures

<table>
<thead>
<tr>
<th>Interrupt cytokine production</th>
<th>Interfere with cytokine action</th>
<th>Interfere with cytokine effector function</th>
</tr>
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<tbody>
<tr>
<td>Interfere with cytokine and chemokine synthesis</td>
<td>Encode homologs of cytokines to block receptors</td>
<td>Alter cytokine signaling pathway</td>
</tr>
<tr>
<td>Inhibit generation of functional cytokines</td>
<td>Encode soluble cytokine receptors to neutralize cytokines</td>
<td></td>
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</tbody>
</table>

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Inflammation usually stimulates potent immune responses

- Cytopathic viruses cause inflammation because they promote cell and tissue damage
  - *Activate the innate response*

- Consequently cytopathic viral genomes encode proteins that modulate this immune response
  - *Adenoviruses, herpesviruses, poxviruses*
Some viruses do not stimulate inflammation

- Typically non-cytopathic viruses
  - Cells are not damaged, no apoptosis/necrosis
  - Low or ineffective innate immune response
  - Do not effectively activate adaptive immune response

- Non-cytopathic viruses have dramatically different interactions with the host immune system
  - Persistent infections: rarely or inefficiently cleared
The lesson

- The classic inflammatory response (heat, swelling, redness, pain) reflects the communication of innate and adaptive immune defense
  - No inflammatory response, ineffective adaptive response
- One reason for using inflammation-stimulating adjuvants for noninfectious vaccines
Not all inflammation is caused by infection!

An important component of smallpox vaccine efficacy!
Viral countermeasures

All viruses must encode at least one regulator of intrinsic/innate defenses

Sensing, IFN production, IFN signal transduction, cytokines, chemokines, NK cells, DCs, complement
Next time: Adaptive immunity