# LONG TERM EFFECTS OF VACCINATION ON THE DEVELOPMENT OF LESIONS IN PSEUDORABIES VIRUS CHALLENGED SWINE*

ROBERTO ALVA V, ROBERT D. GLOCK, and JOHN P. KLUGE
DEPARTMENT OF VETERINARY PATHOLOGY, COLLEGE OF VETERINARY MEDICINE
IOWA STATE UNIVERSITY, AMES, IA 50011

Introduction: The clinical signs, gross lesions and histopathologic changes in young swine infected with pseudorabies virus (PRV) may involve the central nervous system, respiratory system or every major organ in the animal[1]. The use of modified live pseudorabies virus vaccines (MLPRV) and inactivated pseudorabies virus vaccines (IPRV) in swine has been studied by several workers.[2,3,4] Effectiveness of vaccination has been evaluated mainly by reduction in clinical signs and mortality rate. Multiplication and distribution of PRV in pigs immunized with MLPRV or IPRV and later challenged with a virulent strain of PRV has been reported to be the same as in non-vaccinated pigs.[5,6] Multiplication of the virus and the lesions it produces in vaccinated pigs have been reported to be limited to certain areas of the brain.[7]

Objectives: The objectives of this study were to determine the long term effects of declining immunity on clinical signs and lesion development in pigs vaccinated with MLPRV or IPRV vaccines and challenged later with a virulent strain of PRV.

Materials and Methods: Ninety-three-seven-week old castrated male pigs were utilized. They were free of serologic evidence of pseudorabies, transmissible gastroenteritis, leptospirosis and clinical atrophic rhinitis.

Experimental designs: Treatment groups based on time of challenge included groups I,II,III,IV,V,VI and VII. The groups were exposed to PRV at 1,3,5,8,10,12 and 14 months after vaccination respectively. Groups I through IV were composed of 12 pigs each while groups V,VI and VII had 15 pigs each. Groups I,II,III and IV consisted of 4 pigs vaccinated with a MLPRV[a] vaccine, 4 with the IPRV[b] vaccine and 4 with a placebo solution (PI) consisting of sterile distilled water. Groups V,VI and VII consisted of 5 pigs in each category (MLPRV, IPRV and PI). In these groups one pig of each category was left as a negative control (vaccinated non-challenged). All groups were vaccinated simultaneously at 9 weeks of age penned separately for 90 days and then allowed to commingle in 3 large pens containing 31 pigs each. Four ml of an Iowa strain of PRV at the 9th and 10th tissue culture passagg containing $1.6X10^7$ (groups I,II and III) and $5X10^6$ (groups IV,V,VI,VII) plaque forming units respectively was used as the challenge inoculum. Clinical signs were observed for 8 days after challenged (AC). Half the animals in each group were killed at 7 and half at 60 days AC. Tissues were collected in 10% buffered formalin and processed by conventional paraffin techniques. Sections were stained with hematoxylin and eosin. Sections from the central nervous system (CNS), digestive system, hematopoietic system and cardiovascular system were examined. Selected sections were examined with electron microscope. Lesions were scored according to severity and distribution of the following changes: congestion, edema, malacia, necrosis, microglial nodules, perivascular cuffing, neuronophagia, reactive astrocytes, gitter cells, swollen axons, satellitosis, inclusion bodies, presence of lymphocytes, macrophages, plasma cells, eosinophils, neutrophils, fibrin, vasculitis and hemorrhages. A progressive scale of 1 to 5 (in the CNS) and 1 to 4 (in other tissues) was assigned. The data was analyzed by using the analysis of variance procedure.

Results: Body temperature (BT) increased 24 hours AC. The MLPRV vaccinates had an overall significantly lower ($P < .05$) mean BT than the IPRV vaccinates or the PI injected. Pigs were anorexic, prostrate, and sneezing with a purulent exudate flowing from the nostrils at 24 hours AC. Vomiting was noted at 96 hrs. AC in vaccinates and non-vaccinates. Vaccinates began to recover within 7 days AC, while PI injected remained sick for an additional 7 days. Only one MLPRV vaccinate died (48 hrs. AC). PI injected pigs in groups I,III and IV had a higher degree of clinical signs than the MLPRV and IPRV vaccinates. No difference in clinical signs was observed between the MLPRV and IPRV vaccinates. Groups V, VI and VII were equally affected. Gross lesions were observed exclusively at 7 days AC in 18 out of the 93 pigs utilized. The lesions were characterized by purulent exudate in turbinates, areas of pneumonia, and tonsillar necrosis. All PI injected pigs had a highly significant difference ($P < .0001$) in severity of turbinate lesions when compared to IPRV and MLPRV vaccinates. Microscopic lesions were similar in nature but varied in severity between vaccinates and non-vaccinates. The microscopic lesions at 7 days AC were characterized by a lympho-histiocytic-eosinophilic meningoencephalitis with focal areas of edema, demyelination, necrosis, and malacia. These foci contained gitter cells, lymphocytes, reactive astrocytes, plasma cells, eosinophils, and few neutrophils. Vasculitis and necrosis of vascular endothelium was common. Microglial nodules in the brain parenchyma with satellitosis and neuronophagia were usually present. Eosinophilic intranuclear inclusion bodies were rare in neuroglial and endothelial cells. The same type of lesions but in a milder degree were seen at 60 days AC. The turbinates and lungs were characterized by areas of coagulative necrosis with a severe mononuclear cell infiltration. Eosinophilic and basophilic intranuclear inclusion bodies were seen in epithelial cells of the bronchioles, bronchi and in alveolar pneumocytes. Edema was prominent. The tonsils were characterized by multifocal areas of coagulative necrosis with a large number of eosinophilic and basophilic intranuclear inclusion bodies in the epithelial cells of the crypts. Microscopic lesions were significantly higher at 7 days AC ($P < .05$) in the PI injected when compared to the vaccinates. No significant difference in lesion score was observed at 60 days AC among the different treatment groups. Lesions score means in vaccinated pigs were higher in groups V,VI and VII and lower in groups I,III, and IV. No lesions were observed in the non-challenged control pigs. The electron microscopy study demonstrated the presence of mature and immature viral particles in nuclei, cytoplasm an intercellular space of the cells containing the eosinophilic inclusion bodies (tonsils of vaccinated and non-vaccinated challenged pigs).

Conclusions: Vaccination with the MLPRV or IPRV vaccines did not prevent infection or development of microscopic lesions when swine were challenged with a virulent strain of PRV. However vaccination diminished the severity of clinical illness and microscopic lesions for a period of 8 months after vaccination. Both vaccines provided similar protection against development of microscopic lesions.

Selected references:

1. Baskerville, A.: McFerran, J. B.; and Dow, C.: Vet.Bull. 43: 445-480 (1973)
2. Jamrichova, O.; and Skoda, R.: Acta Virol. 13:42-51 (1969)
3. Skoda, R.; Brauner, I.; Sadecky, E.; and Mayer, V.: Acta Virol. 8:1-9 (1964)
4. Skoda, R.; Brauner, I.; Sadecky, E.; and Somogyiova, J.: Acta Virol. 8:123-134 (1964)
5. McFerran, J. B.: Dow, C.; and McCranken, R. M.: Comp. Immun. Microbiol. Infect. Dis. 2:327-334 (1979)
6. Wittman, G.; Jakubik, J.; and Ahl, R.: Arch. Virol. 66:227-240 (1980)
7. McFerran. J. B.; and Dow, C.: Res. Vet. Sci. 19:17-22 (1975)

[a] Norden Laboratories Inc., Lincoln Nebraska 68501 U.S.A.

[b] Salsbury Laboratories, Charles City Iowa 50616 U.S.A.

*