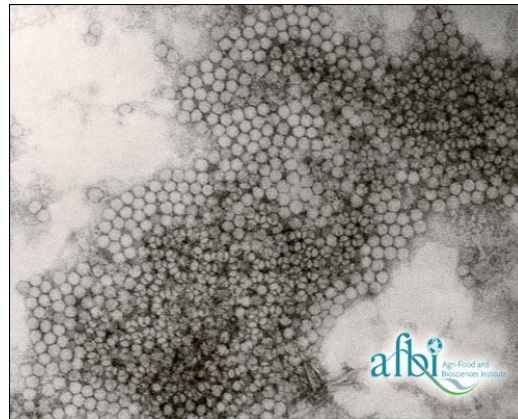


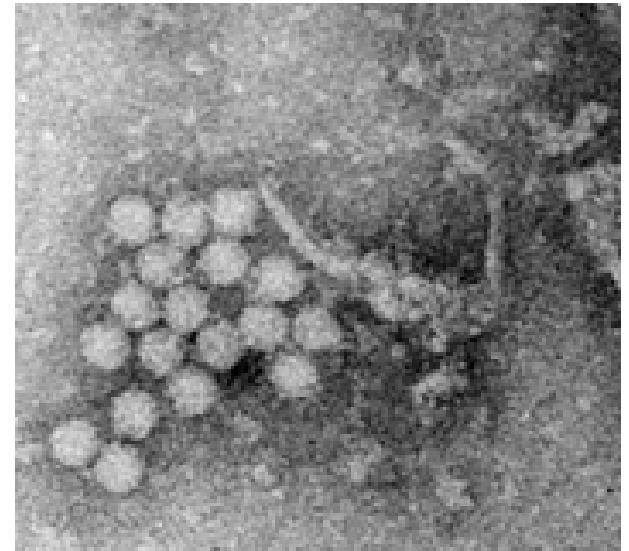
Characterization of a Novel Circovirus from a Stranded Longman's Beaked Whale (*Indopacetus pacificus*)

Nelmarie Landrau Giovannetti, Kuttichantran Subramaniam, Terry Fei Fan Ng, David S. Rotstein, Kristi West, and Thomas B. Waltzek



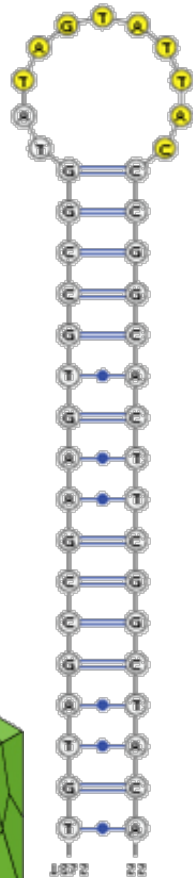
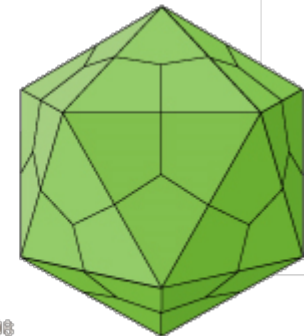
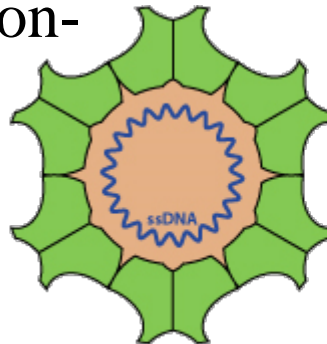
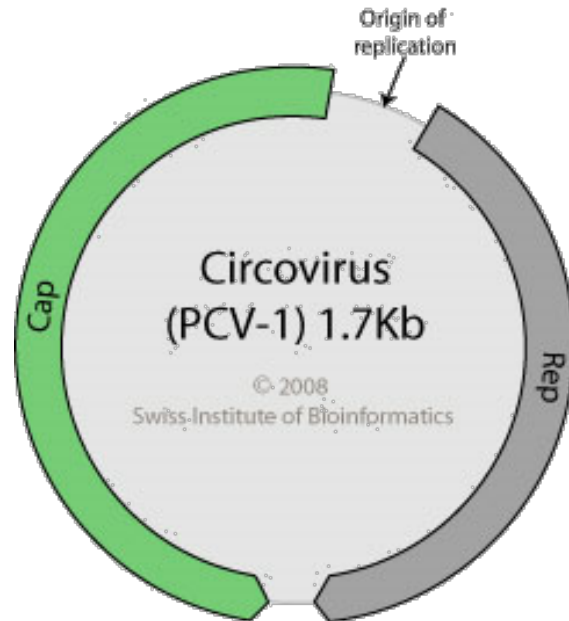
Talk Outline

- Introduction to the family *Circoviridae*
 - Biology
 - Clinical disease
 - Taxonomy
- Objective
- Stranded Hawaiian Longman's beaked whale
 - Clinical case history
 - Viral genomic characterization
 - Beaked whale circovirus (BWCV) genome annotation
 - Phylogenetic analysis
 - Sequence identity matrix
 - BWCV diagnostic PCR
- Conclusions/Future Directions



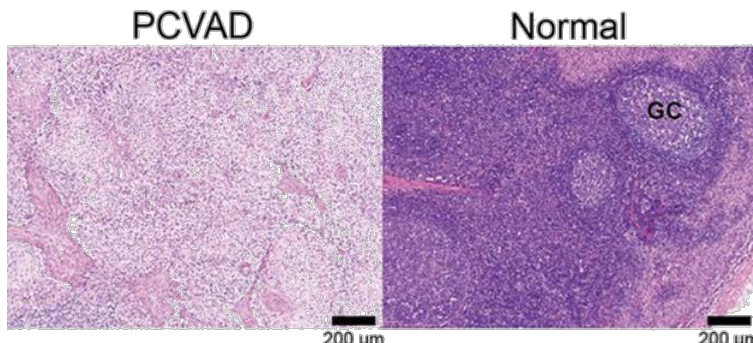
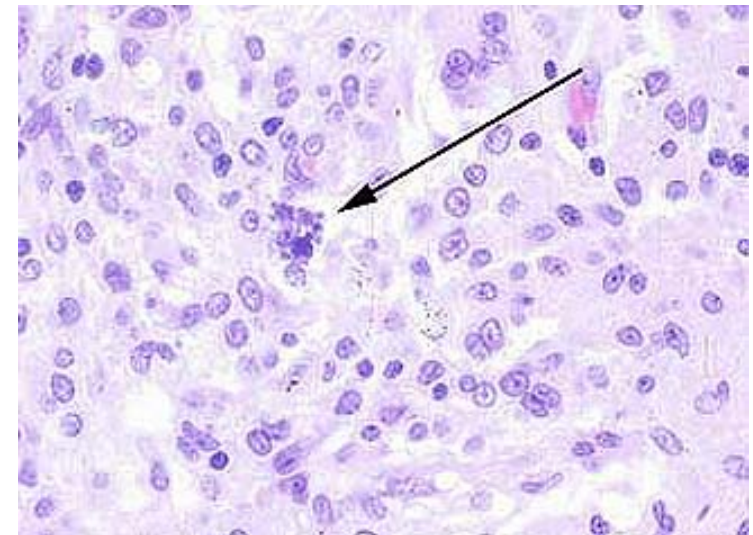
Biology of *Circoviridae*

- Circular, ssDNA
 - 1.8-2 kbp
- Non-enveloped nucleocapsid with icosahedral symmetry
 - 17 nm diameter
- 2 genes: capsid and replication-associated protein
 - Rolling cycle replication



Clinical Disease of Porcine Circovirus 2 (PCV2)

- Associated with the occurrence of postweaning multisystemic wasting syndrome (PMWS) in pigs between 6-18 weeks:
 - Gross pathology
 - Poor body condition
 - Microscopic pathology
 - Lymphoid depletion
 - Cytoplasmic botryoid inclusions
 - Disease outcome
 - Immunocompromised
 - Secondary infection lead to increased mortality



Taxonomy of the family *Circoviridae*

- Genus: *Cyclovirus* (43 species)
 - 16 bat cycloviruses
 - 9 invertebrate cycloviruses
 - 7 mammalian cycloviruses
 - 11 human cycloviruses



Taxonomy of the family *Circoviridae*

- Genus: *Circovirus* (27 species)
 - 2 fish circoviruses (Barbel circovirus – 1st circo reported in fish)
 - 8 bat circoviruses
 - 11 avian circoviruses (beaked and feather disease virus)
 - 5 mammalian circoviruses (porcine circovirus 1-3)

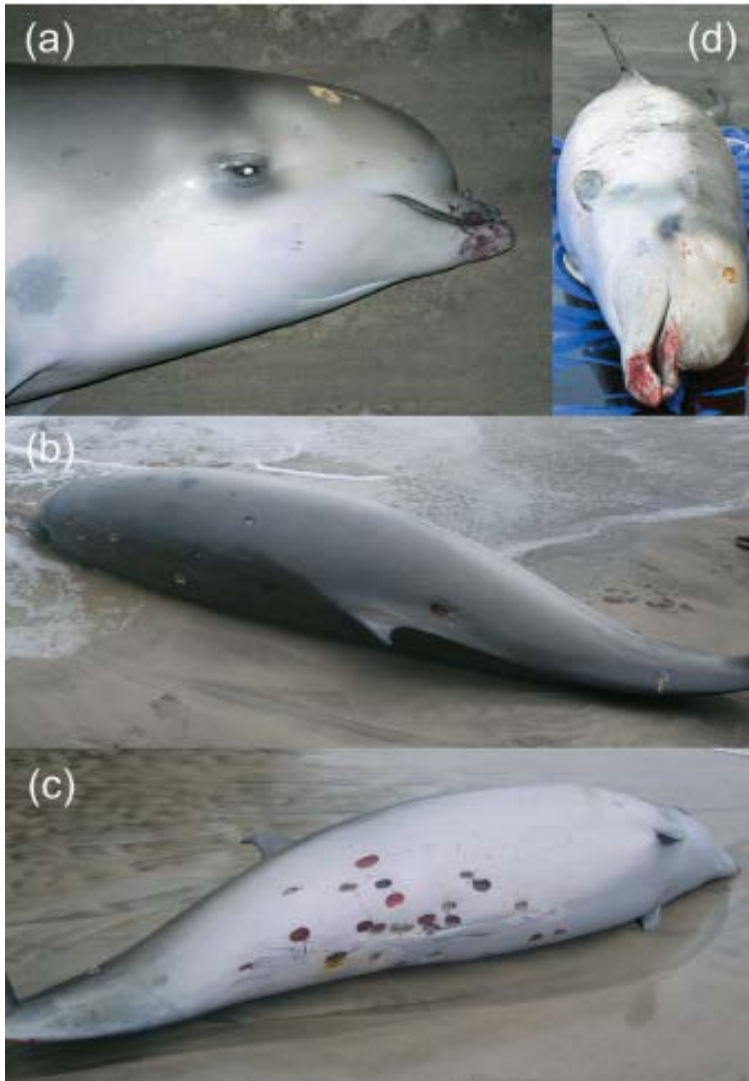


Objective

- To characterize a novel circovirus from a stranded Longman's beaked whale



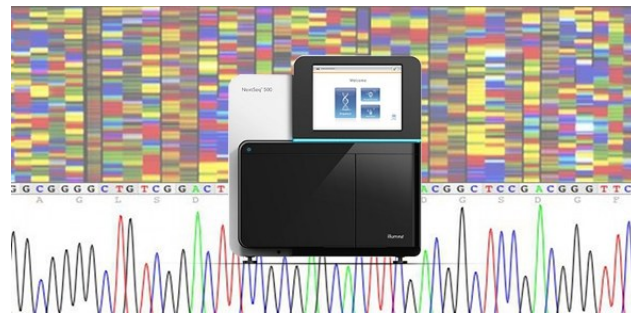
Stranded Hawaiian Longman's Beaked Whale



- Gross pathology
 - Broken jaw
 - Cookie cutter shark bites
- Microscopic pathology
 - Mild lymphocyte depletion
 - Immunocompromised
 - Cerebral encephalitis
 - Pulmonary edema
- Co-infection of an alphaherpesvirus & beaked whale morbillivirus (BWMV)

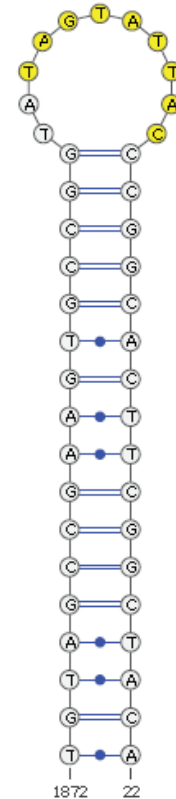
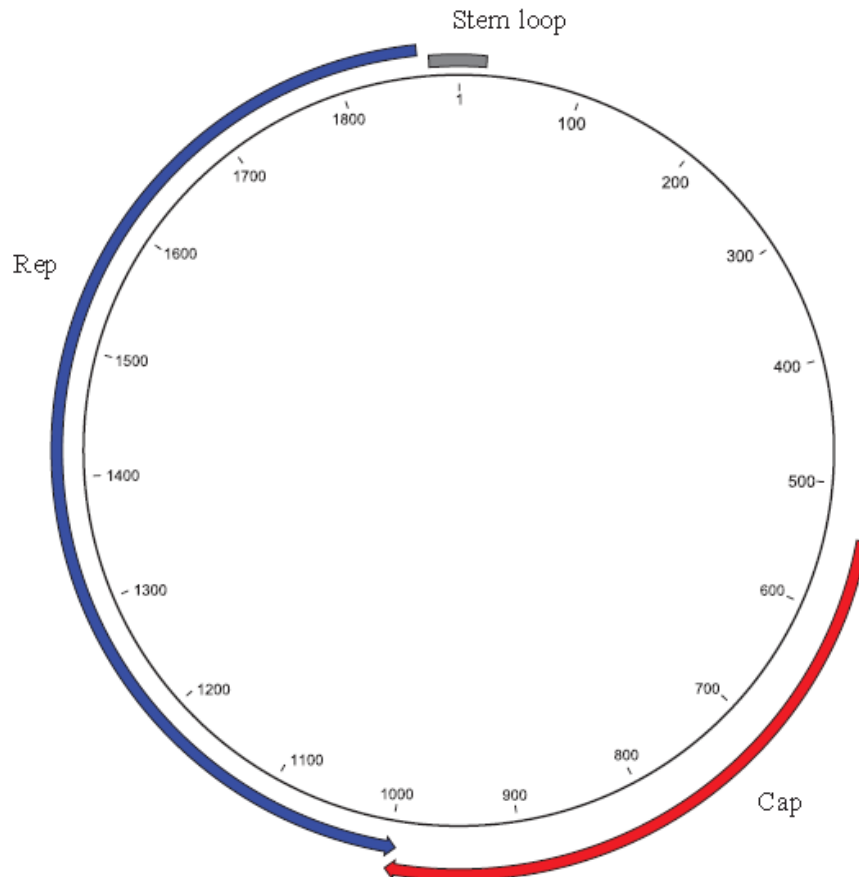
Viral Genomic Characterization

- Illumina MiSeq Next Generation Sequencer (NGS)
 - Beaked whale circovirus (BWCV) was discovered
- Phylogenetic analysis – IQ tree software
- Sequence identity matrix – Sequence Demarcation Tool v1.2
- Development of specific BWCV conventional PCR



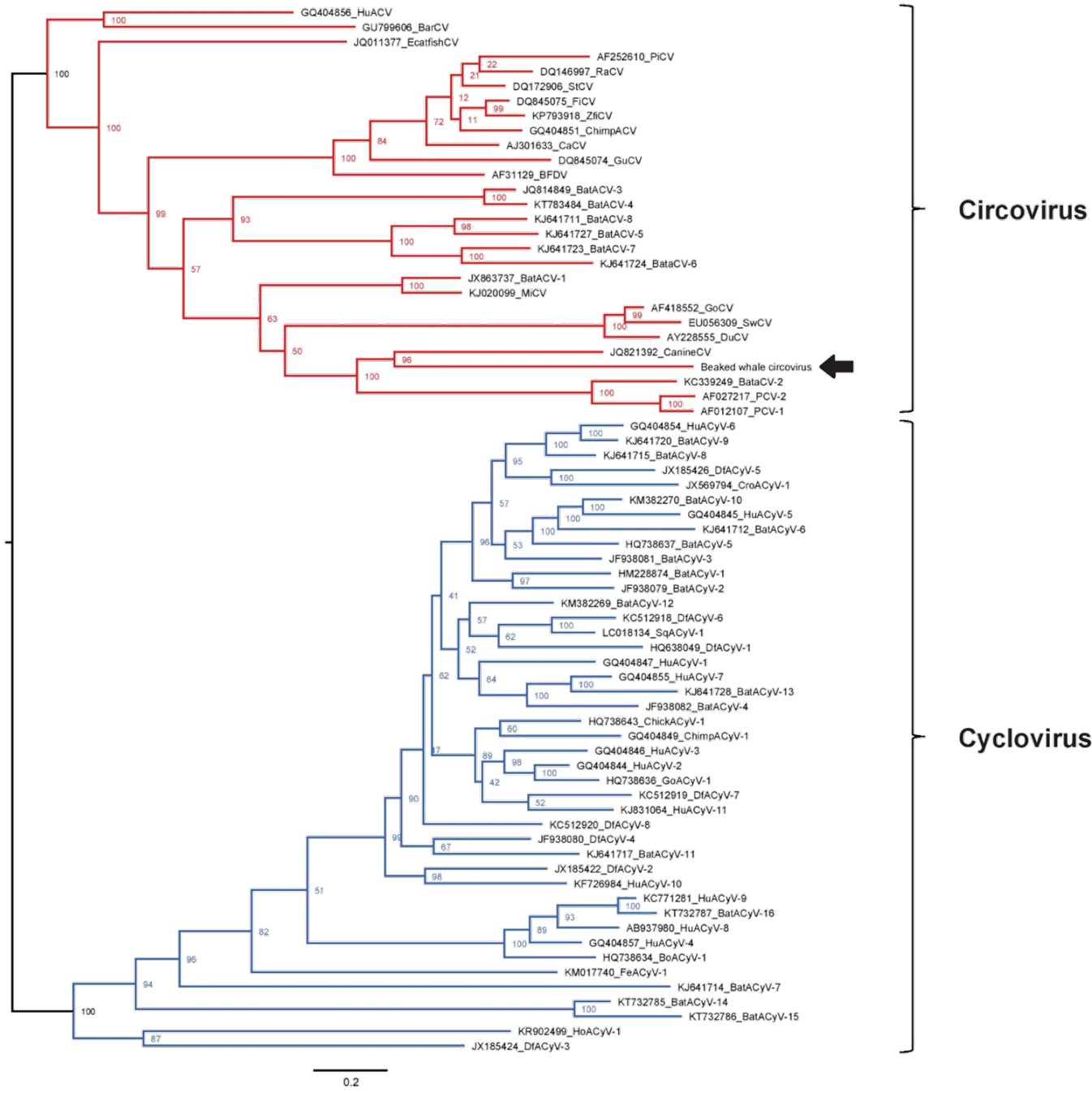
BWCV Genome Annotation

- 2,021 bp, GC content of 46.1%
- Has the nine conserved nonanucleotide (5' - TAGTATTAC -3')



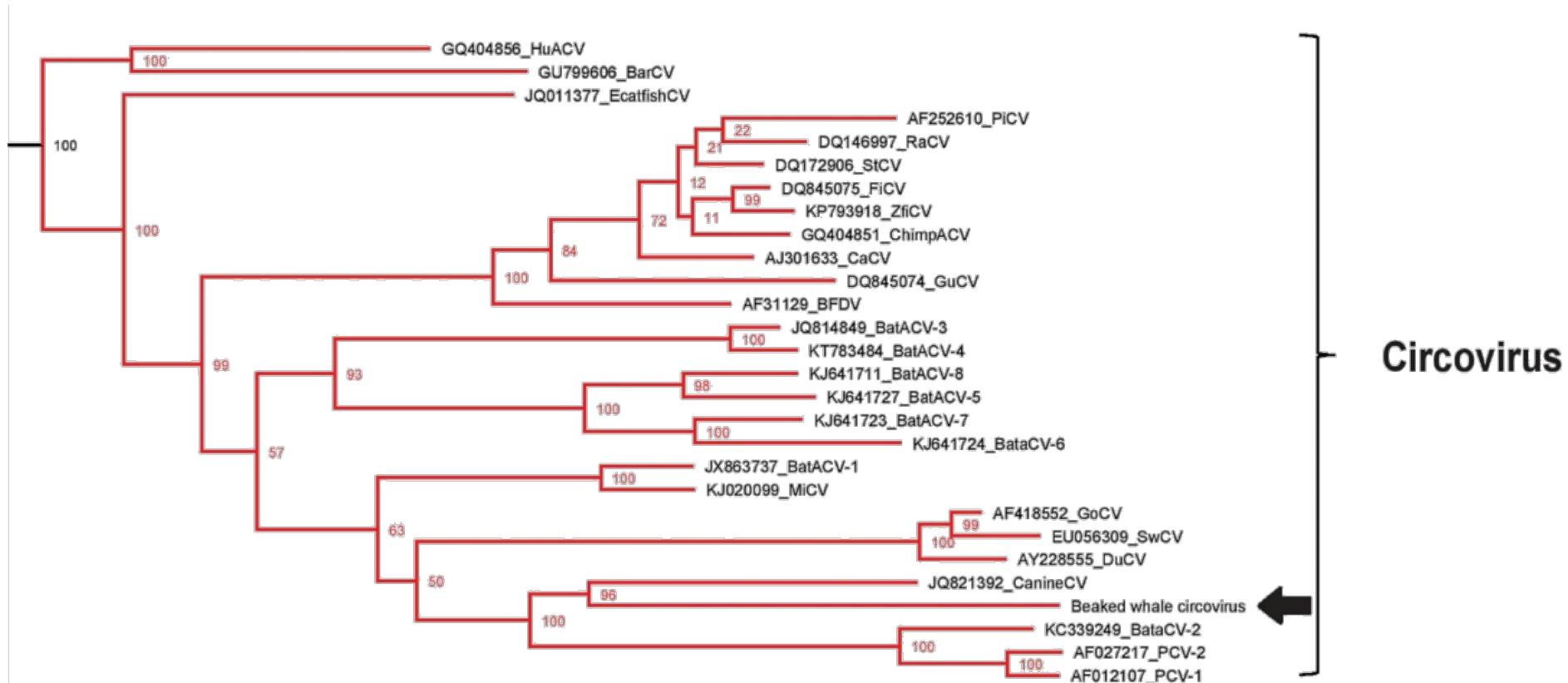
CV Tree (replication gene)

- ML phylogenetic analysis
- Rep AA alignment (351 aligned characters)
- 1000 bootstraps



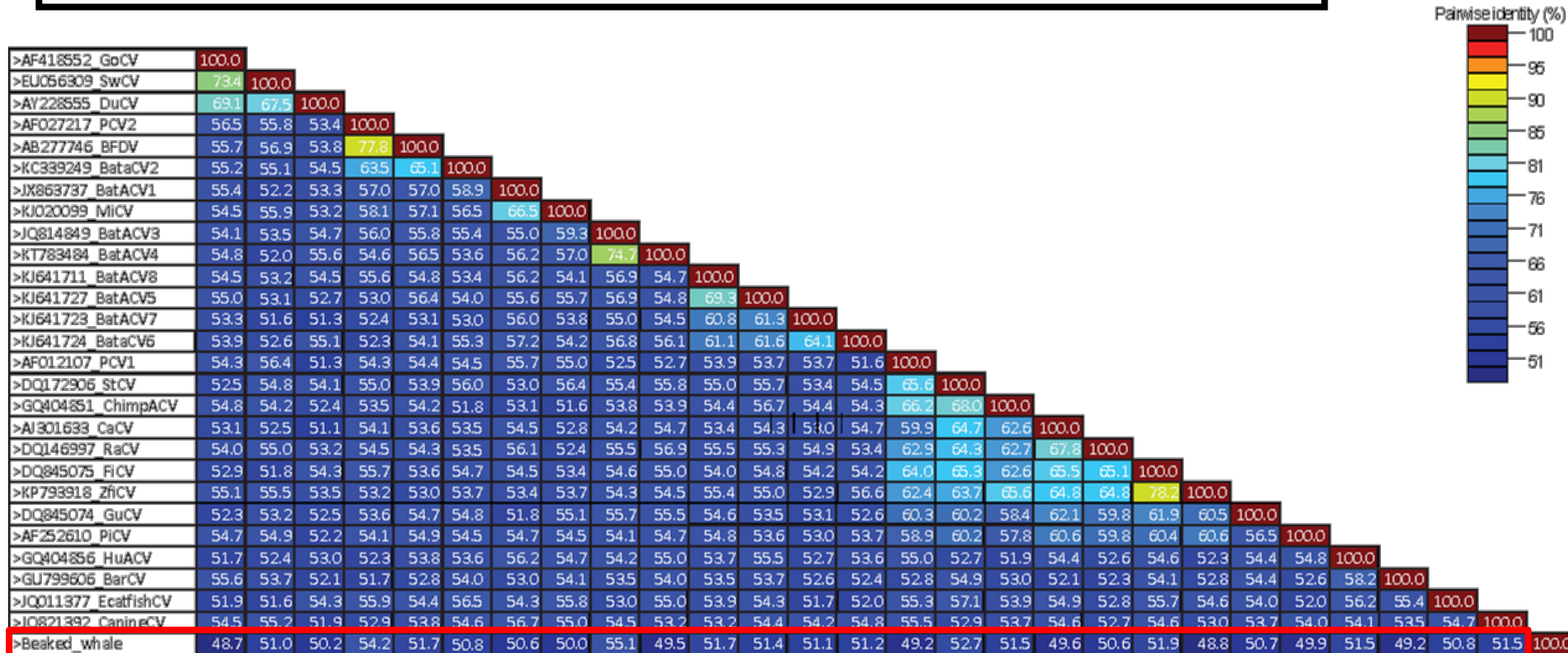
CV Tree (replication gene)

- ML phylogenetic analysis
- Rep AA alignment (351 aligned characters)
- 1000 bootstraps



Circovirus Sequence Identity Matrix

Genus *Circovirus* – full genome nt alignment (2094 aligned characters)



- $X < 80\%$ = new species
- Range of all CVs = 48.7-78.2%
- Range of all CVs to BWCV = 48.7-54.2% → 1st marine mammal CV!

BWCV Diagnostic PCR

1. Lung
2. Cerebrum
3. Cerebellum
4. Scapular LN
5. Mediastinal LN



Conclusions/Future Directions

- Our stranded Longman's beaked whale was co-infected with alphaherpesvirus, BWMV & BWCV
- This is the first description of a circovirus in a marine mammal
 - BWCV can be considered a new species because $X < 80\%$
- The presence of the virus and its role in disease is yet to be proven
 - Positive PCR screening for BWCV on all tissues suggest a systemic infection and/or circulation in the blood
 - Future Direction:
 - *in situ* hybridization (ISH)

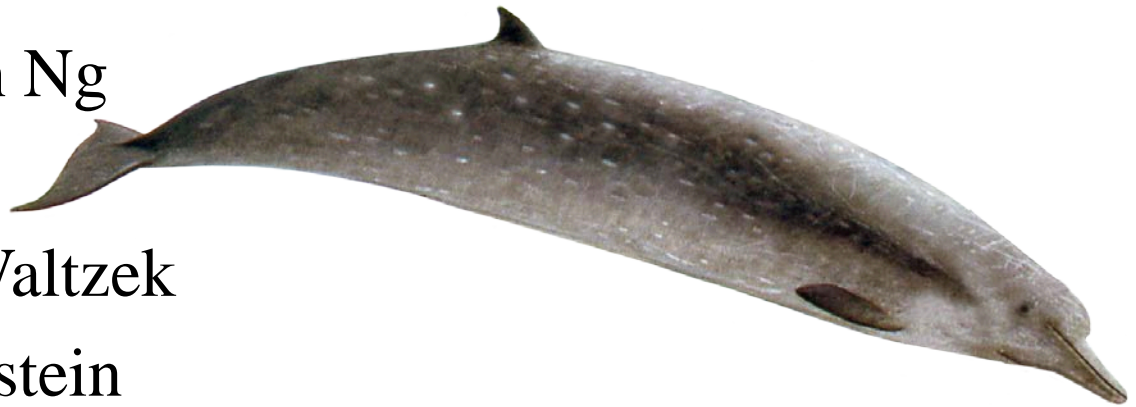


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Acknowledgments

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Thank you!

Questions?

