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Pig viral diseases causing reproductive failure in Korea

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초록 : 1988년부터 1990년 6월까지 전국의 양돈장에서 수집된 돼지 유사산 태아 74복에서 바이러스성 원인체 분리 및 혈청학적 진단을 수행하였던 바 다음과 같은 결과를 얻었다.

공시한 74복의 유사산 태아중 44복의 태아 흉강액에서 면역 globulin이 검출되어 전염성 질병감염에 의한 유사산으로 추정되었다. 이중 37%가 바이러스성 유사산으로 나타났으며 유사산의 원인체별 분포를 살펴보면 돼지 파보바이러스가 21%로 가장 높았으며, 뇌심근염 바이러스가 11%, 일본뇌염 바이러스가 9% 등의 순으로 나타났다. 한편 돼지 콜레라바이러스 및 오제스키병 바이러스에 의한 유사산이 각각 1건씩 검출되었으며 동일 유사산 태아에서 2가지 병원체가 중복감염된 예도 관찰되었다.

Key words : viral infection, swine reproductive failures, porcine parvovirus, encephalomyocarditis virus, Japanese B encephalitis virus, hog cholera virus, pseudorabies virus

Introduction

There are many agents causing reproductive disorders in swine. The agents can be roughly divided as biological infectious factors and non-infectious or environmental disorders, which makes the diagnosis somewhat complicated.[1] Among those agents, it is known that various infectious agents like virus and bacteria are frequently associated with swine reproductive problems[2~6] However, few studies concerning swine reproductive failure were reported in Korea.[7,8] The purpose of present paper was designed to investigate the features of infectious viral disease causing reproductive disorders in swine.

Materials and Methods

Specimens : The fetuses examined in this study were collected from swine farms from 1988 to 1990. Each sample was usually divided into two groups according to days of gestation and the size of fetus. When the fetus had crown-rump length more than 17cm, the thoracic fluid was collected and treated at 56 C for 30 minutes for serological diagnosis. At the same time, the samples less than 17cm were directly examined for isolation of virus. A total of 74 cases(114 fetuses) were examined for reproductive failures.

Serologic examinations : A microtitration of virus neutralization (VN) method was used for the detection of antibody against Hog cholera virus (HCV), Pseudorabies virus (PRV), Porcine enterovirus (SMEDI A-E) using PK-15 cells, primary swine testicle cells and Encephalomyocarditis virus (EMCV) using BHK-21 cells.[9~11]

Hemagglutination inhibition (HI) test was routinely

used for diagnosis for Porcine parvovirus (PPV) and Japanese encephalomyelitis virus (JEV) by the procedures described previously.[12,13]

The presence of immunoglobulins from thoracic fluid was detected by Agar gel precipitation (AGP) test using Rabbit anti-porcine immunoglobulines according to standard method.[14]

Virus isolation : 10% of pooled ground suspension from fetal organs was centrifuged at 1500g for 15 minute and the supernatant was inoculated into primary porcine kidney, primary porcine testicle, PK-15 and BHK-21 cells. The inoculated cells were usually passaged three times until being considered virus free.

A total of 24 separate case was tested for the presence of viral agent. At the same time, 2-3 days old mice were inoculated through intracerebral route for isolation of JEV and 2-3 weeks old mice through intraperitoneal for EMCV.

Results

The results of examining separate 50 cases of aborted litter that were more than 70 days of gestational age are shown in Table 1. Serological diagnosis of viral infections were made in 28 of separate cases examined. The viral infection of aborted samples were commonly turned out to be PPV (16 litters) and EMCV (8 litters) followed by JEV (7 litters), each 1 litter of HCV and PRV. In addition, HI activities against JEV and PPV were detected in two cases. Antibodies against EMCV and JEV were found in two cases, and antibodies against EMCV and PPV were also noted in one case, indicating dual infections of those viruses in the same litter. On the other hand, no case of positive reactions on Enterovirus was detected. Although the exact diagnosis were not made in all fetal cases, 44 from 50 cases, which reported more than 70 days of gestation, showed the presence of immunoglobulins in thoracic fluid, suggesting the cause of reproductive failures came from other infectious agent (Table 2). Since fetal abortions from EMCV infection were not reported previously in this country, serological survey against EMCV in adult swine was further performed. The result showed that the prevalence of EMCV in adult swine increased from 25% in 1987 to 54% in 1988 as indicated in Table 3. Attempts to isolate virus from fetus specimens less than 17 cm were not rewarding even after three times blind passages in tissue culture.

Discussion

The present study provides major virus infections associated with reproductive failures in Korea. Although the exact diagnosis was not possible in all cases of aborted samples, the main viral infections were PPV, JEV, EMCV, HCV, PRV and duel infections of PPV and JEV or EMCV, JEV and EMCV. Compared with the results of previous report[3], the overall rate of viral infections were rather high in this study.

Table 2. Detection of immunoglobulins from aborted fetuses (crown-rump length more than 17 cm)

	No of fetuses(%)
Immunoglobulins detected	44(88)
-with virus infections	26
-unknown etiology	18
Immunoglobulins not detected	6(12)
Total	50(100)

Table 1. Detection of antibodies from thoracic fluids of aborted specimens

Viruses tested	Number of fetuses(Litter)	Distribution of antibody titers			Serological Method
	Diagnosed*	1 : 2-1 : 8	1 : 16-1 : 64	>1 : 64	
PPV	25(16)	2	17	8	HItest
EMCV	10(8)	15	10		VNtest
JEV	11(7)		2	9	HItest
PRV	1(1)		1		VNtest
HCV	1(1)	1			VNtest
Unknown	38(22)				
Total	86(50)**				

* Criteria of infections judged from antibody titer in thoracic fluids of fetuses.
PPV : HItiter(>8), JEV : HItiter(>10). EMCV : VNtiter(>16), HCV : VNtiter(>4).
PRV : VNtiter(>2).

** Dual infection : PPV and JEV(2), JEV and EMCV(2), PPV and EMCV(1).

Table 3. Distribution of neutralizing antibody against EMC virus in Korea

Years	No of pigs	Neutralizing antibody titers*			No of positives(%)
		<1 : 8	1 : 16-1 : 64	>1 : 128	
1987	135	20	10	4	34(25)
1988	287	122	29	6	157(54)

* EMC virus : ATCC VR-129 strain.

The presence of antibody against EMCV was rather unexpected because there was no previous survey on the presence of this viral infection as a causative agent as reproductive failures[7,8] However, EMCV infections were turned out to be as prevalent as JEV through this study. Recently Kim et al[11,15] reported that EMCV infection could be an agent of stillborn pigs supporting these findings. Our result on antibody titers in adult swine also indicated that the EMCV infections were more widespread than expected. Park et al[16] also reported the infection of EMCV in the pigs showing reproductive failures in south-western area. Therefore, the pathogenic role of EMCV as the direct cause of reproductive failure seems to need further studies.

In general, the presence of immunoglobulins in fetus indicates that the fetus was exposed to infectious agents because maternal antibody in pregnant sow does not penetrate fetus and fetus become immunoreactive after 70 days of gestation[1] In this respect, the detection of immunoglobulins can be another way of diagnostic tools supplementing serologic diagnosis. Unlike abortion from viral infections, bacterial infections as well as other miscellaneous environmental factor are known to cause reproductive failures in swine[3] In fact, 44 out of 74 aborted litters indicated that the cause of abortions were related to infectious agents. An attempt to isolate any virus from fetus specimens less than 17cm was not successful. The reason may partly comes from conditions of specimens because it is usually take several days before submitted to laboratory. However, considering the results of present study, the proper vaccination programme on those viral agents seems to be one way of preventing a half of economic losses from productive failure in Korea.

Conclusion

Aborted and stillborn swine fetuses from piggery were investigated for viral infections by fetal serology over three years period. Of 74 aborted litters examined, immunoglobulins were detected in the thoracic fluid of 44 litters of fetuses, indicating that the reproductive failures resulted from infectious agents and twenty eight (37%) were diagnosed as viral infections. The frequency of viral infections involved were most often Porcine parvovirus (21%), followed by Encephalomyocarditis, Japanese encephalitis, Hog cholera and Pseudorabies. Occasionally, dual infections of those viruses were also found in the same litter. A serological survey was conducted to detect virus neutralizing antibody against Encephalomyocarditis virus in field. The results indicated that the incidence of encephalomyocarditis virus infections in pigs were 25% in 1987 and 54% in 1988.

References

1. Lemon AD, Straw B, Glock RD, et al. *Disease of swine* lowa State University Press, 6th ed, lowa, USA 1986 ; 233~238.
2. Littlejohns LR. Encephalomyocarditis virus from stillborn pigs. *Aus Vet J* 1984 ; 61(3) : 93.
3. Kirkbride CA, McAdaragh JP. Infections agents associated with fetal and early neonatal death and abortion in swine. *J AM Vet Med Ass* 1978 ; 172(4) : 480~483.
4. Wonlgemuth K, Leslie PF, Reed DE, et al. Pseudorabies virus associated with abortion in swine. *J AM Vet Med Ass* 1978 ; 172(4) : 478~479.
5. Mengeling WL, Cutlip RC, Wilson RA. Fetal mummification associated with porcine parvovirus infection. *J AM Vet Med Ass* 1975 ; 166(10) : 993~995.
6. Johnson KP, Ferguson LC, Byington DP, et al. Multiple fatal malformations due to persistent viral infection. 1. Abortion, intrauterine death, and gross abnormalities in fatal swine infected with hog cholera vaccine virus. *Lab Inv* 1974 ; 30(5) : 608~617.
7. Joo HS, Lim YM, Lee CK. Isolation of porcine parvovirus from mummified porcine fetus. *Res Rep RDA* 1979 ; 21 : 11~17.
8. Kwon HJ, Lee CK, Kang BJ, et al. Studies on

Japanese encephalitis live vaccine. 1. Isolation of Japanese encephalitis virus (Anyang strain) from a new-born piglet. *Res Rep RDA* (V) 1974 ; 16 : 1~5.

9. Kumagai T, Shimizu T, Ikeda S, et al. A new *in vitro* method (END) for the detection and measurement of hog cholera virus and its antibody by means of effect of HC virus on new castle disease virus in swine tissue culture. I. Establishment of standard procedure. *J Imm* 1961 ; 87 : 245~256.
10. Cottral GE. *Manual of Standardized Methods for Veterinary Microbiology*. Cornell University Press, New York, USA 1978.
11. Kim HS, Joo HS, Bergel ME. Serologic, virologic, and histopathologic observations of encephalomyocarditis virus infection in mummified and stillborn pigs. *J Vet Dia Inv* 1989a ; 1 : 101~104.
12. Joo HS, Donalson CR, Johnson RH. A standardized haemagglutination inhibition test for porcine parvovirus antibody. *Aus Vet J* 1976 ; 52 : 422~424.
13. Clarke DH, Casals J. Techniques for hemagglutination and hemagglutination-inhibition with arthropod-borne viruses. *Am J Tro Med & Hyg* 1958 ; 7 : 561~573.
14. Johnstone A, Thorpe R. *Immunochemisrty in practice* 2nd Edition. Blackwell Scientific Publications. Oxford, Great Britan. 1987 ; 133~135.
15. Kim HS, Christianson WT, Joo HS. Pathogenic properties of encephalomyocarditis virus isolates in swine fetuses. *Arch Virol* 1989b ; 109 : 51~57.
16. Park NY, Chung CY, Ri CY, et al. Encephalomyocarditis virus infection in pigs associated with reproductive failure. *Kor J Vet Sci* 1990 ; 30(4) : 441~446.