Willingness to Approach Behavior and Feed Disappearance of Weaned Pigs Following Vaccination with *Mycoplasma hyopneumoniae*

A.S. Leaflet R2547

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Summary and Implications

In the U.S., pigs during the nursery phase receive vaccinations for circovirus, Mycoplasma hyopneumoniae, erysipelas, salmonellosis and ileitis and these vaccinations are typically administered via intramuscular injection (circovirus, Mhyo +/- erysipelas) or drinking water (ileitis, salmonella +/- erysipelas). It has been noted by swine practitioners (Anon Communication) that approximately 6-h after vaccination via intramuscular injection has been completed, pigs lie down, become more lethargic and reduce their feed consumption. Therefore, the objective of this study was to determine if field observations of markedly reduced pig activity following vaccination with certain Mycoplasma hyopneumoniae vaccines could be confirmed and quantified by observing post-vaccinal pig behavior changes for a 15 sec period over a 28 h period. A total of 1832 weaned crossbred (GPK35 x EB5, Monsanto Choice Genetic, St Louis, MO) pigs 17 to 23 d of age were obtained from a PRRS and SIV-negative, Mycoplasma hyopneumoniae positive production system. Research was conducted over 4 consecutive days in October 2007 at a commercial, mechanically ventilated nursery facility in South Central Missouri. Treatment one; MycoFLEX (n = 36pens), defined as a single intramuscular dose of 1ml MycoFLEX[®] (Boehringer Ingelheim, St. Joseph, MO) vaccine injected into the lateral cervical musculature on the right side of the neck at 5 wks of age using a 16 gauge needle. Treatment two: RespiSure-ONE (n = 36 pens), defined as a single intramuscular dose of 2 ml RespiSure[®]-ONE (Pfizer Animal Health, New York, NY) vaccine injected into the lateral cervical musculature on the right side of the neck at 5 weeks of age using a 16 gauge needle. Willingness to approach observations was performed in the same pen on two occasions 24 h apart. The d -1 (baseline) WTA observation occurred between 3:00 PM and 4:00 PM and was defined as the WTA percentage baseline for each pen (pens were observed in the same order that vaccine was to be administered). On d 0 (post-immunization), vaccination procedures began at 7:00 AM and were completed by 10:00 AM. On d 0, WTA observation occurred

between 1:00 PM and 4:00 PM and was defined as the WTA percentage post-immunization for each pen.

The percentage of pigs willing to approach on d -1 and d 0 were analyzed using a Pearson Chi-square test in JMP v.6.0.0 (Cary, NC) software comparing the percentage of pigs approaching on a pen basis. Results were considered significant when *P* values were ≤ 0.05 .

Baseline willingness to approach was not different (P > 0.05) between the two treatment groups. Six h post vaccination both treatment groups had reduced their willingness to approach to the observer, however nursery aged pigs that received the MycoFLEX[®] vaccination were more willing to approach (P < 0.0001) than their RespiSURE[®]-one counterparts. In conclusion, vaccines do seem to have some short term behavioral effect on the pig and producers and veterinarians should be mindful of this when strategizing herd health management techniques.

Introduction

Pigs over time consider the nursery environment in which they have resided for a few weeks to be their "environmental model" the place where they feel most comfortable and secure and in turn their expectations are based on this place. Novel stimuli infrequently occur during the nursery phase and so when a novel situation is implemented, this could result in innate survival responses by the individual pig. Some novel stimuli could be labeled as "more negative or adverse" to the pig, for example a vaccination procedure. In the U.S., during the nursery phase pigs receive vaccinations for circovirus, Mycoplasma hyopneumoniae, erysipelas, salmonellosis and ileitis and these vaccinations are typically administered via intramuscular injection (circovirus, Mhyo +/- erysipelas) or drinking water (ileitis, salmonella +/- erysipelas). It has been noted by swine practitioners that approximately 6-h after vaccination via intramuscular injection has been completed, pigs lie down, become more lethargic and reduce their feed consumption. These alterations in the pigs' behavioral repertoire have been labeled by swine practioners as the "buzz" response in addition to the aforementioned behavioral alterations. Therefore, the objective of this study was to determine if field observations of markedly reduced pig activity following vaccination with certain Mycoplasma hyopneumoniae vaccines could be confirmed and quantified by observing post-vaccinal pig behavior changes for a 15 sec period over a 28 h period.

Materials and Methods

Animals and location: A total of 1832 weaned crossbred (GPK35 x EB5, Monsanto Choice Genetic, St Louis, MO) pigs 17 to 23 d of age were obtained from a PRRS and SIV-negative, *Mycoplasma hyopneumoniae* positive production system. Research was conducted over 4 consecutive days in October 2007 at a commercial, mechanically ventilated nursery facility in South Central Missouri.

Diets, housing and husbandry: Seventy two pens housing 23 to 26 pigs per pen were used (split sex by side of barn). Each nursery pen measured $1.8 \text{ m width} \times 3.0 \text{ m length}$, providing 0.21 to 0.23 m² per pig respectively. Steel penning dividers were 3.0 m length \times 78.7 cm height. Woven wire flooring (3-guage, Boss Hog, J & L wire, St. Paul, MN) was utilized in all pens and pigs had ad libitum access to a pelleted diet 3413 kcal / kg ME and 24 % CP formulated to meet requirements (NRC, 1998). Diets were provided through a 5-hole dry feeder (14.0 cm depth \times 10.2 cm height \times 91.4 cm length; Automated Production systems, Assumption, IL) that had a pelleted feed capacity of 65 kg. Each pen contained one stainless steel nipple cup drinker $(12.7 \text{ cm depth} \times 25.4 \text{ cm height} \times 16.5 \text{ cm width})$ Farmweld, Teutopolis, IL). Incandescent lights were turned on for one hour at 7:00 AM and again for one hour at 4:00 PM daily during caretaker observation periods, providing a total of 10 hours of natural and/or supplemental light in the pig space each day. This resulted in a 10 h to 14 h light to dark cycle respectively. Farm personnel observed all pigs twice daily at 7:00 AM and 4:00 PM.

Treatments and experimental design: Pigs were identified by body condition (subjectively scored by into light, medium, heavy), sex and then were assigned to pens so that pen weight was even across pens. The experimental unit was the pen (containing the individual pigs) and the experimental design was a complete randomized block design (block defined as body weight). All treatments were in the same room and the pens were alternately vaccinated with one of the two vaccine treatments. Two treatments were compared; Treatment one; MycoFLEX (n = 36 pens), defined as a single intramuscular dose of 1ml MycoFLEX® (Boehringer Ingelheim, St. Joseph, MO) vaccine injected into the lateral cervical musculature on the right side of the neck at 5 wks of age using a 16 gauge needle. Treatment two: RespiSure-ONE (n = 36 pens), defined as a single intramuscular dose of 2 ml RespiSure[®]-ONE (Pfizer Animal Health, New York, NY) vaccine injected into the lateral cervical musculature on the right side of the neck at 5 weeks of age using a 16 gauge needle. The same two technicians (technician defined as the person who held the syringe and had contact with the pigs when administering the vaccine) performed vaccination procedures for both treatments. Pigs were moved by a sort board towards the alley end of their home pen. Pigs were not picked up and individually handled in an effort to avoid any additional handling associated stressors on the pigs. The technicians visually identified and selected a pig amongst the crowed pigs and in 1 sec carefully inserted the needle into the lateral cervical musculature on the right side of the neck and administered the preset dose of the vaccine from a Uni-Matic 2 ml multidose syringe (Air-Tite Products Co., Inc. Virginia Beach, VA). Following vaccination each pig received a mark placed between the scapulas using an animal-safe crayon (Raidex Animal Marking Crayons; Thousand Hill Supply, Walworth, NY) to avoid injecting the same pig twice. This study purposely lacked the inclusion of a placebo-injected group to quantify responses attributable solely to the injection procedures as this study was conducted in the pig flow of a commercial production system and it was not economically feasible to consider a non-immunized portion of this population.

Willingness to approach (WTA) methodology: Two wk post nursery placement (October 16, 2007), individual pigs were evaluated to determine their willingness to approach (WTA) to an observer d -1 (baseline; pre- immunization) and d 0 (post-immunization) using the Swine Welfare Assurance Program[®] (SWAP[®]) behavior protocol. Two different observers (observer defined as having no prior animal contact before observing the pigs) completed the WTA methodology (one observer completed all baseline WTA's and a different observer completed all post immunization WTA's). This methodology was utilized so as to eliminate any familiarity that the pigs may have developed to the baseline observer and thus avoid any positive behavioral bias that may be associated with this observer familiarity. Both observers were blinded to treatment and wore the same clothing, boots, and did not wear any artificial scents. Each observer entered a pen of pigs carrying a pen, a clipboard containing one sheet of paper that had the floor plan diagram of the barn layout (and the pre-recorded pen inventory). Willingness to approach began after the observer quietly entered a pen by stepping over the gating that separated the alleyway from the pen and then immediately crouched down in front of the alley-way gate and extended and held still, one leather gloved hand in the direction of the pigs. During the 15 sec the observer watched the stop watch and avoided eye contact with the pigs. At the conclusion of this 15 sec period the observer raised his head and scanned the pen to record (1) pigs interacting with the observer (defined as pig to human contact) or (2) pigs facing the observer in the semi-circle. Also recorded were the numbers of pigs that took one step toward the observer from any location within the pen, assuming a stance such that both eyes of that pig could be seen. These additional pigs may have been unable to enter the previously described semi-circle of pigs due to space availability within the pen. Pigs not fulfilling any of these criteria were classified as "unwilling to approach" the observer.

Willingness to approach observations: Willingness to approach observations was performed in the same pen on two occasions 24 h apart. The d -1 (baseline) WTA observation occurred between 3:00 PM and 4:00 PM and was defined as the WTA percentage baseline for each pen (pens were observed in the same order that vaccine was to be administered). On d 0 (post-immunization), vaccination procedures began at 7:00 AM and were completed by 10:00 AM. On d 0, WTA observation occurred between 1:00 PM and 4:00 PM and was defined as the WTA percentage postimmunization for each pen. Therefore, there was at least a 6h period of time from the completion of the vaccinations to the re-evaluation for willingness to approach for all pens on trial. In this study, the 6-h period was defined after surveying the U.S. swine industry (two companies that market more than 500,000 nursery-age pigs annually). These producers were asked to define the time period in hours post vaccination when nursery aged pigs were less willing to approach, increased their lying postures, decreased their feed intake and seemed to be exhibiting this "buzz" effect. The majority of respondents indicated that the "buzz" effect could be observed at 6-h post-vaccination.

Willingness to approach calculations: To calculate the percentage of pigs WTA and differences between d -1 (baseline; pre-immunization) and d 0 (post-immunization) for pigs WTA, the following equations were utilized;

1) Percentage of pigs WTA d -1 (base line; preimmunization) = total pigs that approached observer d -1 (baseline; pre-immunization) in each pen / total number of pigs in each pen.

2) Percentage of pigs WTA d 0 (post immunization) = total pigs that approached observer d 0 (post-immunization) in each pen / total number of pigs in each pen.
3) Percentage difference variable between d -0 (baseline; pre-immunization) and d 0 (post immunization) of pigs WTA = WTA d -1 (baseline; pre-immunization) - WTA d 0 (post-immunization) percentage in each pen.

Statistical analysis: The percentage of pigs willing to approach on d -1 and d 0 were analyzed using a Pearson Chi-square test in JMP v.6.0.0 (Cary, NC) software comparing the percentage of pigs approaching on a pen basis. Results were considered significant when *P* values were ≤ 0.05 .

Results and Discussion:

Baseline willingness to approach were not different (P > 0.05) between the two treatment groups. Six h post vaccination both treatment groups had reduced their willingness to approach to the observer, however nursery aged pigs that received the MycoFLEX[®] vaccination were more willing to approach (P < 0.0001) than their RespiSURE[®]-one counterparts (Table 1). In conclusion, vaccines do seem to have some short term behavioral effect

on the pig and producers and veterinarians should be mindful of this when strategizing herd health management techniques. Table 1. Pearson chi-square comparisons for the main effect of treatment (MycoFLEX[®] and RespiSure[®]-ONE) on the percentage of pigs that showed willingness to approach (WTA) to the blinded human observer in the pen, on d -1 (baseline; pre-immunization) and d 0 (post immunization) for 5-wk old nursery aged pigs in October 2007.

Treatments^a

	MycoFLEX	RespiSure-ONE	P-value
Number of pens	36	36	-
Number of pigs per pen, hd	25.3	25.8	0.44
Willingness to approach (WTA) behavior of nursery aged pigs			
Day 0 (post immunization), % willingness	49.2	39.0	< 0.0001
to approach ^c			
Change from d -1 (baseline; pre-			
immunization) to d 0 (post			
immunization), % willingness to			
approach ^d	-11.4	-27.1	< 0.0001
^a Tractment and MussELEV [®] defined as a single intromuscular dass of 1ml MussELEV [®] variation inter-			

^aTreatment one; MycoFLEX[®], defined as a single intramuscular dose of 1ml MycoFLEX[®] vaccine injected into the lateral cervical musculature on the nursery pigs right side of the neck at 5 wks of age using a 16 gauge needle. Treatment two: **RespiSure-ONE** defined as a single intramuscular dose of 2 ml RespiSure[®]-ONE vaccine injected into the lateral cervical musculature on the nursery pigs right side of the neck at 5 weeks of age using a 16 gauge needle. The same 2 people performed vaccination procedures for both treatments. ^bPercentage of pigs WTA base line (pre-immunization) = total pigs that approached observer pre-immunization in each pen / total number of pigs in each pen. ^cPercentage of pigs in each pen. ^dPercentage difference variable between baseline (pre) and post immunization of pigs WTA = WTA baseline (d -1) - WTA post-immunization percentage on d 0 in each pen