

Filoviridae & the Ebola Virus

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Lecture Overview

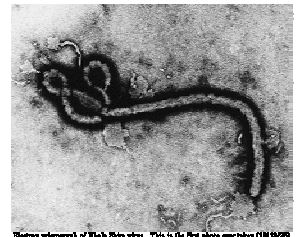
- Structure
- Virus Replication Cycle
 - Protein Synthesis
- Pathogenesis

Filoviridae Family

- *Filo*: from latin meaning *threadlike*
- Structurally & Genetically similar to Rhabdoviridae and Paramyxoviridae
- Two Genera:
 - Marburg-like virus
 - Ebola-like virus

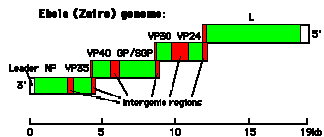
Structure

- Pleimorphic, Filamentous
- Striated
- 80nm diameter
- 130-140,000nm long
- Enveloped



Electron micrograph of Ebola Virus. This is the first photo ever taken (1976) by Dr. T.A. Berger, now at UC Davis, Calif. CDC. Disputed specimen in full culture at 100,000X magnification.

Genome



- Negative Sense ssRNA
- Unsegmented
- 7 proteins
- Gene overlap

Ebola Virus Proteins

- GP- Transmembrane glycoprotein
- NP- Nucleoprotein necessary for capsid assembly
- VP24- Anti-viral inhibitor?
- VP35- Inhibits IFN production
- VP30- Transcription anti-terminator
- VP40- necessary for capsid assembly and budding
- L- Viral Polymerase

Replication Cycle

- 1 Host Entry²
 - Contact with infected bodily fluids
 - Enters through mucous membrane or directly into blood (needle stick)
 - No confirmed spreading of virus by aerosol in nature

Replication cont.

- 2 Adsorption- Glycoprotein (GP₁) binds cellular receptor³
 - Mediated by cellular cofactors. i.e. folate receptor α
 - Mononuclear phagocytic cells & monocytes are primary targets⁴
- 3 Endocytosis
 - pH lowering in endosome
 - GP₂ mediates membrane fusion; release of viral particle into cytoplasm

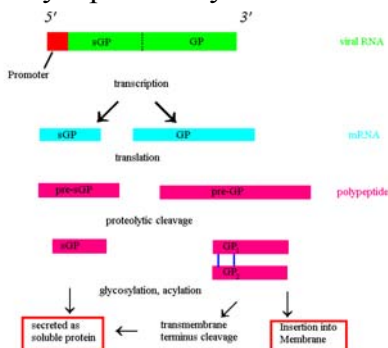
Replication Cont.

- 4 Protein Synthesis
 - requires viral polymerase (L)
 - VP30 anti-terminator allows transcription of genes downstream from first gene (Ebola Only)
 - VP35 prevents anti-viral response to dsRNA⁵
- GP synthesis¹
- Complex post-translational processing
 - O-linked/N-linked glycosylation
 - proteolytic cleaving by proteases
 - acylation

Ebola Glycoprotein

- GP1/GP2- transmembrane protein
 - Binding/Fusion
- sGP- truncated soluble protein (Ebola Only)
 - Secreted
 - Decoy for immune system?
- Give rise to neutralizing & protective antibody

Glycoprotein Synthesis



Replication cont.

- 5 Virus Assembly & release - Exact mechanism not known
 - NP essential for RNA packaging
 - VP40 essential for Budding at membrane
 - VP40 and GP give filamentous morphology

Complete Virion contains:

ssRNA, NP, VP35, VP24, L, VP30 (Ebola Only)

Ebola Virus: Interactions With Immune System

- Innate Immune System
- Monocytes Primary targets
 - Carry virus throughout body
 - Lysis releases cytokines
- Early infection of Dendritic Cells⁶
 - Delays specific immune response

Interaction with Immune System Cont.

- Over expression of proinflammatory signals
 - Cytokines
 - Does not clear infection
- VP35 inhibition of IRF3
 - No transcription of IFN genes
 - No antiviral response to dsRNA
- Inhibition/Destruction of immune cells
 - Neutrophils & macrophages

Pathogenesis

- Glycoprotein responsible for CPE of virus
 - Breakdown of extracellular matrix
 - Rounding and detachment of endothelial cells
 - sGP inhibits Neutrophils
 - Evidence suggests virus does NOT directly cause most of the disease pathology

Pathogenesis

- Massive Immune Response
- Activation of macrophages and monocytes
 - Clumping may cause coagulation observed in some clinical cases
 - Proinflammatory signals released
 - Cytokines, TNF, IFN
 - Breaks down endothelial barrier
 - Blood leaks into tissue- blood pressure drops-Shock most frequent cause of death.

Ebola Hemorrhagic Fever

- 2-21 day incubation period
- Abrupt onset: flu-like symptoms
 - Fever, headache, muscle aches, stomach pains
- Rash, red eyes, internal/external bleeding
- Death (50-90% according to WHO)

Ebola Hemorrhagic Fever

- Not known why some are able to survive
- Larger Early Immune response in those that do
- Virus May remain up to 3 months
 - Convalescents potential human reservoir
 - Virus present in seminal fluid

Treatment/Vaccine

- No effective treatment
 - Research on treating inflammatory immune response
 - Anti-INF, Anti-cytokine antibodies
 - Steroids
- No approved vaccine
 - GP protective antibodies
 - Also immunosuppressive

Ebola as a Biological Weapon

- As late as 1992 Russia was producing large quantities of Ebola virus for use as a weapon.
 - No vaccine
 - No treatment
 - High infectivity (as few as 17 particles necessary to cause disease)
 - Can potentially be spread by aerosols
- Japanese Terrorist Cult, Aum Shinrikyo, unsuccessfully attempted to obtain Ebola virus

References

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- 4 U. Stroher, E. West, H. Bagany, H.D. Klenk, H.J. Schmittler, H. Feldmann. Infection and Activation of Monocytes by Marburg and Ebola Viruses. *J Virol*. 75(22) Nov. 2001: 11025-11033
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- 7 Borio et al. Hemorrhagic fever as biological weapons. *JAMA* 287(18) May 8, 2002, 2391-2405.