



2010 AASV Annual Meeting Proceedings Author Guidelines

QUESTIONS AND ANSWERS:

Q: What is my deadline and how shall I send my paper?

- A:**
- Submit paper by **November 16, 2009**.
 - LABEL each disk with your last name, the file name(s), your email address, and phone number.
 - SEND both electronic and paper copy. You may send your files by mail or email. Please mail a paper copy as this allows us to verify that special characters, tables, and figures are reproduced as intended. Fax copies are unacceptable.
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Q: What file format should I use in creating text, tables, figures and photos?

- A:**
- File #1: Create TEXT in Word and place within its own file separate from tables, figures and photos.
 - File #2: Create TABLES in Word. Do not paste them into the word document containing the text of the manuscript.
 - File #3: Create GRAPHS/FIGURES in Excel[®] and submit as .xls files. NO Power Point files. Do not paste the figures into the word document containing the text of the manuscript. Please submit figures in black and white – do not use color within the figures. Figures may also be submitted as .eps files.
 - File #4: Send PHOTOS as black and white, high resolution .jpeg images or in .tif files. Do not paste photos into the Word document containing the text of the manuscript.

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- A:**
- Format your paper in a one column, single spacing layout. Use sentence capitalization style (capitalize the first letter of the first word only) in title and headings.
 - Do not send paper in outline form.
 - Do not use **bold** or underlined text for title, headings or in the body text. Use *italics* only for genus and species names, titles of books and journals. Titles and headings can be emphasized by using a larger font (see sample).
 - Please do not use the endnote or footnote feature in Word.
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**Submission
Deadline
November 16,
2009**

AASV Formatting Instructions:



File #1: Text

LAYOUT: One column
FONT: Times New Roman • **FONT STYLE:** Regular
SPACING:
• Single line spacing • Use ONE space between sentences.

ARTICLE TITLE:
• Size 24 point type (do not bold or underline)
• Sentence capitalization (capitalize the first letter of the first word only)

Immune response and effect of maternal antibody interference on vaccination with a bivalent swine influenza vaccine

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AUTHOR INFORMATION:
• Font size 12 point
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SUBHEADINGS:
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Introduction
Vaccination against swine influenza virus (SIV) is an important management tool. Maternally-derived antibodies (MDA) can be detected in pigs up to 16 weeks of age and may affect vaccination. The objective of this study was to investigate the immune response to a commercial bivalent SIV vaccine and the effect that the presence of MDA may have immunologically and on vaccine efficacy in pigs experimentally infected with a heterologous H1N1 SIV isolate.

Materials and methods
Ninety-six crossbred pigs were procured at 10-12 days of age from 2 herds serologically negative for PRRSV and *Mycoplasma hyopneumoniae* and allocated to 6 groups of 6 pigs. Pigs in the groups with MDA were obtained from a SIV vaccinated sow herd. Pigs in the MDA groups had hemagglutination inhibition assay (HI) titers of 1:40-1:80 at the time of the first vaccination. The experimental design is summarized in Table 1. Pigs were vaccinated with a commercial vaccine (Flusure[®] bivalent swine influenza vaccine, Pfizer Animal Health, NY, USA) at 3 and 5 wks of age. Two weeks after the second vaccination, a virulent classical H1N1 strain, A/swine/IA/40776/92, with minimal cross-reactivity with vaccine induced antibodies was administered intratracheally at the dose of 10⁵ TCID₅₀/ml (10 ml) to the challenged groups.¹ Clinical signs, including cough, respiratory rate and rectal temperature were evaluated prior to challenge and daily for 7 days post infection (DPI). Pigs were necropsied at 5 and 21 DPI. Lesions consistent with SIV were sketched onto a standard diagram 2 and microscopic analysis of lung tissue and immunohistochemistry (IHC) for SIV antigen were performed. Nasal swabs were collected at -1, 3, 5, and 7 DPI for virus isolation. Bronchoalveolar lavage and nasal washings were assayed for SIV-isotype specific antibodies as previously described.³ Serum was collected prior to each vaccination, prior to challenge and at 4 and 20 DPI. SIV antibodies from serum were measured using HI assay. Analysis of variance was used to analyze the data. A *P* value < 0.05 was considered significant.

Results and discussion
Rectal temperatures on -1, 1-7 DPI were significantly higher in all SIV challenged groups compared to the non-challenged groups (Figure 1). Vaccination in the absence of MDA significantly

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Table/figure numbers need to correspond to the references within text.

reduced the percentage of SIV-induced pneumonia at 5 DPI by macroscopic and microscopic evaluation (Table 2). Interestingly, the percentage of lung lesions was significantly higher in pigs with MDA at the time of first vaccination (group 8). Virus was isolated from all challenged groups up to 5 DPI (Figure 2). At 3 and 5 DPI, the SIV vaccinated and challenged group without MDA (group 6) had significantly lower levels of virus compared to the rest of the challenged groups. Pigs with MDA (group 8) had significantly increased levels of virus compared to group 6.

In the pigs without MDA, antibody levels to both virus antigens increased in response to vaccination and challenge (Figure 3 and 4). The non-vaccinated challenged pigs developed antibodies by 20 DPI and had higher titers to the challenge antigen than the vaccine antigen, although cross reactivity with the vaccine virus occurred. By 20 DPI, the vaccinated MDA positive group had strong antibody responses to both viral antigens demonstrating that an anamnestic response occurred. Vaccination enhanced the local immunity in the respiratory tract of the challenged pigs as measured by SIV-specific IgG and IgA from BALs.

The increased disease observed in vaccinated pigs with MDAs on vaccine was unexpected and the exact mechanism for the increased severity of disease is unknown. This study shows that MDAs are not completely protective against pneumonia and confirm their impact on vaccine efficacy. These results indicate that vaccination in the presence of MDA appears to prime the immune system to respond more quickly to infection. The results of this study provide important information on the immune response to SIV vaccination in the presence of MDAs. The results of this trial demonstrate that a bivalent SIV vaccine administered in the absence of MDA significantly reduced pneumonia and nasal viral load in pigs experimentally infected with a heterologous H1N1 SIV isolate. However, the presence of MDA during vaccination was shown to interfere with vaccine efficacy based on the percentage of SIV-induced pneumonia at 5 DPI and the amount of virus isolated from the nasal cavity.

Acknowledgments
The authors would like to thank Nancy Upchurch for technical support and the students from the Thacker Lab for their assistance with the animal work.

References
1. Thacker, E. L., B. J. Thacker, T. B. Boettcher, and H. Jayappa. 1998. Comparison of antibody production, lymphocyte stimulation, and protection induced by four commercial *Mycoplasma hyopneumoniae* bacterins. *Swine Health and Production* 6:107-112.
2. Thacker, E. L., B. J. Thacker, and B. H. Janke. 2001. Interaction between *Mycoplasma hyopneumoniae* and swine influenza virus. *J Clin Microbiol* 39:2525-30.
3. Larsen, D. L., A. Karasin, F. Zuckermann, and C. W. Olsen. 2000. Systemic and mucosal immune responses to H1N1 influenza virus infection in pigs. *Vet Microbiol* 74:117-31.
4. Jones T. Overview of pork production. Swine nutrition and feeding course, University of Animal Science. <http://www.ansci.usu.edu/web-courses/AnSci101/>. Accessed November 20, 2001.

REFERENCES:
• Superscript and number references (NO ROMAN NUMERALS) Reference numbers at the end of a sentence should be behind the period.
• Use the term References rather than Citations, Literature Cited, Bibliography, etc.
• Please do not use the endnotes feature in Word to format the references.

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Table 1: Experimental design

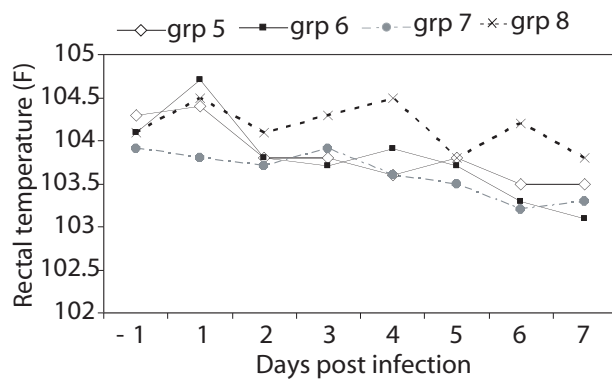
Group	Maternal antibodies	SIV vaccine	SIV challenge
1	No	No	No
2	No	Yes	No
3	Yes	No	No
4	Yes	Yes	No
5	No	No	Yes
6	No	Yes	Yes
7	Yes	No	Yes
8	Yes	Yes	Yes

File #3: Figures

FIGURES:

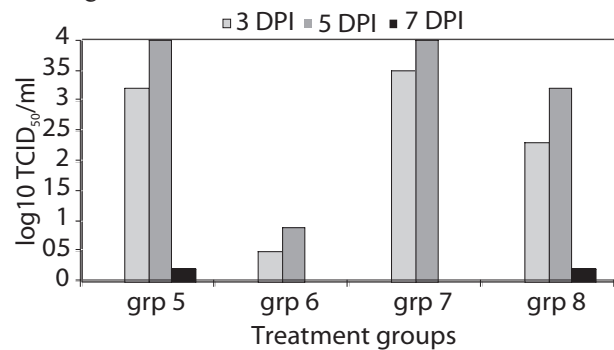
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Figure 1: Rectal temperature from pigs challenged with SIV



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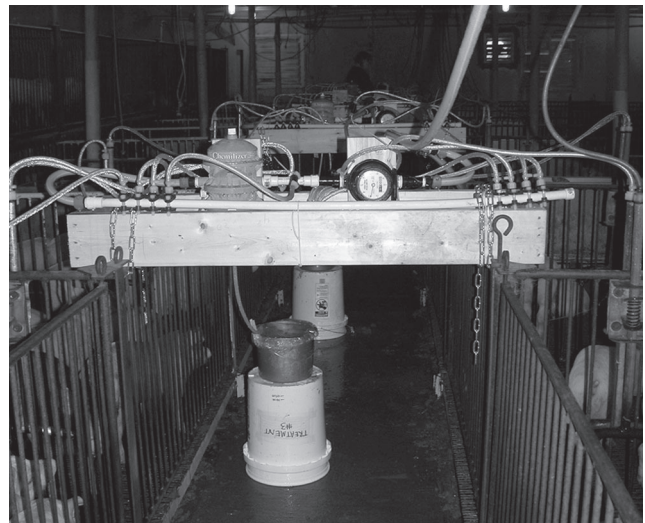
Figure 2: Virus isolation (VI) \pm standard deviation determined by cell culture and immunocytochemistry staining



File #4: Photos

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