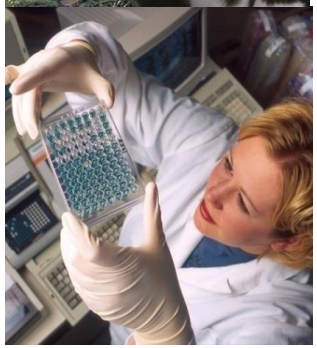
USDA
United States Department of Agriculture

Veterinary Services





Swine Enteric Coronavirus Diseases (SECD)

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Animal and Plant Health Inspection Service
Veterinary Services
STAS/NVSL/DVL/BPA
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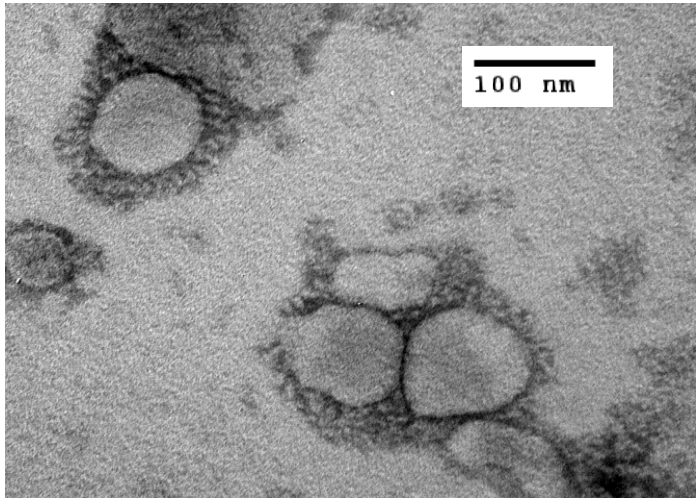
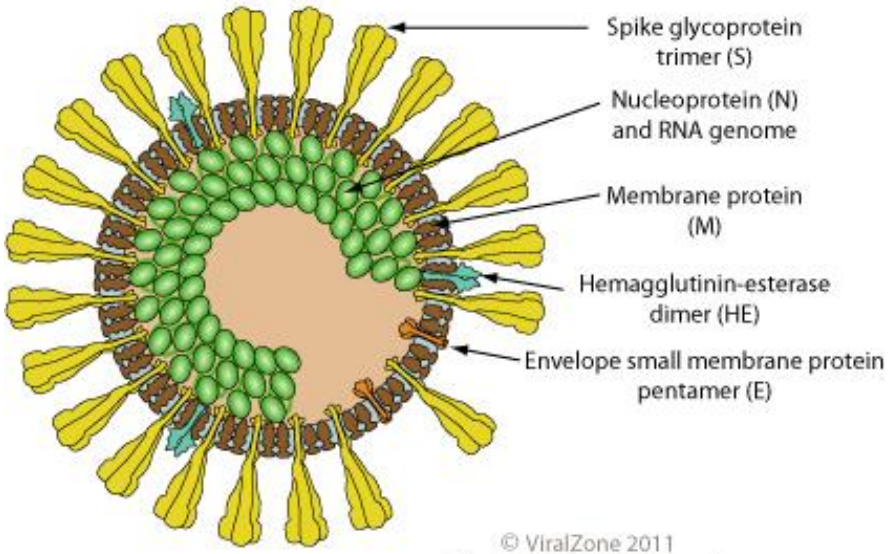
Overview

- **Background on porcine coronaviruses**
- **Porcine epidemic diarrhea virus (PEDV)**
- Porcine deltacoronavirus (PDCoV)
- **Federal Order**



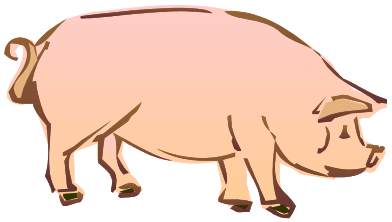
Coronavirinae

- +ss RNA virus
- Enveloped, spike glycoprotein resembles a crown by electron microscopy (EM)



Coronaviridae

- Four genera
  - Alphacoronaviridae
    - Porcine epidemic diarrhea virus (PEDV)
    - Transmissible gastroenteritis virus (TGEV)
    - Porcine respiratory coronavirus (PRCV)-TGE mutant
  - Betacoronaviridae
    - Porcine hemagglutinating encephalomyelitis virus (HEV)
  - Gammacoronaviridae
  - Deltacoronaviridae
    - Porcine deltacoronavirus (PDCoV)



CVB Licensed Products

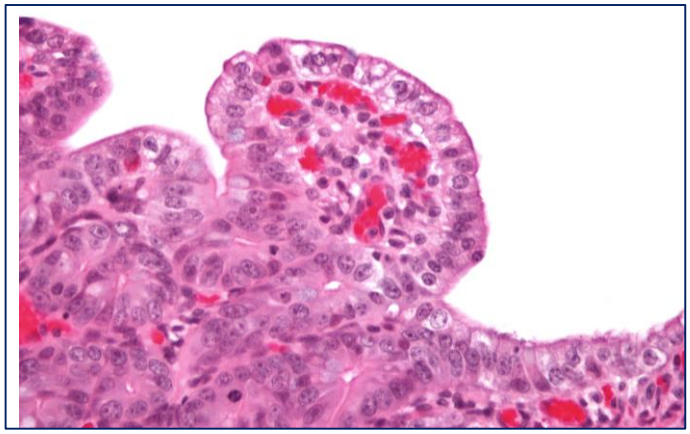
- Vaccines
  - TGEV-Merck/Intervet
  - PEDV
    - Conditionally licensed
    - Harrisvaccines conditional
    - Zoetis conditional
  - PDCoV-none
- Diagnostic assays
  - None

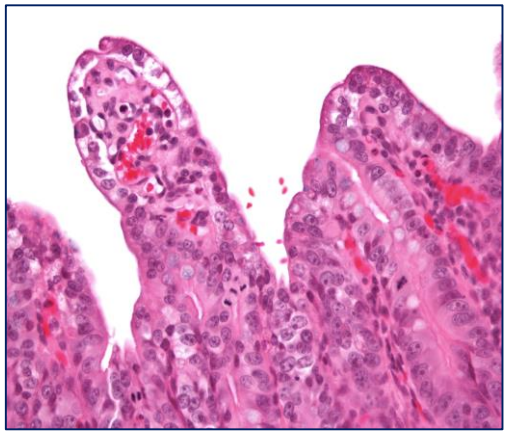
Clinical presentation

- Similar for PEDV, PDCoV, TGEV
  - Diarrhea, vomiting, lethargy, dehydration
  - Diarrhea is the result of malabsorption due to enterocyte infection
- Clinical signs can appear within 24 hours of infection (generally 1-3 days)
- Severity of disease is age dependent with nursing pigs more severely affected
  - Mortality can reach 100% in nursing pigs
  - Mortality rates much lower in grow/finish pigs

Gross & Microscopic lesions

- Pigs may appear gaunt with fecal staining
- Intestines thin and fluid filled
- Viral replication in enterocytes resulting in blunted intestinal villi





Images courtesy of Drs. Lehmkuhl and Predgen, NVSL PL

PEDV History

- 1971 - Large outbreaks in European swine herds
- 1978 - Coronavirus-like agent was identified
- 2010-2012 – Outbreaks of high morbidity and mortality in China
- 2013 – Identification in U.S. herds
- Not reportable to the World Organization for Animal Health (OIE)
- Worldwide distribution with re-emergence
  - Asia, Canada, Columbia, Dominican Republic, Mexico, Ecuador, Peru, Europe, Ukraine, United States…

Headlines

Porcine epidemic diarrhea virus continues to spread

Deadly pig virus slips through US borders
Researchers race to track spread of coronavirus.

'Piglet Smoothie' Keeps Hogs Virus-Free
Humane Society exposes icky practice at Kentucky farm

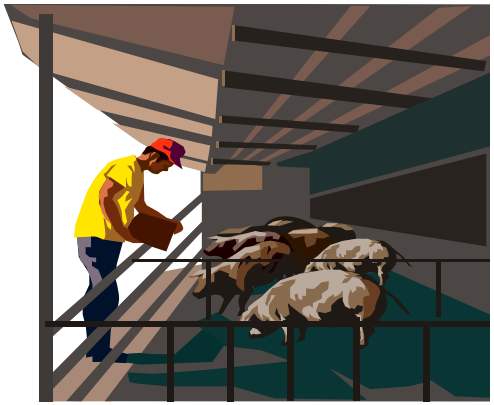
Pig Virus Mysteriously Returns to Indiana Farm

Virus Plagues the Pork Industry, and Environmentalists

USDA to issue federal order in response to Porcine Epidemic Diarrhea virus

Initial PEDV Detection in US

- First diagnosed in U.S. in mid-May, 2013
  - Stevenson et al., 2013
  - Original samples submitted to NAHLN lab
  - Clinical picture similar to TGE
  - Affected all pig ages
  - 90-95% mortality in suckling piglets
  - Submitted to NVSL for confirmatory testing
    - PCR and sequencing
    - Most closely related to 2012 Chinese PEDV



- Retrospective testing indicates PEDV was present in April, 2013

SUNDAY	MONDAY	TUESDAY	WEDNESDAY	THURSDAY	FRIDAY	SATURDAY (TOTALS)
	15 **APRIL**	16 OH – GF	17	18	19	20 1 GF
21	22	23	24	25	26 IN – GF	27 1 GF
28	29 IA (W. Central) – SOW	30 IA (NE) – SOW OH – GF IA – GF	1 **MAY** IA – GF	2 IA – GF	3 IA – GF	4 4 GF 2 SOW
5	6 IA (NW) – SOW	7 IA – GF IA – GF	8 IN – SOW MN – SOW IA – GF	9 IA – GF IA – GF IA – GF	10 CO (Eastern) – SOW IA – GF IN – ?? IN – ?? PA – ??	11 7 GF 4 SOW 3 UNKNOWN
12	13 MN – GF	14 CO – SOW MN – GF	15 MN – SOW MN – GF	16 IA – SOW	17 IA – SOW IA – SOW IA – GF	18 4 GF 5 SOW (31 actual cases)
19	20 IA – SOW IN – SOW IN – SOW IA – GF MN – GF MN – GF	21 CO – GF IA – GF IA – GF IA – GF IA – GF IA – GF IA – GF IA – GF IA – GF MN – GF OH – GF	22 CO – SOW CO – SOW IA – GF IA – GF IA – GF IA – GF IA – GF IA – GF IA – GF OK – GF	23 IA – SOW IA – SOW GF OH MN – ??	24 IA – GF IA – GF IA – GF IA – GF IA – GF CO – GF CO – GF MN – GF MN – GF MO – GF MN – ??	25 34 GF 7 SOW 2 UNKNOWN

Outcome of retrospective testing & on-boarding PEDV PCR – Courtesy Dr. Harry Snelson

Initial Response

- Nonregulatory
- No mandatory reporting
- No movement controls

- Epidemiological studies to investigate inter-herd transmission

- Epidemiological investigations and assessments to discover introductory pathways

- Industry and Academia led research efforts to understand viral ecology and disease dynamics

Experimental PEDV

- RNA detected in serum
- Extended fecal shedding
  - Several weeks
  - Even after clinically normal
- Protection in neonate piglets dependent on colostral intake of IgA
- NOT zoonotic, NO food safety issues
  - Only swine affected

NVSL PEDV Study

- Study design
  - Evaluate infectivity
    - Measured by fecal shedding of PEDV and seroconversion
    - Not designed for pathogenesis purposes
  - Rectal swabs collected twice daily the first 8 days of study
  - Blood collected weekly for 3 weeks to assess antibody response
  - Provide reagents to share

NVSL PEDV Study

- Bioassay (4 wk old pigs)
  - 3 pigs orally inoculated with 10% tissue homogenate (fecal material) from diagnostic PEDV case
  - Day 0: inoculated 3 cc in AM & PM
  - Day 1 AM & PM: vomitus in pen
  - Day 2: vomiting in AM, loose stool in AM & PM

NVSL PEDV Study

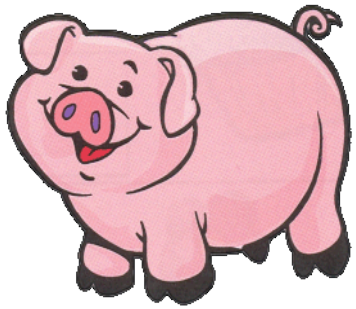
- Bioassay (4 wk old pigs)
  - Day 3: Stool more formed
  - Day 4: Fairly normal stools
  - Day 8: Re-challenged for reagent purposes
- Laboratory testing
  - rRT-PCR Day 1 fecal samples
    - Negative on AM collection
    - Positive on PM collection
    - Remained positive through end of sample collection (day 8)
  - Serum IFA testing—screening assay
    - Negative day 7
    - Positive day 15

Impacts on U.S. Pig Industry

- Loss of pigs
  - ~10% of domestic pig population lost in 1st yr
  - Equates to ~ 7 million piglets (Jung et al., 2015)
- Impact on surviving pigs
  - Field study indicated reduced performance in growing pigs (Alvarez et. al, 2015)
- Export markets
  - Some closed; increased pre-export testing
- Other
  - Decreased slaughter, increased meat prices, employment impacts, etc.

PEDV Transmission

- Fecal-oral route
- Asymptomatic pigs
- Fomites
- Aerosol (Alonso et al., 2014)
- Vehicles (Lowe et al.,2014)



Plasma in Feed

- Definitions
  - SDPP = spray dried porcine plasma
  - SDBP = spray dried bovine plasma or spray dried blood plasma (SDBP=bovine for presentation)
- Source and purpose
  - Blood collected from healthy pigs at slaughter
  - Increased feed intake
  - Better growth performance

PEDV Transmission-Feed/Feed Products

- Can be detected by PCR in feed & SDPP
- Mixed results detecting live virus
- Influenced by # pigs, pig age, sample size, age of sample, strain (?)
- No transmission in bioassay when plasma from viremic pigs or experimentally contaminated plasma spray dried with bench-top laboratory sprayer (Gerber, et al., 2014)

PEDV Transmission-Feed/Feed Products

- PEDV in SDBP was inactivated & SDBP contaminated with PEDV didn't survive past 7 days at room temp or 21 days at refrigerator temp using cell culture (Pujols, 2014)
- PEDV PCR positive pelleted feed (unopened bags) did not transmit virus in bioassay (Bowman, 2015)
- SDPP in feed bioassay did not transmit virus (Opriessnig, 2014)
- NVSL unpublished data-feed, SDPP in bioassay didn't transmit (worked closely with Canada)

Positive results

- Feed collected from positive herd feed bin using paint roller demonstrated infectivity in bioassay (Dee et al., 2014)
- SDPP but not feed resulted in bioassay transmission (Pasick et al., 2014)

PEDV Transmission

- Limit transmission via good biosecurity practices (http://www.pork.org/)
- Effective disinfectants
  - Thorough cleaning a must before disinfection
  - Oxidizing agents (Virkon S)
  - Bleach
  - Phenolic compounds (One-Stroke Environ)
  - Many others (Pospischil et al., 2002)
- Disinfectants (in general) do not disrupt RNA
  - PCR + even if virus inactivated (Bowman et al., 2015)

PEDV Diagnostic Testing

- Virus Detection
  - Virus Isolation
    - Difficult virus to isolate and adapt to cell culture
    - Requires multiple passes (4+)
  - PCR
    - Nested RT-PCR (+/- sequencing)
    - rRT-PCR
- Antibody detection
  - Indirect Fluorescent Antibody
  - ELISA
  - Fluorescent focus neutralization
  - One-way cross reaction in CCIF with Miller TGE antisera (Lin et al., 2015)

PEDV Sequencing

- Isolate or original materials
- Whole genome or gene specific
- PEDV strains (S gene)
  - Sequence typical of index case (non-S INDEL)
  - S INDEL (PEDV variant-INDEL)
    - Multiple insertions and deletions
  - Vlasova et al., 2014, EID, 1620-1628
  - Strain S2aa-del
    - Large amino acid deletion (197 aa) in S gene
    - Marthaler, et al., 2014
    - Oka, et al., 2014

Husbandry Practices

- Strict biosecurity practices
- Immunity measures
  - Goal is for lactogenic immunity
  - Vaccines
  - Feedback…….traditional method but..

http://www.newser.com/story/182670/how-hogs-stay-virus-free-piglet-smoothie.html

Feedback

'Piglet Smoothie' Keeps Hogs Virus-Free

Humane Society exposes icky practice at Kentucky farm (Newser, 2014)

This is about as unappetizing a meal as you could get. The Humane Society has exposed a method of dealing with the fast-spreading porcine epidemic diarrhea virus (PEDV) that's killed more than 2 million piglets since April: Grind up the intestines of piglets that succumbed to it, and feed the "piglet smoothie" to the sows—some of which could be the dead piglets' own mothers, NPR reports.

Major USDA Activities Since May 2013

- Collection of lab testing data
  - Voluntary reporting through NAHLN labs
  - Weekly reports and maps
  - WWW.USDA.GOV (Search for 'SECD')
- Epidemiologic investigations of herds
- Risk pathways assessments
- Diagnostic testing and information sharing
  - Domestic
  - International

Federal Order

- Effective June 5, 2014
- Mandatory reporting
  - SECD-PEDV, SDCoV
- Development of herd plans
- Cost-share approach
  - diagnostic testing
  - biosecurity activities
  - developing herd plans
  - implementing disease control activities
- www.aphis.usda.gov/animal-health/secd

Background on NAHLN

- In 2002, APHIS and CSREES (now NIFA) initiated the Network by entering into cooperative agreements with 12 State and university veterinary diagnostic laboratories. These were funded by Homeland Security appropriations.

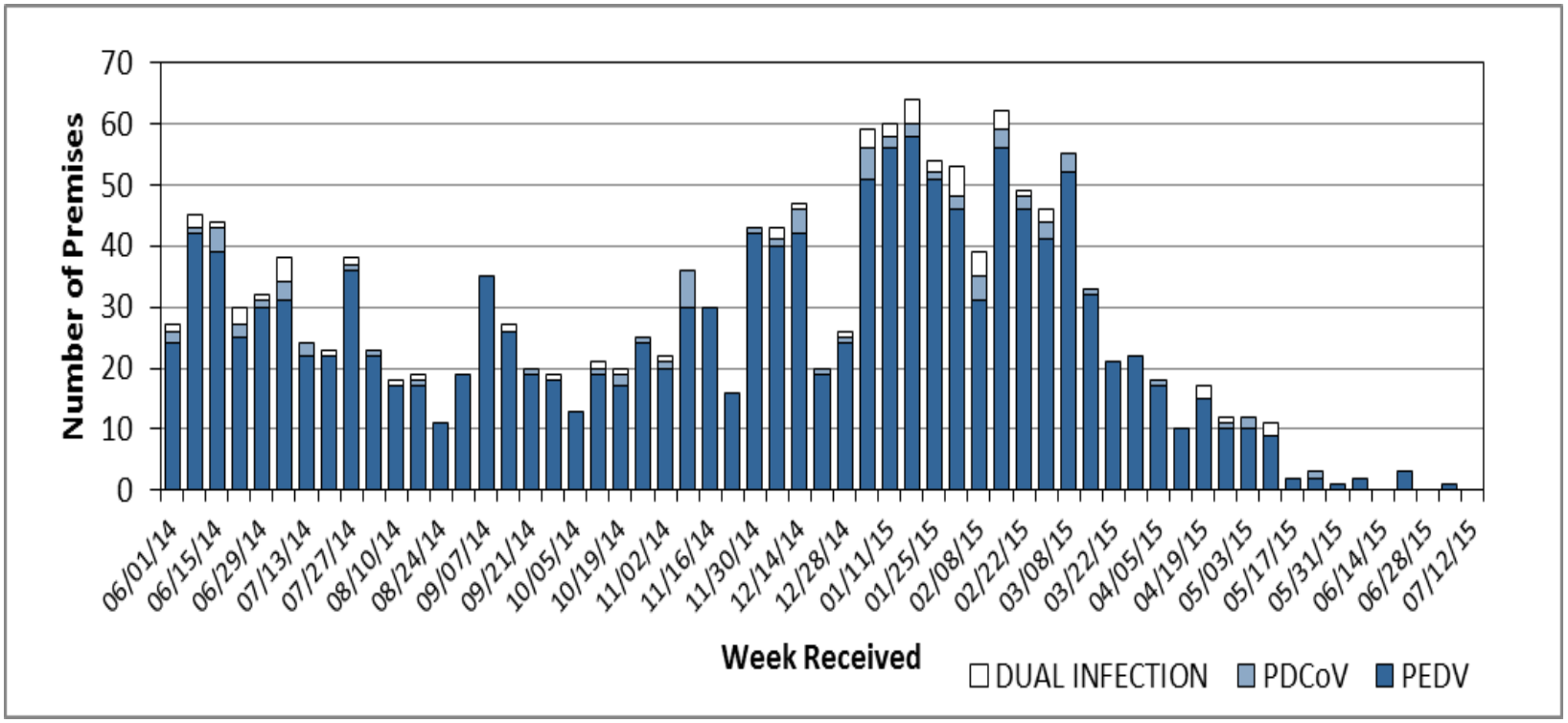
** SECD assays do not currently follow traditional model of other NAHLN diseases; but NAHLN laboratories play a significant role in the identification and testing for PEDV and PDCoV (collectively SECD)

NAHLN laboratories and SECD testing

- June 2014– Several NAHLN labs began reporting SECD results via electronic messaging to VS
- July 2014– NVSL completed a ring trial evaluation/comparison of NAHLN lab testing methods
  - Although different PCR methods were used in the NAHLN labs, NVSL was able to evaluate and provide feedback regarding efficacy of detection

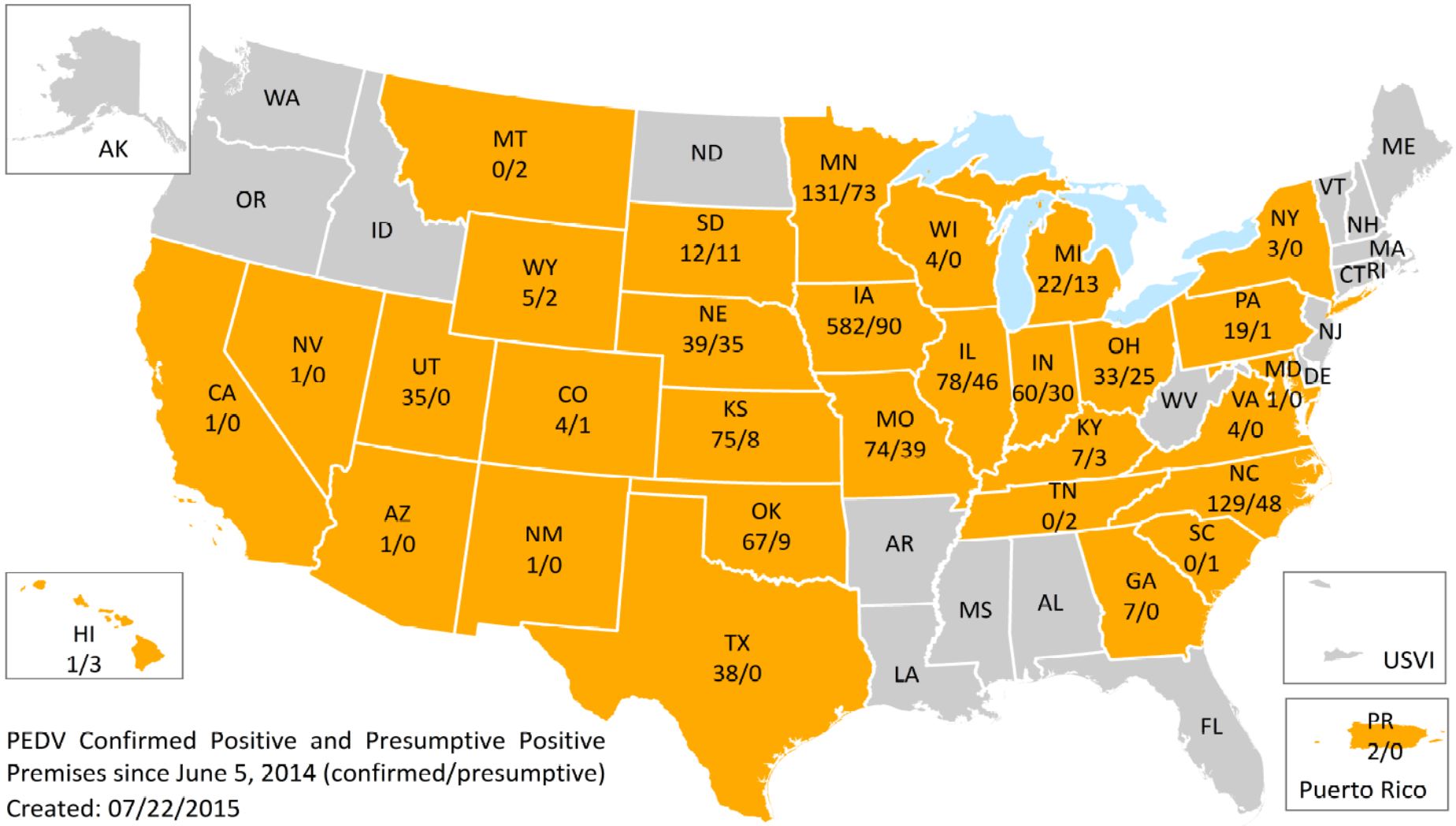
“Epidemic Curve”

Number of Confirmed Positive Premises by Week [a]



[a]Week the sample was received at the laboratory for testing

Geographic Distribution of PEDV



PEDV Confirmed Positive and Presumptive Positive Premises since June 5, 2014 (confirmed/presumptive)
Created: 07/22/2015

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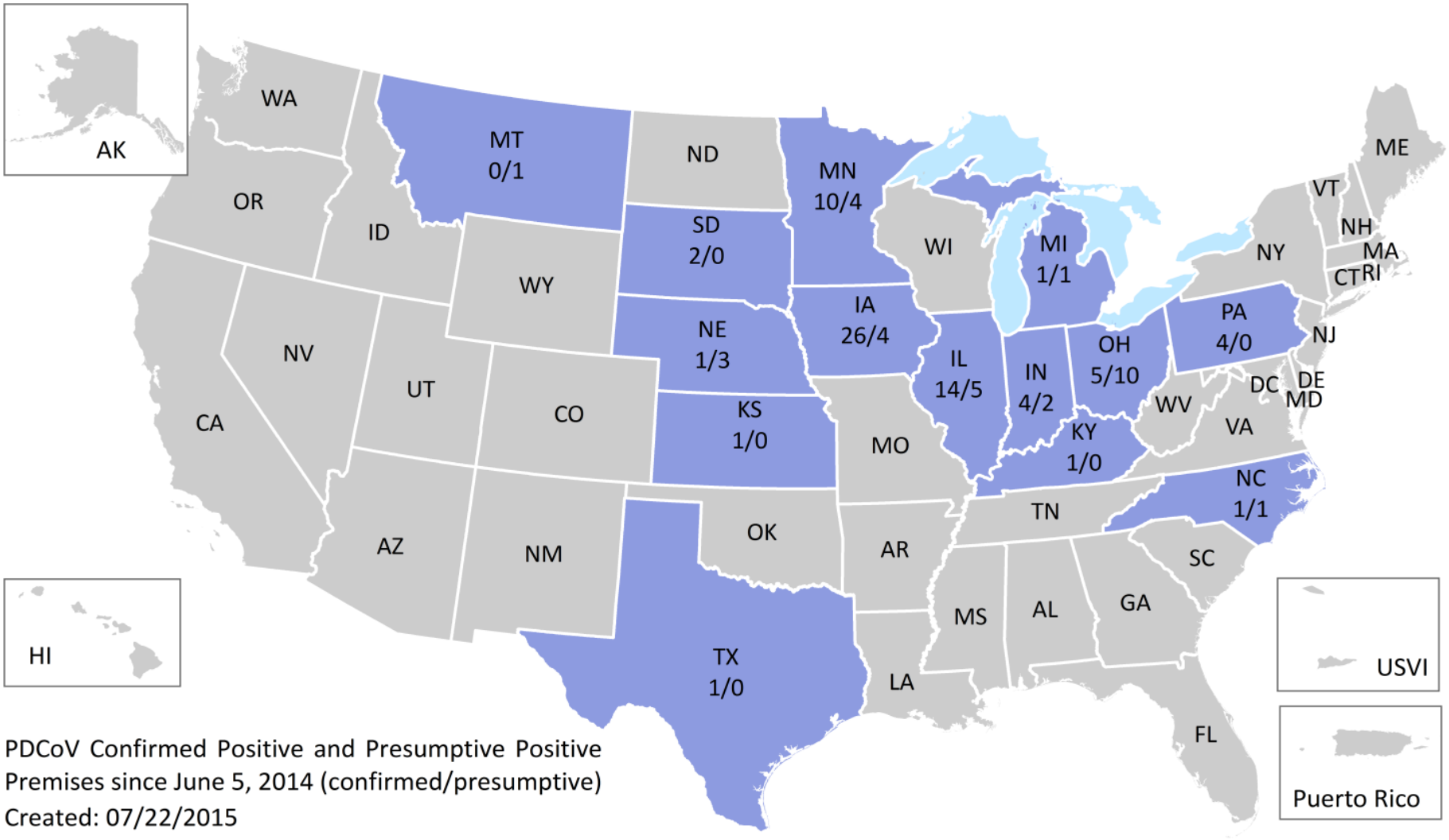
Geographic Distribution of PDCoV



PDCoV Confirmed Positive and Presumptive Positive Premises since June 5, 2014 (confirmed/presumptive)
Created: 07/22/2015

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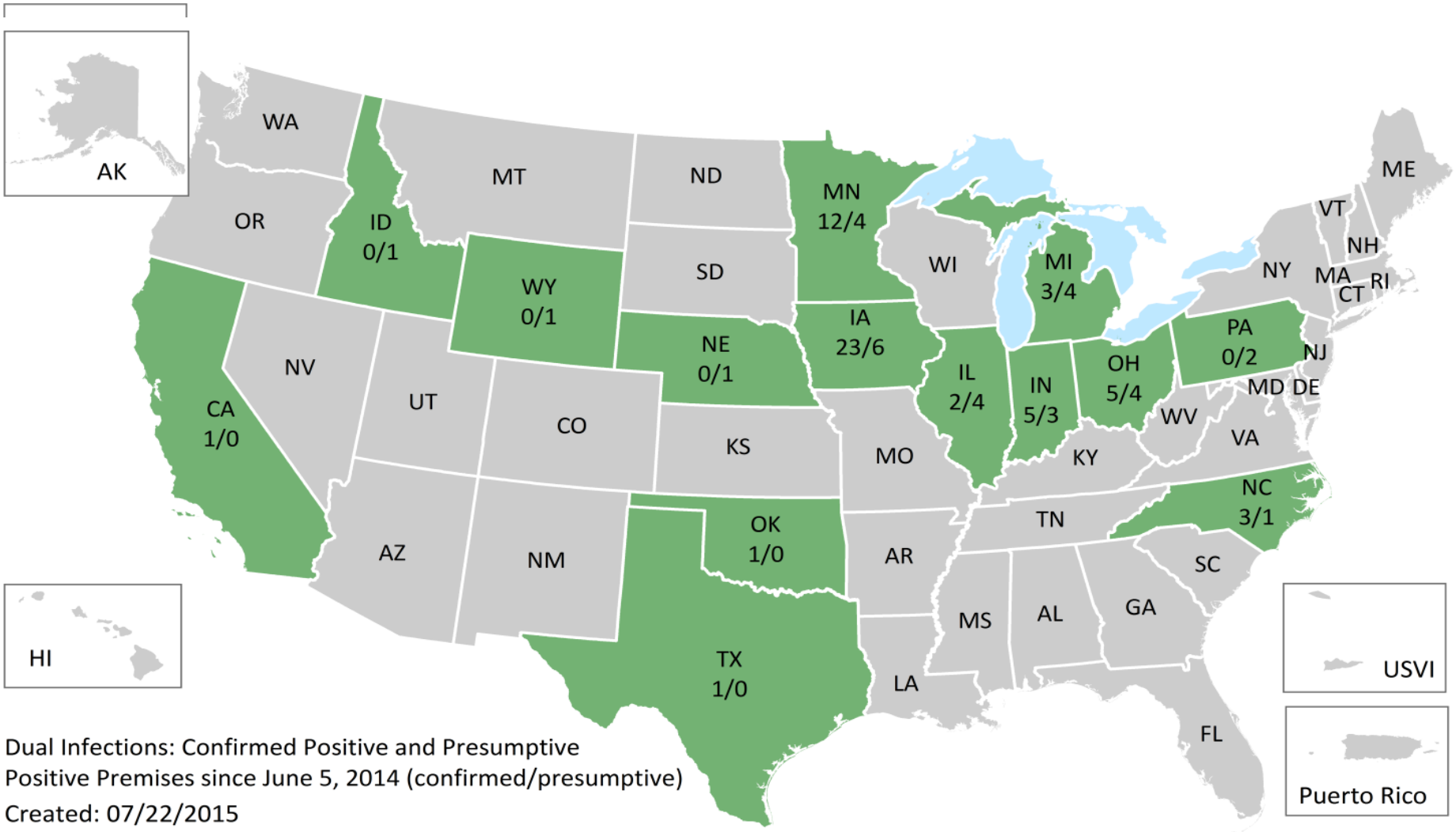
PEDV/PDCoV Dual Infections



Dual Infections: Confirmed Positive and Presumptive
Positive Premises since June 5, 2014 (confirmed/presumptive)
Created: 07/22/2015

It Takes A Village…

- DVL BPA employees
- Dr. Brian McCluskey
- Dr. John Schiltz
- Dr. Eileen Ostlund
- Dr. Nancy Clough
- Dr. Ellen Kasari
- Dr. Harry Snelson

Internet Resources

- www.aasv.org
- www.aphis.usda.gov/animal-health/secd
- http://www.pork.org/pork-checkoff-research/pedv/
- www.oie.int
- Many university veterinary diagnostic labs have SECD information on their websites

Questions?