

**Veterinary Diagnostic Laboratory**

Iowa State University  
College of Veterinary Medicine  
Ames, Iowa 50011-1134

Phone: 515-294-1950  
Fax: 515-294-3564

**Final Report**

Report Date: 6/9/2016 10:04:04

Client Phone: 1-910-592-5771	Species: Porcine	Age: 11
Client Fax: 1-910-592-9552	Breed: Unknown	Weight:
Client Account#: 72372	Sex:	Received:
Date Received: 5/18/2016	Previous Case:	2 Oral Fluids
Sample Taken: 5/13/2016	Farm Type: Grow-Finish (or Wean to Finish)	Reason: General
Diagnostics		
Preliminary Report:		

*The ISU VDL has recently changed the swine influenza A virus (IAV-S) HA gene sequencing report format.*

*For IAV H1 viruses, the Influenza Research Database (IRD, [www.fludb.org](http://www.fludb.org)) has built a swine HA(H1) Clade Classification Tool that involves a reference tree using ~100 HA(H1) sequences representing all of the currently defined U.S. swine H1 clades together with representatives of other H1 clades. A query HA(H1) sequence can be compared to the reference H1 sequences to determine its phylogenetic clusters. When a query sequence is unequivocally within the bounds of a single clade, that clade name is assigned to the query sequences. When classification of a query sequence is less certain, a designation including the suffix '-like' is used to indicate its nearest relation. If a query sequence is not clustered with any reference sequences, it is indicated as 'Not Clustered'. Currently, the output includes possible outcomes such as: (1) H1 alpha clade; (2) H1 beta clade; (3) H1 gamma clade; (4) H1 gamma2 clade; (4) H1 pandemic clade; (5) H1 delta 1 clade; (6) H1 delta2 clade; (7) H1 delta-like clade; (8) Not clustered. Note that this tool classifies HA(H1) sequences regardless of NA subtype.*

*For IAV H3 viruses, a swine HA(H3) Clade Classification Tool is not available online yet; the IRD and collaborators are currently working to define the classification system. We perform HA(H3) sequence analysis and tentatively report the outcomes as: (1) H3 cluster I; (2) H3 cluster II; (3) H3 cluster III; (4) H3 cluster IV; (5) Novel swine human-like H3 IAVs identified since 2012; (6) Not clustered.*

The full-length of the HA, NA and Matrix genes were amplified from the SIV isolate and sequenced. The sequences have been deposited to GenBank as required by the SIV surveillance program. Genetic analysis demonstrated the HA gene of the IAV identified in this case is genetically close to that of the **H3 Cluster IV clade**. Please contact the VDL if you have any questions on this report or if you have other influenza A viruses to compare to this one. (6/3/16 jqz/kl)

**KEY:** Tests: FA = Fluorescent Antibody, IHC = Immunohistochemistry, MLV = Modified Live Virus, ORF = Open Reading Frame, PCR = Polymerase Chain Reaction, RFLP = Restriction Fragment Length Polymorphism, VI = Virus Isolation. Agents: BCV = Bovine Coronavirus, BRSV = Bovine Respiratory Syncytial Virus, BVDV = Bovine Viral Diarrhea Virus, CSF = Classical Swine Fever, HPS = *Haemophilus parasuis*, IAV = Influenza A Virus, IBRV = Infectious Bovine Rhinotracheitis Virus, MHP = *Mycoplasma hyopneumoniae*, MHR = *Mycoplasma hyorhinis*, MHS = *Mycoplasma hyosynoviae*, PCV = Porcine Circovirus, PDCV = Porcine Deltacoronavirus, PEDV = Porcine Epidemic Diarrhea Virus, PPV = Porcine Parvovirus, PRCV = Porcine Respiratory Coronavirus, PRRSV = Porcine Reproductive & Respiratory Syndrome Virus, PRV = Pseudorabies Virus, SVA = Senecavirus A, TGEV = Transmissible Gastroenteritis Virus.

Laboratory Result(s)			
<u>Test Ordered</u>	<u>Order Date</u>	<u>Current Status</u>	<u>Complete Date</u>
PCR - Influenza A - USDA program	5/18/2016	Result Released	5/18/2016
PCR - Influenza A subtype - USDA program	5/19/2016	Result Released	5/19/2016
PCR - Influenza A subtype - USDA program	5/24/2016	Result Released	5/31/2016
Sequencing and Analysis - Influenza A - USDA Prog.	5/19/2016	Result Released	6/7/2016
Virus isolation - Influenza A - USDA program	5/18/2016	Result Released	5/27/2016

## Molecular Diagnostic

### PCR - Influenza A - USDA program

<u>Animal ID</u>	<u>Specimen</u>	<u>Ct/Result</u>	<u>Comment</u>
M&M, Tube #1	Oral fluid	22.1/Positive	
1, Tube #2	Oral fluid	21.9/Positive	

### PCR - Influenza A subtype - USDA program

<u>Animal ID</u>	<u>Specimen</u>	<u>Target</u>	<u>Ct/Result</u>	<u>Comment</u>
M&M, Tube #1	Oral fluid	Influenza H1	>38/Negative	
		Influenza H3	25.1/Positive	
		Influenza N1	>38/Negative	
		Influenza N2	26.5/Positive	

### PCR - Influenza A subtype - USDA program

<u>Animal ID</u>	<u>Specimen</u>	<u>Target</u>	<u>Ct/Result</u>	<u>Comment</u>
1, Tube #2	Oral fluid	Influenza H1	>38/Negative	
		Influenza H3	25.0/Positive	
		Influenza N1	>38/Negative	
		Influenza N2	26.6/Positive	

### PCR - Influenza A subtype - USDA program

<u>Animal ID</u>	<u>Specimen</u>	<u>Target</u>	<u>Ct/Result</u>	<u>Comment</u>
1, Tube #2	Virus isolate	Influenza H1	>38/Negative	
		Influenza H3	16.5/Positive	
		Influenza N1	>38/Negative	
		Influenza N2	18.1/Positive	

### Sequencing and Analysis - Influenza A - USDA Prog.

<u>Animal ID</u>	<u>Specimen</u>	<u>Target Gene</u>	<u>GenBank Ref</u>	<u>Comment</u>
1, Tube #2	Virus isolate	NA	KX358884	

#### Nucleotide

ATGAATCCAAATCAAAGATAATAACAATTGGCTCTGTCTCTCTCATCATTGCCACAATATGCT  
 TCCTTATGCAATTGCCATCCTGGTGACTACTGTAACACTGCATTTCAAGCAACATAATTGCGAT  
 TCCTCCCCAAACAACCAAGTAATGTTTTGCGAACCAACAATAATAGAAAGAAACAAAACGGAGAT  
 TGTGTATTTGACCAACACCACTGTAGAGAAGGAAATATGCCCAAACCAGCAGAATACAGAAATT  
 GGTCAAAGCCTCAATGTAACATTACAGGATTTGCACCTTTTTCTAAGGACAATTCAATTCGGCTT  
 TCTGCTGGTGGGACATCTGGGTGACAAGAGAACCTTAWGTGTCATGCGATCCCGCCAAGTGTTA  
 TCAATTTGCCCTTGGGCAGGGAACAACACTAAACAACGGGCATTCAAATGACACTGTACATGATA  
 GGACCCCTTACCGAACCCCTACTGATGAATGAATTGGGTGTTCCATTTCAATTTGGGAACCAGGCAA  
 GTGTGCATAGCATGGTCCAGTTCAAGTTGTCACGATGGGAAAGCATGGCTGCATGTTTGTATAAC  
 TGGGGATGATAATAATGCAACTGCTAGCTTCATTTACAATGGGAGGCTAGTAGATAGTATTGGTT  
 CATGGTCCAAAAATATACTAAGAACCAGGAGTCGGAATGCGTTTGTATTAATGGAAGTTGTACA  
 GTAGTCATGACTGATGGAAGCGCTTCCGGAAAAGCTGATACTAAAATATTATTCATTGAGGAGGG  
 GAAAATCATTCAATATTAGCACGTTGTCAGGAAGTGCGCAGCATGTCGAGGAGTGCTCTTGTTATC  
 CTCGATATCCTGGTGTGAGATGCGTTTGCAGAGACAAGTGGAAAGGCTCCAATAGGCCCATAGTT  
 GATATAAATGTAAAGGATTATAGCATTGTTTCCAGTTATGTATGCTCTGGACTTGTTGGAGACAC  
 ACCCAGGAAAAACGACAGCTTCAGCAGTAGTCATTGCCTAGATCCTAACAATGAAGAAGGTGGTC

ATGGGGTGAAAGGCTGGGCCTTTGATGATGGAAATGACGTGTGGATGGGAAGAACGATCAGCGAG  
 AAGTCACGCTTAGGCTATGAAACCTTCAAAGTCATCAAAGGATGGTCCACACCCAACTCCAAATT  
 ACAGACAAATAGGCAAGTTATAATTGGTAGAGGTAACAGGTCCGGTTATTCTGGTATTTTCTCCG  
 TTGAAGGCAAAACTGCATCAATAGATGCTTTTATGTGGAGTTGATAAGGGGAAGGAAAGAGGAA  
 ACTAAAGTCTGGTGGACTTCAAACAGTATTGTTGTGTTTTGTGGCACCTCAGGTACGTATGGAAC  
 AGGCTCATGGCCTGATGGGGCGGATATCAATCTCATGCCTATATAA

**Sequencing and Analysis - Influenza A - USDA Prog.**

<u>Animal ID</u>	<u>Specimen</u>	<u>Target Gene</u>	<u>GenBank Ref</u>	<u>Comment</u>
1, Tube #2	Virus isolate	HA	KX358883	

**Nucleotide**

ATGAAGACTATCATTGCTTTTAGCTGCATTCTATGTCTGATTTTTGCTCAAAAACCTCCCGGAA  
 GTGACAACAGCATGGCAACGCTGTGCCTAGGACACCACGCAGTGCCAAACGGAACATTAGTGAAA  
 ACAATCACGGATGACCAAATTGAAGTGACTAATGCTACTGAGCTGGTCCAGAATTCCTCAACAGG  
 TAGAATATGCAACAGTCCTCACCAAATCCTTGATGGGAAAAATTGCACACTGATAGATGCTCTAT  
 TGGGGGACCCTCATTGTGATGACTTCCAAACAAGAAATGGGACCTTTTTGTTGAACGAAGCACA  
 GCCTACAGCAACTGTTACCCTTATTATGTGCCAGATTATGCTACCCTTAGGTCACTAATTGCCTC  
 ATCTGGCAACCTGGAATTTACCCAAGAAAGCTTCAATTGGACTGGAGTTGCTCAAGACGGATCAA  
 GCTATGCCTGCAAAAGGGGTCTGTAAAAAGTTTCTTTAGTAGATTGAATTGGTTATATAACTTG  
 AATTACAAGTATCCAGAACAGAACGTAACATGCCAAACAATGACAAATTTGACAAATTGTACAT  
 TTGGGGGGTTTCACCACCCGGGTACGGACAAAGACCAAAACCAACCTATATGTCCAAGCATCAGGGA  
 GAGTTATAGTCTCTACCAAAGAAGCCAACAACTGTAATCCCGAATATCGGGTCTAGACCCTGG  
 GTAAGGGGTGTCTCCAGCATAATAAGCATCTATTGGACGATAGTAAAACCAGGAGACATACTTTT  
 GATTAACAGCACAGGGAATCTAATTGCCCCTCGGGGTTACTTCAAATACAAAGTGGGGAAAGCT  
 CAATAATGAGATCAGATGCACACATTGATGAATGCAACTCTGAATGCATTACTCCAAATGGAAGC  
 ATTCCCAATGACAAACCTTTTCAAATGTAAACAAGATCACATATGGAGCCTGTCCCAGATATGT  
 TAAGCAAAACACCCTGAAATTGGCAACAGGAATGCGGAATGTACCAGAGAAGCAAACCTAGAGGCA  
 TATTCGGCGCAATTGCAGGTTTCATAGAAAATGGTTGGGAGGGAATGGTAGACGGTTGGTACGGT  
 TTCAGGCATCAGAATTCTGAAGGCACAGGACAAGCAGCAGATCTTAAAGCACTCAAGCAGCAAT  
 CAACCAATCACCGGGAACTAAATAGAGTAATCAAGAAAACGAACGAGAAATTCATCAAATCG  
 AAAAGAATTCTCAGAAGTAGAAGGGAGAATTCAGGACCTAGAGAAATACGTTGAAGACACTAAA  
 ATAGATCTCTGGTCTTACAACGCTGAGCTTCTGTTGCCCTGGAGAACCAACATACAATTGATTT  
 AACCGACTCAGAGATGAGCAAACCTGTTGAAAGAACAAGAAGGCAACTGCGGGAAAATGCTGAGG  
 ACATGGGCAATGGTTGCTTCAAATATACCACAAATGTGACAACGCCTGCATAGGATCAATCAGA  
 AATGGAACCTATGACCATGATGTATACAGAAACGAGGCATTAAACAATCGGTTCCAGATCAAAAG  
 TGTTTCAGCTAAAGTCAGGATACAAAGATTGGATCCTATGGATTTCTTTGCCATATCATGCTTTT  
 TGCTTTGTGTTGTTCTGCTGGGGTTTCATTATGTGGGCCTGCCAAAAGGCAACATTAGGTGCAAC  
 ATTTGCATTTAA

**Virology****Virus isolation - Influenza A - USDA program**

<u>Animal ID</u>	<u>Specimen</u>	<u>Result</u>	<u>Comment</u>
1, Tube #2	Oral fluid	Positive	HA 128 A 01775461

**Veterinary Diagnostic Laboratory**

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College of Veterinary Medicine  
Ames, Iowa 50011-1134

Phone: 515-294-1950  
Fax: 515-294-3564

**Final Report**

Report Date: 1/8/2018 15:40:16

Client Phone: ---	Species: Porcine	Age: 3 Weeks
Client Fax: ---	Breed: Unknown	Weight:
Client Account#: 000-00-00	Sex:	Received:
Date Received: 11/21/2017	Previous Case:	Lung
Sample Taken: 11/17/2017	Farm Type: Nursery	Reason: General Diagnostics
Preliminary Report:	Animal ID(s): A	
Accompanying Cases:		

Swine influenza virus surveillance testing results are listed below. (1/8/18 jqz/cl)

<u>Test Ordered</u>	<u>Laboratory Result(s)</u>	<u>Current Status</u>	<u>Complete Date</u>
PCR - Influenza A subtype - USDA program	11/22/2017	Result Released	11/22/2017
PCR - Influenza A subtype - USDA program	11/30/2017	Result Released	11/30/2017
^ Sequencing and Analysis - Influenza A - USDA Prog.	11/30/2017	Result Released	12/15/2017
Virus isolation - Influenza A - USDA program	11/21/2017	Result Released	11/30/2017
PCR - Influenza A - USDA program	11/22/2017	Result Released	11/22/2017

^ Testing performed in part or in total at a Referral Laboratory.

**Molecular Diagnostic****PCR - Influenza A - USDA program**

<u>Animal ID</u>	<u>Specimen</u>	<u>Ct/Result</u>	<u>Comment</u>
A, Tube #1	Lung	17.5/Positive	

**PCR - Influenza A subtype - USDA program**

<u>Animal ID</u>	<u>Specimen</u>	<u>Target</u>	<u>Ct/Result</u>	<u>Comment</u>
A, Tube #1	Lung	Influenza H1	>40/Negative	
		Influenza H3	19.7/Positive	
		Influenza N1	>40/Negative	
		Influenza N2	20.4/Positive	

**PCR - Influenza A subtype - USDA program**

<u>Animal ID</u>	<u>Specimen</u>	<u>Target</u>	<u>Ct/Result</u>	<u>Comment</u>
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A, Tube #1	Virus isolate	Influenza H1	>40/Negative
		Influenza H3	18.1/Positive
		Influenza N1	>40/Negative
		Influenza N2	20.5/Positive

**Sequencing and Analysis - Influenza A - USDA Prog.**

<u>Animal ID</u>	<u>Specimen</u>	<u>Target Gene</u>	<u>GenBank Ref</u>	<u>Comment</u>
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<u>Animal ID</u>	<u>Specimen</u>	<u>Target Gene</u>	<u>GenBank Ref</u>	<u>Comment</u>
A, Tube #1	Virus isolate	HA	MG681969	

**Sequencing and Analysis - Influenza A - USDA Prog.**

ATGAAGACTATCATTGCTTTTAGCTGCATTCTATGTCTGATTTTTGCTCAAAAACCTCCCGGAAGTGACAACAGCATG  
GCAACGCTGTGCCTAGGACACCACGCAGTACCAAACGGAACATTAGTGAAAACAATCACGGATGACCAAATTGAAGT  
GACTAACGCTACTGAGCTGGTCCAGAGTTCTCAACAGGTAGAATATGCAACAGTCCTACCAAATCCTTGATGGGA  
AAAATTGCACATTGATAGATGCTCTATTGGGGGACCCTCATTGTGATGACTTCCAAAACAAGAAATGGGACCTTTTG  
TTGAACGAAGCACAGCCTACAGCAACTGTTACCCTTATTATGTGCCAGATTATGCTACCCTTAGGTCACTAATTGCCT  
CATCTGGCAACCTGGAATTTACCCAAGAAAGCTTCAATTGGACTGGAGTTACTCAAGACGGATCAAGCTATGCCTGC  
AGAAAGGGTTCTGTAAACAGTTTTTTTTAGTAGATTGAATTGGTTGTATAACTTGAATTACAAGTATCCAGAACAGAACG  
TAACTATGCCAAACAATGACAAATTTGACAACTGTACATTTGGGGGGTTCACCACCCGGGTACGGACAAAGACCAA  
ACCTACCTATATGTCCAAGCACCAGGGAGAGTTATGGTCTCTACCAAAGAAGCCAACAACTATAATCCCGAATATC  
GGGTCTAGACCCTGGGTGAGGGGTGTCTCCAGCATAATAAGCATCCATTGGACGATAGTAAAACCAGGAGACATAC  
TTTTGATTAAACAGCACAGGGAATCTAATTGCCCCTAGGGGTACTTCAAATAACAAGTGGGAAAAGCTCAATAATGA  
GATCAGATGCACACATTGATGAATGCAATTCTGAATGCATTACTCCAAATGGAAGCATTCCCAATGACAAACCTTTTC  
AAAACGTAAACAAGATCACATATGGAGCCTGTCCAGATATGTTAAGCAAAACACCCTGAAATTGGCAACAGGAATG  
CGGAATGTACCAGAGAAGCAAACCAGAGGCATTTTCGGCGCGATTGCAGGTTTCATAGAAAATGGTTGGGAGGGAA  
TGGTAGACGGTTGGTACGGTTTCAGGCATCATAATTCTGAAGGCACAGGACAAGCAGCAGATCTTAAAGCACTCAA  
GCGGCAATCAACCAAAATCACAGGGAACTAAATAGAGTAATCAAGAAAACAAACGAGAAATTCATCAAATCGAAAAA  
GAATTCTCAGAAGTAGAAGGGAGAATTCAGGACCTAGAGAAATACGTTGAAGACACTAAAATAGATCTCTGGTCTTAC  
AACGCTGAGCTTCTTGTGGCCTGGAGAACCAACATACAATTGATTTAACCGACTCAGAGATGAGCAAACTGTTCGA  
AAGAACAAGAAGGCAACTGCGGGAAAATGCTGAGGACATGGGCAATGGTTGCTTCAAATATACCACAAATGTGACA  
ACGCCTGCATAGGATCAATCAGAAATGGAACCTTATGACCATGATGTATACAGAAGCGAGGCATTAAACAATCGGTTTC  
CAGATCAAAGGTGTTTCAGCTAAAGTCAGGATACAAAGATTGGATTCTATGGATTTCTTTGCCATATCATGCTTTTTG  
CTTTGTGTTGTTCTGCTGGGGTTCATTATGTGGGCCTGCCAAAAGGCAACATTAGATGCAACATTTGCATTTAA

**Veterinary Diagnostic Laboratory**

Iowa State University  
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 Ames, Iowa 50011-1134  
 Phone: 515-294-1950  
 Fax: 515-294-3564

**Final Report**

Report Date: 4/4/2018 13:13:12

Client Phone: 1-608-445-7149	Species: Porcine	Age: Adult
Client Fax: ---	Breed: Unknown	Weight:
Client Account#: 404207	Sex:	Received:
Date Received: 3/2/2018	Previous Case:	12 Nasal Swabs
Sample Taken: 3/1/2018	Farm Type: Breeding Herd	Reason: General Diagnostics
Preliminary Report:	Animal ID(s): , , ,	
Accompanying Cases:		

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New coordination statements for IAV-S HA sequencing report (updated on 10May2017)

*The ISU VDL has recently changed the swine influenza A virus (IAV-S) HA gene sequencing report format.*

*For IAV H1 viruses, the Influenza Research Database (IRD, [www.fludb.org](http://www.fludb.org)) has built a swine HA(H1) Clade Classification Tool that involves a reference tree using ~100 HA(H1) sequences representing all of the currently defined U.S. swine H1 clades together with representatives of other H1 clades. A query HA(H1) sequence can be compared to the reference H1 sequences to determine its phylogenetic clusters. When a query sequence is unequivocally within the bounds of a single clade, that clade name is assigned to the query sequences. When classification of a query sequence is less certain, a designation including the suffix '-like' is used to indicate its nearest relation. If a query sequence is not clustered with any reference sequences, it is indicated as 'Not Clustered'. Currently, the output of the U.S. swine H1 classification includes possible outcomes such as: (1) H1 alpha clade; (2) H1 beta clade; (3) H1 gamma clade; (4) H1 gamma2 clade; (4) H1 pandemic clade; (5) H1 delta 1 clade; (6) H1 delta2 clade; (7) H1 delta-like clade; (8) Not clustered. The output of the global swine H1 classification includes 1A classical swine lineage (1A.1, 1A.2, and 1A.3 and their subclades), 1B human seasonal lineage (1B.1 and 1B.2 and their subclades), and 1C Eurasian avian lineage (1C.1 and 1C.2 and their subclades). We report both the U.S. swine H1 lineages and the global swine H1 lineages with the latter in parenthesis. Note that this tool classifies HA(H1) sequences regardless of NA subtype.*

*For IAV H3 viruses, a swine HA(H3) Clade Classification Tool is not available online yet; the IRD and collaborators are currently working to define the classification system. We perform HA(H3) sequence analysis and tentatively report the outcomes as: (1) H3 cluster I; (2) H3 cluster II; (3) H3 cluster III; (4) H3 cluster IV; (5) Novel human-to-swine H3 IAVs since 2011/2012; (6) Novel human-to-swine H3 IAVs since 2016/2017; (7) Not clustered.*

The full-length of the HA and NA genes were amplified from the IAV isolate and sequenced. The sequences have been deposited to GenBank as required by the IAV surveillance program. Genetic comparison to the Influenza Research Database reference sequences revealed that the H1 gene of the IAV identified in this case is genetically close to that of the ????. Please contact the VDL if you have any questions on this report or if you have other influenza A viruses to compare to this one. (03/29/18 jqz/ks)

**KEY:** Tests: FA = Fluorescent Antibody, IHC = Immunohistochemistry, ISH = *in situ* hybridization, MALDI = Matrix-assisted laser desorption/ionization, MLV = Modified Live Virus, ORF = Open Reading Frame, PCR = Polymerase Chain Reaction, RFLP = Restriction Fragment Length Polymorphism, VI = Virus Isolation. Agents: BCV = Bovine Coronavirus, BRSV = Bovine Respiratory Syncytial Virus, BVDV = Bovine Viral Diarrhea Virus, CSF = Classical Swine Fever, HPS = *Haemophilus parasuis*, IAV = Influenza A Virus, IBRV = Infectious Bovine Rhinotracheitis Virus, MHP = *Mycoplasma hyopneumoniae*, MHR = *Mycoplasma hyorhinis*, MHS = *Mycoplasma hyosynoviae*, PCV = Porcine Circovirus, PDCV = Porcine Deltacoronavirus, PEDV = Porcine Epidemic Diarrhea Virus, PPV = Porcine Parvovirus, PRCV = Porcine Respiratory Coronavirus, PRRSV = Porcine Reproductive & Respiratory Syndrome Virus, PRV = Pseudorabies Virus, SVA = Senecavirus A, TGEV = Transmissible Gastroenteritis Virus.

Laboratory Result(s)				
<u>Test Ordered</u>		<u>Order Date</u>	<u>Current Status</u>	<u>Complete Date</u>
PCR - M. hyopneumoniae		3/2/2018	Result Released	3/2/2018
PCR - Influenza A subtype - USDA program		3/5/2018	Result Released	3/5/2018
PCR - Influenza A subtype - USDA program		3/15/2018	Result Released	3/15/2018
^ Sequencing and Analysis - Influenza A - USDA Prog.		3/6/2018	Result Released	4/3/2018
Virus isolation - Influenza A - USDA program		3/6/2018	Result Released	3/15/2018
PCR - Influenza A subtype		3/5/2018	Result Released	3/5/2018
PCR - Influenza A - USDA program		3/2/2018	Result Released	3/2/2018

^ Testing performed in part or in total at a Referral Laboratory.

## Molecular Diagnostic

### PCR - Influenza A subtype

<u>Animal ID</u>	<u>Specimen</u>	<u>Target</u>	<u>Ct/Result</u>	<u>Comment</u>
GA [1 - 3]	Nasal swab	Influenza H1	28.4/Positive	
		Influenza H3	>40/Negative	
		Influenza N1	29.0/Positive	
		Influenza N2	>40/Negative	

### PCR - Influenza A subtype

<u>Animal ID</u>	<u>Specimen</u>	<u>Target</u>	<u>Ct/Result</u>	<u>Comment</u>
GB [4 - 6]	Nasal swab	Influenza H1	30.0/Positive	
		Influenza H3	>40/Negative	
		Influenza N1	30.3/Positive	
		Influenza N2	>40/Negative	

### PCR - Influenza A subtype

<u>Animal ID</u>	<u>Specimen</u>	<u>Target</u>	<u>Ct/Result</u>	<u>Comment</u>
GC [7 - 9]	Nasal swab	Influenza H1	26.9/Positive	
		Influenza H3	>40/Negative	
		Influenza N1	27.6/Positive	
		Influenza N2	>40/Negative	

### PCR - Influenza A subtype - USDA program

<u>Animal ID</u>	<u>Specimen</u>	<u>Target</u>	<u>Ct/Result</u>	<u>Comment</u>
GD [10 - 12]	Nasal swab	Influenza H1	25.9/Positive	
		Influenza H3	>40/Negative	
		Influenza N1	26.8/Positive	
		Influenza N2	>40/Negative	

### PCR - Influenza A subtype - USDA program

<u>Animal ID</u>	<u>Specimen</u>	<u>Target</u>	<u>Ct/Result</u>	<u>Comment</u>
GD [10 - 12]	Virus isolate	Influenza H1	19.5/Positive	
		Influenza H3	>40/Negative	
		Influenza N1	19.0/Positive	
		Influenza N2	>40/Negative	

**Sequencing and Analysis - Influenza A - USDA Prog.**

<u>Animal ID</u>	<u>Specimen</u>	<u>Target Gene</u>	<u>GenBank Ref</u>	<u>Comment</u>
GD [10 - 12]	Virus isolate	NA	MH156834	

**Nucleotide**

ATGAATCCAAACCAAAAGATAATAACCATTGGTTTCGATCTGTATGACAATTGGAATGGCTAACT  
 TAATATTACAAATTGGAAACATAATCTCAATATGGGTTAGCCACTCAATTCAAATTGGAAATCAA  
 AGCCAGATTGAAACATGCAATCAAAGCGTCATTACTTATGAAAACAACACTTGGGTAAATCAGAC  
 ATATGTTAACATCAGCAACACCAACTTTGCTGCTAGACAGTCAGTGGCTTCCGTGAAATTAGCGG  
 GCAATTCCTCTCTCTGCCCTGTTAGTGGATGGGCTATATACAGTAAAGACAACAGTGTAAGAATC  
 GGTTCCAAGGGGGATGTGTTTGTCTAAGGGAACCATTTCATATCATGCTCTCCCTTGGAATGCAG  
 AACCTTCTTCTTGACTCAAGGGGCTTTGCTAAATGACAAACATTCCAATGGAACCATTAAGACA  
 GGAGCCCATATCGAACCTAATGAGCTGTCCTATTGGTGAAGTTCCTCTCCATACAACTCAAGA  
 TTTGAGTCAGTCGCTTGGTCAGCAAGTGCTTGTCTATGATGGCACCATTGGCTAACAATTGGAAT  
 TTCTGGCCAGACAGTGGGGCAGTGGCTGTGTTAAATACAATGGCATAATAACAGACACTATCA  
 AGAGTTGGAGGAACAATATATTGAGAACAAGAGTCTGAATGTGCATGTGTAAATGGTTCTTGC  
 TTTACCATAATGACCGATGGACCAAGTGATGGACAGGCCTCATACAAAATCTTCAGAATAGAAAA  
 GGGAAAGATAATCAAATCAGTCGAAATGAAAGCCCCCTAATTATCACTATGAGGAATGCTCCTGTT  
 ATCCTGATTCTAGTGAATCAGTGTGTGTGCAGGGATAACTGGCATGGCTCGAATCGACCGTGG  
 GTGTCTTTCAACCAGAATCTGGAATATCAGATGGGATACATATGCAGTGGGGTTTTTCGGAGACAA  
 TCCACGCCCTAATGATAAGACAGGCAGTGTGTGGTCCAGTATCGTCTAATGGAGCAAATGGAGTAA  
 AAGGATTTTCATTCAAATACGGCAATGGTGTGGTATAGGAGGAGAACTAAGAGCATTAGTTCAAGA  
 AAAGGTTTTGAGATGATTTGGGATCCGAATGGATGGACTGGGACTGACAATAAATTCTCAATAAA  
 GCAAGATATCGTAGGAATAAATGAGTGGTCAGGGTATAGCGGGAGTTTTGTTTCAGCATCCAGAAC  
 TAACAGGGCTGGATTGTATAAGACCTTGCTTCTGGGTTGAACTAATAAGAGGACGACCCGAAGAG  
 AAAACAATCTGGACTAGCGGGAGCAGCATATCCTTTTGTGGTGTAGACAGTGACACTGTGGGTTG  
 GTCTTGGCCAGACGGTGCTGAGTTGCCATTTACCATTGACAAGTAA

**Sequencing and Analysis - Influenza A - USDA Prog.**

<u>Animal ID</u>	<u>Specimen</u>	<u>Target Gene</u>	<u>GenBank Ref</u>	<u>Comment</u>
GD [10 - 12]	Virus isolate	HA	MH156833	

**Nucleotide**

ATGAAGGCAATACTAGTAGTTCTGCTGTATACATTTACAACCGCAAATGCAGACACATTATGTA  
 TAGGTTATCATGCGAACAATTCAACAGACACTGTAGACACAGTACTAGAAAAGAATGTAACAGTA  
 ACACACTCTGTAAATCTTCTGGAAGACAAGCATAACGGAAAACCTATGCAAACCTAGGAGGGGTAGC  
 CCCATTGCATTTGGGTAAATGTAACATTGCTGGCTGGATCCTGGGAAATCCAGAGTGTGAATCAC  
 TCTCCACAGCAAGATCATGGTCCTACATTGTGGAACATCTAATTCAGACAATGGAACGTGTTAC  
 CCAGGAGATTTTCATCAATTATGAGGAGCTAAGAGAGCAACTGAGCTCAGTGTCTCATTTGAAAG  
 GTTTGAAATATTTCCCAAGACAAGTTCATGGCCTAATCATGACTCGAACAAAGGTGTAACGGCAG  
 CATGTCCTCACGCTGGAGCAAAAAGCTTCTACAAAACTTGATATGGCTGGTTAAAAAAGGAAAT  
 TCATACCCAAAGCTCAACCAAACCTACATTAATGATAAAGGGAAAGAAAGTCTCTGCTGTGGGG  
 CATTACCATCCACCTACTACTGCTGACCAACAAAGTCTCTATCAGAATGCAGATGCATATGTTT  
 TTGTGGGGACATCAAGATACAGCAAGAAGTTCAAGCCGGAAATAGCAACAAGACCCAAAGTGAGG  
 GATCAAGAAGGGAGAATGAACCTATTACTGGACACTAGTAGAGCCGGGAGACAAAATAACATTCTGA  
 AGCAACTGGAAATCTAGTGGTACCGAGATATGCATTACCAATGGAAAGAAATGCTGGATCTGGTA  
 TTATCATTTTCAGATACGCCAGTCCACGATTGCAATACAACCTTGTGAGACAACCGAGGGTGCTATA  
 AACACCAGCCTCCCATTTTCAGAATGTACATCCGGTCACAATTGGGAAATGTCCAAAGTATGTAAA  
 AAGCACAAAATTGAGGCTGGCCACAGGATTGAGGAATGTCCCGTCTATTCAATCTAGAGGCCTAT  
 TCGGGGCCATTGCCGGCTTCATTGAAGGGGGGTGGACAGGGATGGTAGATGGATGGTACGGTTAT  
 CACCATCAAAATGAGCAGGGGTGAGGATATGCAGCCGATCTGAAGAGCACACAAAATGCCATTGA  
 CAAGATTACTAACAAAGTAAATTCTGTTATTGAAAAGATGAACACACAGTTCACAGCAGTGGGTA  
 AAGAGTTCAACCACCTTGAAAAAGAATAGAGAATCTAAATAAAAAAGTTGATGATGGTTTCCTG  
 GACATTTGGACTTACAATGCCGAACCTGTTGGTTCTACTGGAAAATGAAAGAACTTTGGACTATCA  
 CGATTCAAATGTGAAGAACTTGATGAAAAAGTAAGAAACCAGTTAAAAACAATGCCAAGGAAA  
 TTGGAACGGCTGCTTTGAATTTTATCACAATGCGATAACACATGCATGGAAAGTGTCAAGAAT  
 GGGACTTATGACTACCCAAAATACTCAGAGGAAGCAAAATTAACAGAGAAAAAATAGATGGAGT  
 AAAGCTGGAATCAACAAGGATCTACCAGATTTTGGCGATCTATTCAACTGTCGCCAGTTCATTGG  
 TACTGGTAGTCTCCCTGGGGGCAATCAGCTTCTGGATGTGCTCTAATGGGTCTCTACAATGTAGA  
 ATATGTATTTAA



**PCR - Influenza A - USDA program**

<u>Animal ID</u>	<u>Specimen</u>	<u>Ct/Result</u>	<u>Comment</u>
GA [1 - 3]	Nasal swab	26.5/Positive	
GB [4 - 6]	Nasal swab	28.1/Positive	
GC [7 - 9]	Nasal swab	25.3/Positive	
GD [10 - 12]	Nasal swab	24.1/Positive	

**Virology****Virus isolation - Influenza A - USDA program**

<u>Animal ID</u>	<u>Specimen</u>	<u>Result</u>	<u>Comment</u>
GD [10 - 12]	Nasal swab	Positive	HA:64 A02154738

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Patient pool's member(s), # means tube number:

GA [1 - 3] : (#1 ) , (#2 ) , (#3 )  
GB [4 - 6] : (#4 ) , (#5 ) , (#6 )  
GC [7 - 9] : (#7 ) , (#8 ) , (#9 )  
GD [10 - 12] : (#10 ) , (#11 ) , (#12 )

**Veterinary Diagnostic Laboratory**

Iowa State University  
College of Veterinary Medicine  
Ames, Iowa 50011-1134  
Phone: 515-294-1950  
Fax: 515-294-3564

**Final Report**

Report Date: 9/18/2018 13:04:03

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Client Phone: 1-479-290-7740	Species: Porcine	Age: 6 Weeks
Client Fax: 1-479-290-3815	Breed: Unknown	Weight:
Client Account#: 88893	Sex:	Received:
Date Received: 8/22/2018	Previous Case:	Fresh & Fixed tissue
Sample Taken: 8/21/2018	Farm Type: Other	Reason: General Diagnostics
Preliminary Report:	Animal ID(s): 1	
Accompanying Cases:		

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**History:** Respiratory signs**Gross Pathology:** None reported**Histopathology:**

- Lung: Moderate congestion and rare intra-alveolar hemorrhage are present in all sections. No inflammatory infiltrate is apparent in any section.
- Lymph node: Moderate hemorrhage is present within the parenchyma in one section.
- Liver, kidney, spleen: Evaluated sections are unremarkable.

**Ancillary Diagnostic Tests:** See completed results below

- Influenza PCR: **Positive**

**Laboratory Diagnosis:**

- Influenza detected in lung via PCR

**Comments:**

- Changes in submitted tissues are mild and non-specific, but do not correlate with reported respiratory disease. Influenza was detected in the lung via PCR, but lesions suggestive of an epitheliotropic viral infection are not apparent. Please take impressions of clinical disease into account when interpreting this finding.
- Please correlate clinically and contact the laboratory if further testing is desired or questions arise. (9/18/18 drl/clm)

**SUPPLEMENTAL REPORT**

*The ISU VDL has recently changed the swine influenza A virus (IAV-S) HA gene sequencing report format.*

For IAV H1 viruses, the Influenza Research Database (IRD, [www.fludb.org](http://www.fludb.org)) has built a swine HA(H1) Clade Classification Tool that involves a reference tree using ~100 HA(H1) sequences representing all of the currently defined U.S. swine H1 clades together with representatives of other H1 clades. A query HA(H1) sequence can be compared to the reference H1 sequences to determine its phylogenetic clusters. When a query sequence is unequivocally within the bounds of a single clade, that clade name is assigned to the query sequences. When classification of a query sequence is less certain, a designation including the suffix '-like' is used to indicate its nearest relation. If a query sequence is not clustered with any reference sequences, it is indicated as 'Not Clustered'. Currently, the output of the U.S. swine H1 classification includes possible outcomes such as: (1) H1 alpha clade; (2) H1 beta clade; (3) H1 gamma clade; (4) H1 gamma2 clade; (4) H1 pandemic clade; (5) H1 delta1 clade; (6) H1 delta2 clade; (7) H1 delta-like clade; (8) Not clustered. The output of the global swine H1 classification includes 1A classical swine lineage (1A.1, 1A.2, and 1A.3 and their subclades), 1B human seasonal lineage (1B.1 and 1B.2 and their subclades), and 1C Eurasian avian lineage (1C.1 and 1C.2 and their subclades). We report both the U.S. swine H1 lineages and the global swine H1 lineages with the latter in parenthesis. Note that this tool classifies HA(H1) sequences regardless of NA subtype.

For IAV H3 viruses, a swine HA(H3) Clade Classification Tool is not available online yet; the IRD and collaborators are currently working to define the classification system. We perform HA(H3) sequence analysis and tentatively report the outcomes as: (1) H3 cluster I; (2) H3 cluster II; (3) H3 cluster III; (4) H3 cluster IV; (5) Novel human-to-swine H3 IAVs since 2011/2012; (6) Novel human-to-swine H3 IAVs since 2016/2017; (7) Not clustered.

The full-length of the HA and NA genes were amplified from the IAV isolate and sequenced. The sequences have been deposited to GenBank as required by the IAV surveillance program. H3 gene of the IAV identified in this case is genetically close to that of the ????. Please contact the VDL if you have any questions on this report or if you have other influenza A viruses to compare to this one. (09/13/18 jqz/ks)

**KEY:** Tests: FA = Fluorescent Antibody, IHC = Immunohistochemistry, ISH = *in situ* hybridization, MALDI = Matrix-assisted laser desorption/ionization, MLV

= Modified Live Virus, ORF = Open Reading Frame, PCR = Polymerase Chain Reaction, RFLP = Restriction Fragment Length Polymorphism, VI = Virus Isolation. Agents: BCV = Bovine Coronavirus, BRSV = Bovine Respiratory Syncytial Virus, BVDV = Bovine Viral Diarrhea Virus, CSF = Classical Swine Fever, HPS = *Haemophilus parasuis*, IAV = Influenza A Virus, IBRV = Infectious Bovine Rhinotracheitis Virus, MHP = *Mycoplasma hyopneumoniae*, MHR = *Mycoplasma hyorhinis*, MHS = *Mycoplasma hyosynoviae*, PCV = Porcine Circovirus, PDCV = Porcine Deltacoronavirus, PEDV = Porcine Epidemic Diarrhea Virus, PPV = Porcine Parvovirus, PRCV = Porcine Respiratory Coronavirus, PRRSV = Porcine Reproductive & Respiratory Syndrome Virus, PRV = Pseudorabies Virus, SVA = Senecavirus A, TGEV = Transmissible Gastroenteritis Virus.

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Laboratory Result(s)			
Test Ordered	Order Date	Current Status	Complete Date
Culture Summary	8/23/2018	Result Released	8/24/2018
Routine Culture - Tissue	8/22/2018	Result Released	8/24/2018
PCR - <i>Haemophilus parasuis</i>	8/22/2018	Result Released	8/23/2018
PCR - Influenza A subtype - USDA program	8/22/2018	Result Released	8/23/2018
PCR - Influenza A subtype - USDA program	8/30/2018	Result Released	8/31/2018
^ Sequencing and Analysis - Influenza A - USDA Prog.	8/24/2018	Result Released	9/14/2018
Virus isolation - Influenza A - USDA program	8/23/2018	Result Released	8/31/2018
Hematoxylin and Eosin Slides	8/22/2018	Result Released	8/22/2018
PCR - Influenza A - USDA program	8/22/2018	Result Released	8/22/2018

-

^ Testing performed in part or in total at a Referral Laboratory.

#### PCR - Influenza A subtype - USDA program

Animal ID	Specimen	Target	Ct/Result	Comment
1, SID #1	Lung	Influenza H1	>=40/Negative	
		Influenza H3	26.8/Positive	
		Influenza N1	>=40/Negative	
		Influenza N2	25.0/Positive	

#### PCR - Influenza A subtype - USDA program

Animal ID	Specimen	Target	Ct/Result	Comment
1, SID #1	Virus isolate	Influenza H1	>=40/Negative	
		Influenza H3	16.6/Positive	
		Influenza N1	>=40/Negative	

Influenza N2

16.4/Positive

**PCR - Influenza A - USDA program**

<u>Animal ID</u>	<u>Specimen</u>	<u>Ct/Result</u>	<u>Comment</u>
1, SID #1	Lung	23.6/Positive	

**Sequencing and Analysis - Influenza A - USDA Prog.**

<u>Animal ID</u>	<u>Specimen</u>	<u>Target Gene</u>	<u>GenBank Ref</u>	<u>Comment</u>
1, SID #1	Virus isolate	HA	MH910359	

**Nucleotide**

ATGAAGACTATCATTGCTTTGAGCTACATTCTATGTCTGGTTTTCGCTCAAAAAATTCCTGGAA  
 ATGACAATGGCACAGCAACGCTGTGCCTTGGGCACCATGCAGTACCAAACGGAACGATAGTGAAA  
 ACAATCACAGATGACCGAATTGAAGTTACTAATGCTACTGAGTTGGTTCAGAATTCCTCAATAGG  
 TGAAATATGCGACAGTCCTCATCAGGTCTCGATGGAGAGAACTGCACACTAATAGATGCTCTAT  
 TGGGGGACCCTCAGTGTGACGGCTTTCAAATAAGAAATGGGACCTTTTTGTTGAACGAAGCAGA  
 GCCTACAGCAACTGTTACCCTTATGATGTGCCGGATTATGCCTCCCTTAGGTCACTAGTTGCCTC  
 ATCCGGCACACTGGAGTTTAAAAATGAAAGCTTCAATTGGACTGGAGTCACTCAAAACGGAAAAA  
 GTTCTTCTTGATAAGGGGATCTAGTAGTAGTTTCTTTAGTAGATTAAATTGGTTGACCCACTTA  
 AACTACACATATCCAGCACTGAACGTGATTATGCCAAACAGGGAAAAATTTGACAAATTGTACAT  
 TTGGGGGGTCCACCACCCGGGTACGGACAATGACCAAATCTTCCTGTATGCTCGATCATCAGGAA  
 GAATCATAGTATCTACCAAAAAGAAGCCAACAAGCTGTAATCCCAAATATCGGATCTAGACCCAGA  
 ATAAGGGATATCCCCAGCAGAATAAGCATCTATTGGACAATAGTAAAACCGGGAGACATACTTTT  
 GATTAACAGCACAGGGAATCTGATTGCTCCTAGGGGTTACTTCAAATACGAAGTGGGAAAAGCT  
 CAATAATGAGATCAGATGCACCCATTGGCAAATGCAATCTGAATGCATCACTCCAAATGGAAGC  
 ATTCCCAATGACAAACCATTCCAAAATGTAACAGGATTACATACGGGGCCTGTCCCAGATATGT  
 TAAGCAAAGCACTCTGAAATTGGCAACAGGAATGCGAAATGTACCAGAGAAACAACTAGAGGCA  
 TATTTGGCGCAATAGCGGGTTTCATAGAAAATGGTTGGGAGGGAATGGTGGATGGTTGGTACGGT  
 TTCAGGCATCAAAATTCTGAGGGAAGAGGACAAGCAGCAGATCTCAAAAGCACTCAAGCAGCAAT  
 CGATCAAATCAATGGGAAGCTGAATCGATTGATCGGAAAAACCAACGAGAAATTCCATCAGATTG  
 AAAAGAATTCTCAGAAGTAGAAGGAAGAGTTCAAGACCTTGAGAAATATGTTGAGGACACTAAA  
 ATAGATCTCTGGTCATACAACGCGGAGCTTCTTGTGGCCCTGGAGAACCAACATACAATTGATCT  
 AACTGACTCAGAAATGAACAACTGTTTGAACAAAACAAAGCAACTGAGGGAAAATGCTGAGG  
 ATATGGGAAATGGTTGTTTCAAATATACCACAAATGTGACAATGCCTGCATAGGATCAATAAGA  
 AATGAACTTATGACCACAATGTGTACAGGGATGAAGCATTAAACAACCGGTTCCAGATCAAGGG  
 AGTTGAGCTGAAGTCAGGGTACAAAGATTGGATCCTATGGATTTCTTTGCCATATCATGTTTTT  
 TGCTTTGTGTTGCTTTGTTGGGGTTCATCATGTGGGCCTGCCAAAAAGGCAACATTAGATGCAAC  
 ATTTGCATTTGA

**Sequencing and Analysis - Influenza A - USDA Prog.**

<u>Animal ID</u>	<u>Specimen</u>	<u>Target Gene</u>	<u>GenBank Ref</u>	<u>Comment</u>
1, SID #1	Virus isolate	NA	MH910360	

**Nucleotide**

ATGAATCCAAATCAAAGATAATAACGATTGGCTCTGTTTCTCTCACCATTTCACAATATGCT

TCTTCATGCAAATTGCCATCTTAATAACTACGGTAACATTGCATTTCAAGCAATATGAATTC AAC  
 TCCCCCCCCAAACAACCAAGTAATGCAGTGTGAACCAACAATAATAGAAAGAAACATAACAGAGAT  
 AGTGTATTTGACCAACACCACTATAGAGAAGGAAATATGCCCTAAACCAGCAGAATACAGAAATT  
 GGTCAAACCGCAATGTGGCATTACAGGATTTCGCACCATTCTCTAAGGACAATTTCGATTAGGCTT  
 TCCGCTGGTGGGGACATCTGGGTGACAAGAGAACCTTATGTATCATGCGATCCTGAAAAGTGTTA  
 TCAATTTGCCCTTGGACAGGGAACAACACTAAACAACGTGCATTCAAATAACACAGTACGTGRTA  
 GGACCCCTTATCGGACTCTATTAATGAATGAGTTGGGTGTTCCCTTTCCATCTGGGGACCAAGCAA  
 GTGTGCATAGCATGGTCCAGCTCAAGTTGTCACGATGGAAAAGCATGGCTGCATGTTTGTATAAC  
 GGGGGATGATAAAAAATGCAACTGCTAGCTTCATTTACAATGGGAGGCTTGTAGATAGTGTTGTTT  
 CATGGTCCAAAGATATCCTCAGGACCCAGGAGTCAGAATGCGTTTGTATCAATGGAACCTTGTA  
 GTAGTAATGACTGATGGAAGTGCTACAGGAAAAGCTGATACTAAAATATTATTCATTGAGGAGGG  
 GAAAATAGTTCATACTAGCAAATTGTCAGGAAGTGCTCAGCATGTCTGAAGAGTGCTCTTGCTATC  
 CTCGATATCCTGGTGTGATGTGTATGCAGAGACAACTGGAAAGGATCCAACCGGCCTATCGTA  
 GATATAAACATAAAGGATCATAGCATTGTTTCCAGTTATGTGTGTTTCAGGACTTGTTGGAGACAC  
 ACCCAGGAAAAACGACAACCTCCAGTAGTAGCCATTGTTTGGATCCTAATAATGAAGAAGGTGGTC  
 ATGGAGTGAAAGGCTGGGCCTTTGATGATGGAAATGACGTGTGGATGGGGAGAACAAATCAACGAG  
 GCGTCACGCTTAGGGTATGAAACCTTCAAAGTCGTTGAAGGCTGGTCCAACCCTAAGTCCAAATT  
 GCAGATAAATAGGCAAGTCATAGTTGACAGAGGTGATAGGTCCGGTTATTCTGGTATTTTCTCTG  
 TTGAAGGCAAAAAGCTGCATCAATCGGTGCTTTTATGTGGAGTTGATTAGGGGAAGAAAAGAGGAA  
 ACTGAAGTCTTGTGGACCTCAAACAGTATTGTTGTGTTTTGTGGCACCTCAGGTACATATGGAAC  
 AGGCTCATGGCCTGATGGGGCGAACCTCAATCTCATGCATATATAA

## Virology

### Virus isolation - Influenza A - USDA program

<u>Animal ID</u>	<u>Specimen</u>	<u>Result</u>	<u>Comment</u>
1, SID #1	Lung	Positive	A02256610 HA: 32

**Veterinary Diagnostic Laboratory**

Iowa State University  
College of Veterinary Medicine  
Ames, Iowa 50011-1134

Phone: 515-294-1950  
Fax: 515-294-3564

**Final Report**

Report Date: 10/12/2018 08:32:04

Client Phone: 1-910-289-2111	Species: Porcine	Age: NA
Client Fax: ---	Breed: Unknown	Weight:
Client Account#: 63709	Sex:	Received:
Date Received: 9/25/2018	Previous Case:	1 Oral Fluid
Sample Taken: 9/7/2018	Farm Type: Other	Reason: General Diagnostics
Preliminary Report:	Animal ID(s): 2	
Accompanying Cases:		

*The ISU VDL has recently changed the swine influenza A virus (IAV-S) HA gene sequencing report format.*

*For IAV H1 viruses, the Influenza Research Database (IRD, [www.fludb.org](http://www.fludb.org)) has built a swine HA(H1) Clade Classification Tool that involves a reference tree using ~100 HA(H1) sequences representing all of the currently defined U.S. swine H1 clades together with representatives of other H1 clades. A query HA(H1) sequence can be compared to the reference H1 sequences to determine its phylogenetic clusters. When a query sequence is unequivocally within the bounds of a single clade, that clade name is assigned to the query sequences. When classification of a query sequence is less certain, a designation including the suffix '-like' is used to indicate its nearest relation. If a query sequence is not clustered with any reference sequences, it is indicated as 'Not Clustered'. Currently, the output of the U.S. swine H1 classification includes possible outcomes such as: (1) H1 alpha clade; (2) H1 beta clade; (3) H1 gamma clade; (4) H1 gamma2 clade; (4) H1 pandemic clade; (5) H1 delta 1 clade; (6) H1 delta2 clade; (7) H1 delta-like clade; (8) Not clustered. The output of the global swine H1 classification includes 1A classical swine lineage (1A.1, 1A.2, and 1A.3 and their subclades), 1B human seasonal lineage (1B.1 and 1B.2 and their subclades), and 1C Eurasian avian lineage (1C.1 and 1C.2 and their subclades). We report both the U.S. swine H1 lineages and the global swine H1 lineages with the latter in parenthesis. Note that this tool classifies HA(H1) sequences regardless of NA subtype.*

*For IAV H3 viruses, a swine HA(H3) Clade Classification Tool is not available online yet; the IRD and collaborators are currently working to define the classification system. We perform HA(H3) sequence analysis and tentatively report the outcomes as: (1) H3 cluster I; (2) H3 cluster II; (3) H3 cluster III; (4) H3 cluster IV; (5) Novel human-to-swine H3 IAVs since 2011/2012; (6) Novel human-to-swine H3 IAVs since 2016/2017; (7) Not clustered.*

The full-length sequence of the HA gene was determined from the oral fluid. Genetic comparison to the Influenza Research Database reference sequences revealed that the H1 gene of the IAV identified in this case is genetically close to that of the ????. Please contact the VDL if you have any questions on this report or if you have other influenza A viruses to compare to this one. (10/1/18 jqz/ks)

**SUPPLEMENTAL REPORT**

Swine influenza virus isolation was attempted in MDCK cells and was negative. (10/11/18 jqz/clm)

**KEY:** Tests: FA = Fluorescent Antibody, IHC = Immunohistochemistry, ISH = *in situ* hybridization, MALDI = Matrix-assisted laser desorption/ionization, MLV = Modified Live Virus, ORF = Open Reading Frame, PCR = Polymerase Chain Reaction, RFLP = Restriction Fragment Length Polymorphism, VI = Virus Isolation. Agents: BCV = Bovine Coronavirus, BRSV = Bovine Respiratory Syncytial Virus, BVDV = Bovine Viral Diarrhea Virus, CSF = Classical Swine Fever, HPS = *Haemophilus parasuis*, IAV = Influenza A Virus, IBRV = Infectious Bovine Rhinotracheitis Virus, MHP = *Mycoplasma hyopneumoniae*, MHR = *Mycoplasma hyorhinis*, MHS = *Mycoplasma hyosynoviae*, PCV = Porcine Circovirus, PDCV = Porcine Deltacoronavirus, PEDV = Porcine Epidemic Diarrhea Virus, PPV = Porcine Parvovirus, PRCV = Porcine Respiratory Coronavirus, PRRSV = Porcine Reproductive & Respiratory Syndrome Virus, PRV = Pseudorabies Virus, SVA = Senecavirus A, TGEV = Transmissible Gastroenteritis Virus.

Laboratory Result(s)			
Test Ordered	Order Date	Current Status	Complete Date
PCR - Influenza HA gene	9/26/2018	Result Released	10/1/2018
^ Sequencing and Analysis - Influenza HA gene	9/26/2018	Result Released	10/1/2018
PCR - Influenza A	9/25/2018	Result Released	9/25/2018
PCR - Influenza A subtype	9/25/2018	Result Released	9/26/2018
Virus isolation - Influenza A	9/26/2018	Result Released	10/10/2018

^ Testing performed in part or in total at a Referral Laboratory.

## Molecular Diagnostic

### Sequencing and Analysis - Influenza HA gene

Animal ID	Specimen	Target Gene	Comment
2, SID #1	Oral fluid	HA	
Nucleotide			
ATGAAAGTAAACTAATGGTTCTGTTATGTACATTTACAGCTACGTATGCAGACACAATATGCG			
TGGGCTACCATGCCAACAACTCAACTGACATTGTTGACACAGTACTTGAGAAGAATGTGACAGTG			
ACACACTCTGTCAACCTACTTGAAGACAGCCACAATGGGAACTATGTCTACTAAAAGGGGTAGC			
TCCACTACAAGTAGGTAGTTGCAGCGTTGCCGGATGGATCTTAGGAAACCCAGAGTGCGAATTGC			
TGATTTCCAAGGAATCATGGTCTTACATTGTAGAGGCACCAATCCTGAGAATGGAACATGTTAC			
CCAGGGTATTTTGAAGACTATGAAGAACTGAGGGAGCAATTGAGTTCAGTATCTTCATTCAAGAG			
ATTCGAAATGTTTCCCAAAGGAGCTCATGGCCCAACCACTGTAACCGGAGTGTCATCATCAT			
GCTCCCATAGCGGGAACAGCAGCTTCTACAGAAATTTGCTATGGCTGACGGTGAAAAACAATTTG			
TACCCAAACCTGAGCAAGTCTATACAAACAAAAAAGAGAAAGAAATCCTTGACTGTGGGGTGT			
TCATCACCCATCTAACATGGAGGACCAAGGACCCTCTATCGTACTGAAAATACTTATGTCTCTG			
TAGTGTCTTCACATTATAGCAGAAGATTCACCCAGAAATAGCCAAGAGACCCAAGGTGAGAAAT			
CAGGAAGGGAGAATAAACTATTATTGGACTCTGCTAGAACCCGGGGATACAATAATATTTGAGGC			
AAGTGGAAATCTAATAGCACCAAGGTATGCCCTTGAAGTGAAGTAGGGGTTTTGGATCAGGAATCA			
TCACATCAAATGCACCAATGGGAGAATGCAATACAAAATGTCAAACACCTCAGGGGGCTATAAAC			
AGCAGTCTTCTTTCCAGAATGTACACCCAGTAACAATAGGAGAGTGTCCAAATATGTCAGGAG			
TGCAAAATTAAGGATGGTTACAGGACTAAGGAACACCCCATCCATCCAATCCAGAGGTTTGTGTTG			
GAGCTATTGCCGGTTTTATTGAAGGAGGATGGACTGGAATGGTAGATGGATGGTATGGTTACCAT			
CATCAGAATGAGCAAGGGTCTGGATATGCTGCAGATCAACAAAGCACACAAAATGCCATTAATGG			
GATTACAAACAAGGTGAATTCTGTAATGAAAAAATGAACACTCAATTCACAGCTGTGGGCAAG			
AATTCAACAAATTTGAAAGAAAGAAATGGAATACTAAATAAAAAAGTTGATGATGGGTTTTCTAGAC			
ATTTGGACATATAATGCAGAATTGTTAGTTCTACTGGAAAATGAAAGGACTTTGGATTTCCATGA			
CTCCAACGTGAAGAATCTGTATGAGAAAGTAAAGAGGCAACTAAAAATAATGCCAAAGAAATAG			
GAAACGGGTGTTTTGAATTCTATCATAAGTGTGACGATGAATGCATGGATAGTGTGAAAAATGGA			
ACTTATGACTATCCAAAATATTCCGAAGAATCAAACTAAACAGGGAGAAAATTGATGGAGTGAA			
ATTGGAATCAATGGGAGTCTATAATATTCTGGCGATCTACTCAACAGTCGCCAGTTCCTAGTTC			
TTTTAGTCTCCCTGGGGGCAATCTGCTTCTGGATGTGTTCCAATGGGTCTTTACAGTGTAGAATA			
TGCATCTAA			

**PCR - Influenza A**

<u>Animal ID</u>	<u>Specimen</u>	<u>Target</u>	<u>Ct/Result</u>	<u>Comment</u>
2, SID #1	Oral fluid	Influenza general	25.8 / Positive	

**PCR - Influenza HA gene**

<u>Animal ID</u>	<u>Specimen</u>	<u>Result</u>	<u>Comment</u>
2, SID #1	Oral fluid	Positive	

**PCR - Influenza A subtype**

<u>Animal ID</u>	<u>Specimen</u>	<u>Target</u>	<u>Ct/Result</u>	<u>Comment</u>
2, SID #1	Oral fluid	Influenza H1	30.3/Positive	
		Influenza H3	>=40/Negative	
		Influenza N1	>=40/Negative	
		Influenza N2	31.1/Positive	

**Virology****Virus isolation - Influenza A**

<u>Animal ID</u>	<u>Specimen</u>	<u>Result</u>	<u>Comment</u>
2, SID #1	Oral fluid	Negative	



**Veterinary Diagnostic Laboratory**

Iowa State University  
College of Veterinary Medicine  
Ames, Iowa 50011-1134

Phone: 515-294-1950  
Fax: 515-294-3564

**Final Report**

Report Date: 10/9/2018 09:05:03

Client Phone: 1-217-357-2811	Species: Porcine	Age: NA
Client Fax: 1-217-357-6665	Breed: Unknown	Weight:
Client Account#: 17581	Sex:	Received:
Date Received: 10/2/2018	Previous Case:	1 ORAL FLUID
Sample Taken:	Farm Type:	Reason: General Diagnostics
Preliminary Report:	Animal ID(s): pooled	
Accompanying Cases:		

*The ISU VDL has recently changed the swine influenza A virus (IAV-S) HA gene sequencing report format.*

*For IAV H1 viruses, the Influenza Research Database (IRD, [www.fludb.org](http://www.fludb.org)) has built a swine HA(H1) Clade Classification Tool that involves a reference tree using ~100 HA(H1) sequences representing all of the currently defined U.S. swine H1 clades together with representatives of other H1 clades. A query HA(H1) sequence can be compared to the reference H1 sequences to determine its phylogenetic clusters. When a query sequence is unequivocally within the bounds of a single clade, that clade name is assigned to the query sequences. When classification of a query sequence is less certain, a designation including the suffix '-like' is used to indicate its nearest relation. If a query sequence is not clustered with any reference sequences, it is indicated as 'Not Clustered'. Currently, the output of the U.S. swine H1 classification includes possible outcomes such as: (1) H1 alpha clade; (2) H1 beta clade; (3) H1 gamma clade; (4) H1 gamma2 clade; (5) H1 pandemic clade; (6) H1 delta1 clade; (7) H1 delta2 clade; (8) H1 delta-like clade; (9) Not clustered. The output of the global swine H1 classification includes 1A classical swine lineage (1A.1, 1A.2, and 1A.3 and their subclades), 1B human seasonal lineage (1B.1 and 1B.2 and their subclades), and 1C Eurasian avian lineage (1C.1 and 1C.2 and their subclades). We report both the U.S. swine H1 lineages and the global swine H1 lineages with the latter in parenthesis. Note that this tool classifies HA(H1) sequences regardless of NA subtype.*

*For IAV H3 viruses, a swine HA(H3) Clade Classification Tool is not available online yet; the IRD and collaborators are currently working to define the classification system. We perform HA(H3) sequence analysis and tentatively report the outcomes as: (1) H3 cluster I; (2) H3 cluster II; (3) H3 cluster III; (4) H3 cluster IV; (5) Novel human-to-swine H3 IAVs since 2011/2012; (6) Novel human-to-swine H3 IAVs since 2016/2017; (7) Not clustered.*

The full-length sequence of the HA gene was determined from the oral fluid. Genetic comparison to the Influenza Research Database reference sequences revealed that the H1 gene of the IAV identified in this case is genetically close to that of the ????. Please contact the VDL if you have any questions on this report or if you have other influenza A viruses to compare to this one. (10/8/18 jqz/ks)

**KEY:** Tests: FA = Fluorescent Antibody, IHC = Immunohistochemistry, ISH = *in situ* hybridization, MALDI = Matrix-assisted laser desorption/ionization, MLV

= Modified Live Virus, ORF = Open Reading Frame, PCR = Polymerase Chain Reaction, RFLP = Restriction Fragment Length Polymorphism, VI = Virus Isolation. Agents: BCV = Bovine Coronavirus, BRSV = Bovine Respiratory Syncytial Virus, BVDV = Bovine Viral Diarrhea Virus, CSF = Classical Swine Fever, HPS = *Haemophilus parasuis*, IAV = Influenza A Virus, IBRV = Infectious Bovine Rhinotracheitis Virus, MHP = *Mycoplasma hyopneumoniae*, MHR = *Mycoplasma hyorhinis*, MHS = *Mycoplasma hyosynoviae*, PCV = Porcine Circovirus, PDCV = Porcine Deltacoronavirus, PEDV = Porcine Epidemic Diarrhea Virus, PPV = Porcine Parvovirus, PRCV = Porcine Respiratory Coronavirus, PRRSV = Porcine Reproductive & Respiratory Syndrome Virus, PRV = Pseudorabies Virus, SVA = Senecavirus A, TGEV = Transmissible Gastroenteritis Virus.

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Test Ordered	Laboratory Result(s)	Order Date	Current Status	Complete Date
PCR - Influenza HA gene		10/2/2018	Result Released	10/8/2018
^ Sequencing and Analysis - Influenza HA gene		10/2/2018	Result Released	10/8/2018
PCR - Influenza A		10/2/2018	Result Released	10/2/2018
PCR - Influenza A subtype		10/2/2018	Result Released	10/3/2018

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^ Testing performed in part or in total at a Referral Laboratory.

**Molecular Diagnostic****Sequencing and Analysis - Influenza HA gene**

<u>Animal ID</u>	<u>Specimen</u>	<u>Target Gene</u>
	<u>Comment</u> pooled, SID #1	
	Oral fluid	HA

**Nucleotide**

ATGAAGGCAACTAGTAGTCCTGCTATATACATTTACAGCAGCAAATGCCGACACATTATGTA  
TAGGTTATCATGCAAACAATTCAACTGACACCGTAGACACAGTACTAGAAAAGAATGTAACAGTA  
ACACACTCTGTCAACCTTCTAGAAAACAGGCATAATGGGAACTATGTAACTAAGAGGGGTAGC  
TCCATTGCACTTGGGTAAATGTAGCATTGCTGGATGGCTTCTGGGAAATCCAGAGTGTGAATCAT  
TCTCCACAGCAAGATCATGGTCCTACATTGTGGAAACATCTAAGTCAGAAAATGGGACGTGTTAC  
CCAGGGGATTTTCATAAATTATGAGGAGCTGAGAGAGCAGTTGAGCTCAGTGTTCATCATTTGAAAG  
ATTTGAGATATTTCCCAAGACAAGTTCATGGCCCAATCATGACACGAACAGAGGTGTGACGGCAG  
CATGTCCTCATGCTGGGACAAACAGCTTCTACAGGAATTTAGTATGGCTAGTGCAAAAGGAAAAT  
TCATACCCAAAAATCAACAAATCCTACATTAACAATAGAGAGAAGGAAGTTCTCGTGCTATGGGC  
CATTACCATCCATCCACAGTGCCGACCAACAGAGTCTCTATAAAAATGCAGATGCCTATGTGT  
TTGTGGGGTTCATCAAAATACAGCAAGAAGTTCGAGCCAGAAATAGCTACAAGACCTAAAGTGAGA  
GACCAAGCAGGGAGAATGAATTATTACTGGACACTAGTTGAGCCTGGAGACAAGATAACATTCTGA  
AGCAACTGGAAATCTAGTGGCACCGAGATATGCCTTCGCATTGAAAAGAAATTCTGGATCTGGTA  
TTATCATTTTCAGATACATCAGTCCACGATTGTGATACAACCTTGTCAAACACCCAATGGTGCTATA  
AACACCAGCCTCCCATTTCAAATGTACATCCAGTCACAATCGGAGAATGTCCAAAATATGTAAA  
AAGTACTAACTGAGAATGGCCACAGGTTTAAGGAATATCCCGTCTATTCAAACCTAGAGGCCTGT  
TTGGTGCCATTGCTGGCTTTATCGAAGGGGGTGGACAGGAATGATAGATGGATGGTACGGTTAT  
CACCATCAAAATGAGCAGGGATCAGGATATGCAGCCGACCTGAAGAGCACACAAAATGCCATTGA  
CGGGATCACTAACAAGGTGAACTCTGTTATTGAAAAGATGAACACCCAATTCACGGCAGTAGGTA  
AAGAGTTCAGCCACTTGGAAGAAGAATAGAGAATTTAAATAAAAAGGTTGATGATGGTTTTCTA  
GATATTTGGACTTACAATGCAGAACTGTTGGTCTATTGGAAAATGAACGAACCTTTGGATTACCA  
CGACTCAAATGTTAAAACTTATATGAAAAAGTAAGAAGCYAACTAAAAACAATGCCAAGGAAA  
TTGGGAATGGCTGCTTTGAATTTTACCACAAATGTGATGACATGTGCATGGAAAGCGTCAAAAAT  
GGAACCTTATGATTACCCTAAATACTCAGAGGAAGCAAACTAAACAGAGAGGAAATAGATGGGGT  
AAAGTTGGAATCAACAAGAATTTACCAAATTTGGCGATCTATTCAACGGTCGCCAGTTTCATTGG  
TACTGGTAGTCTCCCTGGGGGCAATCAGTTTCTGGATGTGCTCTAATGGGTGCTACAGTGCAGA  
ATATGTATTWAA

**PCR - Influenza A**

<u>Animal ID</u>	<u>Specimen</u>	<u>Target</u>	<u>Ct/Result</u>	<u>Comment</u>
pooled, SID #1	Oral fluid	Influenza general	28.1 / Positive	

**PCR - Influenza HA gene**

<u>Animal ID</u>	<u>Specimen</u>	<u>Result</u>	<u>Comment</u>
pooled, SID #1	Oral fluid	Positive	

**PCR - Influenza A subtype**

<u>Animal ID</u>	<u>Specimen</u>	<u>Target</u>	<u>Ct/Result</u>	<u>Comment</u>
pooled, SID #1	Oral fluid	Influenza H1	29.5/Positive	
		Influenza H3	>=40/Negative	
		Influenza N1	27.4/Positive	
		Influenza N2	>=40/Negative	

**Veterinary Diagnostic Laboratory**

Iowa State University  
College of Veterinary Medicine  
Ames, Iowa 50011-1134

Phone: 515-294-1950  
Fax: 515-294-3564

**Final Report**

Accessed Date: 10/26/2018 1:59 pm

Client Phone: 1-402-564-0407	Species: Porcine	Age: 21 Days
Client Fax: 1-402-564-5471	Breed: Unknown	Weight:
Client Account#: 403814	Sex:	Received:
Date Received: 10/18/2018	Previous Case:	30 bronchial swabs
Sample Taken: 10/17/2018	Farm Type: Breeding Herd	Reason: General Diagnostics
Accompanying Cases:	Animal ID(s): gilt ..., gilt ..., gilt ..., gilt ...	
gilt ... Final Report(s): 10/26/2018 1:59 pm		

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**Laboratory Result(s)**

<u>Test Ordered</u>	<u>Order Date</u>	<u>Current Status</u>	<u>Complete Date</u>
PCR - Influenza HA gene	10/19/2018	Result Released	10/24/2018
^ Sequencing and Analysis - Influenza HA gene	10/19/2018	Result Released	10/24/2018
PCR - M. hyopneumoniae	10/18/2018	Result Released	10/18/2018
PCR - Influenza A - USDA program	10/18/2018	Result Released	10/18/2018
PCR - Influenza A subtype	10/18/2018	Result Released	10/19/2018

-

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## Molecular Diagnostic

### Sequencing and Analysis - Influenza HA gene

<u>Animal ID</u>	<u>Specimen</u>	<u>Target Gene</u>	<u>Comment</u>
GB [6 - 10]	Bronchial swab	HA	
<b>Nucleotide</b>			
ATGAAGGCAATATTGTTGGTCTTGCTACATACATTTGCAGCCGCAAATGCAGACACAATATGTA TAGGCTATCATGCGAATAATTCAACTGACACTGTTGATACAGTGTTAGAGAAGAATGTAACAGTA ACACACTCTGTCAACCTTCTAGTAGACAAACATAATGGGAAATTATGCAAACCTAGGGGGAAAAGC CCCATTGTACTTGGGTAAATGTAACATTGCCGATGGCTTTTGGGAAACCCAGAGTGTGAATCAA TATTCACAGTAAGCTCATGGTCTTACATTGTGGAACATCTAACTCAGACAATGGGACATGTTAT CCTGGAAGTTTCGCCAATTATGAAGAGCTGAGAGAGCAATTGAGCTCAGTGTCATCATTTGAAAA ATTTGAGATATTTCCCATAGAAAATTCATGGTCCGACTATAAACAGGTGTGACAGCATCATGCC CTTATGCTGGAGAAAACAGTTTCTACAGAAATTTAATGTGGCTGGTGAAAAAAGGAGATTTCATAC CCAAGGCTCAACATATCCTATGTTAACAATAAGGGGAAAGAAGTCCTTGTGCTATGGGGCATTCA TCACCCACCTACTGGTAATGATCAACAATGGCTCTACCAGAATTCAAATGCCTCTGTTTTTGTGG GAACATCAACGTACAGCCAGAAATTCAAACCAGAAATAGCGACAAGACCCAAGGTGAGAGGTCAA ACAGGGAGGATGAACTATTACTGGACACTATTAGAACCCGGAGACACAATAACATTTGAAGCAAC TGGAAACTTGATGGTACCGAAGTATGCTTTCGCAATGAATAGAGGATCTGGGTCTGGTATTATAG TTTCGGATGCCCAATCCACGATTGTAATACGACTTGTCAGACACCTAAAGGTGCCCTAAACACC AGCCTCCCATTTTCAAGATGTACATCCAGTCACAATTGGAGAATGTCCAAAGTATGTCAAAGCAC AAGACTAAAAATGGCTACAGGATTACGAAATATCCCATCTATTCAATCTAGAGGTCTATTTGGAG CCATAGCTGGTTTTATTGAAGGGGGATGGACAGGAATGATAGATGGATGGTATGGTTATCATCAC CAAAATGACCAGGGATCGGGGTATGCCGCTGACCAGAAGAGCACACAGAGAGCCATTGATGGGA T CACAAACAAAGTAAATTCCGTTATTGAAAAAATGAACACGCAGTTCACAGCAGTAGGTAAAGAAT TCAACAAATTAGAAAGGAGAATAGAGAACTTGAATAGAAAAGTTGACGATGGCTTTTGGATGTT TGGACCTACAATGCCGAAGTGTGATACTCTTGAAAAATGAAAGGACTTTGGATTTCATGATTTC AAATGTGAAGAATCTATATGAGAAAGTAAGAAACCAGCTAAGGAACAATGCCAAGGAAATTGGGA ATGGGTGTTTTGAATTCTACCACAGATGTGATAACACATGTATGGAAAGTGTCAAAAATGGGACT TATAATTACCTAAAATACTCAGAAGAATCGAACTAAACAGAGAGGAAATAGATGGAGTGAAGTT GGATTCAACAAGGGTTTATCAGATTTTGGCGATATATTCAACTGTCGCCAGTTCATTGGTACTAT TAGTCTCCCTGGGGGCACTCAGTTTCTGGATGTGTTCCAATGGGTCTTTGCAATGCAGAATATGT ATTTAA			

### PCR - Influenza HA gene

<u>Animal ID</u>	<u>Specimen</u>	<u>Result</u>	<u>Comment</u>
GB [6 - 10]	Bronchial swab	Positive	

### PCR - Influenza A subtype

<u>Animal ID</u>	<u>Specimen</u>	<u>Target</u>	<u>Ct/Result</u>	<u>Comment</u>
GA [1 - 5]	Bronchial swab	Influenza H1 Influenza H3 Influenza N1 Influenza N2	24.7/Positive ≥40/Negative ≥40/Negative 25.0/Positive	

### PCR - Influenza A subtype

<u>Animal ID</u>	<u>Specimen</u>	<u>Target</u>	<u>Ct/Result</u>	<u>Comment</u>
GB [6 - 10]	Bronchial swab	Influenza H1 Influenza H3 Influenza N1 Influenza N2	24.1/Positive ≥40/Negative ≥40/Negative 24.4/Positive	

### PCR - Influenza A subtype

<u>Animal ID</u>	<u>Specimen</u>	<u>Target</u>	<u>Ct/Result</u>	<u>Comment</u>
GC [11 - 15]	Bronchial swab	Influenza H1 Influenza H3	≥40/Negative ≥40/Negative	

Influenza N1  
Influenza N2>=40/Negative  
39.3/Suspect**PCR - Influenza A subtype**

<u>Animal ID</u>	<u>Specimen</u>	<u>Target</u>	<u>Ct/Result</u>	<u>Comment</u>
GF [26 - 30]	Bronchial swab	Influenza H1	26.5/Positive	
		Influenza H3	>=40/Negative	
		Influenza N1	>=40/Negative	
		Influenza N2	26.4/Positive	

**PCR - Influenza A - USDA program**

<u>Animal ID</u>	<u>Specimen</u>	<u>Ct/Result</u>	<u>Comment</u>
GA [1 - 5]	Bronchial swab	21.2/Positive	
GB [6 - 10]	Bronchial swab	20.3/Positive	
GC [11 - 15]	Bronchial swab	37.9/Positive	
GD [16 - 20]	Bronchial swab	>40/Negative	
GE [21 - 25]	Bronchial swab	>40/Negative	
GF [26 - 30]	Bronchial swab	22.4/Positive	

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Patient pool's member(s), # means tube number:

GA [1 - 5] : (#1 gilt - rm 7, 21 Days) , (#2 gilt - rm 7, 21 Days) , (#3 gilt - rm 7, 21 Days) , (#4 gilt - rm 7, 21 Days) , (#5 gilt - rm 7, 21 Days)

GB [6 - 10] : (#6 gilt - rm 7, 21 Days) , (#7 gilt - rm 7, 21 Days) , (#8 gilt - rm 7, 21 Days) , (#9 gilt - rm 7, 21 Days) , (#10 gilt - rm 7, 21 Days)

GC [11 - 15] : (#11 sow - rm 27, 21 Days) , (#12 sow - rm 27, 21 Days) , (#13 sow - rm 27, 21 Days) , (#14 sow - rm 27, 21 Days) , (#15 sow - rm 27, 21 Days)

GD [16 - 20] : (#16 sow - rm 27, 21 Days) , (#17 sow - rm 27, 21 Days) , (#18 sow - rm 27, 21 Days) , (#19 sow - rm 27, 21 Days) , (#20 sow - rm 27, 21 Days)