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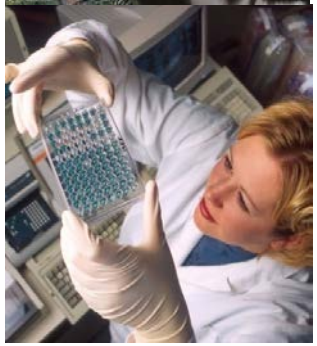

Update on *Theileria equi* genotyping at NVSL

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EP genotyping at NVSL

- NVSL's approach
- Progress
- Next steps
- Potential pitfalls

Whole genome sequencing

- NVSL has developed expertise in next-gen sequencing and bioinformatics
  - TB and *Brucella* programs
  - Organism typing moving towards WGS
- Can we successfully sequence the larger genome of *T. equi* using same approach?

Progress

- Proof of concept
  - *In vitro*-derived *T. equi*
  - Ct value 29.1
  - Captured 97% of the *T. equi* genome at a coverage of 200x
  - Sequencing results were 30% *T. equi* and 70% equine
- Sample inventory
  - Approx. 600 whole blood samples archived

Next steps

- Build database of sequences
  - Compare sequence data to known epi info
  - Compare WGS to other published genotyping studies
- Evaluate methods of concentrating *T. equi* DNA
  - Minimize horse DNA
  - Decreased cost
  - Increased sensitivity

Next steps (cont.)

- Archived vs. fresh samples
  - Archived stored in EDTA, significant lysis of cells
  - Two different approaches for DNA concentration
    - Affinity purification for archived samples
    - WBC depletion for fresh samples
      - Enrich by *in vitro* culture?

Potential pitfalls

- Sensitivity
  - Can’t be cultured/isolated like bacteria
  - Low parasitemia in chronic carriers
  - Extraction of sufficient amount of *T. equi* for sequencing
- Exchange of genetic information

Questions?