RANAVIRUS TAXONOMY AND PHYLODYNAMICS

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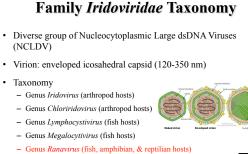
Talk Objectives

- Introduction to the family Iridoviridae Taxonomy and basic biology
- Introduction to the genus Ranavirus Taxonomy and basic biology
- Phylogenomic patterns
 - Gene trees • Genome organization



- Phylodynamic modeling
- Expanding geographic and host ranges
- Recombination

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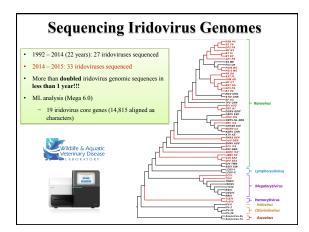


- FV3 BIV EHNV ATV ECV SCRV
- Unassigned members: CMTV, REV, ADRV, CoIV/RMAX, SERV, SGIV/GIV
- Unassigned members: EIVs (fish, amphibian, & reptilian hosts)

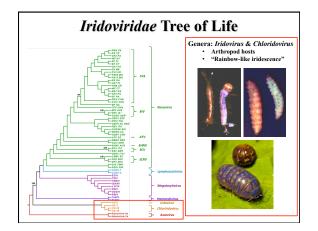
Sequencing Iridovirus Genomes

- 33 iridovirus samples contributed by the international ranavirus community
- Cell culture on EPC/BF-2
- Freeze thaw x 3, slow spin, fast spin, DNAse treatment,
- sucrose cushion, DNA extracted via commercial Qiagen kit
- DNA sheared via sonication, dual indexed libraries via Illumina TruSeq (no PCR)
- · Each library prepared in a hood on separate days, MiSeq bleached between runs
- Typical V3 chemistry (2 x 300 bps), 40 MRs
- Typical virus (Index) 1.9 MRs / 2 MRs = 95% pure!
- De novo assembly in SPAdes, Velvet, CLC Genomics
 Greater than 1000x coverage

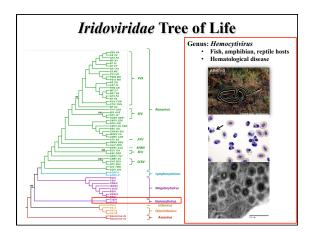




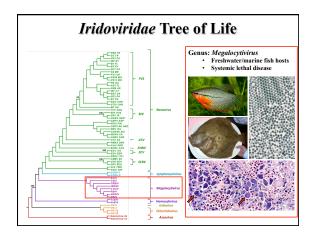




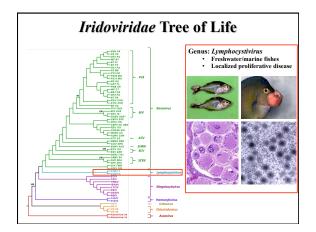




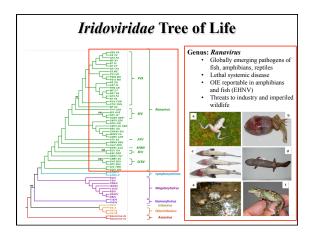




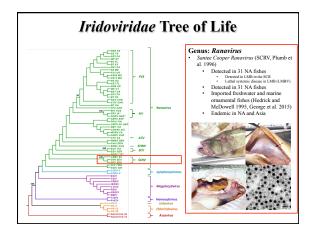




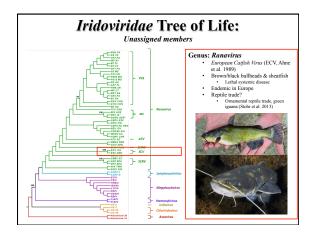




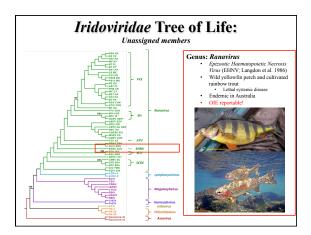




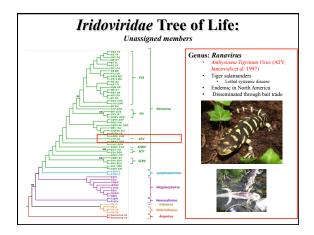




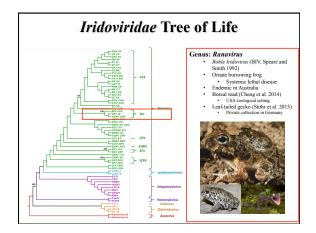


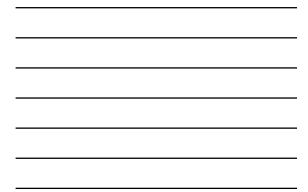


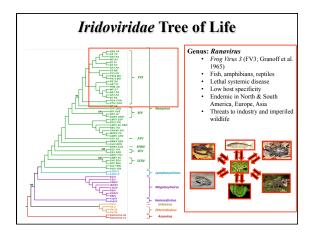




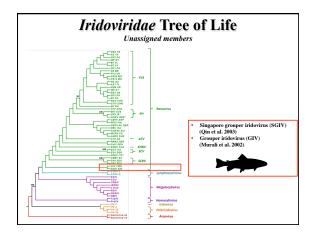




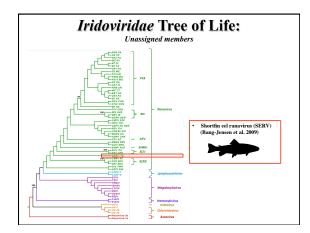




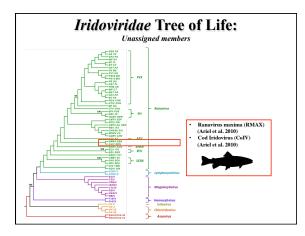




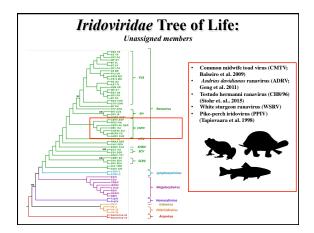




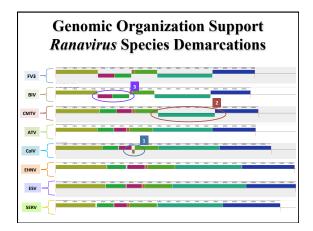




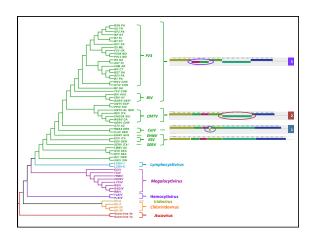




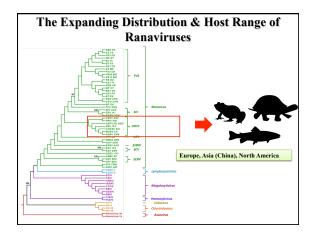




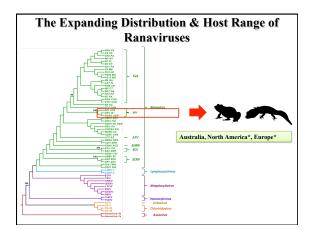




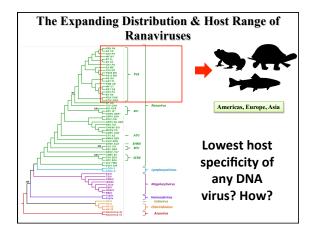














Ranavirus Host Range Evolution

- Could recombination be a driver of host range evolution and virulence? Virus outpaces host through recombination...
- Two bullfrog ranavirus isolates from the same aqua/ ranaculture facility sequenced
- Genomic sequencing reveals 1st isolate is CMTV, 2nd is FV3
- Recombination analyses (RdRP4.46b) reveals significant recombination between the FV3 and CMTV strains!

Conclusions

- + NGS & global collaboration facilitated sequencing of 33 iridoviruses $<1~\rm{yr}$
- ML analysis (19 iridovirus core genes) produced the most comprehensive *Iridoviridae* tree to date
- Phylogenomic analysis supports the creation of a novel genus for EIVs (Hemocytivirus)
- Phylogenomic analysis supports the current *Ranavirus* species demarcations and the potential creation of new species (CMTV, SGIV/GIV, CoIV/RMAX, SERV)
- The geographic and host ranges of certain ranaviruses appears to be broader than previously realized (FV3, BIV, CMTV)
- Preliminary phylogeographic analysis suggests that the unregulated international movement of live animals (bullfrogs) and their products may explain the global distribution of FV3
- Recombination among ranaviruses may be a driver of host range evolution and virulence!









