大韓獸醫學會誌 (2015) 第 55 卷 第 1 號
Korean J Vet Res(2015) 55(1) : 61~63
http://dx.doi.org/10.14405/kjvr.2015.55.1.61

<Short communication>

Seroprevalence of porcine proliferative enteropathy before initiating vaccine marketing in Korea

Jung-Yong Yeh[1,2,*]

[1]*Division of Life Sciences, Incheon National University, Incheon 406-772, Korea*
[2]*Animal and Plant Quarantine Agency, Anyang 430-757, Korea*

(Received: July 15, 2014; Revised: November 22, 2014; Accepted: January 9, 2015)

Abstract : Proliferative enteropathy caused by *Lawsonia intracellularis* is one of the most common enteric diseases in pigs. The objective of this study was to determine the prevalence of serum antibodies against *L. intracellularis* in the general swine population of Korea from 2005 to 2008. In total, 8,008 swine serum samples obtained from 1,001 herds were tested. The samples were analyzed with an immunoperoxidase monolayer assay to detect anti-*L. intracellularis* antibodies. The overall 4-year average true prevalence was 40.0% (CI: 39.4 - 40.6%) at the individual animal level and 71.9% (CI: 70.3-73.4%) at the herd level.

Keywords : Korea, *Lawsonia intracellularis*, prevalence, proliferative enteropathy, serology

In the pig farming industry, enteric diseases are a problem of increasing importance. Porcine proliferative enteropathy (PPE) caused by *Lawsonia* (*L.*) *intracellularis* is one of the most common enteric diseases in grower and finisher pigs. Information regarding the distribution of this pathogen in swine herds at the country level would be beneficial for the development of control protocols. In the Republic of Korea (ROK), the reported prevalence of PPE has varied by study, ranging from 3.3% to 56% at the pig level and from 20% to 100% at the herd level [6, 7, 12]. However, all studies on the prevalence of PPE in the ROK have targeted only selected herds with a history of diarrhea or only pigs with diarrhea. The objective of our study was to determine the prevalence of serum antibodies to *L. intracellularis* in the general swine population in the ROK. It is important to note that the avirulent live *L. intracellularis* vaccine (Enterisol Ileitis; Boehringer Ingelheim Animal Health, Germany), which is the first and only vaccine for the control of PPE, was not available for use in pigs in the ROK until the time of sampling; thus, the detected antibodies could not have been elicited by the *L. intracellularis* vaccine.

The swine serum samples were randomly obtained from serum specimen sources obtained under the Korea National Animal Health Monitoring Project, which was conducted from 2005 through 2008. The study included 1,001 randomly selected herds distributed throughout the country. Samples were taken from 8 pigs from each herd. The serum samples were analyzed by an immunoperoxidase monolayer assay (IPMA) to detect the *L. intracellularis* antibody. Using the pathogenic isolate PHE/KK421 [15], IPMA was performed. Briefly, the acetone-methanol-fixed *L. intracellularis* culture plate was incubated with sera diluted 1 : 30 in phosphate-buffered saline (PBS) at a volume of 50 µL per well for 30 min at 37°C and washed 5 times with PBS, pH7.2. Peroxidase-labeled goat anti-porcine IgG was diluted 1 : 1000 (KPL, USA) in 2% bovine serum albumin and 0.08% Tween 80 in PBS and then added at a volume of 50 µL per well. The plate was incubated for 45 min at 37°C. The plate was washed again, and a chromogenic (3-amino-9-ethyl-carbazole; Dako, USA) solution was added to each well. The plate was then incubated at room temperature for 20 min. The plate was washed with distilled water three times, allowed to dry, and examined using an inverted light microscope (Olympus, Japan). *L. intracellularis*-positive and *L. intracellularis*-negative antiserum controls and a secondary antibody control were included on each plate. Positive samples contained red-labeled bacteria, in both the cytoplasm of infected McCoy cells and the supernatant. Negative control plates using mock-infected cells were included for each individual serum sample to avoid false-positive results. True prevalence (TP) was estimated, as described by Marchevsky *et al*. [4, 9, 10], using published test sensitivity and specificity of 100% and 90%, respectively [2]. The formula used to determine TP was: TP = (apparent prevalence + specificity –1)/(sensitivity + specificity – 1). Statistical analyses were performed with the NCSS 2007 Statistical Software package for Win-

*Corresponding author
Tel: +82-32-835-8249, Fax: +82-32-835-0763
E-mail: yehjy@incheon.ac.kr

Table 1. Herd- and pig-level seroprevalence of antibodies to *Lawsonia intracellularis* in pigs in the Republic of Korea, 2005-2008

Year	Herd/Pig	Gangwon	Gyeonggi[†]	Chungbuk	Chungnam[‡]	Jeonbuk	Jeonnam[§]	Gyeongbuk[‖]	Gyeongnam[¶]	Jeju	Total
Total	Tested herds	129 (557[*])	141 (1,681)	51 (497)	147 (1,800)	102 (1,842)	74 (1,841)	69 (1,147)	240 (2,489)	48 (246)	1,001 (12,100)
	Positive herd (AP%)	82 (63.6)	109 (77.3)	27 (52.9)	131 (89.1)	39 (38.2)	59 (79.7)	63 (91.3)	115 (47.9)	23 (47.9)	648 (64.7)
	Herd TP[**]% (95% CI)	70.7	85.9	58.8	99.0	42.4	88.6	100.0	53.2	53.2	71.9
	95% CI of herd TP	66.3-75.0	82.3-89.5	51.6-66.0	96.3-100	37.5-47.4	83.7-93.4	96.5-100	49.9-56.5	45.8-60.7	70.3-73.4
	Tested pig	1,032	1,128	408	1,176	816	592	552	1,920	384	8,008
	Positive pig (AP%)	480 (46.5)	393 (34.8)	108 (26.5)	522 (44.4)	251 (30.8)	238 (40.2)	347 (62.9)	389 (20.3)	156 (40.6)	2,884 (36.0)
	Pig TP% (95% CI)	51.7	38.7	29.4	49.3	34.2	44.7	69.9	22.6	45.1	40.0
	95% CI of herd TP	50.1-53.3	37.2-40.1	27.2-31.7	47.8-50.8	32.6-35.9	42.6-46.7	67.8-72.0	21.6-23.5	42.5-47.7	39.4-40.6

[*]total herd number in each province (2005 National Statistical Office). [†]Seoul metropolis and Incheon metropolitan city included; [‡]Daejeon metropolitan city included; [§]Gwangju metropolitan city included; [‖]Daegu metropolitan city included; [¶]Ulsan and Busan metropolitan cities included. [**]TP = (AP + specificity − 1)/(sensitivity + specificity − 1) [4, 9, 10]. (Marchevsky *et al*, 1974). Specificity and sensitivity of the test employed in this study were according to a previous report (Collins *et al*., 2012). AP: apparent prevalence, TP: true prevalence, CI: confidence interval.

dows (NCSS, USA) and the Survey Toolbox (ver. 1.04; AusVet Animal Health Services, Australia).

In total, 8,008 swine serum samples obtained from 1,001 herds were tested in this study. The yearly breakdown included 2,064 samples from 258 herds in 2005, 1,888 from 236 herds in 2006, 2,088 from 261 herds in 2007, and 1,968 from 246 herds in 2008. The overall four-year averaged TP of *L. intracellularis* antibodies was 40.0% (confidence interval [CI]: 39.4-40.6%) at the pig level and 71.9% (CI: 70.3-73.4%) at the herd level, as shown in Table 1. Yearly overall serological TP by head were 42.8% (CI: 41.7-43.9%) in 2005, 38.4% (CI: 37.3-39.6%) in 2006, 36.7% (CI: 35.6-37.7%) in 2007, and 42.2% (CI: 41.1-43.4%) in 2008, respectively, while at the herd level, 76.7% (CI: 73.7-79.6%) in 2005, 66.3% (CI: 63.0-69.6%) in 2006, 68.6% (CI: 65.5-71.7%) in 2007, and 75.9% (CI: 72.8-78.9%) in 2008. The prevalence of *L. intracellularis* infection differed by geographic region, as shown in Table 1. The overall TP of anti-*L. intracellularis* IgG in pigs was 69.9% (CI: 67.8-72.0%) in Gyeongbuk Province but only 22.6% (CI: 21.6-23.5%) in the Gyeongnam region. The results of this study confirm that *L. intracellularis* is endemic in ROK pig herds, as more than half of the tested herds were found to be positive for *L. intracellularis*-specific antibodies.

In the swine industry, PPE is considered one of the most economically significant diseases among pigs worldwide. A subclinical form of PPE in which pigs become infected and have some intestinal lesions but do not exhibit clear diarrhea or weight loss has been recognized. Serological tests are able to detect subclinical disease and may therefore be one of the most accurate measures of *L. intracellularis* infection in terms of recognizing a chronic infection that persists in the host, making the control of PPE difficult in affected farms. The prevalence of *L. intracellularis* infection at the herd level has previously been reported to vary between 15% and 93.7% for 85 and 79 herds investigated, respectively [1, 3, 5, 8, 11, 13, 14]. In a previous Danish study, the prevalence of infection was 75% in 72 farrow-to-finish herds (samples taken from 10 growing pigs per herd with signs of diarrhea and weighing between 15 and 80 kg) and 39% in 26 herds without diarrhea [8]; our results were within this range. Although the study by Lee *et al*. [7] included the serological monitoring of PPE in 2000, their results needed more information to reflect overall seroprevalence in the general swine population in the ROK. That study had a limitation on sampling regions because of the failure to collect sufficient samples from major segments of the Korean swine industry, *e.g*., Gangwon, Chungbuk, Chungnam, Jeju Provinces. Our study investigated the serological prevalence of PPE by country-level monitoring from all the provinces by random sampling. Further investigation of the dynamics of *L. intracellularis* in the Korean swine population should be carried out and supplemented with bacterial analysis data and serological data after beginning marketing of a PPE vaccine to better understand the epidemiology of this disease.

Acknowledgments

The authors are very grateful to the local government veterinary officers for the help in collecting swine serum samples from all over the country. This work was supported by the Incheon National University Research Grant in 2014.

References

1. **Chang WL, Wu CF, Wu Y, Kao YM, Pan MJ.** Prevalence of *Lawsonia intracellularis* in swine herds in Taiwan. Vet Rec 1997, **141**, 103-104.

2. **Guedes RM, Gebhart CJ, Deen J, Winkelman NL.** Validation of an immunoperoxidase monolayer assay as a serologic test for porcine proliferative enteropathy. J Vet Diagn Invest 2002, **14**, 528-530.
3. **Herbst W, Willems H, Baljer G.** Distribution of *Brachyspira hyodysenteriae* and *Lawsonia intracellularis* in healthy and diarrhoeic pigs. Berl Munch Tierarztl Wochenschr 2004, **117**, 493-498.
4. **Hilden J.** A further comment on "Estimating prevalence from the results of a screening test". Am J Epidemiol 1979, **109**, 721-723.
5. **Holyoake PK, Cutler RS, Caple IW.** Prevalence of proliferative enteritis on pig farms in Australia. Aust Vet J 1994, **71**, 418-422.
6. **Kim O, Kim B, Chae C.** Prevalence of Lawsonia intracellularis in selected pig herds in Korea as determined by PCR. Vet Rec 1998, **143**, 587-589.
7. **Lee SW, Kim TJ, Park SY, Song CS, Chang HK, Yeh JK, Park HI, Lee JB.** Prevalence of porcine proliferative enteropathy and its control with tylosin in Korea. J Vet Sci 2001, **2**, 209-212.
8. **Møller K, Jensen TK, Jorsal SE, Leser TD, Carstensen B.** Detection of *Lawsonia intracellularis*, *Serpulina hyodysenteriae*, weakly beta-haemolytic intestinal spirochaetes, *Salmonella enterica*, and haemolytic *Escherichia coli* from swine herds with and without diarrhoea among growing pigs. Vet Microbiol 1998, **62**, 59-72.
9. **Rogan WJ, Gladen B.** Estimating prevalence from the results of a screening test. Am J Epidemiol 1978, **107**, 71-76.
10. **Shoukri MM, Edge VL.** Statistical methods for health sciences. CRC Press, Boca Raton, 1996.
11. **Stege H, Jensen TK, Møller K, Bækbo P, Jorsal SE.** Prevalence of intestinal pathogens in Danish finishing pig herds. Prev Vet Med 2000, **46**, 279-292.
12. **Suh DK, Song JC.** Prevalence of *Lawsonia intracellularis, Brachyspira hyodysenteriae* and *Salmonella* in swine herds. J Vet Sci 2005, **6**, 289-293.
13. **Thomson JR, Smith WJ, Murray BP.** Investigations into field cases of porcine colitis with particular reference to infection with *Serpulina pilosicoli*. Vet Rec 1998, **142**, 235-239.
14. **van der Heijden HMJF, Bakker J, Elbers ARW, Vos JH, Weyns A, de Smet M, McOrist S.** Prevalence of exposure and infection *of Lawsonia intracellularis* among slaughter-age pigs. Res Vet Sci 2004, **77**, 197-202.
15. **Yeh JY, Kim TJ, Park SY, Song CS, Yoon YD, Kim SK, Lee JB, Choi IS.** Isolation of *Lawsonia intracellularis* in Korea and reproduction of proliferative enteropathy in pigs and hamsters. J Vet Med Sci 2006, **68**, 499-501.