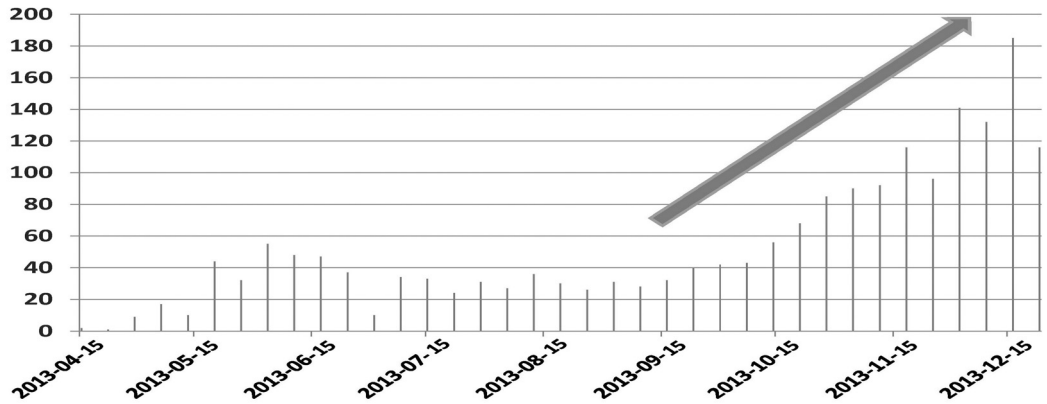
돼지 질병분야 신기술과 동향
PED (Porcine Epidemic Diarrhea)

김 현 일 수의사
(주)옵티팜 대표이사

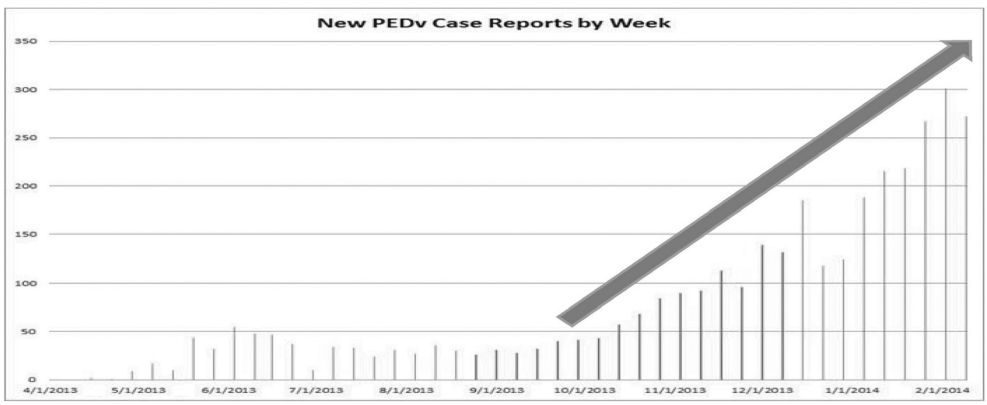
Table 1. Total number of PEDv positive laboratory swine accessions / premises tested per week

Week of	Total Number of Swine Accessions/Diagnostic Case Submissions Testing Positive for PEDv	Farm Type / Age Class				
		Suckling	Nursery	Grower / Finisher	Sow / Boar	Unk
4/15/2013	2	0	0	1	0	0
4/22/2013	1	0	0	1	0	0
4/29/2013	9	0	0	5	1	0
5/6/2013	17	0	0	7	4	3
5/13/2013	10	0	0	3	5	1
5/20/2013	44	0	0	33	7	3
5/27/2013	32	0	0	22	6	1
6/3/2013	55	0	0	34	13	5
6/10/2013	48	0	0	24	11	7
Subtotals *	218	Not Reported	Not Reported	130	47	20
6/16/2013	47	7	5	13	11	14
6/23/2013	37	7	7	13	1	9
6/30/2013	10	3	3	3	1	0
7/7/2013	34	2	5	15	10	3
7/14/2013	33	4	10	10	4	6
7/21/2013	24	2	2	8	4	9
7/28/2013	31	7	6	9	5	7
8/4/2013	27	12	1	9	6	1
8/11/2013	36	20	7	4	6	1
8/18/2013	30	11	12	2	3	2
8/25/2013	26	9	8	0	6	3
9/1/2013	31	6	10	8	4	3
9/8/2013	28	16	6	2	4	3
9/15/2013	32	11	8	7	1	5
9/22/2013	40	6	6	11	10	8
9/29/2013	42	8	10	11	6	8
10/6/2013	43	14	10	12	5	2
10/13/2013	56	22	11	14	2	7
10/20/2013	68	26	7	15	15	5
10/27/2013	85	23	16	28	17	4
11/3/2013	90	17	28	28	7	12
11/10/2013	92	16	28	36	4	12
11/17/2013	116	34	20	39	6	21
11/24/2013	96	21	23	15	3	34
12/1/2013	141	29	26	36	11	42
12/8/2013	132	22	36	37	9	29
12/15/2013	185	27	37	41	10	73
12/22/2013	116	22	21	35	7	34
TOTAL	1946	404	369	591	225	377

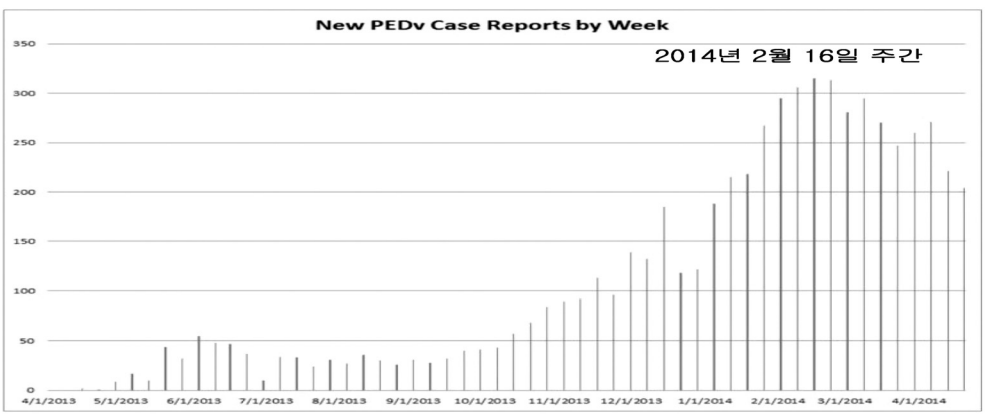
미국에서의 PED cases (2013년 12월)



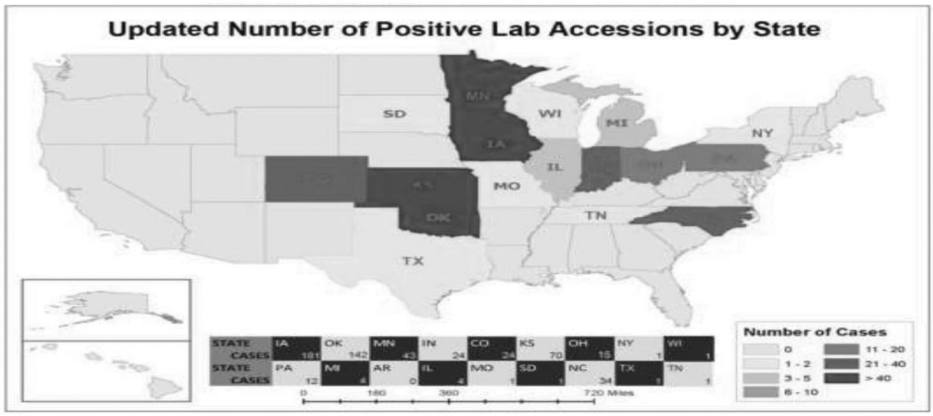
미국에서의 PED cases (2014년 2월)



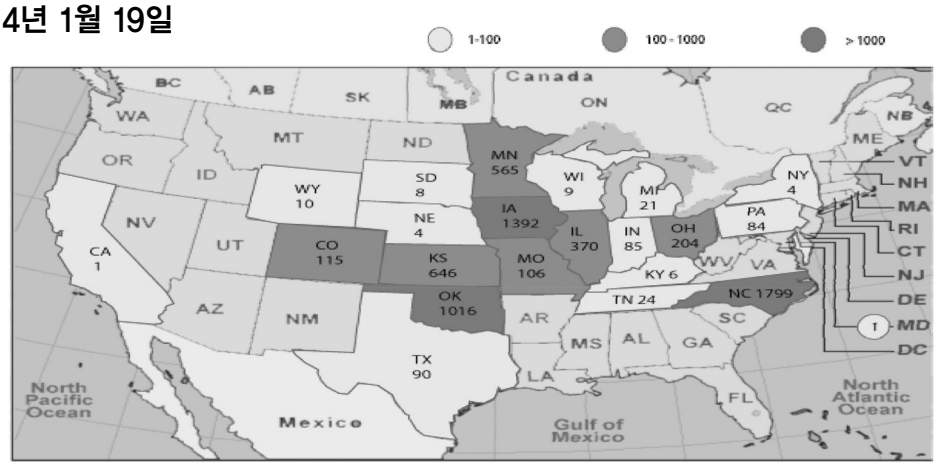
미국에서의 PED cases (2014년 4월)



2013년 9월 13일



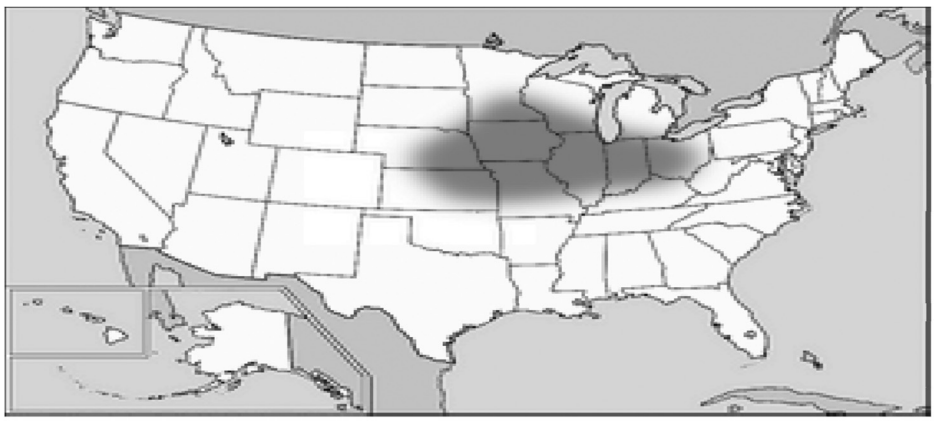
2014년 1월 19일



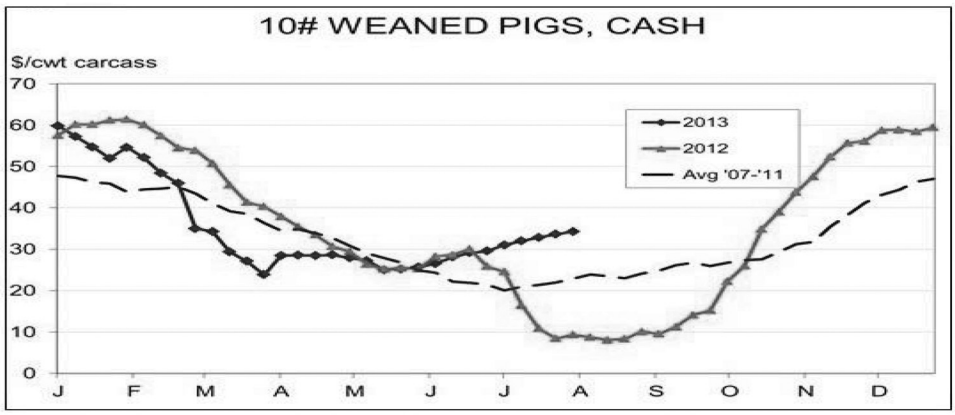
IA(1887)>MN(1018)>NC(613)>IL(608)

미국 내 양돈장 분포

US Corn Belt



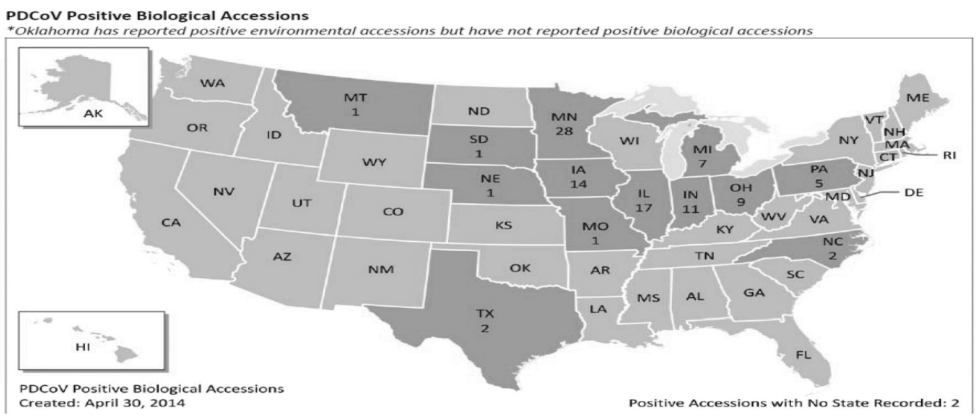
2013년 8월 미국 돈가



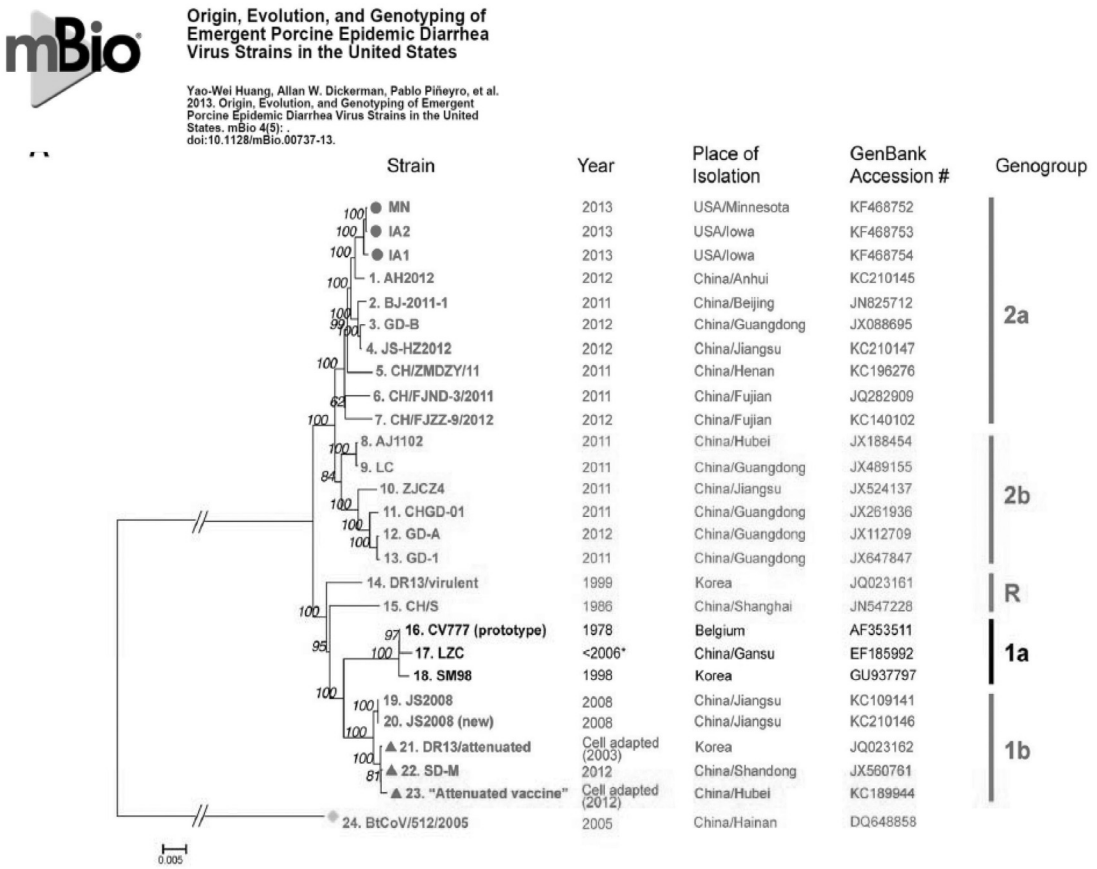
PED 발생 농가와 비발생 농가 간의 차이

요인	감염농장	비감염농장
최근 2주 이내 다른 농장에서 트레일러를 빌려옴	11%	0%
최근 2주 이내 모돈/후보돈/자돈 도태 및 이송	82%	38%
최근 2주 이내 자돈/육성돈 이동	89%	42%
농장 사람 가족이 도축장이나 다른 농장에 근무	37%	17%
농장 사람 가족이 다른 양돈 관련 산업에서 근무(사료)	11%	0%
최근 2주 이내 사체 처리업자가 농장 방문	93%	57%
최근 2주 이내 농장에서 야생 조류 목격	18%	4%
최근 2주 이내 다른 야생동물 목격	89%	59%
독수리 등 조류와 관련된 문제 발생	29%	13%
최근 2주 이내 자선단체에 돼지 이송	11%	38%

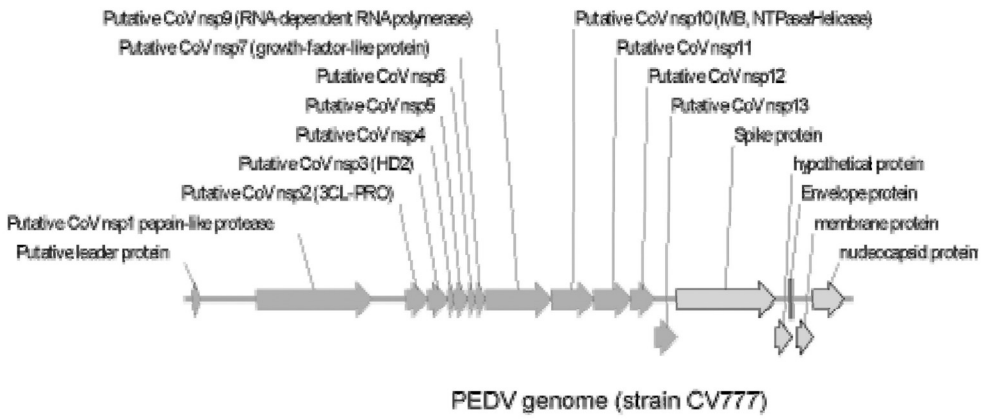
PDCoV (Porcine Delta Corona virus)



미국 PED 바이러스는 어디서 왔을까? 한국은?



Spike protein으로 유전자 분석



미국 PED 발생 원인?

OBSERVATION

Origin, Evolution, and Genotyping of Emergent Porcine Epidemic Diarrhea Virus Strains in the United States

Yao-Wei Huang,[a,b] Allan W. Dickerman,[c] Pablo Piñeyro,[a] Long Li,[d] Li Fang,[d] Ross Kiehne,[e] Tanja Opriessnig,[f,g] Xiang-Jin Meng[a]

Department of Biomedical Sciences and Pathobiology, College of Veterinary Medicine, Virginia Polytechnic Institute and State University, Blacksburg, Virginia, USA[a]; College of Animal Sciences, Zhejiang University, Hangzhou, China[b]; Virginia Bioinformatics Institute, Virginia Polytechnic Institute and State University, Blacksburg, Virginia, USA[c]; Hangzhou Beta Veterinary Diagnostic Laboratory, Hangzhou, China[d]; Swine Veterinary Center, P.A., St. Peter, Minnesota, USA[e]; Department of Veterinary Diagnostic and Production Animal Medicine, College of Veterinary Medicine, Iowa State University, Ames, Iowa, USA[f]; The Roslin Institute and The Royal (Dick) School of Veterinary Studies, University of Edinburgh, Edinburgh, United Kingdom[g]

ABSTRACT Coronaviruses are known to infect humans and other animals and cause respiratory and gastrointestinal diseases. Here we report the emergence of porcine epidemic diarrhea virus (PEDV) in the United States and determination of its origin, evolution, and genotypes based on temporal and geographical evidence. Histological lesions in small intestine sections of affected pigs and the complete genomic sequences of three emergent strains of PEDV isolated from outbreaks in Minnesota and Iowa were characterized. Genetic and phylogenetic analyses of the three U.S. strains revealed a close relationship with Chinese PEDV strains and their likely Chinese origin. The U.S. PEDV strains underwent evolutionary divergence, which can be classified into two sublineages. The three emergent U.S. strains are most closely related to a strain isolated in 2012 from Anhui Province in China, which might be the result of multiple recombination events between different genetic lineages or sublineages of PEDV. Molecular clock analysis of the divergent time based on the complete genomic sequences is consistent with the actual time difference, approximately 2 to 3 years, of the PED outbreaks between China (December 2010) and the United States (May 2013). The finding that the emergent U.S. PEDV strains share unique genetic features at the 5′-untranslated region with a bat coronavirus provided further support of the evolutionary origin of PEDV from bats and potential cross-species transmission. The data from this study have important implications for understanding the ongoing PEDV outbreaks in the United States and will guide future efforts to develop effective preventive and control measures against PEDV.

미국 PED 발생 근원지로 Anhui 주목!

한국 PED 새로운 Strain의 유입?

Viruses **2013**, *5*, 1991-2004; doi:10.3390/v5081991

OPEN ACCESS

viruses

ISSN 1999-4915

www.mdpi.com/journal/viruses

Article

Molecular Characterization and Phylogenetic Analysis of New Variants of the Porcine Epidemic Diarrhea Virus in Gansu, China in 2012

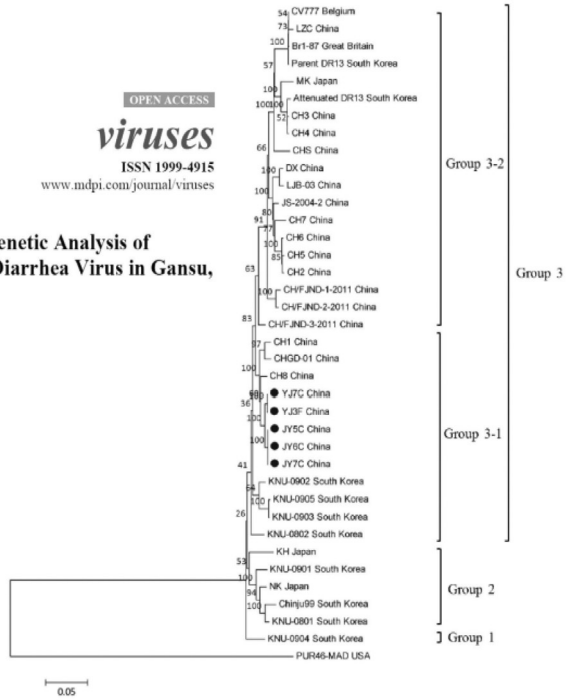
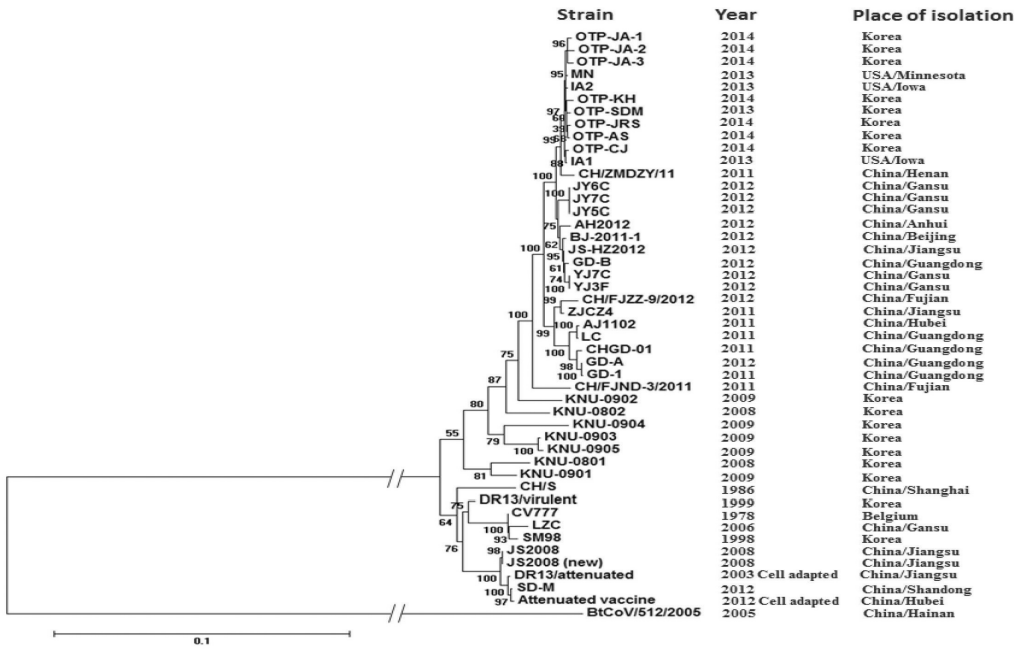
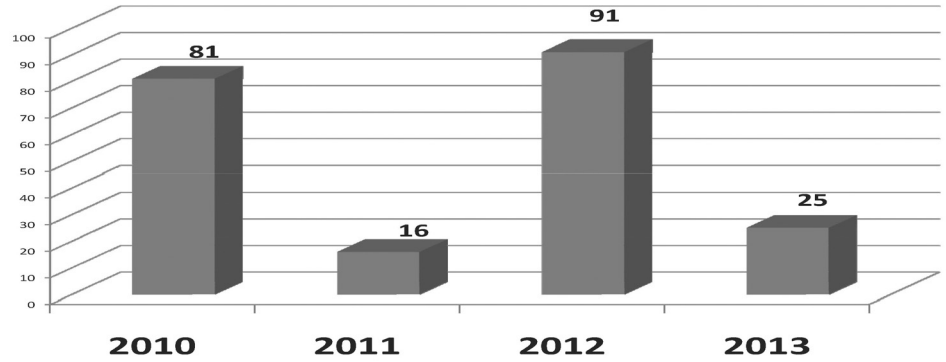


Fig. 1. Phylogenetic analysis of the nucleotide sequences of the complete spike gene of forty-seven PEDV strains



2년마다 발생 주기를 보이는 PED



PED 진단 시 주의사항

Infective material, concepts and procedures for intentional sow herd exposure to Porcine Epidemic Diarrhea virus

Kent Schwartz, DVM, MS
Iowa State University Veterinary Diagnostic Laboratory, Ames IA
Steve Henry, DVM, Dipl ABVP
Lisa Tokach, DVM, Dipl ABVP
Megan Potter, DVM, PhD
Abilene Animal Hospital PA, Abilene KS
Don Davidson, DVM, MS
Cody Egnor, DVM
PFFJ, LLC Taylor AZ

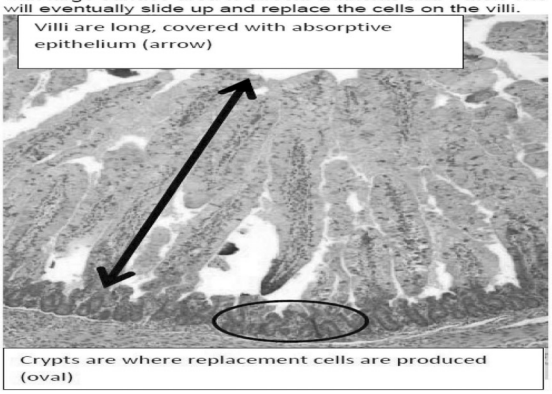
A philosophic preamble:
"We believe, regardless of the following words offered, that no one knows or can accurately predict how present clinical responses to PEDv infections will manifest over time. There are no tested, objective protocols. There are, at best, vague indices which are often used to define 'success' or 'failure'. Moreover, variation is a consistent feature of biology and swine production. Herd exposure to farrowing materials has evolved on farms to be a procedure that is commonly performed without consideration of context, clarity of objectives or process design. Based on our education and experience, we believe the following represents a logical, empirical, reasoned and balanced approach for herd exposure to PEDv."

포유자돈이 심하게 감염되는 이유

- Suckling piglets have greater risk for severe disease for several very important reasons :
  - **More susceptible cells** □ The villi of the small intestine of neonates are very long with many more mature or permissive enterocytes available to be infected by virus.
  - **Heal more slowly** □ The regeneration time for new epithelial cells (enterocytes) produced in the crypts is longer in neonates than in older pigs.
  - **Immature homeostasis** □ The colon of neonates is functionally immature and less able to compensate for **acid-base and** electrolyte imbalances or to resorb water.
  - **Osmotic pressures from undigested milk** : Damage to epithelium reduces enzyme activity and disrupts digestion of milk which increases the osmotic pressures within the intestine, effectively sucking water out of the piglet's body and into the intestine and feces

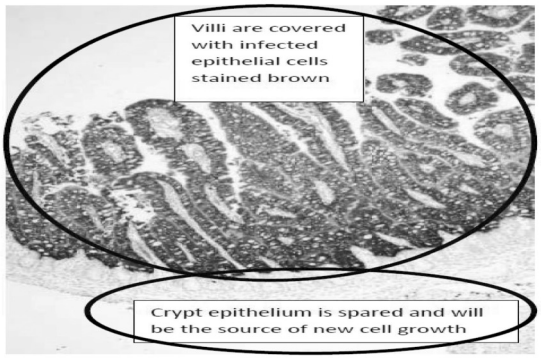
정상적인 자돈의 장 융모

Figure 1 shows the intestinal villi from a normal neonatal pig. Note that the villi are very long (black arrow) with large surface area to absorb nutrients and water. The crypts are where new cells are produce which will eventually slide up and replace the cells on the villi.



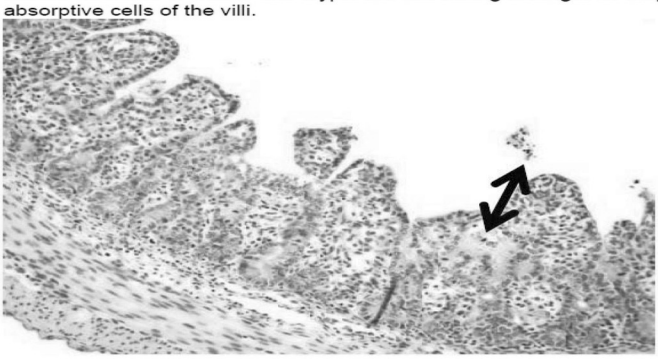
PED 감염 8시간 후 장 융모 면역염색 결과

Figure 2 shows early PED infection in a neonatal pig. The brown staining is infected cells lining the villi in a neonatal piglet roughly 8 hours after infection. All of the brown cells will die, releasing billions of virus particles to infect more cells and more pigs.



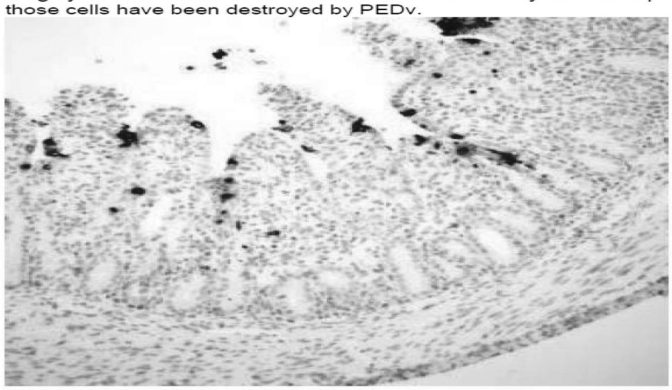
PED 감염 후 36시간 후의 장 융모

Figure 3 shows severe decrease in height of villi (villus atrophy – black arrow) and loss of absorptive epithelium approximately 36 hours after infection of a neonatal pig. There is much less surface area to absorb nutrients or water. The crypts are increasing in length as they produce more cells to rebuild the absorptive cells of the villi.



PED 감염 후 36시간 후의 장 융모 면역염색 사진

Figure 4 shows just a few brown-staining PEDv-infected cells remaining in the gut of a neonatal piglet roughly 36 hours after infection. Note that there is very little absorptive surface remaining since most of those cells have been destroyed by PEDv.

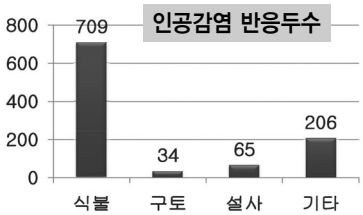


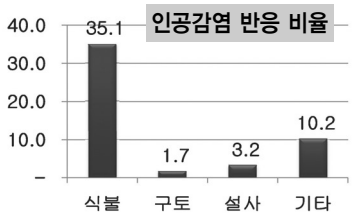
인공감염

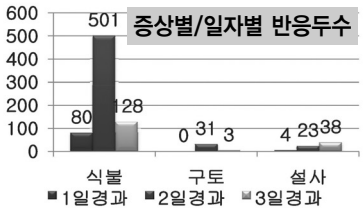
실제 농장 PED 발병 현황

- 7/23(목) : 1차 인공감염 실시 2,000두 (소장1⇒모돈 7두분)
- 7/24(금) : 각 동별 4~5두씩 반응 시작
- 7/26(일) : 50% 전후 반응
- 7/27(월) : 2차 인공감염 실시 1,000두
  - (증상 발현 미진한 개체 / 후보돈 전체)
- 7/30(목) :
  - 거의 100% 증상 발현(심한 증상 50% 수준)
  - 식불돈 30여두 남아 있는 상태
- 8/5(수) :
  - 번식돈 대부분 정상화 / 백신 스트레스 완화됨
  - 7/30 이후 분만자돈 설사 비율 감소, 발생일령 지연

인공감염 후 반응 데이터





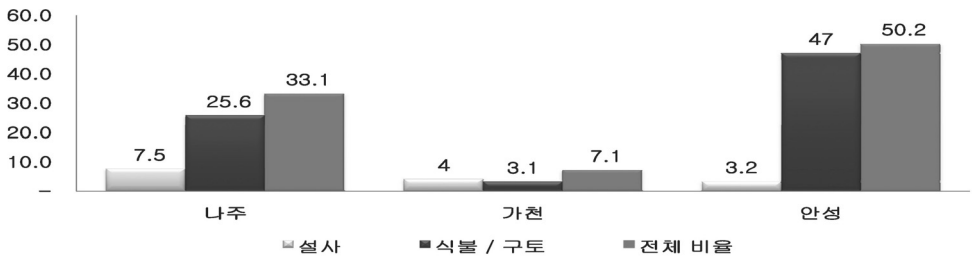


❖ 1, 2차 인공감염 후 심하게 증상 발현된 개체만 표기(50%)
⇒ 거의 100% 증상 발현
(사료꺼림 / 고열 / 식불)
⇒ 후보돈 의무적으로 2차 감염
⇒ 전체 두수 : 2,020두

효과적으로 인공감염 시키는 전략

- Expose each sow's gut to virulent **PEDv in sufficient concentrations** to cause infection, replication, and a strong mucosal immune response; therefore **higher doses of virus** in inoculum are desirable.
- Expose all breeding animals within a narrow window or short time-frame.
  - The best opportunity for an effective immune response is in na□ve animals with no preexisting immunologic 'memory'.
  - The narrow window of exposure establishes a projection-to-immunity calendar for prognostic purposes. □ Implement a post-exposure strategy to accurately and objectively monitor immune response, viral presence and PEDv associated pathology.

농장별 인공감염 발현 비율 비교



❖ 나주 : 08년 5월 발생 ⇒ 인공감염
❖ 가천 : 08년 4월 발생 / 08년 10월 재발 ⇒ 인공감염
❖ 안성 : 09년 7월 발생 ⇒ 인공감염
⇒ 이전 발생 경력 (만성 여부) ⇒ 바이러스 유전형 차이 (?) / 백신 상태 (?)
⇒ 반응 비율이 높을수록 조기 종결 가능성이 높다

설사를 막 시작한 자돈을 이용해야 하는 이유

- Neonatal pigs have very long villi with much higher populations of mature intestinal enterocytes which are most permissive for PEDv replication (Figures 1 and 2).
- □ In contrast, post-weaned and adult pig intestines contain far **fewer permissive mature enterocytes** so total amount of virus that can be produced is much less than in neonates.
- □ Infected enterocytes fill with viral particles until they explode, shedding huge numbers of virus particles in liquid feces.
  - o Live neonates with diarrhea will be shedding most of the virus prior to death. PCR testing demonstrates virus concentrations to **be 10,000 times higher* in piglet feces than in sow feces.** That means that 1 gallon of piglet feces has same number of virus particles as 10,000 gallons of sow feces.
  - Most of the infectious virus generated with PED in neonates is shed via fecal excretion in the first 12–18 hours of diarrhea, with less virus remaining at the time of death.
  - Enterocytes and virus are sloughed and expelled in feces. Intestines of piglets at death have much less virus remaining in infected enterocytes and feces than in feces of acutely affected neonatal piglets pigs (Figures 3 and 4).

백신

농장 상황에 따른 중화항체

심한 설사농장

개체 구분	중화항체가
42-34-1	64
42-34-2	128
42-78	<4
42-82	<4
43-18	32
43-21	8
43-44	<4
44-10	128
44-11	<4
46-63-1	32
46-63-2	<4
46-72	64
46-87	4
47-17	4
47-23	8
47-31	16
50-32	32
51-52	128
51-56	512
51-75	16

재발농장 : 부분 방어

개체 구분	중화항체가
설사하는 자돈의 모돈	
Y21-2733 2산 A	8
Y21-1695 4산 A	64
F1-2052 1산 A	<4
F1-1183 1산 A	128
F1-1976 1산 B	128
설사안하는 자돈의 모돈	
F1-2047 1산 B	512
Y13-3255 7산 B	512
Y21-526 5산 B	512
Y22-1082 3산 B	512
F1-1709 1산 B	32

인공감염 + 3회 보강접종

개체 구분	중화항체가
1산 F1-1085 2/11	512
1산 F1-1272 2/5	512
1산 F1-1638 2/5	512
1산 F1-1941 2/5	512
1산 F1-1976 2/11	512
1산 F1-2037 1/30	512
1산 F1-2117 2/11	512
1산 F1-2196 2/11	512
1산 F1-2280 2/5	512
1산 F1-2321A 2/11	512
2산 F1-2383 2/7	512
1산 F1-2455 2/11	512
1산 F1-2496 2/5	512
1산 F1-2593 2/11	512
1산 F1-2634 2/11	512
2산 F1-3323 2/4	32

모돈 산차 & 중화항체

PED 중화항체가 검사 결과서

접수번호	08-2594	의뢰인	김현백
농장정보			혈액 42

저산차/노산차
모돈의
항체가가 낮다

개체구분	중화항체가
1산 0345	128
1산 08-96	4
1산 08-153	4
1산 10-104	16
1산 33-37	256
1산 63-45	64
2산 08-198	<4
2산 1730	64
2산 2320	256
2산 33-10	64
2산 43-16	512
2산 53-00	512
3산 06-62	512
3산 1351	64
3산 26-45	512
3산 85-15	16
4산 25-49	128
4산 73-23	256
4산 84-23	128
4산 XL85-26	128
5산 3-12	128

PED 중화항체가 검사 결과서

의뢰인	김현백부장님(이지바이오)
시료	혈액 42점

5산 52-24	256
5산 8399	64
5산 92-86	64
6산 LY12-6	64
6산 LY22-47	64
6산 LY32-79	256
6산 45-91	128
6산 YL63-52	8
6산 85-75	64
7산 7-3	64
7산 7348	64
7산 94-22	32
8산 LY34-39A	16
8산 LY43-74	16
8산 LY64-92	64
8산 131-185	32
9산 YL2-31	8
9산 4-28	4
9산 198-164	128
10산 135-143	64
10산 172-168	64

자돈 상태에 따른 모돈 중화항체

개체 구분	중화항체가
설사하는 자돈의 모돈	
Y21-2733 2산 A	8
Y21-1695 4산 A	64
F1-2052 1산 A	<4
F1-1183 1산 A	128
F1-1976 1산 B	128
설사안하는 자돈의 모돈	
F1-2047 1산 B	512
Y13-3255 7산 B	512
Y21-526 5산 B	512
Y22-1082 3산 B	512
F1-1709 1산 B	32

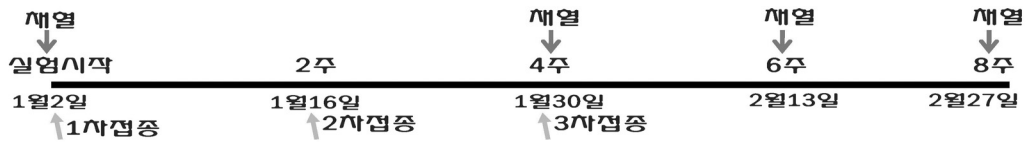
- PED 만성농장에서 포유자돈이 설사를 나타 내는 복의 모돈과 포유자돈 설사가 없는 복의 모돈의 PED 중화항체 비교
  ⇒ 중화항체가 높으면 폭발적 설사 발현이 안 된다.
  ⇒ 어느 정도 수치면 발병을 안 하는가? (옵티팜 자체 결론 : 최소 256 이상)

검사 이후에 설사 시작

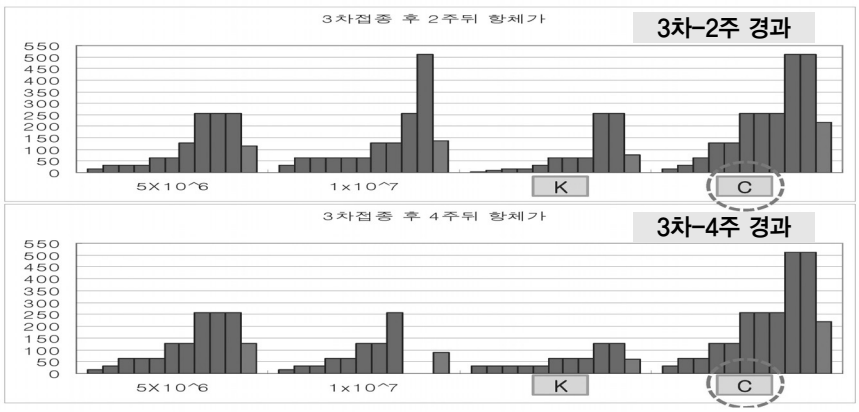
백신별 중화항체 유도

야외 비육돈 접종 테스트 (사독 백신별 항체가 비교)

- 4개 그룹으로 구분하여 PED 중화항체가 차이 비교(각 10두)
  - A군 : (옵티팜 제조) 1×10^7 접종군, 2㎖/두
    - KPEDV-9+SM98+DR13 =1x10^7 pfu/㎖
  - B군 : (옵티팜 제조) 5×10^6 접종군, 2㎖/두
    - KPEDV-9+SM98+DR13 =5×10^6 pfu/㎖
  - C군 : K PT 3㎖/두
    - SM98주 = 10^7 TCID50/㎖ 이상
  - D군 : C PT- 3㎖/두
    - SM98주 = 10^7 TCID50/㎖ 이상



백신별 중화항체 유도



SMART OPTIPHARM!

Q&A

경청해 주셔서 감사합니다.

MEMO