

# WOAH Collaborative Centre Reports Activities 2023

## Activities in 2023

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### Centre Information

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<b>Name (including Title and Position) of Head of the Collaborating Centre (WOAH Contact Point):</b>	Dr. Suelee Robbe-Austerman Director, National Veterinary Services Laboratories USDA, APHIS, VS, DB
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### TOR1 AND 2: SERVICES PROVIDED

1. Activities as a centre of research, expertise, standardisation and dissemination of techniques within the remit of the mandate given by WOA

Category	Title of activity	Scope
Diagnosis, biotechnology and laboratory (true)	African Swine Fever (ASF)	Direct diagnostic test methods: PCR, VI, and Sequencing. Performed 68,114 Nationally; 88,358 Internationally Indirect test methods: ELISA, IP, and IFA. Performed 65,058 Nationally; 4,229 Internationally Supplied rtPCR amplification control, ASFV antiserum, and IPT plates to WOA member country Developed new diagnostic methods Collected epidemiological data relevant to Dominican Republic (DR) and US Virgin Islands ASF/CSF surveillance of domestic and feral pigs. WGS was conducted on field specimens from DR ASF outbreak and information including animal location and testing data were assembled. Received ASF positive blood samples from the DR to support National Veterinary Services Laboratories (NVSL) diagnostic assay evaluations.
Diagnosis, biotechnology and laboratory (true)	Anthrax	Direct diagnostic test methods: Isolation and Identification. Performed 6 Nationally. Reference materials produced/provided: Gamma phage reagent for use in the gamma phage lysis diagnostic test

<p>Diagnosis, biotechnology and laboratory (true)</p>	<p>Avian Influenza (AI)</p>	<p>Direct: RT-PCR (IAV, subtyping), Virus Isolation, Molecular pathotype (Sanger), In vivo pathotype (IVPI), WGS. Performed 24,842 nationally; 178 internally. Indirect: Agar gel immunodiffusion (AGID), Hemagglutination-inhibition (HI) antibody subtype identification (H1-16). Performed 2,647 nationally; 133 internationally. Produced/provided reference antigen/antisera, AGID reagents, positive amplification controls, extraction controls, and PT panels to member countries. NVSL works with another unit within USDA for distribution of analysed data. Over 1,000 avian influenza full genomes deposited into GISAID Proficiency testing rounds for approved-NAHLN laboratories alternate yearly for AI/ND. 2023 is an ND year, out of cycle AI PTs were also provided as needed.</p>
<p>Diagnosis, biotechnology and laboratory (true)</p>	<p>Chronic Wasting Disease (CWD)</p>	<p>Direct diagnostic test method: Immunohistochemistry. Performed 5,709 Nationally.</p>
<p>Diagnosis, biotechnology and laboratory (true)</p>	<p>Contagious equine metritis (CEM)</p>	<p>Direct: Identification of the agent, RT-PCR, and WGS. Performed 1,325 nationally; 138 internationally. Indirect: Complement Fixation. Performed 2242 nationally; 168 internationally. Produced/provided culture control isolates to member countries. NVSL works with another unit within USDA for distribution of analysed data. Proficiency test administered by NVSL and required to conduct official testing in the U.S.; shipped internationally by request.</p>
<p>Diagnosis, biotechnology and laboratory (true)</p>	<p>Foot and mouth disease (FMD)</p>	<p>Direct diagnostic test methods: Virus Isolation (VI), Antigen ELISA, PCR, Sequencing. Performed 3,043 Nationally. Indirect diagnostic test methods: 3 ABC ELISA, Vaccine Matching, Virus Neutralization, VIAA AGID. Performed 450 Nationally. Supplied PCR Control for FMDV rRT-PCR to member country Developed new diagnostic methods Supplied PCR control reagent for FMDV rTR-PCR to produce positive amplification control to member country Organize diagnostic performance quality in the United States National Animal Health Laboratory Network for FMD testing</p>
<p>Diagnosis, biotechnology and laboratory (true)</p>	<p>Leptospirosis</p>	<p>Direct: Microscopic Agglutination, Fluorescent Antibody, Isolation, RT LipL32 PCR, WGS, 16S and secY PCR, Serogroup Testing. Performed 5,801 nationally; 128 internationally. Supplied reference positive control sera, reference cultures, multivalent fluorescent antibody conjugate, and Leptospira medium to WOAHA member countries.</p>
<p>Diagnosis, biotechnology and laboratory (true)</p>	<p>Newcastle disease (ND)</p>	<p>Direct: RT-PCR (matrix, fusion), virus isolation, molecular pathotype (Sanger), Invivo pathotype (ICPI), and WGS. Performed 3,397 nationally; 18 internationally. Produced/provided reference antigen and antisera, positive amplification controls, extraction controls, and PTs to member countries. NVSL works with another unit within USDA for distribution of analysed data. Proficiency testing rounds for approved-NAHLN laboratories alternate yearly for AI/ND. 2023 is an ND year, out of cycle AI PTs were also provided as needed.</p>
<p>Diagnosis, biotechnology and laboratory (true)</p>	<p>Rinderpest</p>	<p>Direct diagnostic test methods: RT-PCR, VI. Performed 2 Nationally. Indirect diagnostic test methods: RPV freedom tests (RT-PCR). Performed</p>

		202 Nationally.
Diagnosis, biotechnology and laboratory (true)	Swine influenza (SI)	<p>Direct: RT-PCR (IAV, subtyping), WGS, and repository propagation. Performed 6,160 Nationally.</p> <p>Produced/provided reference antigen and antisera, reference/surveillance viruses, positive amplification controls, and PTs to member countries. NVSL works with another unit within USDA for distribution of analysed data. Proficiency testing rounds for approved-NAHLN laboratories are offered every other year. 2023 is an off year, out of cycle PTs were provided as needed.</p>
Diagnosis, biotechnology and laboratory (true)	Tuberculosis Mammalian	<p>Direct: Culture and Direct PCR for Livestock/Wildlife/Zoo Species; Histopathology. Performed 31,882 nationally; 32,686 internationally</p> <p>Indirect: Interferon-gamma release Assay, Lateral Flow – Cervid and Zoo Species. Performed 12,222 nationally; 16 internationally. Supplied</p> <p>Mycobacterium antigen – Mycobacterium bovis purified protein derivative Supplied Mycobacterium antigen – purified protein derivative (PPD) avian balanced tuberculin and bovis balanced tuberculin Supplied Mycobacterium bovis direct PCR control, negative, bovine tissue to Malaysia Supplied Mycobacterium bovis direct PCR control, positive, bovine tissue with BCG to Columbia Supplied Mycobacterium bovis direct PCR control, positive, bovine tissue with Mtb H37a to Malaysia and Columbia Supplied Mycobacterium bovis serum panel Supplied Mycobacterium species DNA to Malaysia Supplied Mycobacterium reference culture to Malaysia Supplied M. bovis vaccine strain BCG to Mexico NVSL participates in diagnosis of mammalian TB, however, epidemiological data such as animal movements and field test data are stored outside of the laboratory, but within the parent agency of NVSL, USDA-APHIS-Veterinary Services. The Laboratory does work directly with our counterparts in other countries to coordinate strain/genotype information to inform investigations between our countries.</p> <p>Whole genome sequencing of cultured mammalian TB isolates were analysed to determine the phylogenetic relationships between new isolates and isolates from previous outbreaks or detections. The analysis was distributed to federal and state animal health officials and epidemiologists to aid in disease tracing.</p>
Diagnosis, biotechnology and laboratory (true)	Vesicular stomatitis virus (VSV)	<p>Direct: Virus isolation, RT-PCR, and sequencing. Performed 1,343 nationally. Indirect: Competitive enzyme-linked immunosorbent assay (cELISA; Indiana-1 and New Jersey serotypes), Complement fixation (Indiana-1 and New Jersey serotypes), and Virus Neutralization (Indiana-1 and New Jersey serotypes). Performed 4,520 nationally; 880 internationally. Produced/provided antigen, reference virus, recombinant antigen, polyclonal ascites, complement, CF test panel, cELISA test panel, PCR test panel, and antiserum to member countries. NVSL works with another unit within USDA for distribution of analysed data. Proficiency testing rounds for approved-NAHLN laboratories are offered every other year. 2023 is an off year, out of cycle PTs were provided as needed. PT administered by NVSL and required to conduct official testing in the U.S.; shipped internationally by request.</p>

## TOR3: HARMONISATION OF STANDARDS

2. Proposal or development of any procedure that will facilitate harmonisation of international regulations applicable to the main focus area for which you were designated

Proposal title	Scope/Content	Applicable area
ASF/CSF multiplex PCR	Implemented in the APHIS laboratory in Dorado, Puerto Rico and NAHLN laboratory	Laboratory expertise
Evaluating enzyme systems, from various vendors, to use with the FADDL ASF real-time PCR assay	Initial stage	Laboratory expertise
Evaluating the MIC qPCR platform to use with the FADDL ASF real-time PCR assay	Initial stage	Laboratory expertise
Direct RNA FMDV Sequencing	Sequencing of FMDV on the Nanopore platform directly from the viral RNA without the reverse transcription step to reduce time and cost to acquiring the whole genome sequence for characterization.	Laboratory expertise
FMDV P1 Sequencing	Sequencing of FMDV P1 on Nanopore using Amplicon approach of P1 and Flongle flow cells to reduce cost and time for rapid characterization.	Laboratory expertise
Evaluation of diagnostic sensitivity of the FMDV singleplex real-time PCR assay with two different probes	The results of this study indicate that the current TAMRA quenched probe and the suggested ZEN/ IABk quenched probe perform comparably with the sample set tested.	Laboratory expertise
Chemical inactivation method of peste des petits ruminants virus (PPRV used as a surrogate virus of RPV)	Amaresh Das, Zaheer Ahmed, Lizhe Xu, 1 Wei Jia (2023). Assessment and verification of chemical inactivation of peste des petits ruminants virus by virus isolation following virus capture using Nanotrap magnetic virus particles. Microbiology Spectrum, September/October 2023 Volume 11 Issue 5. 0.1128/spectrum.00689-23. <a href="https://doi.org/10.1128/spectrum.00689-23">https://doi.org/10.1128/spectrum.00689-23</a>	Laboratory expertise
Quality control test method of chemical inactivated peste des petits ruminants virus (PPRV used as a surrogate virus of RPV)	Amaresh Das, Zaheer Ahmed, Lizhe Xu, 1 Wei Jia (2023). Assessment and verification of chemical inactivation of peste des petits ruminants virus by virus isolation following virus capture using Nanotrap magnetic virus particles. Microbiology Spectrum, September/October 2023 Volume 11 Issue 5. 0.1128/spectrum.00689-23. <a href="https://doi.org/10.1128/spectrum.00689-23">https://doi.org/10.1128/spectrum.00689-23</a>	Laboratory expertise
Real-Time PCR detection of Mycobacterium bovis DNA in bovine milk samples	Zeineldin, M. M., Lehman, K., Camp, P., Farrell, D., & Thacker, T. C. (2023). Diagnostic Evaluation of the IS1081-Targeted Real-Time PCR for Detection of Mycobacterium bovis DNA in Bovine Milk Samples. Pathogens, 12(8), 972.	Laboratory expertise
Whole Genome Sequencing of Mycobacterium bovis direct from tissue samples without culture	Zeineldin, M., Camp, P., Farrell, D., Lehman, K., & Thacker, T. (2023). Whole genome sequencing of Mycobacterium bovis directly from clinical tissue samples without culture. Frontiers in Microbiology, 14, 1141651.	Laboratory expertise

3. In exercising your activities, have you identified any regulatory research needs\* relevant for WOA?H?

No

4. Did your Collaborating Centre maintain a network with other WOA?H Collaborating Centres (CC), Reference Laboratories (RL), or organisations designated for the same specialty, to coordinate scientific and technical studies?

Yes

Name of WOA?H CC/RL/other organisation(s)	Location	Region of networking Centre	Purpose
Mexico (SENE?ICA)	Baja California, Mexico	Americas	Evaluation of BCG vaccine efficacy in naturally infected dairy cattle in Mexico
Global Partnership for Animal and Zoonotic Disease Surveillance (GPAZDS)	The Philippines, Gambia, Ghana, Senegal, Nigeria, Uganda, Cameroon	Africa Asia and Pasific Europe	Sequencing and Bioinformatic analysis, ASF
African Center of Excellence for Genomics of Infectious Diseases	Nigeria	Africa	Genomic sequencing and surveillance of ASFV in West Africa
Central Veterinary Laboratory (LAVECEN)	Dominican Republic	Americas	Diagnosis and genomic surveillance of ASFV and CSFV
CISA-INIA	Spain	Europe	ASF Interlaboratory Comparison Test
Pirbright Institute	United Kingdom	Europe	FMD Proficiency Testing Scheme - January and October 2023
WOAH Rinderpest Virus Holding Facilities (RHF)	France and Japan	Asia and Pasific Europe	Standardize RPV RT-PCR method and apply the non-pathogenic RPV control and PT panel developed by FADDL. France and Japan participated and passed the PT. Two remaining RHF?s, China Institute of Veterinary Drug Control (IVDC) and the Pan African Veterinary Vaccine Centre of the African Union (AU-PANVAC), expressed their interest in 2023 to resume or participate in the project. FADDL answered inquiries of the RHF?s in IVDC and AU-PANVAC.
Leptospirosis Reference Center	The Netherlands	Europe	Serological typing of Leptospiral isolates

USDA ARS National Poultry Center Southeast Poultry Research Laboratory	USA	Americas	Studies in Poultry Transmission, Airborne Spread and Mitigation Tools for Avian Influenza and Newcastle Disease in the USA
WOAH Reference Laboratories for Animal Influenza	Multiple countries	Africa Americas Asia and Pasific Europe MiddleEast	Genetic characteristics of zoonotic influenza viruses Data contributions to OFFLU for the twice yearly WHO Vaccine Composition Consultations
WOAH Reference Laboratories for Vesicular Stomatitis Virus	USA/Mexico	Americas	Vesicular Stomatitis Virus Grand Challenge project. Cooperative agreement among APHIS, ARS, and academic partners.

## TOR4 AND 5: NETWORKING AND COLLABORATION

5. Did your Collaborating Centre maintain a network with other WOAHO Collaborating Centres, Reference laboratories, or organisations in other disciplines, to coordinate scientific and technical studies?

Yes

Name of WOAHO CC/RL/other organisation(s)	Location	Region of networking Centre	Purpose
CISA-INIA	Spain	Europe	PT and reagent production
NAFMDVB (PIADC/CFIA)	Canada	Americas	FMD Vaccine Potency Test

## TOR6: EXPERT CONSULTANTS

6. Did your Collaborating Centre place expert consultants at the disposal of WOAHO?

Yes

NAME OF EXPERT	KIND OF CONSULTANCY	SUBJECT
Ginger Harvey	Virtual review	Updated WOAHO Terrestrial Code, Anthrax Chapter
Vivan O'Donnell	Virtual review	Response to FMDV VLP vaccines to be included into Terrestrial Manual
Tyler Thacker	Meeting attendance	Tuberculosis

Nichole Hines-Bergeson	Regional evaluation	Avian Influenza
Mia Torchetti	Regional WOAHA seminars	Swine Influenza

## TOR7: SCIENTIFIC AND TECHNICAL TRAINING

7. Did your Collaborating Centre provide advice/services to requests from Members in your main focus area?

Yes

*Provided anthrax vaccine assessment to Kazakhstan*

*Provided literature for inactivation of anthrax agent and nucleic acids to Austria*

*Provided a resource for ordering a proficiency test for anthrax to Georgia*

*Provided technical and logistical ASF support to the Dominican Republic*

*Provided ASF PCR diagnostic testing for DR and Haiti*

*Provided Direct PCR, culture, and whole genome sequencing for Guatemala and Mexico*

*USDA-APHIS SAGARPA project to conduct slaughter surveillance testing in Baja CA Mexico. Samples are split between the laboratories of USA and Mexico for test harmonization and whole genome sequencing database development. Methods and test comparisons done virtually.*

*Provided MAT testing for Belgium, Curacao, Columbia, and The Netherlands*

*Provided avian influenza molecular and genomic testing to Costa Rica, Panama, Honduras, Guatemala, and Peru*

8. Did your Collaborating Centre provide scientific and technical training, within the remit of the mandate given by WOAHA, to personnel from WOAHA Members?

Yes

a) Technical visit : 45

b) Seminars : 0

c) Hands-on training courses: 207

d) Internships (>1 month) : 0

TYPE OF TECHNICAL TRAINING PROVIDED (A, B, C OR D)	CONTENT	COUNTRY OF ORIGIN OF THE EXPERT(S) PROVIDED WITH TRAINING	NO. PARTICIPANTS FROM THE CORRESPONDING COUNTRY
C	Nanopore Rapid and Ligation kits for ASFV library preparation for sequencing. Sample type and quality controls.	National Veterinary Research Institute, Vom, Nigeria	2
C	Realtime detection of ASFV, sequencing on Nanopore platform and Introduction to Basic Bioinformatics.	Laboratory, College of Veterinary Medicine Animal Resources And Bio Security Makerere University, P.o Box 7062, Kampala, Uganda	30
C	Realtime detection of ASFV, Sequencing on oxford platform, basic bioinformatics	Central Luzon State, Philippine	50
C	Foreign Animal Disease Diagnostician Training Course	United States	125
C	TB sample processing, colony identification, acid-fast staining, direct PCR, real-time PCR detection, interpretation of whole genome sequencing results	Guatemala	10
A	Real Time PCR training for ASF	Dominican Republic	35

A	TB testing	Mexico/USA	10
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## TOR8: SCIENTIFIC MEETINGS

9. Did your Collaborating Centre organise or participate in the organisation of scientific meetings related to your main focus area on behalf of WOA?H

Yes

NATIONAL/INTERNATIONAL	TITLE OF EVENT	CO-ORGANISER	DATE (MM/YY)	LOCATION	NO. PARTICIPANTS
International	FMD Reference Lab Network Meeting	WOAH	2023-10-01	Canada	1
International	Rinderpest Virus Holding Facility Network Meeting	WOAH	2023-02-01	UNK	1
International	Rinderpest Virus Holding Facility Network Meeting	WOAH	2023-08-01	UNK	1
International	Rinderpest Virus Holding Facility Network In-Person Meeting	WOAH	2023-12-01	France	1

## TOR9: DATA AND INFORMATION DISSEMINATION

10. Publication and dissemination of any information within the remit of the mandate given by WOA?H that may be useful to Members of WOA?H

a) Articles published in peer-reviewed journals:

22

*Full genome sequence for the African swine fever virus outbreak in the Dominican Republic in 1980; Scientific Reports, January 19, 2023. 2023, 13: 1024.*

<https://pubmed.ncbi.nlm.nih.gov/36658154/> E. Spinard, V. O'Donnell, E. Vuono, A. Rai, C. Davis, E. Ramirez-Medina, N. Espinoza, A. Valladares, M. Borca, D. P Gladue.

*The 2022 Outbreaks of African Swine Fever Virus Demonstrate the First Report of Genotype II in Ghana, Viruses, 2023, 15, 1722.*

<https://www.ncbi.nlm.nih.gov/pmc/articles/PMC10459280/> E. Spinard, A. Rai, J. Osei-Bonsu, V. O'Donnell, P. Ababio, D. Tawiah-Yingar, D. Arthur, D. Baah, E. Ramirez-Medina, N. Espinoza, A. Valladares, B. Faburay, A. Ambagala, T. Odoom, M. V Borca, D. P Gladue.

*Schneider D, Lehmkuhl AD, Spraker TR, Dittmar RO, Lockwood MA, Rollo S, Nichols TA. Tonsil biopsy to detect chronic wasting disease in white-tailed deer (Odocoileus virginianus) by immunohistochemistry. PLoS One. 2023 Mar 30; 18(3):e0282356. doi: 10.1371/journal.pone.0282356. eCollection 2023.*

*Amaresh Das, Zaheer Ahmed, Lizhe Xu, Wei Jia (2023). Assessment and verification of chemical inactivation of peste des petits ruminants virus by virus isolation following virus capture using Nanotrap magnetic virus particles. Microbiology Spectrum, September/October 2023 Volume 11 Issue 5. 0.1128/spectrum.00689-23.*

<https://doi.org/10.1128/spectrum.00689-23>

*Vaccination of White-Tailed Deer with Mycobacterium bovis Bacillus Calmette-Guérin (BCG): Effect of Mycobacterium avium ssp. paratuberculosis Infection; Mitchell V Palmer, Carly Kanipe, Kimberly A Lehman, Tyler C Thacker, Ellie J Putz, Paola M Boggiatto. PMC PubMed Central National Library of Medicine National Center for Biotechnology Information. PubMed Central*

*Advancing animal tuberculosis surveillance using culture-independent long-read whole-genome sequencing; Ghielmetti G, Loubser J, Kerr TJ, Stuber T, Thacker T, Martin LC, O'Hare MA, Mhlophe SK, Okunola A, Loxton AG, Warren RM, Moseley MH, Miller MA, Goosen WJ. Front Microbiol. 2023 Nov 21;14:1307440. doi: 10.3389/fmicb.2023.1307440. eCollection 2023. PMID: 38075895*

*Hamond C, LeCount K, A Springer Browne AS, Anderson T, Stuber T, Hicks J, Camp P, Fernandes LGV, van der Linden H, Goris MGA, Bayles DO, Schlater LK, Nally JE. Concurrent colonization of rodent kidneys with multiple species and serogroups of pathogenic Leptospira. Applied and environmental microbiology vol. 89, 10 (2023): e0120423. doi:10.1128/aem.01204-23*

*Stone NE, McDonough RF, Hamond C, LeCount K, Busch JD, Dirsmith, KL, Rivera-Garcia S, Soltero F, Arnold LM. Weiner Z, Galloway RL, Schlater LK, Nally JE, Sahl JW, Wagner DM, DNA Capture and Enrichment: A Culture-Independent Approach for Characterizing the Genomic Diversity of Pathogenic Leptospira Species. Microorganisms 2023, 11, 1282.*

*Anderson T, Hamond C, Haluch A, Toot K, Nally JE, LeCount K, Schlater LK. Animals Exposed to Leptospira Serogroups Not Included in Bacterins in the United States and Puerto Rico. Trop Med Infect Dis. 2023 Mar 22;8(3):183. doi: 10.3390/tropicalmed8030183. PMID: 36977184; PMCID: PMC10051158.*

*LeCount K, Fox K, Anderson T, Bayles DO, Stuber T, Hicks J, Schlater LK, Nally JE. Isolation of Leptospira kirschneri serovar Grippotyphosa from a red panda (Ailurus fulgens) after antimicrobial therapy: Case report. Front Vet Sci. 2023 Feb 2;9:1064147. doi: 10.3389/fvets.2022.1064147. PMID: 36819120; PMCID: PMC9932277.*



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Feng A, Bevins S, Chandler J, DeLiberto TJ, Ghai R, Lantz K, Lenocho J, Retchless A, Shriner S, Tang CY, Tong SS, Torchetti M, Uehara A, Wan XF. Transmission of SARS-CoV-2 in free-ranging white-tailed deer in the United States. *Nat Commun.* 2023 Jul 10;14(1):4078. doi: 10.1038/s41467-023-39782-x. PMID: 37429851; PMCID: PMC10333304.

Youk S, Torchetti MK, Lantz K, Lenocho JB, Killian ML, Leyson C, Bevins SN, Dilione K, Ip HS, Stallknecht DE, Poulson RL, Suarez DL, Swayne DE, Pantin-Jackwood MJ. H5N1 highly pathogenic avian influenza clade 2.3.4.4b in wild and domestic birds: Introductions into the United States and reassortments, December 2021-April 2022. *Virology.* 2023 Oct;587:109860. doi: 10.1016/j.virol.2023.109860. Epub 2023 Aug 2. PMID: 37572517.

Nguyen HT, Chesnokov A, De La Cruz J, Pascua PNQ, Mishin VP, Jang Y, Jones J, Di H, Ivashchenko AA, Killian ML, Torchetti MK, Lantz K, Wentworth DE, Davis CT, Ivachtchenko AV, Gubareva LV. Antiviral susceptibility of clade 2.3.4.4b highly pathogenic avian influenza A(H5N1) viruses isolated from birds and mammals in the United States, 2022. *Antiviral Res.* 2023 Sep;217:105679. doi: 10.1016/j.antiviral.2023.105679. Epub 2023 Jul 24. PMID: 37494978; PMCID: PMC10508830.

Ip HS, Uhm S, Killian ML, Torchetti MK. An Evaluation of Avian Influenza Virus Whole-Genome Sequencing Approaches Using Nanopore Technology. *Microorganisms.* 2023 Feb 19;11(2):529. doi: 10.3390/microorganisms11020529. PMID: 36838494; PMCID: PMC9967579.

Elsmo EJ, Wünschmann A, Beckmen KB, Broughton-Neiswanger LE, Buckles EL, Ellis J, Fitzgerald SD, Gerlach R, Hawkins S, Ip HS, Lankton JS, Lemley EM, Lenocho JB, Killian ML, Lantz K, Long L, Maes R, Mainenti M, Melotti J, Moriarty ME, Nakagun S, Ruden RM, Shearn-Bochsler V, Thompson D, Torchetti MK, Van Wettere AJ, Wise AG, Lim AL. Highly Pathogenic Avian Influenza A(H5N1) Virus Clade 2.3.4.4b Infections in Wild Terrestrial Mammals, United States, 2022. *Emerg Infect Dis.* 2023 Dec;29(12):2451-2460. doi: 10.3201/eid2912.230464. PMID: 37987580; PMCID: PMC10683806.

Patyk KA, Fields VL, Beam AL, Branan MA, McGuigan RE, Green A, Torchetti MK, Lantz K, Freifeld A, Marshall K, Delgado AH. Investigation of risk factors for introduction of highly pathogenic avian influenza H5N1 infection among commercial turkey operations in the United States, 2022: a case-control study. *Front Vet Sci.* 2023 Aug 30;10:1229071. doi: 10.3389/fvets.2023.1229071. Erratum in: *Front Vet Sci.* 2023 Nov 23;10:1336351. PMID: 37711433; PMCID: PMC10498466.

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Wünschmann A, Franzen-Klein D, Torchetti M, Confeld M, Carstensen M, Hall V. Lesions and viral antigen distribution in bald eagles, red-tailed hawks, and great horned owls naturally infected with H5N1 clade 2.3.4.4b highly pathogenic avian influenza virus. *Vet Pathol.* 2024 Jan 10:3009858231222227. doi: 10.1177/03009858231222227. Epub ahead of print. PMID: 38197395.

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2.3.4.4 Viruses. *Viruses*. 2023 Nov 18; 15(11):2273. doi: 10.3390/v15112273. PMID: 38005949; PMCID: PMC10674317.

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Andrea Fortin, Andrea Laconi, Isabella Monne, Siamak Zohari, Kristofer Andersson, Christian Grund, Mattia Cecchinato, Marika Crimauo, Viviana Valastro, Valeria D'Amico, Alessio Bortolami, Michele Gastaldelli, Maria Varotto, Calogero Terregino, Valentina Panzarin, A novel array of real-time RT-PCR assays for the rapid pathotyping of type I avian paramyxovirus (APMV-1), *Journal of Virological Methods*, Volume 322, 2023, 114813, ISSN 0166-0934, <https://doi.org/10.1016/j.jviromet.2023.114813>.

b) International conferences:

9

Rinderpest Virus Holding Facility Network meetings February and August 2023

Rinderpest Virus Holding Facility Network in-person meeting December 2023

WOAH sponsored conference on tuberculosis in Birmingham, UK

Hamond C, Adam E, LeCount K, Anderson T, Camp P, Hicks J, Stuber T, Morningstar-Shaw BR, Schlater LK, Nally JE. Thoroughbred mares as reservoir hosts of *Leptospira*. *International Symposium of the World Association of Veterinary Laboratory Diagnosticians*, June 2023, Lyon, France.

Hamond C, LeCount K, Morningstar-Shaw BR, Schlater LK, Nally JE. Optimization of PCR for the detection of pathogenic *Leptospira* in bovine semen. *International Symposium of the World Association of Veterinary Laboratory Diagnosticians*, June 2023, Lyon, France.

Hamond C. Overview of Animal Leptospirosis and Diagnostics. *5th European Congress on Infectious Diseases*, October 9-10, 2023, London, UK.

Hamond C. Leptospirosis: risk factors, diagnosis and control challenges. *International Forum on Public Health and Health Care Management 2023*, September 7-11, 2023, virtual.

Global consultation on Highly Pathogenic Avian Influenza (HPAI) FAO virtual meeting

World Organisation for Animal Health and Food and Animal Organization (OFFLU) Swine influenza virus technical activity teleconferences and WHO Vaccine Composition meeting preparation

c) National conferences:

13

Post presentation of ASF/CSF multiplex at the 2023 CRWAD meeting

US Animal Health Association, American Association of Veterinary Laboratory Diagnosticians, 2023

Hamond C, LeCount K, Morningstar-Shaw BR, Schlater LK, Nally JE. Optimization of PCR for the detection of pathogenic *Leptospira* in bovine semen. *United States Animal Health Association 2023 Annual Meeting*. October 12-18, 2023, National Harbor, MD.

Hamond C, Adam E, LeCount K, Anderson T, Camp P, Hicks J, Stuber T, Morningstar-Shaw BR, Schlater LK, Nally JE. Thoroughbred mares as reservoir hosts of *Leptospira*. *2023 American Association of Equine Practitioners Convention*. December 2023, San Diego, CA.

Live Bird Market Working Group Meeting February 2023

National Poultry Improvement Program Biennial General Conference Committee Meeting June 2023

NPIP Diagnostic Workshop August 2023

Animal Influenza Viruses Gap Analysis Workshop

AVMA American Association of Avian Pathologist Conference August 2023

AAVLD Highly pathogenic avian influenza virus infection of raptors in the upper Midwest June 2023

American College of Veterinary Pathologists November 2023

Alaska Bird Conference December 2023

USDA IAV-S Stakeholder Workshop March 2023

d) Other (Provide website address or link to appropriate information):

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*Home | Animal and Plant Health Inspection Service (usda.gov)*

*PPT presentations on forums which we are invited and requested laboratory tours*

*NVSL NAHLN African Swine Fever Laboratory Preparedness Week, June 12-15, 2023, Manhattan, Kansas*

*Monthly TB summary reports and affected herd maps: (<https://www.aphis.usda.gov/aphis/ourfocus/animalhealth/animal-disease-information/cattle-disease-information/tuberculosis-brucellosis-monthly-report/tb-bruc-reports>)*

*HPAI Epidemiologic Analysis of Poultry Flocks (usda.gov)*

*World Organisation for Animal Health and Food and Animal Organization (OFFLU) Swine influenza virus technical activity teleconferences and WHO Vaccine Composition meeting preparation*

*ACVM: HPAI Update: <https://www.acvm.us/>*

*HPAI sequencing and genetics at the monthly NASAHO call*

*Avian Influenza panel at Iowa Egg Producers meeting*

*HPAI Wildlife Phylongentic Update for Wildlife Services Annual Meeting*

*Abstract: Alexander Morris ER, Torchetti MK, Killian ML, Suarez D, Dimitrov KM. Development of a Multiplex rRT-PCR for Simultaneous Detection of all Pigeon-adapted Type 1 Avian Paramyxoviruses and first detection of Asian lineage viruses in the US. Oct 2023 American Association of Veterinary Diagnostic Laboratories, National Harbor, MD*

*Participated with international regionalization evaluation for Newcastle disease*

*<https://www.aphis.usda.gov/aphis/ourfocus/animalhealth/animal-disease-information/equine/cem/contagious-equine-metritis>*

11. What have you done in the past year to advance your area of focus, e.g. updated technology?

*Improved various laboratory techniques as reported above*

12. Additional comments regarding your report:

*ANTHRAX: This is a well-controlled disease within the United States. There were very few premises with animals that tested positive for anthrax in 2023. The positive cases occurred in states that are endemic for anthrax and have staff and laboratories that are well prepared to confirm their own samples. The NVSL did not receive any requests for international diagnostic testing in 2023. NVSL maintains our availability and proficiency and are happy to accept samples from other countries. The NVSL does produce and distribute reagents internationally and provide scientific guidance to our international partners. We do not see the demand for testing increasing and since NVSL's mission is primarily diagnostics and not research, we do not expect our activities to change.*

*We have completed RVCV sequencing and destruction project according to the WOA and FAO approved project proposal (sequencing data analysis is ongoing).*

*There is no network of WOA or FAO Rinderpest Reference Laboratories. There is a network of WOA/FAO Rinderpest Virus Holding Facility. We have participated in all the activities of the RHF Network such as providing updates, participating in discussions, exchanging information, and answering RP related inquiries of other RHF.*

*Currently, there are no WOA recognized reference reagents for Mammalian Tuberculosis.*

*At present there is no internationally recognized approved vaccine for mammalian tuberculosis. NVSL is involved in a vaccine study (mentioned in TOR5: Collaborative Scientific and Technical Studies) to evaluate the use of a vaccine for bovine tuberculosis in dairies. This project is still ongoing and production of an international vaccine for use in animals around the world is premature. However, vaccine production has been taken into consideration and the USDA has worked with a production facility to prepare for large scale production if the efficacy of the vaccine is confirmed.*

*Though we did not participate in specific TB training events, we have continued contact with the countries we have trained the year before and continued to provide support. This support is through continued dialog and not specifically defined training events.*

*Our laboratory participates in proficiency testing to maintain our ISO 17025:2017 accreditation. We currently use commercially available proficiency tests for Mammalian TB testing. The proficiency testing is performed twice each year. We were not contacted by the other reference laboratories to determine if we would offer a proficiency test, nor were aware of any proficiency tests offered by other reference laboratories.*

*We did not receive any requests from WOA to attend meetings, review standards relating to mammalian tuberculosis, or provide responses to technical queries or papers though we are very willing to participate and would appreciate the opportunity.*

*Participated in multiple Lepto surveillance studies that greatly increased variety of tests and testing numbers this year. A post-doc is currently on staff that has increased testing as well.*