Does vaccination with Vibrovax® affect semen quality in young bulls?

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Introduction

Vibriosis is a venereal disease caused by infection with *Campylobacter fetus* subsp. *venerealis*. It can cause transient infertility, early embryonic death and sporadic abortion in cattle (Hum et al., 1994). McGowan et al (2014) found that overall 62% of beef producers in northern Australia vaccinated their bulls to prevent vibriosis with a further 7% vaccinating both bulls and heifers. Further vaginal mucus serology demonstrated that the annual prevalence of heifer mobs considered at high risk of vibriosis varied between 4 and 14%. Vibrovax® is the only vaccine registered in Australia to protect cattle against vibriosis. It is a formalin killed suspension of two biotypes of *Campylobacter (Vibrio) fetus* subsp. *venerealis* with a mineral oil adjuvant.

As part of the routine pre-sale breeding soundness examination of approximately 20 month old bulls on a central Queensland property it was observed that the incidence of bulls having percent morphologically normal sperm <70% at the first examination (typically conducted in June) varied markedly between years. A review of management of these bulls indicated that they typically received a booster vaccination for bovine ephemeral fever, received their first vaccination for vibriosis and were vaccinated for botulism either immediately before conducting the BBSE’s or some weeks before. The question that arises is ‘could vaccination of these bulls cause a decrease in percent normal sperm’.

The immune response following any vaccination varies with respect to the type of vaccine and the adjuvant used. A component of the immune response to vaccination is the acute phase reaction, which includes inflammation and the production of acute phase proteins (Horadagoda et al 1999). It is important to understand whether there is an acute inflammatory response following routine vaccinations, and whether this is associated with any adverse impact on semen quality. Given that herd bulls are commonly vaccinated immediately prior to mating with females, even transient effects of vaccination-induced inflammation could impact mating outcomes.

The aim of this study was to evaluate the impact on the semen quality of young tropically adapted bulls of vaccination with Vibrovax® to prevent vibriosis.

Materials and Methods

The study was conducted in early winter on a commercial beef cattle station near Theodore, Queensland. Bulls selected for inclusion in the study were selected from a cohort of Santa Gertrudis Bulls (n = 82) that were 21 to 23 months of age. During the study, bulls were managed in a paddock that was approximately 405ha in size and grazed pastures comprised of approximately 80% Buffel grass and 20% mixture of native pastures. As part of routine herd health, all bulls had been vaccinated for clostridial diseases as calves (four to six months of age), and for ephemeral fever and tick fever at weaning (eight to 10 months of age). All booster vaccinations were administered to the bulls three months prior to commencement of the trial. None of the bulls had been previously vaccinated against vibriosis.
Twenty-four days prior to the commencement of the study (Day -24), all bulls underwent a BBSE, as recommended by the Australian Cattle Veterinarians. The BBSE included collection of semen samples for gross and microscopic examination. At the same time, all bulls were body condition scored (BCS; 1 = thin to 5 = fat) and weighed. Bulls were excluded from the study if they had < 70% morphologically normal spermatozoa (n = 25), a semen sample suitable for assessment of sperm morphology could not be obtained (n = 1), or they were diagnosed with evidence of testicular hypoplasia (n = 1) or unilateral cryptorchidism (n = 1). Bulls selected for the study (n = 52; two bulls were excluded from the study after initial selection as they failed to present to the handling facility on Day 0 for treatment) were allocated to either a treatment or control group using a randomised block design.

All bulls were treated with two subcutaneous injections of either 5mL of the commercially available vaccine Vibrovax® (Zoetis Australia, Sydney, NSW, Australia) (treatment group) or 5mL saline (0.9% sodium chloride; Baxter, Sydney, Australia) (control group). Treatments were administered using a 16 G x ½” needle attached to a multi-dose vaccination gun. Vaccination sites were located on the left neck adjacent to the dorsal midline just cranial to the ‘hump’ on Day 0 (vaccination site 1; V1) and just caudal to the ‘hump’ on Day 28 (vaccination site 2; V2). The vaccination sites were assessed by manual palpation to detect the presence of any swelling and if present to estimate its size, on Day 0 and 14 for V1 and Day 28, 42 and 57 for both V1 and V2.

Semen samples were collected by electro-ejaculation (Lane Pulsator IV- Auto Adjust™) on days 14, 28, 42, and 57. On day 28, semen collection and treatment for parasites (12mL subcutaneous injection of VETMEC (10mg/mL Abamectin; Chemvet Australia P/L, Port Melbourne, Victoria)) was performed prior to the second vaccination. At each semen collection, samples were examined crush side for density and percentage progressively motile spermatozoa according to the guidelines published by the Australian Cattle Veterinarians, and an aliquot of raw semen was stored in buffered formol saline. Stored semen samples were sent to the Queensland Sperm Morphology Laboratory for determination of the percentage of morphologically normal sperm.

Statistical analysis
A total of 259 sperm samples were collected and measured during the study period between Day -24 and Day 57. The data were arranged so that each time point relative to the day of first Vibrovax® vaccination or saline treatment was a single observation. The per cent of normal spermatozoa were analysed within a mixed linear model that included a random intercept term for each bull.

Results
Descriptive data
Bulls selected for the study had an average BCS 2.7 (range 2.0 to 4.0) and weighed an average of 416 kg (range 345 to 550kg). The BCS remained constant over the trial period and a steady increase was observed in the mean bodyweight of the bulls. Most bulls developed some swelling after the 1st and 2nd Vibrovax® vaccination, but none showed any evidence of ulceration or abscessation of the vaccination sites (Table 1).
Table 1: Incidence and estimated size of swellings after vaccination with Vibrovax®

<table>
<thead>
<tr>
<th>Day</th>
<th>Measurement</th>
<th>0</th>
<th>14</th>
<th>28</th>
<th>42</th>
<th>57</th>
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<tr>
<td>1st vaccination</td>
<td>% swelling</td>
<td>0</td>
<td>72.0</td>
<td>92.3</td>
<td>76.9</td>
<td>61.5</td>
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<tr>
<td></td>
<td>(0/26)</td>
<td>(18/25)</td>
<td>(24/26)</td>
<td>(20/26)</td>
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<tr>
<td>Mean length*</td>
<td>-</td>
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<td>3.3</td>
<td>4.4</td>
<td>4.3</td>
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<tr>
<td>2nd vaccination</td>
<td>% swelling</td>
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<td>-</td>
<td>0</td>
<td>88.5</td>
<td>80.8</td>
</tr>
<tr>
<td></td>
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<td>(23/26)</td>
<td>(21/26)</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Mean length</td>
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<td>-</td>
<td>-</td>
<td>2.5</td>
<td>5.9</td>
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*Visual estimate in centimetres using index finger diameter

Semen quality
The mean percentage of morphologically normal spermatozoa (Figure 1), mean percentage of progressively motile spermatozoa, mean mass activity score and mean density score did not differ significantly between bulls that were injected with either Vibrovax® or saline on Days 14, 28, 42 and 57. There were also no significant interactions observed between treatment and day of semen collection for any of the semen quality measures. Further, the BCS and bodyweight of bulls did not have a significant effect on the percentage of morphologically normal spermatozoa.

At Days 14, 28, 42 and 57 a small proportion (about 10%) of bulls were detected as having <70% morphologically normal spermatozoa, but this was not significantly different between Vibrovax® or Saline treated bulls (Table 2). In bulls in which the percentage normal spermatozoa decreased below 70% at one or more consecutive samplings, the percentage normal sperm generally increased to ≥70% at the next sampling.

Table 2: The proportion of rising 2 year-old Santa Gertrudis bulls that had < 70% morphologically normal spermatozoa after vaccination with Vibrovax® or Saline on Day 0 and Day 28. Note on Day 14 one bull failed to be mustered.

<table>
<thead>
<tr>
<th>Treatment</th>
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<th>42</th>
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<td>7.7</td>
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<td>(3/26)</td>
<td>(2/26)</td>
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<tr>
<td>Vibrovax®</td>
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<td>15.4</td>
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</tr>
<tr>
<td></td>
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<td>P value</td>
<td>-</td>
<td>0.710</td>
<td>0.825</td>
<td>0.905</td>
<td>0.975</td>
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Figure 1 Mean ± SD percentage of morphologically normal spermatozoa in rising 2-year-old Santa Gertrudis bulls vaccinated with Vibrovax® (n = 26) or Saline (n = 26) on Day 0 (V1) and Day 28 (V2). Note no semen collections were performed on Day 0.

Discussion
This study demonstrated that Vibrovax® vaccination does not affect the quality of ejaculated spermatozoa from rising 2-year-old Santa Gertrudis bulls that had previously passed a BBSE and had ≥ 70% morphologically normal spermatozoa. The results are reassuring as herd sires are commonly vaccinated against vibriosis just prior to mating with females. Historically, it was not known whether this husbandry practice had any adverse impact on semen quality. However, it is important to note that this study only assessed the impact on semen quality of vaccination with Vibrovax® alone, and it is common for beef cattle producers in northern Australia to vaccinate their bulls against other diseases such as ephemeral fever, tick fever and clostridial diseases. Further research needs to be done to determine whether administration of multiple different vaccines at the one time adversely affects semen quality.

It is of interest that for 13 bulls (25% of all bulls) on at least one sampling day the percentage normal sperm was <70%. Decreases below the 70% threshold occurred on all sampling days with similar frequency. For 8 of these bulls (1 saline and 7 Vibrovax®) there was a single decline below the threshold and in 6 of these percent normal sperm was >70% on day 57. Four bulls (3 saline and 1 Vibrovax®) had declines below the threshold at 2 sampling with 2 subsequently having >70% on day 57 and one was borderline (68% normal), and in 1 bull (saline) declines below the threshold occurred at 4 samplings. For 6 bulls the major cause of the decrease in percentage normal was a high percentage vacuoles/teratoids (21 to 59%), for 3 bulls it was a high percentage proximal droplets (22 to 35%) and for 2 bulls it was a high percentage of midpiece abnormalities.

This study also demonstrated that when Vibrovax® vaccination is conducted according to label directions there is no associated development of vaccination site abscesses. However, mild to moderate swelling at the vaccination site was detected in most bulls 2 to 4 weeks post-vaccination.
Acknowledgements
The authors would like to thank Dr John Al-Alawneh (School of Veterinary Science, The University of Queensland) for conducting all the statistical analysis. This work was funded by Zoetis Australia Research and Manufacturing Pty Ltd.

References