Family: Togaviruses
Genus: Alphaviruses

- from Latin “toga” = a cloak
- “Alpha” refers to “Group A arbovirus”, (arthropod-borne)
- “chikungunya” means “to walk bent over” in Makonde

The alphaviruses consist of >30 members grouped into 7 serocomplexes (cross-reactivity of antibodies between viruses in same serocomplex).

Rubella is the only member of the Rubivirus genus. aka “German Measles”. Part of the MMR vaccine given to (nearly) all children and required for admission to UW
General properties of alphaviruses

- Form enveloped spherical particles
- ~11.7kb, single-stranded, non-segmented (+) sense RNA
- Expression of viral proteins and replication of genome in the cytoplasm
- Short replication cycle of ~4 hour
- Broad host range, grow in both mammalian and insect cell lines
- All are arthropod-borne viruses (with the possible exception of the fish viruses)
- Can get into the CNS and infect neurons, leading to encephalitis

Alphaviruses geographically segregate into New World and Old World Viruses

**Examples**

- **Venezuelan equine Encephalitis (VEE)**
  - Febrile illness encephalitis
  - Central and South America
- **Eastern equine Encephalitis (EEE)**
  - Febrile illness encephalitis
  - North, Central and South America
- **Semliki Forest Virus**
  - Febrile illness
  - Africa
- **Ross River Virus**
  - Febrile illness Arthralgia, rash
  - Australia, Oceana
- **Chikungunya (CHIK)**
  - Febrile illness Arthralgia, rash
  - Africa, Asia

**New World alphaviruses** generally cause encephalitis while **Old World alphaviruses** generally cause immune-mediated pathology
Transfers of virus between Old and New World

Transmission cycles of arboviruses

Reservoir hosts

Vectors

Occasional intersection of cycles

Spillover to humans
Alphaviruses make two polyproteins which get cleaved by proteases to make additional proteins

Non-structural proteins

Structural proteins

![Genome organization of the alphaviruses with major protein functions listed below.](image)

E2 = attachment   E1 = fusion

How is this similar and different from the picornaviruses and coronaviruses?

Weaver, Antiviral Research 94 (2012) 242–257

The structural proteins are made from a subgenomic message

![Diagram showing the replication process of (-) strand and (+) strand.](image)

Replication of (+) viral genome for packaging

Internal promoter for structural proteins from (-) strand

Translation of non-structural proteins initially

Remaining cleavages are host proteases in ER

nsP2 has protease activity for the non-structural polyprotein cleavage sites

Capsid contains protease activity and self-cleaves from polyprotein

Fields Virology 4th Ed
Temporal Regulation of (-) and (+) strand RNA synthesis

This complex makes (-) strands

nsP4 encodes the polymerase
nsP2 has the protease activity

trans-cleavage at the nsP1-nsP2 site of the polyprotein causes conversion to efficient (+) strand synthesis

Once concentration of P123 is high enough, then switches to + strand synthesis only

Both genomic RNA and subgenomic RNA are made from minus RNA, but subgenomic RNA is favored

Host protein transcription and translation also inhibited, but by different mechanisms for different alphaviruses
Evolutionary consequences of mandatory alternating host switches between very different kinds of hosts

- Alphaviruses seem to have lower mutation frequency than many RNA viruses
- One hypothesis for alphavirus genetic stability → strong purifying selection imposed by the alternating host transmission cycle
- Viruses passaged in the vertebrate cells show a drop in fitness in the mosquito cells and vice versa
- Single-host-cell adaptation results in more mutations than alternating cell passages

The fitness landscape of vertebrate infection is different from mosquito infection for the most part

What do you think the alternating host landscape looks like?
Alphavirus infections in vertebrates

*Alphavirus infections in vertebrates*

**Aedes aegypti**

**Aedes albopictus**

(usual)

Other species in non-human transmissions

Replication of alphavirus in mosquitoes

- The infection in the mosquito begins in **midgut epithelial cells** and spreads to the salivary glands.
- The **time from mosquito infection to transmission** is relatively short (2–7 days) for alphaviruses.
- Susceptibility of mosquitoes to viral infection is mostly determined by how well the virus binds and infects the epithelial cells.
- **Once infected, the mosquito remains infectious for life.**
The important parameters of transmission occur in the mosquito

Incubation time = period between taking an infected blood meal and ability to transmit virus.

Determined by rapidity of virus replication and dissemination to mosquito salivary gland

Higher temperatures accelerate the transmission cycle in warm months

*Tight control of infection process in the mosquito (shutoff of minus-strand synthesis)*

Chikungunya Epidemic in 2006

1.4 to 6.5 million suspected cases total (2006-07)

La Reunion island
270,000 cases (40% of population), 2006-07
The CHIKV epidemic in the Indian Ocean came from Africa

Rural Africa
Cycle between non-human primates and mosquitoes

Urban Asia and Africa
Cycle between humans and mosquitoes

Evolution of CHIKV during the Indian Ocean outbreak

E1-226V mutation increases infectivity of CHIKV for Ae. albopictus
Synchronization of Vector Activity Increases the Probability of CHIKV epidemics

Adapted from Charrel et al. The Lancet Infectious Diseases 2008.

Chikungunya is now in the Western Hemisphere since 2013

> 300,000 suspected and ~10,000 laboratory-confirmed chikungunya cases

The vector is probably the *Aedes aegypti* mosquito
Volume of travelers from Chikungunya Areas of the Caribbean to the US in July and corresponding vector activity

As of July 2017, ~1000 cases in the US, most travel-associated, but some Florida cases are locally acquired
Things You Should Know About Alphaviruses

- Similarities and differences between positive sense RNA viruses we have talked about in class
- Alternating transmission between vertebrates and mosquitoes
- Replication strategy including mechanism of regulation of plus and minus strands
- Indian Ocean and now Caribbean epidemics of CHIKV
- Mechanisms of disease emergence by expanded host range of vectors

**Paper for discussion:**
- OID50 = Oral dose of virus necessary to infect 50% of the mosquitoes after a blood meal (lower numbers mean more infectious)
- *Epistatsis*
- What is the argument for why the mutation in the glycoprotein that enhances infection of a new mosquito did not arise before?
- Try to understand this in terms of fitness landscapes that describe the limitations of virus evolution