



United States Department of Agriculture

Animal and
Plant Health
Inspection
Service

Veterinary
Services

July 2018

Influenza A Virus in Swine Surveillance

Fiscal Year 2018 Quarterly Report

Surveillance Summary for Second Quarter: Jan. 1 – Mar. 31, 2018

Report Summary¹

- This report covers the second quarter (Q2) of FY 2018, from Jan. 1 – Mar. 31, 2018.
- Where relevant, the report also includes previous years' data for historical perspective.
- The report provides data from both national and regional levels.
- In FY 2018 Q1-Q2, a total of 7,284 samples were submitted for influenza A virus in swine (IAV-S) surveillance from 2,448 accessions.
- H1N1 continues to be the predominant subtype reported in USDA data.
- Over the past 8 quarters, H1N2 predominated in Regions 1, 3, 4 and 5. H1N1 predominated in Region 2. When regions are recorded as "unknown," there was an even mix between H1N2 and H1N1.
- Limited accessions from a region can skew data and lead to misinterpretation. Therefore, less inference can be applied to results from Regions 3, 4, and 5.
- All IAV-S submissions are voluntary and based on clinical case submissions to veterinary diagnostic labs. These data are not a statistically representative sampling of the U.S. swine population.
- Due to the voluntary nature of this surveillance, the information in this report cannot be used to determine regional and/or national incidence, prevalence, or other epidemiological measures, but it may help identify IAV-S trends.

Introduction

This report, based on data received as of July 2, 2018, provides a brief update on the status of national surveillance for IAV in swine for producers, swine practitioners, diagnosticians, and the public. Summaries in this report may differ from those in past reports due to the regular addition of data from participating laboratories. Reporting months are based on the month the sample was collected. The USDA-APHIS website provides general information about the [IAV-S surveillance program](https://www.aphis.usda.gov/aphis/ourfocus/animalhealth/animal-disease-information/swine-disease-information/influenza-a-virus) (<https://www.aphis.usda.gov/aphis/ourfocus/animalhealth/animal-disease-information/swine-disease-information/influenza-a-virus>).

¹ In November 2016, VS modernized the process that prepares and stages laboratory results data for reporting. Consequently, VS recognizes there is a small difference in previously reported summary numbers for IAV-S surveillance. The results in this report reflect updated and corrected numbers achieved with the modernized data process.

The IAV-S surveillance program is voluntary and, as a result, the accessions and samples submitted represent a subset of the swine population. Submitted samples should only be collected from animals displaying influenza-like illness. Due to its voluntary nature, this surveillance system does not entirely represent the total U.S. domestic swine population. Therefore, the data cannot be used to determine IAV-S prevalence or other epidemiologic measures in the swine population. However, the data may help identify trends in influenza in swine.

When the submitter does not report relevant information, data are recorded as “unknown.” Summaries in this report may differ from those in past reports due to the ongoing addition of data from participating labs. Reporting months are based on the month when the sample was collected.

A laboratory accession is considered a set of samples collected at a single premises on a single day and received at the laboratory. While a nasal swab or lung tissue sample represents a single animal within the herd, a single oral fluid sample may represent one to two pens of animals in a herd. A positive sample status is based on the screening matrix real-time reverse transcriptase polymerase chain reaction (rRT-PCR). The subtype is based on the rRT-PCR- subtyping assays and sequencing if the sample is sequenced. Virus isolation (VI) and sequencing are only attempted on rRT-PCR positives meeting criteria listed in the testing algorithm posted on the web.

On a monthly basis, USDA performs whole genomic sequencing on a selected subset of isolates received through surveillance. On a quarterly basis, a phylogenetic analysis is performed; phylogenetic analyses are based on all successful USDA surveillance sequencing results, with sequences deposited into GenBank®, the public sequence database.

IAV-S Surveillance Objectives

USDA’s National Surveillance Plan for Swine Influenza Virus in Pigs (July 2010) describes the current surveillance system for IAV-S in detail. The surveillance objectives are to:

1. Monitor genetic evolution of endemic IAV in swine to better understand endemic and emerging influenza virus ecology;
2. Make influenza isolates from swine available for research and establish a data management system to facilitate genetic analysis of these isolates and related information; and
3. Select proper isolates for the development of relevant diagnostic reagents, updated diagnostic assays, and vaccine seed stock products.

Objective 1. Monitoring genetic evolution of endemic IAV in swine to better understand endemic and emerging influenza virus ecology

Objective 1 is met through the submission of diagnostic laboratory samples to the surveillance system, collection of the viruses that are isolated from the samples, and analysis of the HA and NA sequences that are generated at the NAHLN laboratories and posted into GenBank®. Each month selected viruses undergo whole genomic sequencing by NVSL. Phylogenetic analysis of the genetic sequences is provided through an interagency agreement with the USDA-ARS National Animal Disease Center (NADC).

National Surveillance Data Summary

From FY 2011 through FY 2016, the total number of accessions and samples submitted rose over time. Changes initiated in FY 2016 have resulted in decreased laboratory accessions and samples, but a higher percentage of accessions that yield a virus that can be sequenced and analyzed. For FY 2018’s second quarter, 7,284 samples have been tested from 2,448 accessions (Figure 1). Figure 2 shows the overall trends in total accessions, PCR-positive accessions, subtyped accessions, and VI positive accessions.



Figure 1. Number of IAV-S laboratory accessions and samples tested in swine, FY 2011 through FY 2018 Q2

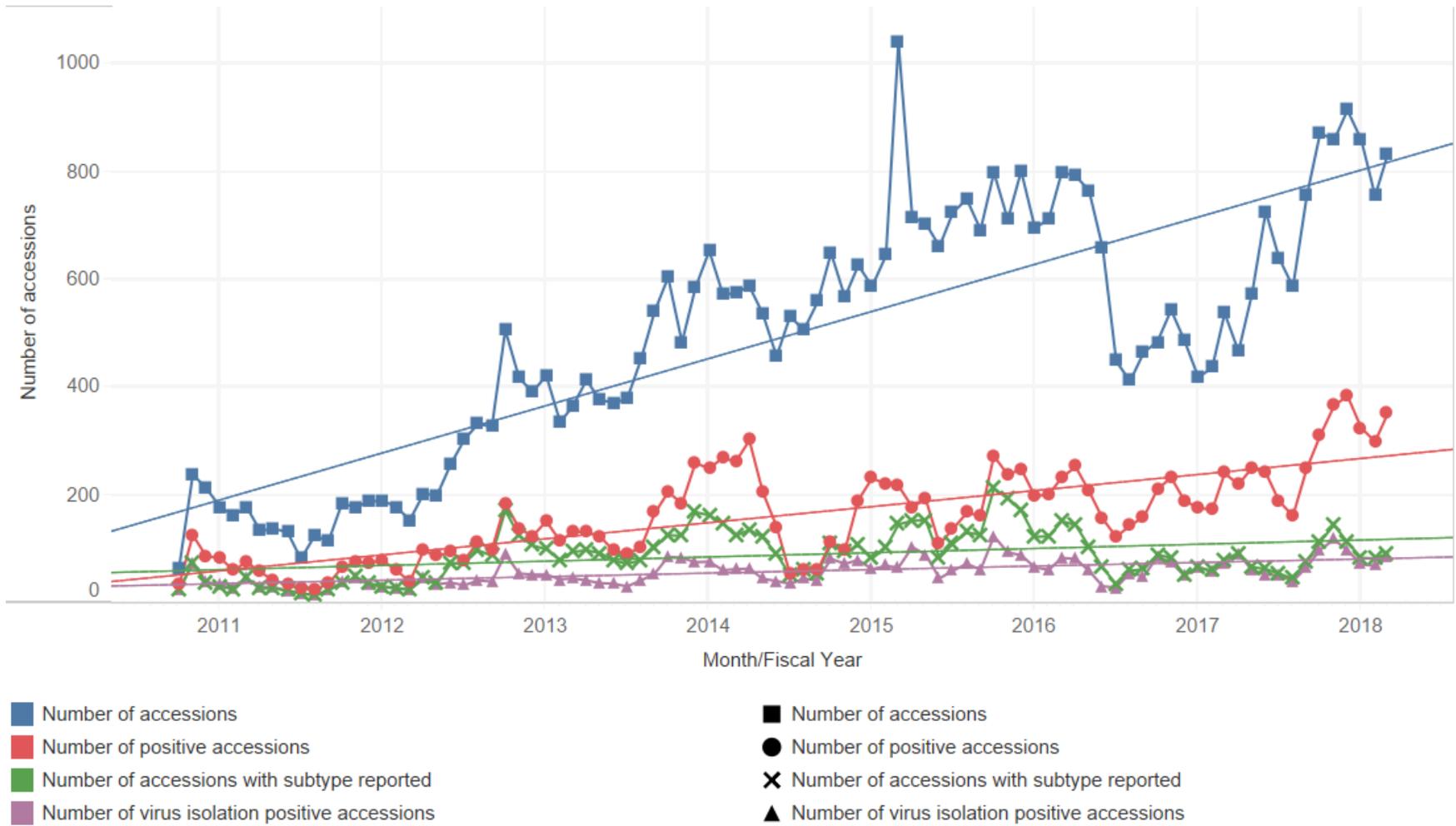


Figure 2. Accessions submitted, subtyped accessions, positive accessions, and VI positive accessions over time with trend lines, FY 2011 through FY 2018 Q2

Figure 3 shows the number of subtype detections in FY 2018 Q2. The total number of samples subtyped was 253, including 110 H1N1, 73 H1N2, 53 H3N2, 1 H3N1, and 16 mixed.

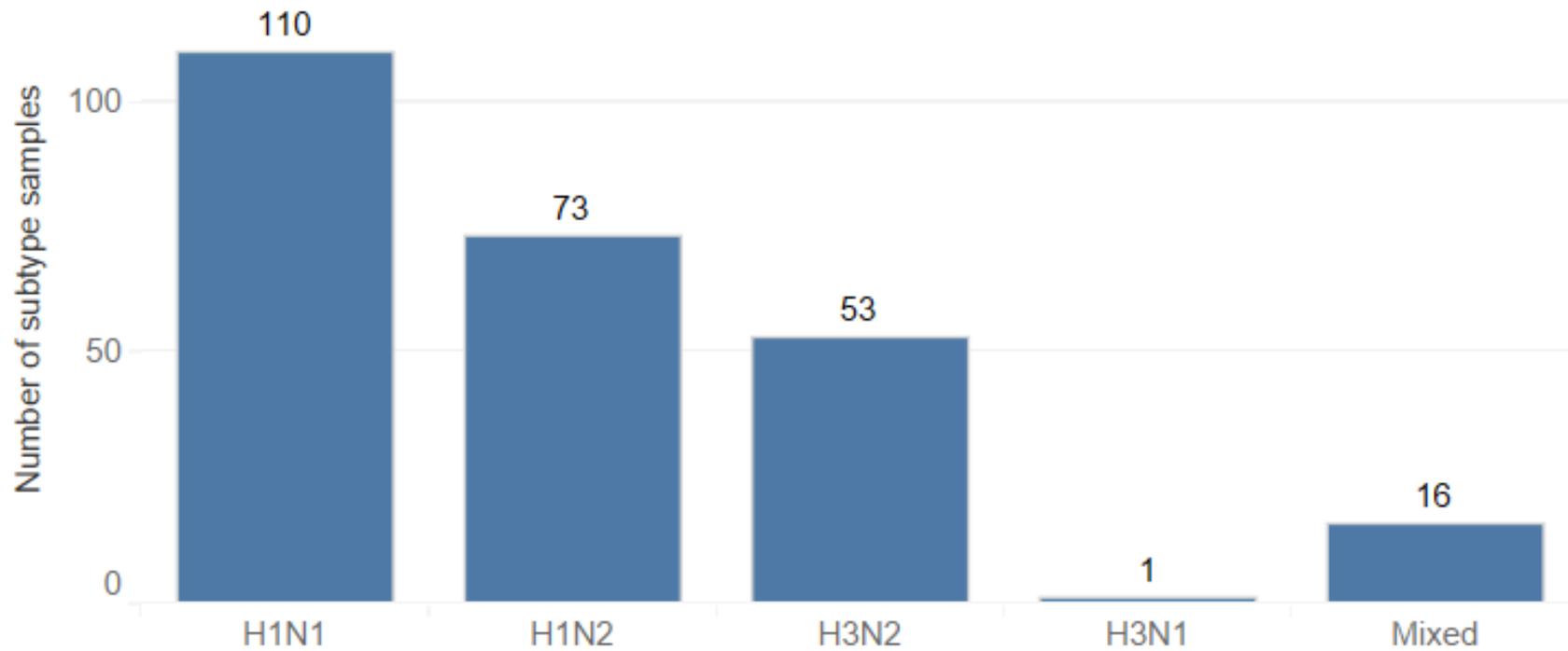


Figure 3. Number of subtype detections in FY 2018 Q2

Figure 4 breaks down accessions by rRT-PCR subtype from FY 2011 to FY 2018 Q2. H1N1 remains the major subtype over the course of the surveillance; however, H1N2 and H3N2 detections have increased substantially since 2012 and H1N1 was the most predominant subtype in FY 2018 (to date, July 2, 2018).

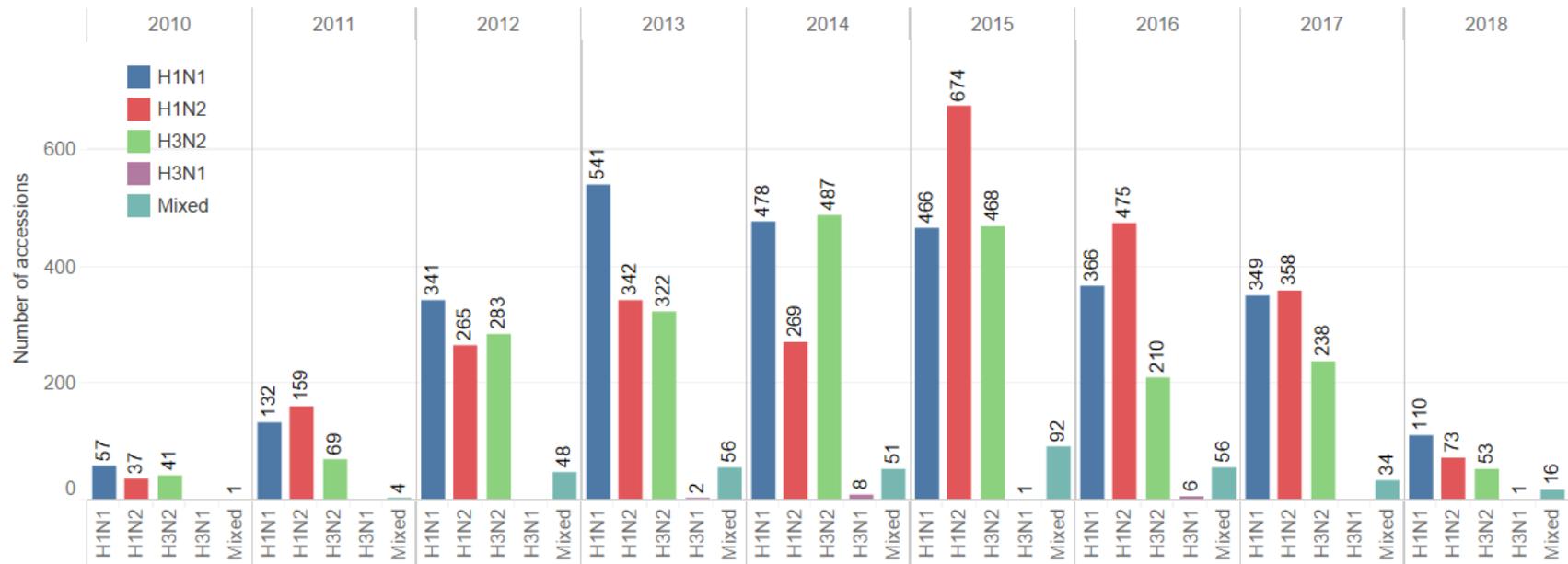


Figure 4. Number of subtypes, FY 2011 through FY 2018 Q2

Figure 5 displays the number of VIs attempted in blue, the number of those attempts that were positive in red, and the number of positive VIs that are submitted to GenBank in green. Since the implementation of the June 2016 modifications to the program, almost all VIs attempted now yield a virus and the sequences are submitted to Genbank for analysis.

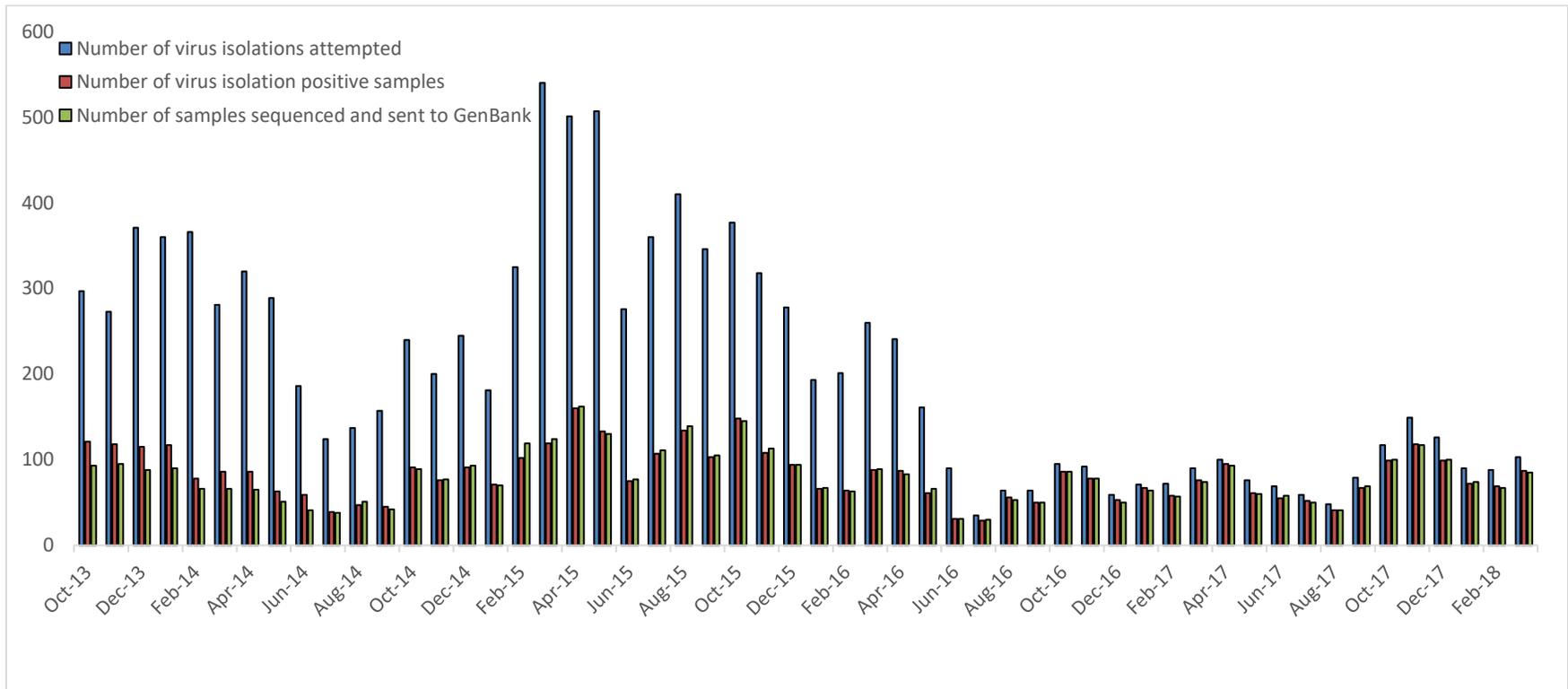


Figure 5. Number of virus isolations attempted, positive virus isolations, and GenBank submissions from FY 2014 through FY 2018 Q2

When accessions were evaluated by age-class for the second quarter, the following observations were noted. H1N2 was the most common subtype among suckling pigs. H1N1 was the most common subtype among nursery and grower/ finisher pigs. Sow/boar had limited testing, with only four accessions of which one was H1N1, one was H1N2, and two were H3N2. Among accessions for which the age class was unknown or not recorded, H1N1 was the predominant subtype (Table 1). Nasal specimens (swab or wipes) were the most successful at providing positive virus isolation and submission to GenBank (Table 2).

Table 1. Number of positive accessions tested for IAV-S by age class and by viral subtype, Q2 FY 2018

Age Class (group)	Number of accessions with subtype reported	Number of H1N1	Number of H1N2	Number of H3N1	Number of H3N2	Number of Mixed
Suckling	68	21	22	0	17	7
Nursery	75	31	24	0	14	6
Sow/Boar	4	1	1	0	2	0
Grow/Finish	48	34	8	0	5	1
Unknown	64	23	18	1	15	2

Table 2. Number of positive accessions tested for IAV-S by specimen type and by viral subtype, Q2 FY 2018.

Specimen Type (group)	Number of accessions with subtype reported	Percent of subtyped accessions with positive virus isolation	Number of H1N1	Number of H1N2	Number of H3N1	Number of H3N2	Number of Mixed	Number of samples sequenced and sent to GenBank
Lung	199	86%	91	60	0	36	11	169
Nasal or Nasal Swab	48	102%	16	10	1	16	4	46
Oral Fluid	4	50%	1	1	0	1	1	4
Other Specimen	8	63%	2	2	0	0	0	7

*Accessions may include samples with multiple specimen types. In these cases, individual accessions are counted in more than one specimen type category.

**Other includes specimen types recorded as swab, mixed tissue, or unknown.

Regional surveillance data

In this section, we present data in five different regions (Figure 6) to parse the analysis across regions. These regions are based on current USDA administrative districts for simplicity; these divisions do not represent specific industry distribution. Submissions are voluntary, as is any identifying information accompanying the submission (except the State of animal origin), and therefore no sampling strategies can be applied to the regions.

Figure 6. A map of the regions for national IAV-S surveillance



Summary of Regional Data from ARS

Table 4. Summary of predominant subtypes in each region for FY 2016 Q3 through FY 2018 Q2

Most Predominant HA/NA phylo-types overall:

H1N1 (Gamma H1/Classical N1)
 H1N2 (Delta1a H1/2002-N2)
 H3N2 (hu-like_2010.1 H3/2002-N2)

Region 1 (Total HA/NA: 274)

Gamma H1/Classical N1
 Delta2 H1/1998-N2
 Alpha H1/2002-N2

Region 2 (Total HA/NA: 1,146)

Most diversity of all regions
 Gamma H1/Classical N1
 Delta1a H1/2002-N2
 Hu-like_2010.1 H3/2002-N2

Region 3 (Total HA/NA: 137)

Delta1a H1/2002-N2
 Gamma H1/Classical N1
 Hu-like_2010.1 H3/2002-N2

Region 4 (Total HA/NA: 159)

Delta1b H1/2002-N2
 IV-A H3/2002-N2
 Gamma H1/Classical N1

Region 5 (Total HA/NA: 11)

Delta 1b H1/2002-N2
 Hu-like_2010.1 H3/2002 N1
 Delta1a H1/1998-N2

Figure 7 shows the distribution of rRT-PCR subtyped accessions among the five regions for Q2 FY 2016 through Q2 FY 2018. Regions 1, 3, 4, and 5 demonstrate H1N2 as the predominant subtype. Region 2 demonstrates H1N1 as the predominant subtype. For regions recorded as “unknown,” there was an even mix between H1N2 and H1N1.

Figure 7. Distribution of rRT-PCR subtyped accessions among the five regions FY 2016 Q2 through FY 2018 Q2.

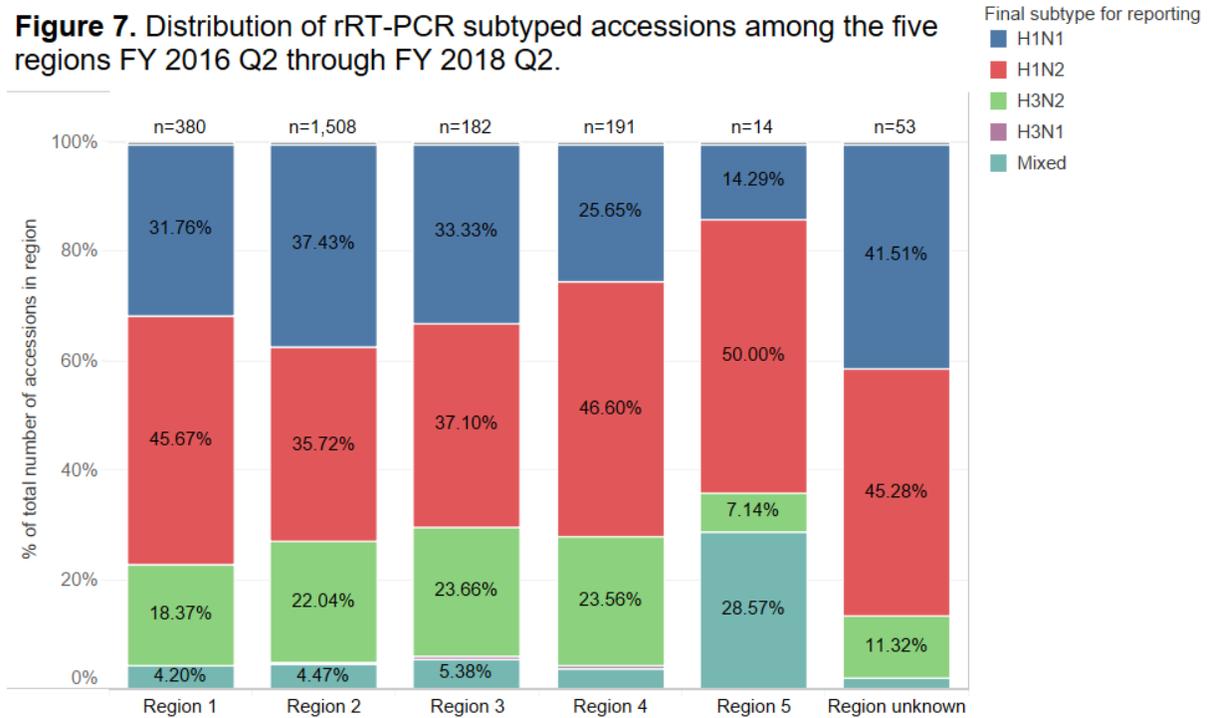


Figure 7. Percentage of subtyped accessions by region for FY 2016 Q2 through FY2018 Q2

Regional phylogenetic analysis

Phylogenetic analysis of sequences from the IAV-S surveillance system

Phylogenetic analysis is conducted on gene sequences of the influenza A virus in swine to further examine the genetic changes that occur in HA, NA, and M genes, when available, of this rapidly changing virus. Through collaboration with ARS, a dataset² of 473 isolates with published sequences in GenBank[®] was characterized by phylogenetic analysis for the Q2 FY 2018 report. This analysis provides information on the genetic diversity and evolution patterns of influenza in swine and allows for inferences about population and/or vaccine immunity.

The following bar charts parse the data into an approximate 2-year window by quarters and subtypes for each region, followed by charts further parsing the H1 and H3 subtypes into phylogenetic clades. Regional charts depicting the various combinations of HA and NA are available in Appendix 1.

² The ARS dataset is comprised of IAV-S surveillance isolate sequences that were posted in Genbank. This represents only a subset of the complete IAV-S surveillance dataset that includes PCR diagnostic test-based results as well as sequencing results. Therefore, ARS dataset results, such as subtype percentages, differ from the complete IAV-S dataset results provided in other sections of this report.

Virus Type by Region

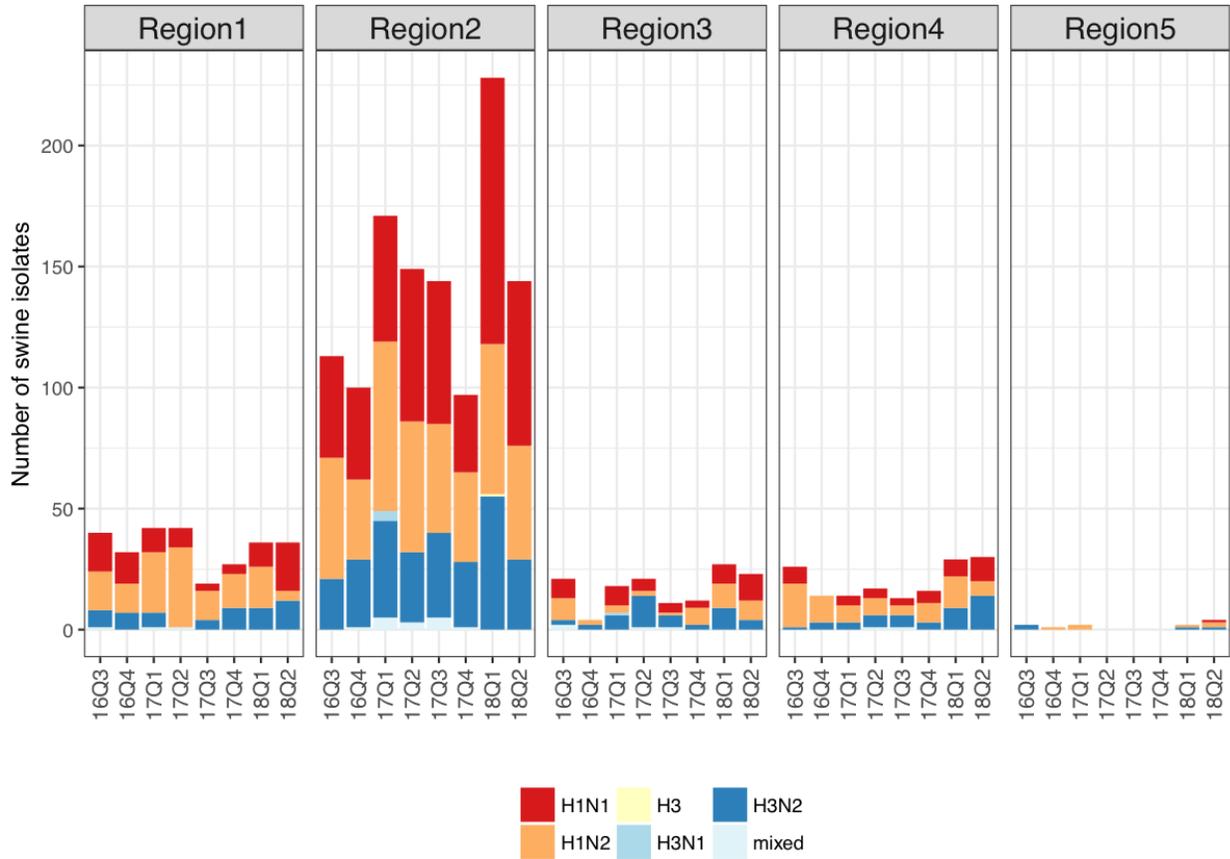


Figure 8. Virus type by region 2-year summary Q3 FY 2016 to Q2 FY 2018

Figure 8 demonstrates the four subtypes H1N1, H1N2, H3N1, H3N2, and mixed subtypes across the five regions. Regions 1 and 2 reported the most submissions, with a mixture of mostly H1N1, H1N2, and H3N2. Limited accessions from a region can skew data and lead to misinterpretation and therefore, less inference can be applied to results from Regions 3, 4, and 5.

National phylogenetic HA gene information

HA genes from H1 subtype viruses are classified as alpha, beta, gamma, delta-1, delta-2, or pandemic H1N1 2009 (H1N1pdm09) phylogenetic clades based on a previously published nomenclature system. Similarly, H3 subtype viruses are classified as Cluster IV, Cluster IV-A, Cluster IV-B, Cluster IV-C, Cluster IV-D, Cluster IV-E, Cluster IV-F, or human-like. In the H1 subtypes (see Figure 9), there continued to be detections of alpha with two aa deletions (n=10). Delta 1a, delta 2, and gamma viruses are the predominant H1.

H1 phylogenetic-clades by Region

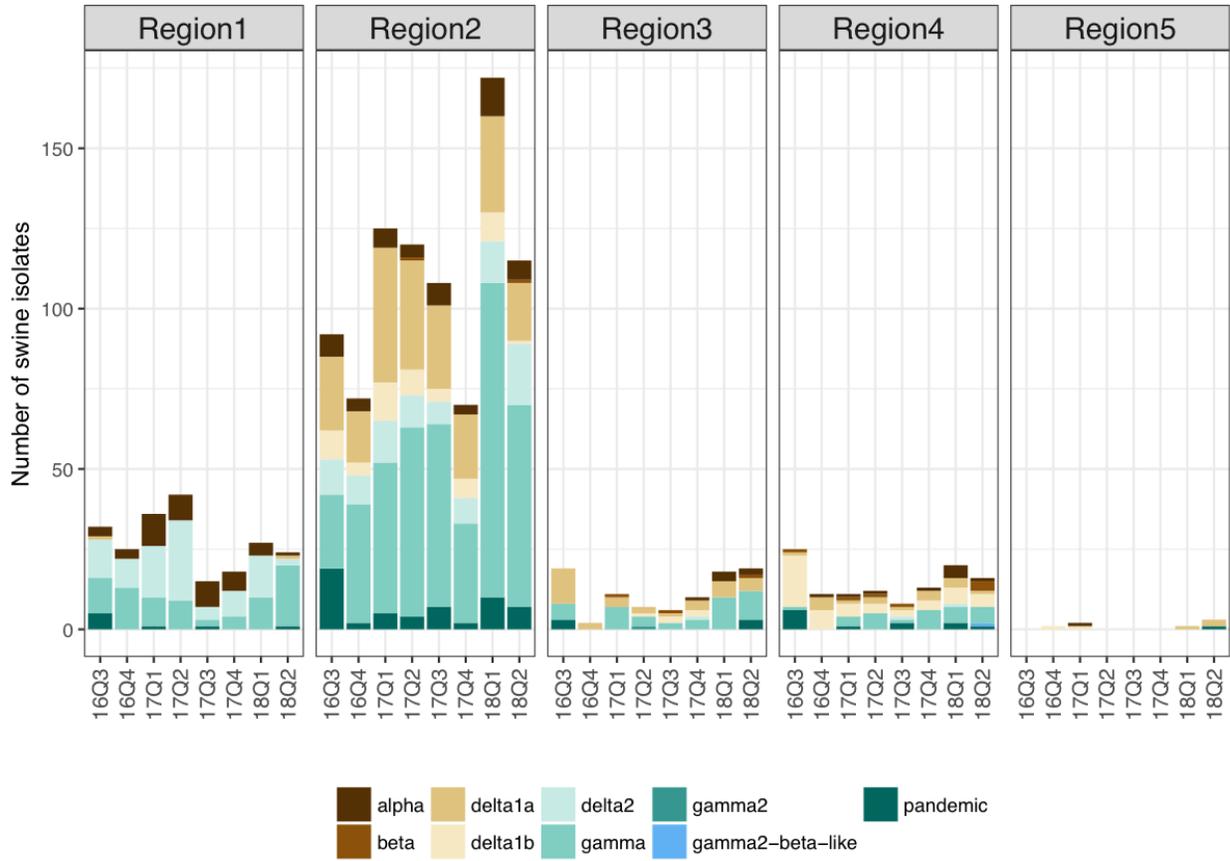


Figure 9. H1 phylo-cluster by region – 2-year summary Q3 FY 2016 to Q2 FY 2018

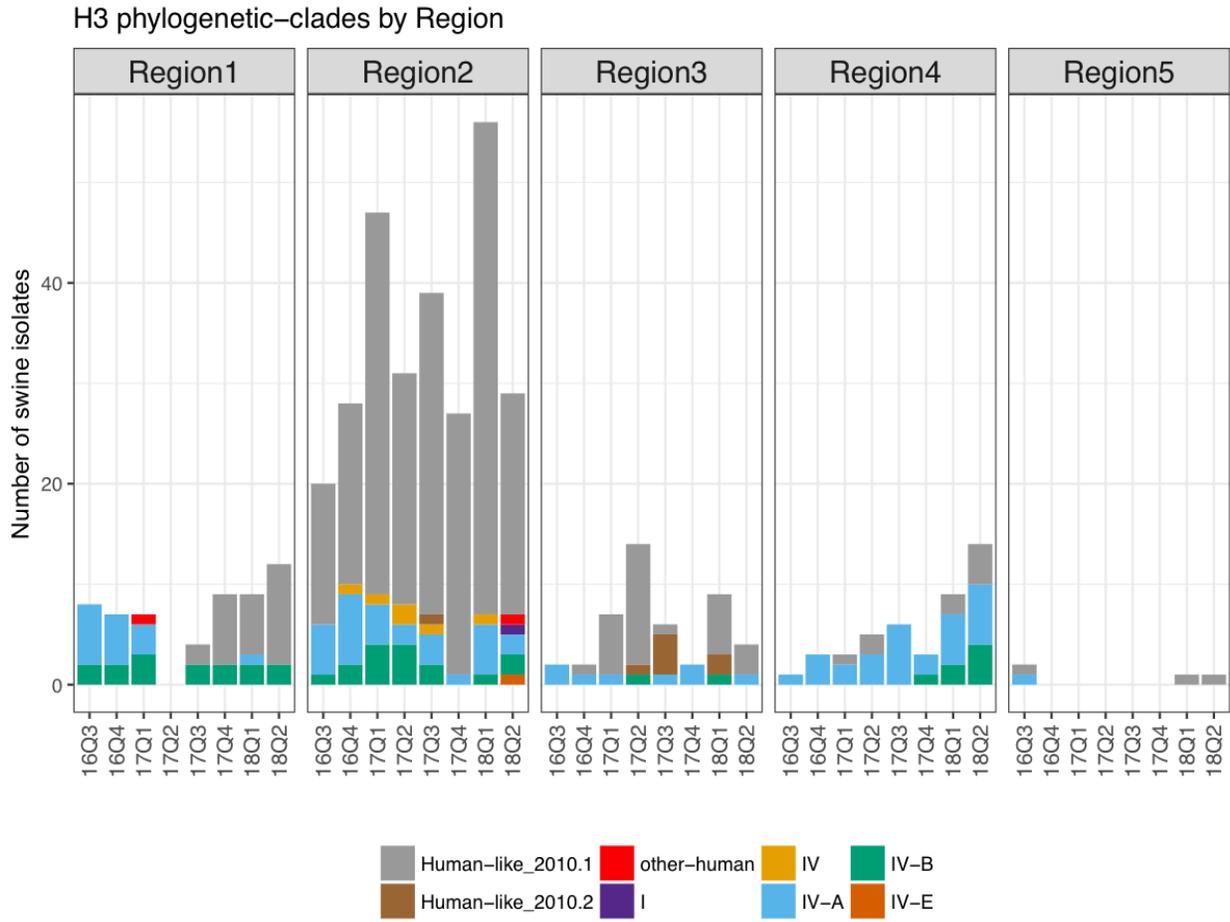


Figure 10. H3 phylo-cluster by region, 2-year summary Q3 FY 2016 to Q2 FY 2018

In Q2 FY 2018, there were 60 H3s. Human-like_2010.2 H3 was detected in Illinois (North Carolina, Oklahoma, Indiana, and Illinois). Cluster I H3 was detected in Iowa, paired with 1998N2. Human-like_2010.1 H3 is the predominant H3.

National phylogenetic NA gene information

Both the N1 and N2 subtypes are found in circulating swine viruses. Classical N1 continued to be the dominant cluster. The 2002-lineage N2 represents 82 percent of N2 collections.

Objective 2. Make influenza isolates from swine available for research and establish a data management system to facilitate genetic analysis of these isolates and related information

A primary goal of IAV-S surveillance is to share selected virus isolates obtained through the surveillance system with public health, animal health, and academic researchers to facilitate genetic analysis and research on viruses of interest.

The USDA’s National Veterinary Services Laboratories (NVSL) in Ames, IA, receives virus isolates acquired through USDA’s IAV-S surveillance and deposits the isolates into the IAV-S virus repository. NAHLN testing labs post the genomic sequences from the isolates into GenBank®, a publically accessible database maintained by the National Institutes for Health. USDA swine surveillance influenza A virus isolates can be identified in GenBank® by their eight-digit barcode designations in the virus name, e.g. A/swine/NY/A01104005/2011(H3N2). The information in GenBank® will usually list “USDA Swine Surveillance” or similar in the Consortium field. The majority of available isolates will have H and N sequenced, but some will also have full genome sequencing completed, including the M gene. The repository contains over 3,000 viruses. NVSL makes these viruses available to the public upon request. Instructions for repository IAV-S isolate requests can be downloaded from here: https://www.aphis.usda.gov/animal_health/lab_info_services/downloads/OrderingIAV-SRepositoryIsolates.pdf

In the second quarter of FY 2018, the NVSL Diagnostic Virology Laboratory provided 64 isolates to five institutions: one governmental, two pharmaceutical, and two international (government). NVSL received 274 isolates into the repository in Q2 for a total of 554 in FY 2018 YTD (Table 4). Table 5 reports the total number of isolates available in the repository by subtype for sharing.

Table 4. Virus isolates received in repository

Virus isolates in the repository	
2018	554
2017	844
2016	1,046
2015	883
2014	765
TOTAL TO DATE	3,818

Objective 3. Select proper isolates for development of relevant diagnostic reagents, updating diagnostic assays, and vaccine seed stock products

USDA makes IAV-S isolates available in the public domain for further research. ARS-NADC conducts research on isolates obtained from the repository and sequences generated from the surveillance system. Genetic sequencing reported to GenBank is available for private corporations, government entities, academia, and other scientific community partners for research and vaccine strain selection and efficacy testing. NVSL and ARS staff are consulted as subject matter experts when necessary.

Table 5. Total number of subtyped isolates available through repository

Subtyped isolates available through repository	
H3N2	1,545
H3N1	16
H1N1	2,027
H1N2	1,828
Mixed	298
TOTAL	5,714

Conclusion

The IAV-S voluntary surveillance system continues to provide insight into the genetic makeup of circulating influenza virus in limited populations of commercial pigs. Genetic information and virus isolates are made publicly available for further research and possible vaccine strain selection and efficacy testing. Influenza A virus in swine remains a dynamic virus with high levels of genetic variability in the hemagglutinin and neuraminidase genes.

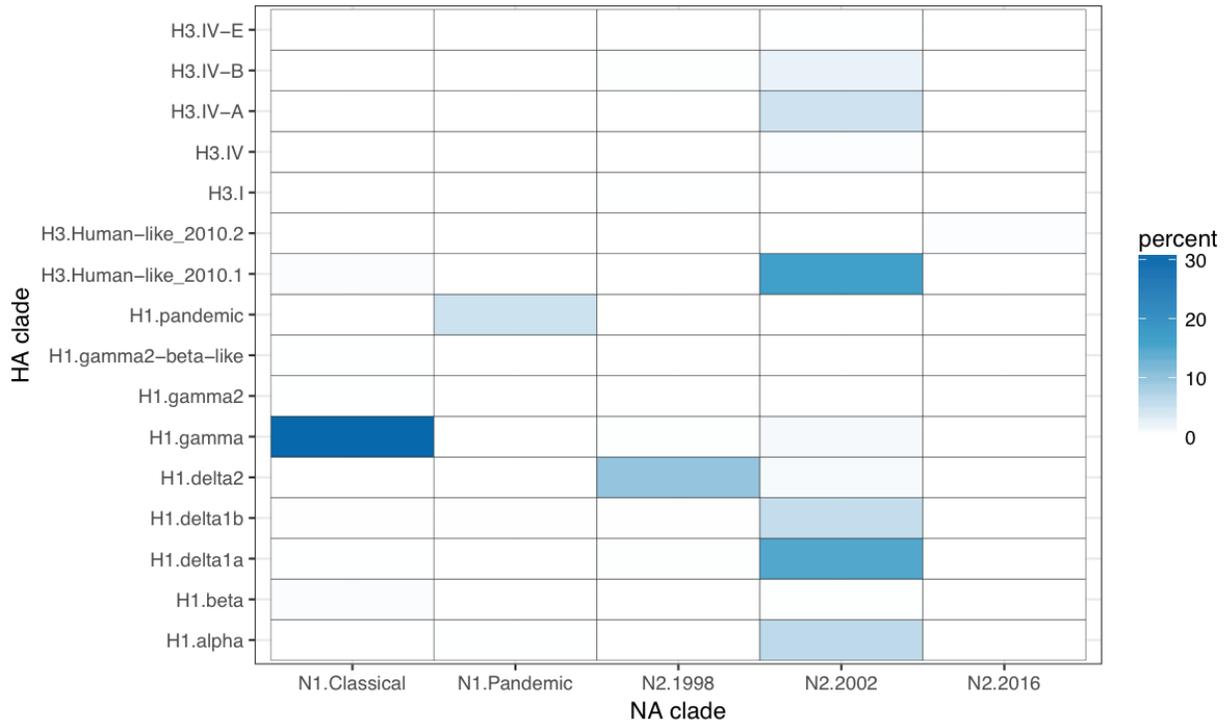
Appendix 1. Regional Charts of HA and NA Combinations by Percentage

The following charts present the percentages of combinations of HA and NA on the national and regional scales based on ARS-NADC phylogenetic analyses. The results are reported from April 2016 to March 2018. These “heat maps” represent the percentage of combinations by using a color gradient; a deeper gradient represents a greater percentage occurrence for a particular HA-NA combination. HA clusters are listed on the left vertical axis of the chart and NA clusters are listed on the bottom horizontal axis. Line up the HA cluster with the corresponding NA cluster to determine the percentage of occurrence of that particular combination.



Total HA & NA combinations – 1,721

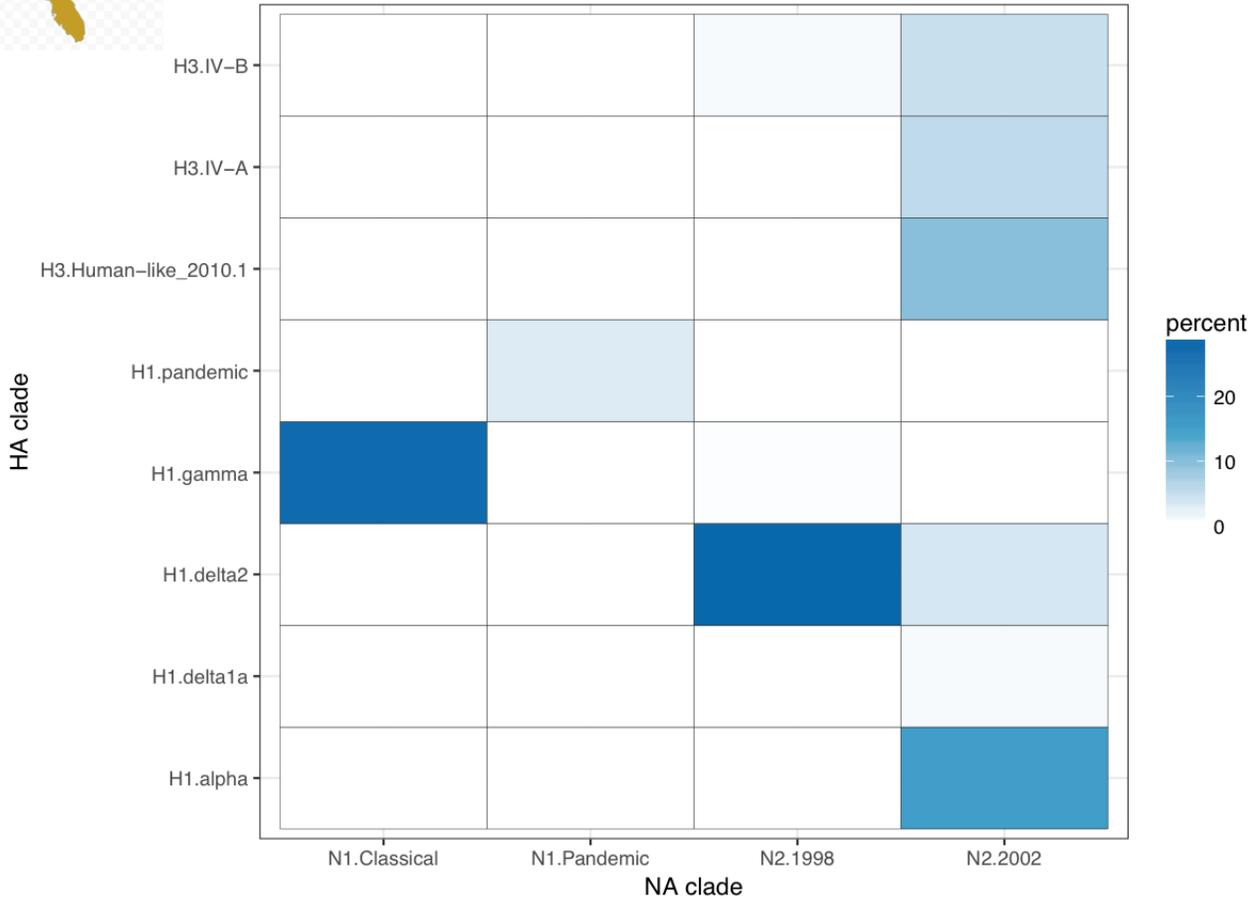
Percentage of HA and NA combinations – Apr 2016 to Mar 2018





Region 1. Total HA & NA combinations – 276

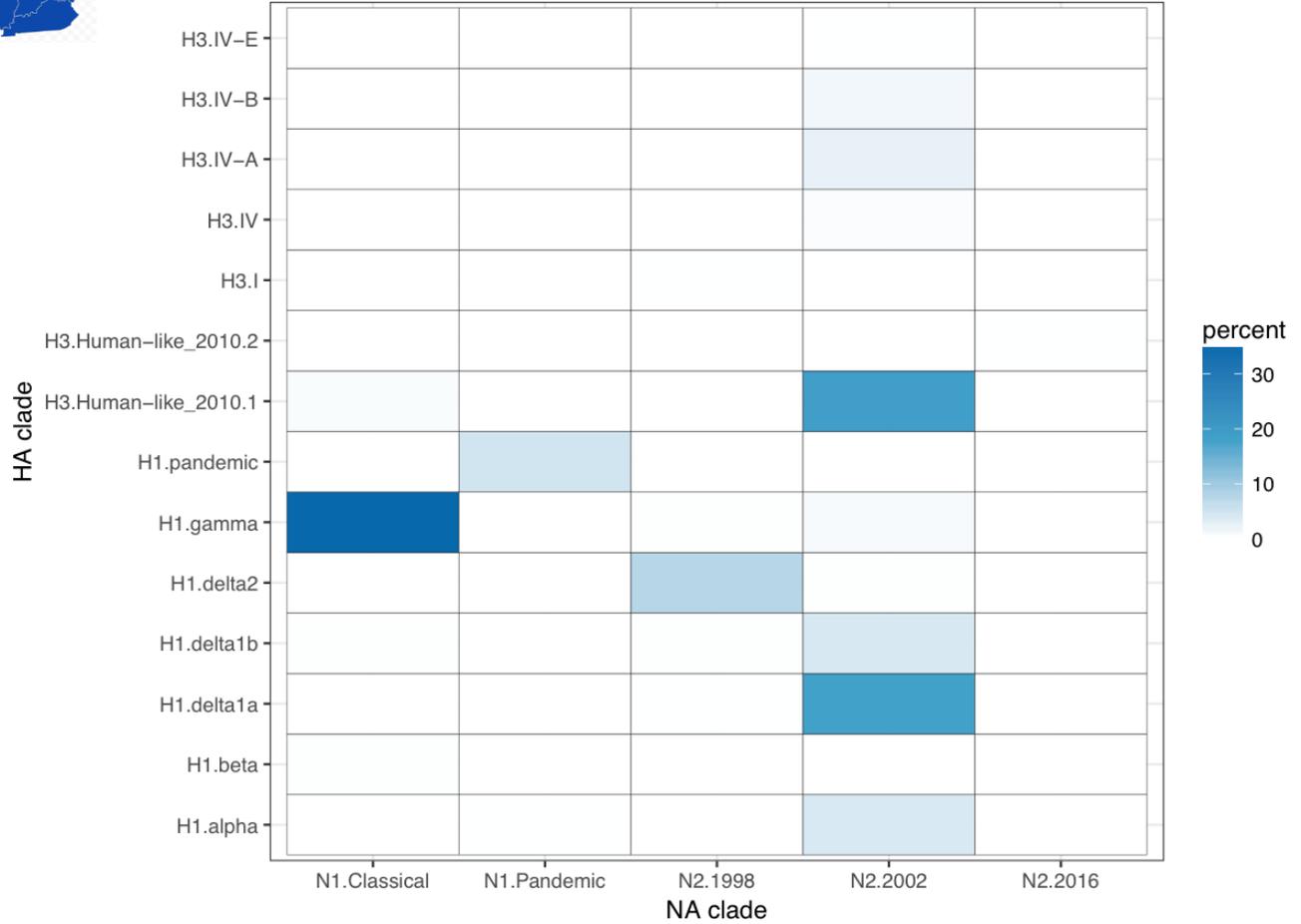
Percentage of HA and NA combinations – Apr 2016 to Mar 2018



Region 2. Total HA & NA combinations – 1,136



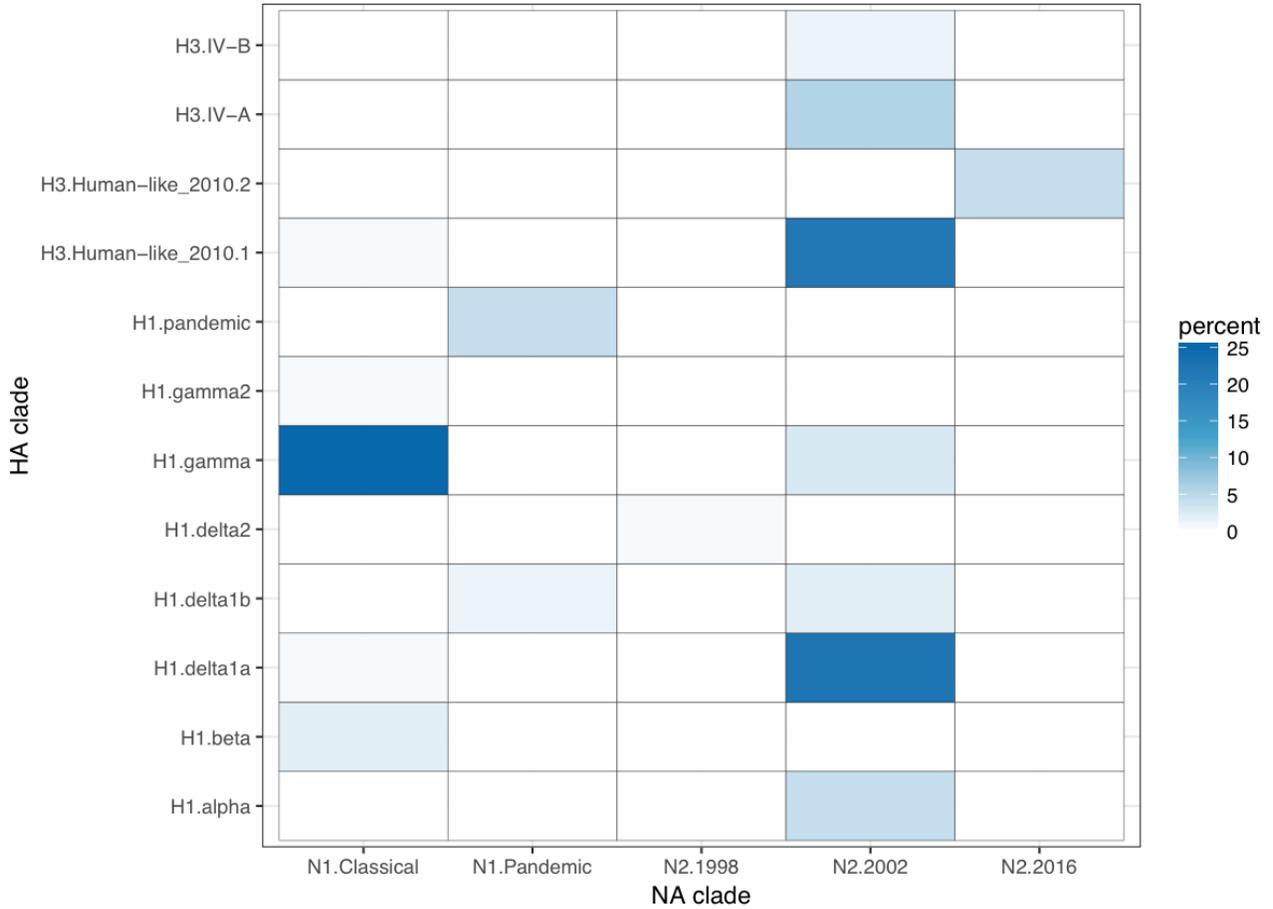
Percentage of HA and NA combinations – Apr 2016 to Mar 2018





Region 3. Total HA & NA combinations – 138

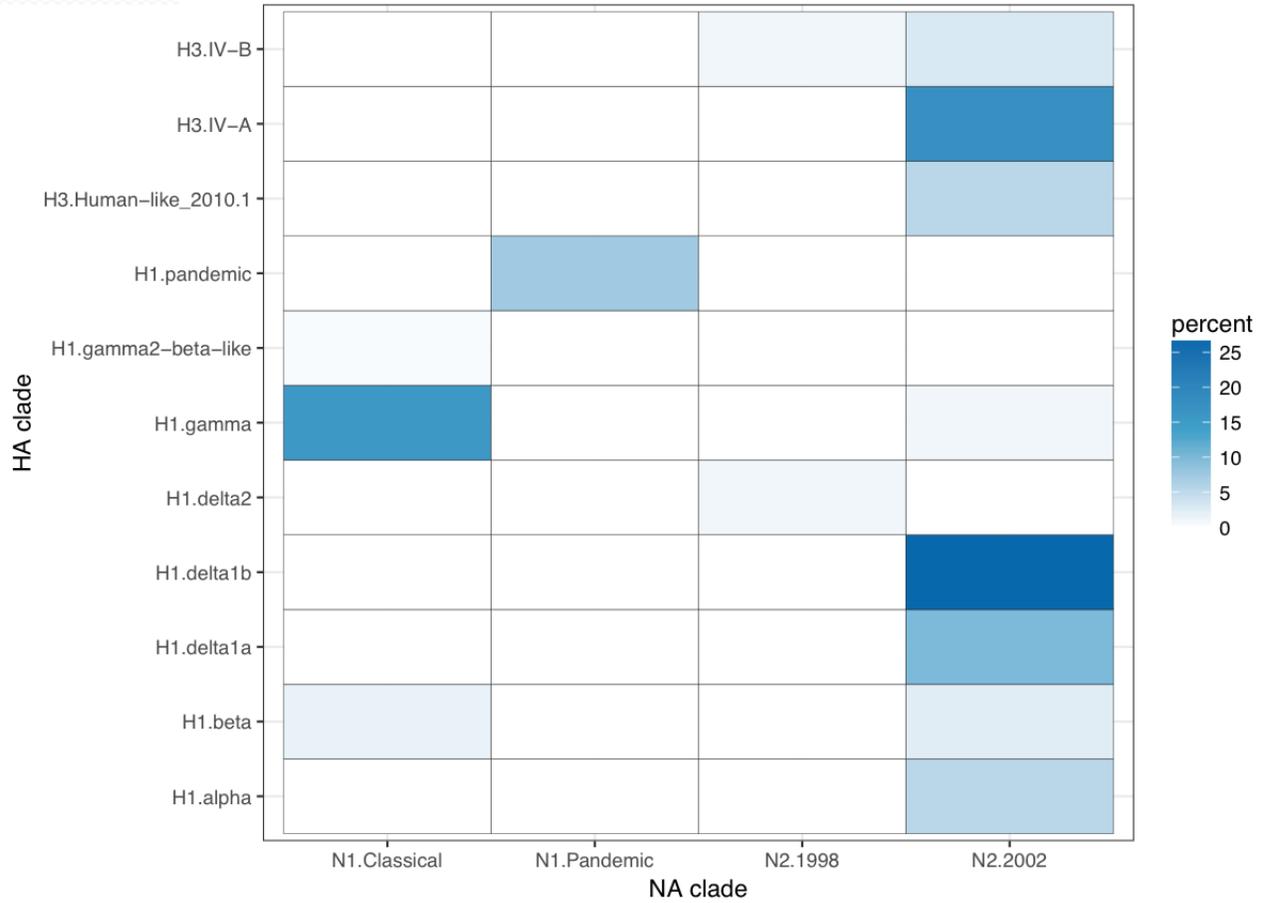
Percentage of HA and NA combinations – Apr 2016 to Mar 2018



Region 4. Total HA & NA combinations – 154



Percentage of HA and NA combinations – Apr 2016 to Mar 2018



Region 5. Total HA & NA combinations – 11



Percentage of HA and NA combinations – Apr 2016 to Mar 2018

