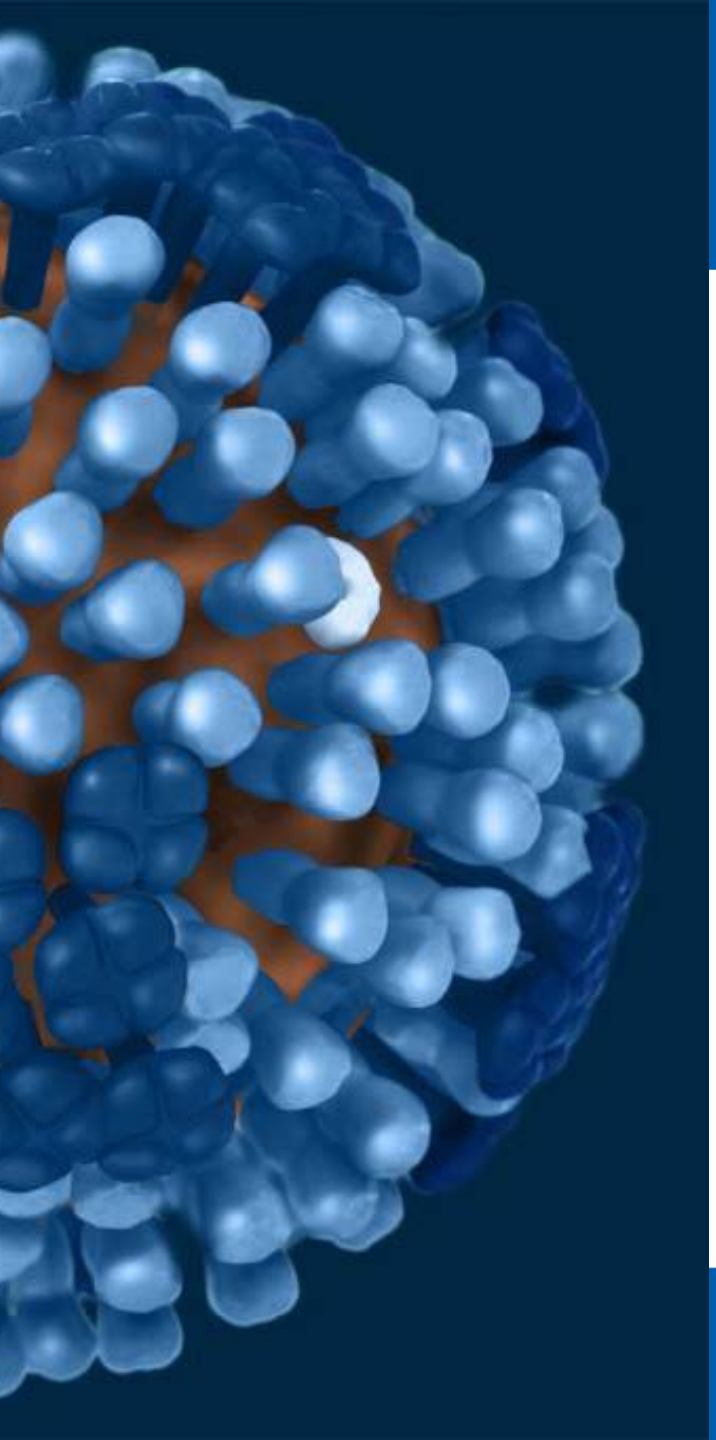
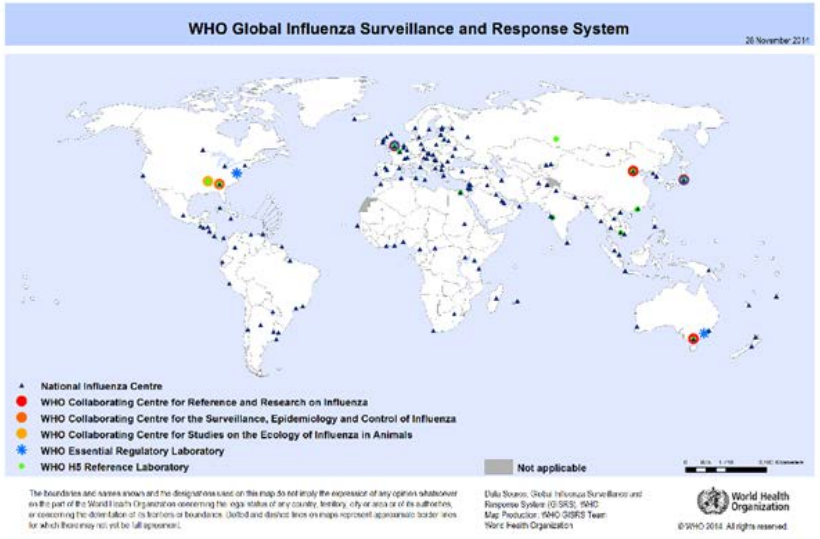


Enhancing Influenza Surveillance with AMD

John Barnes, PhD
CDC Influenza Division

AMD Impact on Influenza Vaccine Strain Selection

- CDC Influenza division is important player in surveillance and epidemiology of influenza
  - Serves as US National Influenza Center and WHO Collaborating Center for Surveillance, Epidemiology and Control of Influenza
  - Analyses 8000-12,000 influenza samples /yr in support of surveillance and selection of vaccine strains
    - Vaccine is produced in a “just in time” fashion
    - 150 Million vaccine doses/year in the US
- Evolution of influenza is very rapid
  - Critical to find variants quickly
  - Antigenic Drift
  - Reassortment
- AMD improves characterization
  - High throughput NGS sequencing for influenza surveillance
  - Antigenic inference



Transforming the Surveillance Paradigm:

1. Specimen collection

N ≈ 10,000/yr

2. Isolate and propagate

N ≈ 10,000/yr

3. Phenotypically analyze

N ≈ 10,000/yr

4. Genetic analysis: a subset

N ≈ 2,000/yr

AMD/Genomics Drive phenotypic analysis

Advantages

- **Faster**
- **Cheaper**
- **More samples**
- **More data**
- **Better data**

1. Specimen collection

N ≈ 10,000/yr

2. Genetic analysis

N ≈ 10,000/yr

3. Isolate and propagate

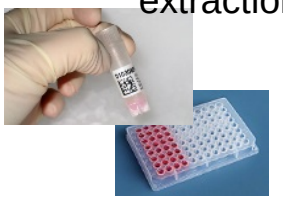
N ≈ 2,000/yr

4. Phenotypically analyze

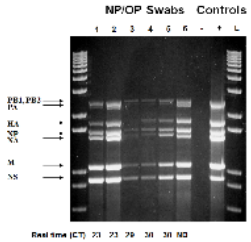
N ≈ 2,000/yr

CDC Influenza Virus NGS Pipeline

1 RNA extraction



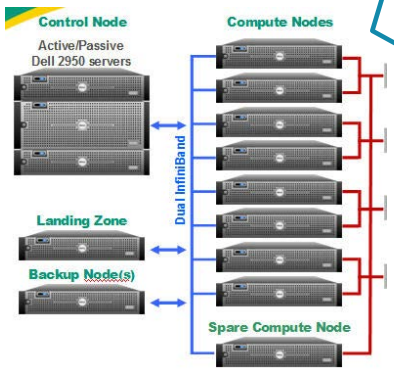
2 Universal genomic amplification



3 **NGS library construction**

4 Next Generation Sequencing

Iterative Refinement MiSeq Assembler

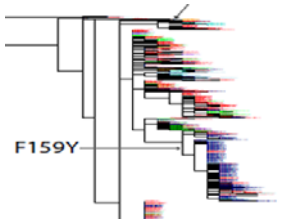


5 Genome assembly and curation

6 Data integration and storage

DISC

7 Analysis

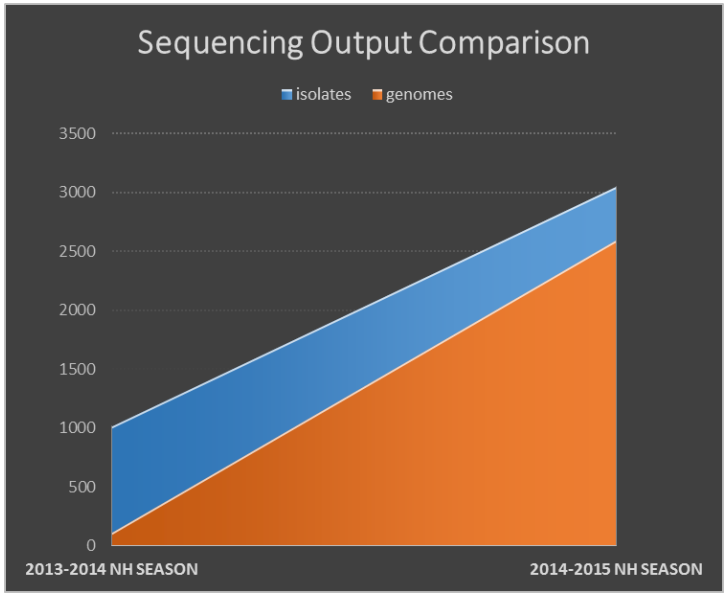


8 Data Base

GISAID

NCBI

Impact of AMD on Influenza Genomics



Complete genomes were generated via the same technique for H1N1, H3N2, H5Nx, H7N9, and Influenza B

Informatics pipelines established

- Large gains in genetic information
  - 2,192% increase in genomes
  - 200% increase in isolates sequenced
- Lower cost of WGS production
  - Reagent cost is ~1/3 of Sanger per sample
- NGS data aided in vaccine match reporting to FluView in 2014-15 season
- Genetic data regularly uploaded to public databases GISAID and NCBI
  - Used by scientists worldwide

Wisconsin AMD Pilot

Goal: Install full Influenza NGS pipeline in Wisconsin State Lab of Hygiene (WSLH)

- WSLH is a lab utilized by CDC
  - Virus isolation, propagation, submission to CDC
- Pilot for implementation of CDC pipeline in NIC or public health laboratory
  - Establish laboratory workflow
    - RNA isolation, genomic amplification, library construction, NGS
  - Transfer data to CDC for analysis

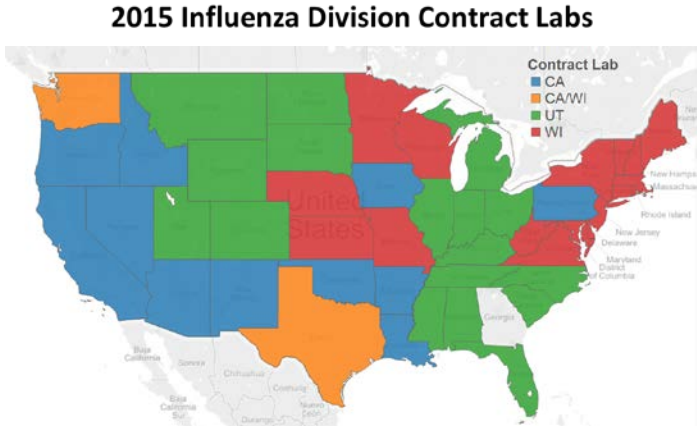
NGS training held at WSLH March 8-19

- Installed full wet lab workflow with WSLH staff
- Operational: First three runs have matched CDC generated data 100%
- Currently developing cloud based data transfer and assembly

CDC Fostering Influenza Virus Sequencing and Analysis Capacity

Robust next generation sequencing scheme
- Wisconsin AMD pilot

Future directions
- State and local
- WHO-Collaborating Centers
- National Influenza Centers



Influenza Genomic Amplification

Library Construction

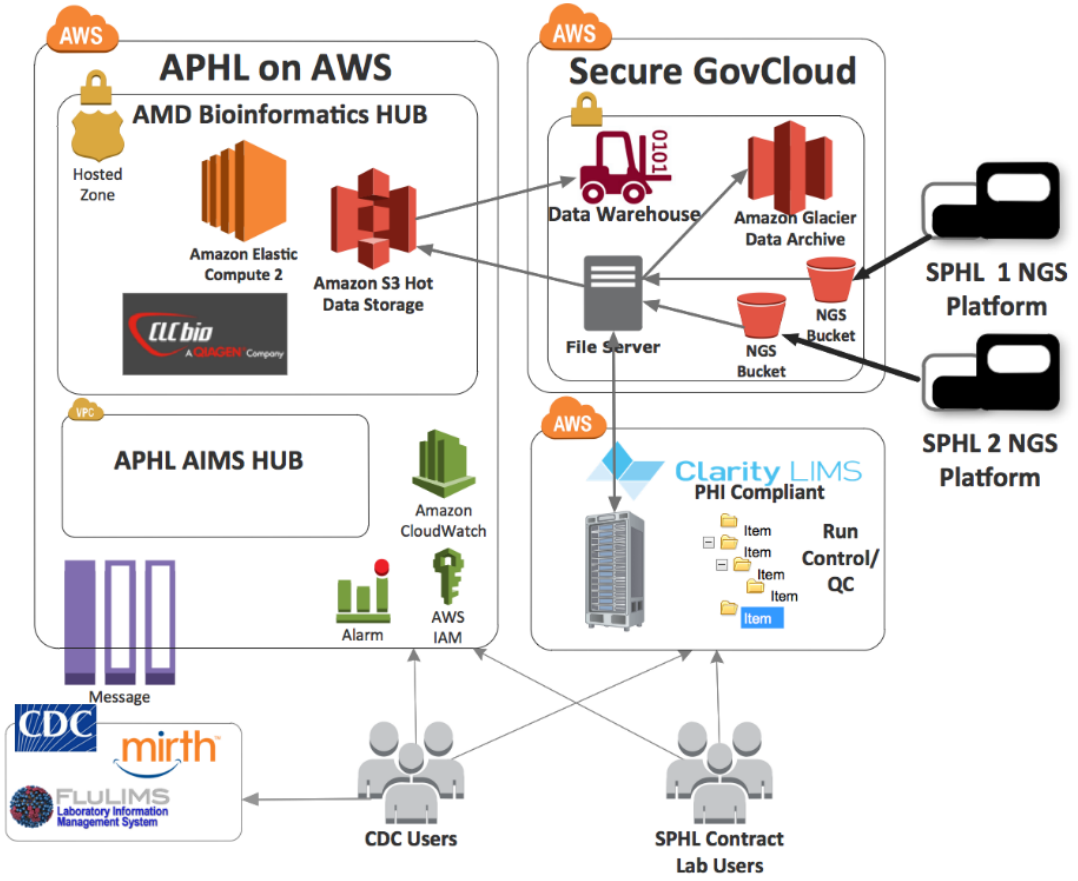
Next Generation Sequencing

Cloud assembly and analysis

Sequence databases

Developing Critical Cloud Based Analytics for Future Influenza NGS Expansion

- Cloud based analytic NGS
  - Important for implementing at many NIC's and PHL
- Collaborating with APHL
  - analytic resource to assemble analyze and store NGS data



Thank you