TECHNICAL BULLETIN

February 2013

Comparative Serologic Responses of Horses Vaccinated with Commercial Equine West Nile Virus Vaccines

Key Points

- A study compared the serological responses to West Nile virus (WNV) in 280 horses after vaccination with 6 different regimens of commercial equine vaccines.[1]
- Three vaccination regimens involved vaccine formulations with WNV in combination with equine encephalomyelitis viruses (Eastern equine encephalomyelitis (EEE) and Western equine encephalomyelitis (WEE) antigens) and tetanus antigens, while 3 vaccination regimens had the WNV component separated from the EEE, WEE and tetanus antigens.
- All vaccines stimulated both primary and anamnestic WNV responses to vaccination.
- WNV antibody responses were consistently reduced ($P \leq 0.0001$) when the WNV component was formulated in combination with EEE, WEE and tetanus antigens compared to similar vaccination regimens where the WNV fraction was concurrently administered with EEE, WEE and tetanus containing vaccines.
- When developing WNV protection protocols, practitioners should consider the implications of lower WNV antibody responses in horses vaccinated with WNV combination vaccines.

Horse owners rely on equine practitioners to recommend and implement vaccine protocols that will help protect their animals from disease threats like West Nile virus (WNV), Western equine encephalomyelitis (WEE) virus, Eastern equine encephalomyelitis (EEE) virus, tetanus, equine influenza virus, equine herpesvirus (rhinopneumonitis), and other pathogens. Many vaccine choices are available, often involving antigen combinations in a single formulation intended to help optimize convenience. However, research is needed to ensure that combination products do not cause antigen interferences that might inhibit immune responses, particularly for critical disease threats like WNV.

A recent research study compared the effects of vaccination with 6 different vaccination regimens of commercial equine vaccines on subsequent WNV serological responses,

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specifically investigating outcomes when WNV was formulated in combination with tetanus and equine encephalomyelitis virus antigens (including both WEE and EEE).[1] It should be noted that WNV antibody titers are not indicative of protection but are just indicators of a immune response to vaccination.

Experiment Design

The study involved 280 adult quarter horses representing a wide age range (2-17 years) that were pastured at 2 commercial ranches in Oregon. All animals were determined to be serologically negative for WNV (antibody titer <4) at 15 days before study initiation. Horses were blocked (age, gender, site) and then randomly assigned to 1 of 7 treatment groups.

Six different vaccine regimens that delivered similar viral components were evaluated in the study, two from each of 3 major equine vaccine manufacturers [Zoetis, Boehringer Ingelheim, and Merck]. Three treatment groups received a vaccine regimen that contained the WNV antigen with EEE, WEE and tetanus antigens in a single vaccine formulation, while 3 groups received regimens with the WNV component administered concurrently with vaccines containing the EEE, WEE and tetanus antigens. The remaining group served as non-vaccinated (saline) controls. The vaccines used in each treatment group are summarized in Table 1 (n=40/group).

Table 1: Treatment groups and vaccine regimens.

Treatment group	Vaccine regimen
T01	WEST NILE-INNOVATOR®+EWT and FLUVAC INNOVATOR® EHV-4/1
T02	Saline
T03	Fluvac Innovator® 5, and West Nile-Innovator®
T04	Vetera® Gold
T05	Vetera® EWT + EIV/EHV, and Vetera® WNV
T06	Prestige® V + WNV
T07	Prestige® V, and EquiNile™

Horses received their respective treatment group vaccine(s) twice, 21 days apart, with the first vaccination day representing day 0 of the 42-day study. All vaccines were administered intramuscularly in the neck and treatment groups were commingled during the study.

Blood samples were collected from each horse on days 0, 7, 14, 21, 28, and 42 for serologic analysis of WNV antibody titers by an enzyme-linked immunosorbent assay (ELISA). This testing was performed by the Cornell Diagnostic Laboratory on all serum samples to evaluate the serological response to vaccination.

Statistical analyses of the serological responses of the various treatment groups were performed, with distinctions between treatment groups deemed significant at $P \leq 0.05$. Five horses were excluded from data analyses due to injury or receiving partial doses of vaccine.

Results

Horses in the control group (T02, Saline controls) remained seronegative for WNV at all sample dates. WNV serologic results over the course of the entire study are summarized in Figures 1, 2, and 3 for each manufacturer's respective WNV combination and concurrently administered WNV vaccines. On day 0, no significant differences were seen between any of the treatment groups. Beginning on day 7 post-vaccination (1 week after the first vaccination) and continuing through the duration of the study, all vaccine groups had significantly ($P \leq 0.05$) higher WNV antibodies titers than the saline control group. At day 7, with the exception of the Boehringer Ingelheim concurrently administered WNV vaccine (T05), no distinct trends between the WNV combination groups and the WNV concurrently administered groups were detected, though the greatest mean WNV antibody titer at day 7 was demonstrated by the Zoetis combination vaccine (T01).

Serologic titers for all vaccinated groups further increased by day 21 and peaked at day 28, one week after the second vaccination. WNV antibody titers were consistently elevated in all vaccinated groups at days 21, 28, and 42 compared to controls ($P \leq 0.05$), and all vaccine groups clearly displayed anamnestic antibody responses following the second vaccination on day 21. However, approximately 3-times

greater antibody titers were consistently generated at day 28 in the WNV concurrently administered treatment groups compared to horses that received the WNV combination vaccine regimens ($P \leq 0.0001$; Figures 1-3). It appears, the inclusion of WNV in vaccines containing EEE, WEE and tetanus antigens significantly decreased WNV antibody responses ($P \leq 0.0001$) compared to similar vaccines where the WNV vaccine was concurrently administered with the EEE, WEE and tetanus antigens. Similar outcomes were observed at day 42 though antibody titers had subsided in all treatment groups.

These reductions in WNV antibody titers observed in the WNV combination vaccine treatment groups vs the WNV concurrently administered vaccine treatment groups may be due to antigen interference, antigen load, or some other factors. Regardless of the attributing cause, the implications of lower WNV antibody responses in horses vaccinated with WNV combination vaccine regimens must be considered by practitioners when developing and implementing vaccine protocols in client herds.

Figure 4 summarizes WNV antibody titer outcomes for the WNV combination vaccine regimens at day 7 (7 days after first vaccination) and day 28 (7 days after second vaccination). At both time points the WEST NILE-INNOVATOR® + EWT combination vaccine regimen generated significantly ($P \leq 0.0001$) greater WNV antibody titers than the VETERA® Gold combination product. In addition, a significant ($P \leq 0.0001$) antibody titer increase was observed by the WEST NILE-INNOVATOR+EWT -combination vaccine regimen relative to the PRESTIGE® V + WNV combination product.

Figure 1.

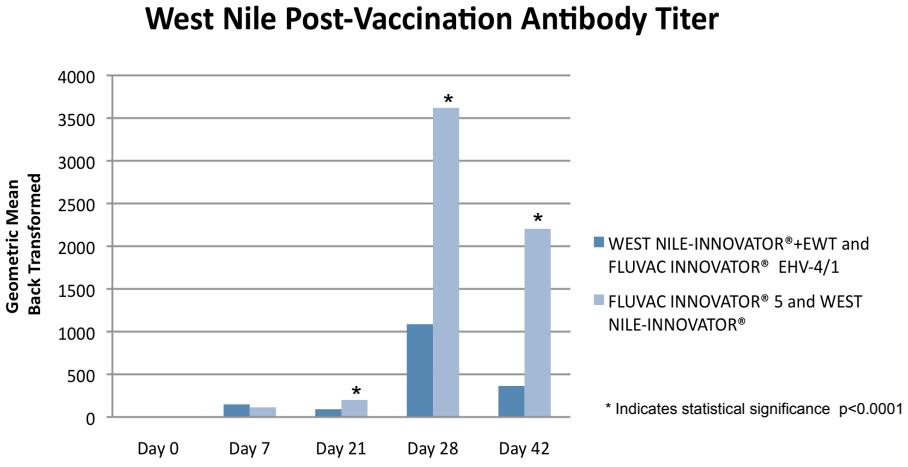


Figure 2.

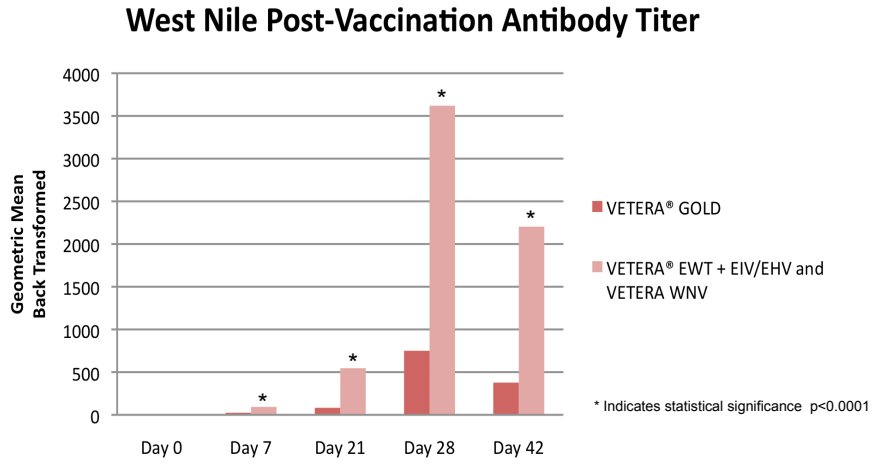


Figure 3.

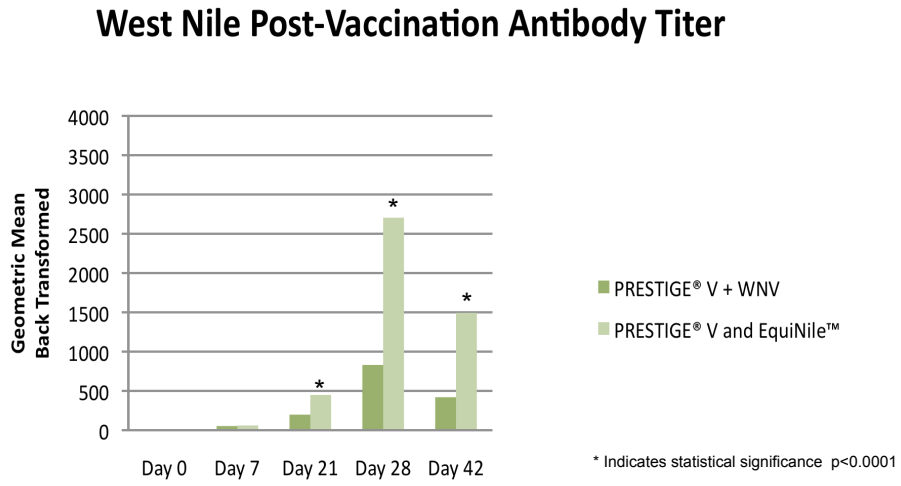
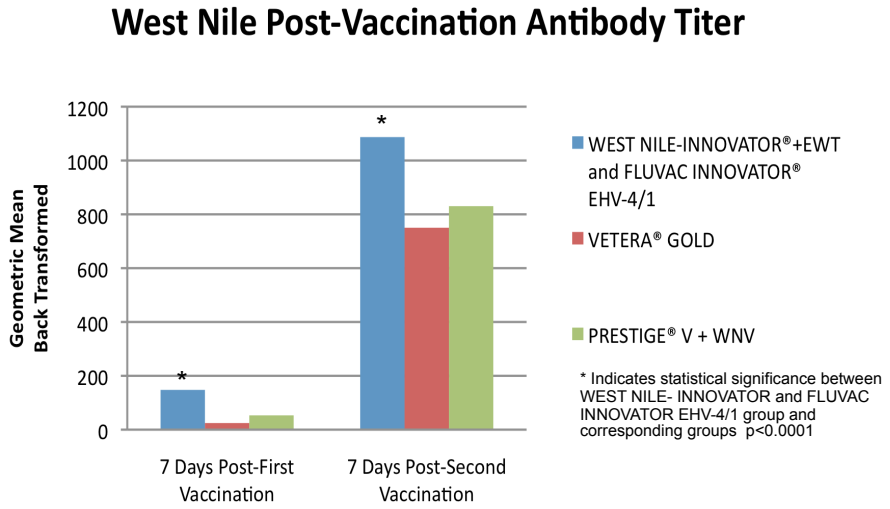


Figure 4.



Conclusion

Vaccination regimens that combined WNV with EEE, WEE and tetanus antigens in a single formulation appeared to exert an impact on the WNV antibody response compared to vaccination regimens that employed concurrent administration of the WNV component and the EEE, WEE, and tetanus antigens. When developing vaccine protocols for WNV protection, practitioners should consider the implications of lower WNV antibody responses in horses vaccinated with some WNV combination vaccines. Though combination vaccines may provide some convenience, optimization of WNV immune response could likely pose a substantial criterion for vaccine selection.

References

1. Data on file, Study Report No. 12PETBIOEQ01, Zoetis Inc.