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Short communication

Effect of a quadrivalent vaccine against respiratory virus on the incidence of respiratory disease in weaned beef calves

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Abstract

We investigated the effect of vaccination of male beef calves (mean age ± S.D.: 158 ± 31days) against bovine herpes virus (BHV-1 or IBR virus), bovine respiratory syncitial virus (BRSV), bovine viral diarrhea (BVD) virus and para-influenza (PI_3) virus on the incidence of respiratory disease during the first forty days after weaning and entering a feed-lot in Portugal.

In May 2003, Mertolenga, Preta and mixed-breed calves from 10 different beef herds, were systematically assigned (by order of entrance in a chute) to two treatment groups, before moving to a common feed-lot. One hundred and twenty five male calves were vaccinated with a quadrivalent vaccine (Rispoval 4®) and revaccinated after 21–27 days while 148 herdmates were injected with saline (0.9% NaCl) on the same occasions. The incidence and severity of clinical cases of ''bovine respiratory disease'' (BRD) were evaluated every day during the first 40 days after entering the feed-lot. Morbidity (3% vs. 14%) and mortality (0% vs. 4%) due to BRD were significantly lower in the vaccinated group. Ten days after revaccination, the calves were treated with an antimicrobial – ending the study – after an outbreak of BRD caused a high incidence of disease in the non-vaccinated group.

In conclusion, our results showed that Rispoval 4®, a quadrivalent vaccine against respiratory viruses, under field conditions, reduces morbidity and mortality due to BRD in beef calves after weaning.

Keywords: Clinical trial; Vaccination; Bovine respiratory virus; IBR; BVD; BRSV; PI

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1. Introduction

Respiratory disease in weaned feedlot calves (''bovine respiratory disease complex''; BRD), is the leading cause of morbidity and mortality in feed-lots worldwide (Griffin, 1997). This syndrome has a complex and multifactorial aetiology that usually is divided into three major categories: environmental factors, host factors and infectious factors (Stilwell, 2003; Ellis, 2001; Dyer, 1993). Bacteria, such as *M. haemolytica* and *P. multocida*, cause a severe illness when environmental factors (stress, lack of ventilation, crowding and others) and/or viruses reduce the capacity of the animal to control the infection (Storz et al., 2002; Thomson, 1993).

Although environmental factors are crucial in the origin of BRD and should be addressed in a preventive program (Engelken, 1997), their control is usually harder to achieve than the management of the infectious element of the disease. This is the main reason why, although the etiopathogenesis of the disease is well known, the beef producer depends so much on antimicrobials and vaccines.

The control of BRD through the continuous use of antimicrobials has many disadvantages (Ellis, 2001; Brumbaugh, 1996): expense, inefficiency, risk of antimicrobial resistance, and threats to animal welfare (because it only controls the bacterial stage of the disease). As a consequence of this, it follows that the administration of antimicrobials (even in a prophylactic approach) should be kept for situations when the control of BRD is not possible through other means.

Vaccination against respiratory viruses seems to be a sensible and prudent approach, especially when we can forecast moments of important immunity suppression associated with these viruses, such as during the weaning and grouping of young animals from different herds (Dyer, 1993). However, the use of vaccination for the control of BRD in animals on arrival to feed-lots is still controversial (Ellis, 2001) and only occasionally used in Europe and very seldom used in Portugal (personal observations). The economic benefit of vaccination against BRD is not fully demonstrated (Smith et al., 1996) although there are some important factors (e.g. animal welfare) that are not considered in many of these studies (Tizard, 2000). Most vaccine trials are conducted under controlled conditions or followed by experimental infection. Field studies, on the contrary, are still scarce (Engelken, 1997).

The respiratory viruses commonly responsible for BRD include bovine herpes virus-1 (Infectious Bovine Rinotracheitis or IBR), bovine respiratory syncitial virus (BRSV), bovine viral diarrhea (BVD) virus and para-influenza (PI_3) virus (Storz et al., 2002). Other viruses that have been implicated in the pathogenesis of BRD include coronavirus, adenovirus, parvovirus and rhinovirus (Storz et al., 2002; Smith et al., 1996). All of these viruses are known to be involved in BRD solely or in synergism with each other and bacteria. Some affect the lung parenchyma directly (BRSV and PI_3), while others act on the immune system (BVD and BRSV) or local defences (IBR), like the ciliated epithelium. The characteristics and role of these viruses in the pathogenesis of BRD are well documented (Ellis, 2001; Baker et al., 1997; Smith et al., 1996; Thomson, 1993). However, in the field, it is usually difficult to identify the virus or viruses implicated in an outbreak of BRD especially in the feed-lot conditions. This is the main reason why multivalent vaccines are preferred.

Our objective was to evaluate the benefit of a multivalent vaccine (Rispoval 4®; Pfizer Animal Health) against the four respiratory viruses, by comparing the morbidity incidence and the mortality of clinical cases of BRD between groups of vaccinated and non-vaccinated male beef calves, during the 40 days in a feed-lot.

2. Material and methods

2.1. Study animals

The weaned calves were from 10 different herds from the Ribatejo region of Portugal. The calves' mean age at weaning (overall and by breed) is presented in Table 2.

Weaning of calves born during winter takes place in May and on the day of weaning the calves are all transported to one nearby feed-lot. Morbidity due to BRD has always been ~10% in this unit and in 2002 mortality reached 6% in the month following weaning. All calves are kept in the same open barn with slated concrete floor. Feeding includes commercial corn-based concentrate and alfalfa hay. Water is always available.

The history of these herds showed that no vaccination against the respiratory viruses was ever performed.

2.2. The vaccination

The best vaccination program would be to administer the first dose three weeks before weaning and revaccinate on the day of weaning (Engelken, 1997). However, our main objective was to test the efficacy of this vaccination in field conditions, where vaccination on the weaning day and revaccination 15–28 days later is the most practical schedule.

The vaccine used, Rispoval 4®—Pfizer, contains the following:

- Inactivated BHV-1 (IBR virus).
- Modified live BRS virus.
- Inactivated BVD virus.
- Modified live PI_3 virus.

On weaning day, 126 male calves were vaccinated intramuscularly with 5 ml Rispoval 4® and 147 received a 5-ml intramuscular injection of saline solution. No calf showed any signs of illness and all were in excellent body condition.

In each herd, all males to be weaned were moved into a race and were alternately allocated to the two treatment groups by order of entrance (the first was always allocated to the non-vaccinated group). With two herds, due to vaccine unavailability, the last calves of the last chute were all included in the non-vaccinated group. In contrast, the last four calves in two Mertolenga herds were all vaccinated to use the last doses of already open bottles of vaccine.

The first dose of vaccine was administered between the 15th and 22nd of May 2003 (weaning of the 10 herds occurred during this period) and the second on the 12th of June. With this schedule we had calves revaccinated 21–27 days after first dose.

The distribution of calves vaccinated and non-vaccinated are presented in Table 1.

2.3. Disease evaluation and treatment

Animals were inspected daily by herdsmen (blind to the study) and sick or suspected calves were separated. Clinical evaluation and selection of animals for antimicrobial treatment were made by the feed-lot veterinarian (blind to the study). Clinical signs used in the selection of BRD-affected animals were: isolation from herdmates, decreased appetite, depression (first signs detected by herdsman) and dyspnea, cough, nasal and ocular discharge and hyperthermia

Table 1
Number and breed of calves vaccinated and affected by bovine respiratory disease (BRD) in June 2003

	Calf breeds		
	Mertolenga	Preta	Unknown
Vaccination			
Mean age ± S.D.[a]	156 ± 30	161 ± 28	150 ± 29
Non-vaccinated (*n*)	48	56	44
Vaccinated (*n*)	56	39	30
BRD incidence			
Mean age ± S.D.[b]	130 ± 12	218 ± 34	156 ± 20
Morbidity			
Non-vaccinated (*n*, %)	4/48	12/56	4/44
	8	14	9
Vaccinated (*n*, %)	2/56	0/39	2/30
	4	0	7
Mortality			
Non-vaccinated (*n*, %)	0/48	4/56	2/44
	0	7	5
Vaccinated (*n*, %)	0/56	0/39	0/30
	0	0	0

[a] At weaning day.
[b] On first day of disease.

(>39.5 °C), that resulted from the vet-conducted physical examination. Only animals showing all of these signs (albeit with different severities) were included in the BRD-affected group. Four sudden deaths occurred and were also considered BRD after blinded post-mortem confirmed extensive pulmonary lesions.

Those animals that were considered BRD positive were isolated and treated with antimicrobials (Tilmicosin, 10 mg/kg BW, subcutaneous) and non-steroid anti-inflammatory drugs (Flunixin Meglumine, 2 mg/kg BW, subcutaneous).

The lungs of three dead animals were sent for microbiology and histopathology exams at the appropriate laboratories of the Faculdade de Medicina Veterinaria de Lisboa (all lab personnel were blind to the study).

2.4. Statistical analysis

Numbers of morbidity and mortality due to BRD were submitted to a logistic-regression analysis through PROC LOGISTIC and PROC GENMOD, respectively, using SAS software (SAS, 2004). Effect of vaccination, breed, calf's age at vaccination (co-variable) and their respective interactions, were included in the logistic multiple regression models. The Wald Chi-square test was used to assess the importance of each factor. Because a significant effect ($P > 0.05$) was not shown for interactions these were removed from the model.

The need to use PROC GENMOD for mortality was due to the fact that data showed some ''quasi-complete separation'' problems. If the data are completely or partially separated, it may not be possible to obtain reliable maximum likelihood estimates because convergence may not occur. Convergence does not occur because one or more parameters in the model become theoretically infinite. Such is the case if the model perfectly predicts the response or if there are more parameters in the model than can be estimated because the data are sparse (Webb et al., 2004).

Table 2
Logistic regression analysis: Effect of vaccination, breed and age of calves weaned on morbidity and mortality due to BRD.

Effect	d.f.	Morbidity		Mortality	
		Wald χ^2 values	P	Wald χ^2 values	P
Vaccination	1	7.62	0.0057	7.46	0.0063
Breed	2	2.98	0.2253	3.27	0.1949
Age at vaccination	1	3.87	0.0498	0.11	0.7444
AIC		157.457		54.860	

d.f., Degree of freedom; AIC, Akaike Information Criterion for Intercept and Covariate.

3. Results

Table 1 shows the data concerning morbidity and mortality during the first 40 days after weaning and first vaccination. Mean age at weaning and at onset of disease is not comparable because calves got sick at different days during the 40-day study.

Logistic-regression analysis (Tables 2 and 3) shows that vaccination did have a significant effect ($P < 0.01$) on morbidity and mortality due to BRD. The Odds ratio between non-vaccinated and vaccinated animals showed that the former are 4.822 more likely to get BRD.

Age at weaning/vaccination also had a significant effect on respiratory disease incidence ($P < 0.05$), with a slope coefficient of -0.0133 ± 0.0066 that represent the change in log Odds of the liability per unit increase in age (days). Odds ratio for age showed that older calves are less likely (0.987) to get BRD (Fig. 1).

Breed did not have any effect on morbidity or mortality.

Calves showed signs of BRD throughout the study period, but mortality was especially high after a few very hot days (>40 °C). During this outbreak (20–24th of June, 40 days after the first group was weaned), it was decided to finish the trial and the majority of the animals were treated with antibiotics (10 mg/kg BW of Tilmicosin) to try to control mortality. From that moment onwards the study comparing susceptibility to BRD was considered closed.

There was no report of treatments or deaths due to BRD after the outbreak previously described.

3.1. Post-mortem exams

Post-mortem examination was performed on the four animals that died at three different times—3, 5 and 8 days after second vaccination.

Gross lesions included: pleurisy, emphysema and oedema (especially evident in the dorsal lobes), thickness of interlobular septae and signs of bronchopneumonia in the cranioventral lung.

Table 3
Logistic regression coefficients and Odds ratio (OR) estimates

Effect	$b \pm$ S.E.	OR estimate	OR 95% confidence	Wald limits
Vaccination	0.7866 ± 0.2848	4.822	1.579	14.722
Age at vaccination	-0.0133 ± 0.0066	0.987	0.972	1.002

b, Regression coefficient; S.E., standard error; OR, Odds ratio.

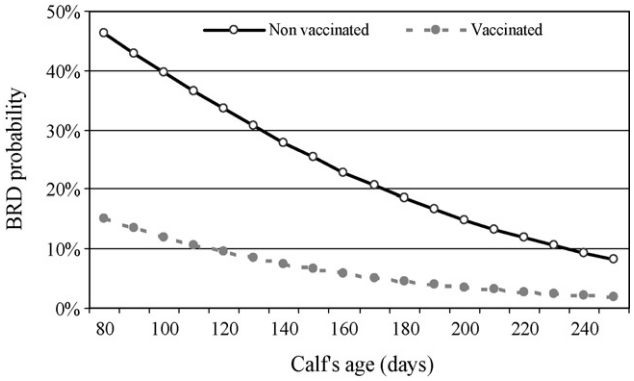

Fig. 1. Respiratory disease morbidity—probable evolution in relation to age and treatment (vaccinated vs. non-vaccinated) in calves.

The lungs of three dead animals were sent for histopathology exam (the fourth animal had been dead for long and it was felt the laboratory analyses were not going to be useful). Microscopic exam showed pleurisy lesions, traqueitis with pseudo-membranes, intense proliferation of the sub-mucosa lymphoid tissue, pronounced hyperplasia of the peribronchic lymphoid tissue (BALT), hyperplasia of bronchiolar and alveolar epithelium, oedematous and emphysematosous alveolitis showing large number of multinucleated giant cells (syncytial cells).

No bacterium was found in two samples sent for microbiology and one revealed *P. multocida*. Virus isolation was not attempted.

4. Discussion

The study was slightly affected in its duration by the need to treat animals, after 40 days of permanence in the feed-lot, because of the outbreak of clinical respiratory disease that severely affected the non-vaccinated population (Table 2). This outbreak and the end of the study occurred 13 days after the second dose of the vaccine was given to the last weaned calves and 20 days after the revaccination of the first weaned calves.

Our study promoted more adverse conditions for the vaccinated group than would occur if all animals were to be vaccinated. Virus shedding and circulation is heavier when more than half the population has not been vaccinated. In the same perspective, the non-vaccinated calves were favoured because of the relatively lower circulation of virus compared to the situation where no animal had been vaccinated.

In spite of this drawback, vaccine efficacy was demonstrated by reducing the incidence of BRD and mortality due to respiratory disease in calves recently weaned (Table 2). The Odds ratio analysis also shows that animals that are weaned at an older age are less likely to show respiratory disease. This is especially evident in the non-vaccinated group (Fig. 1).

Clinical signs, necropsy lesions and histopathology exams suggest that at least one of the respiratory viruses (BRSV) was involved in the BRD cases. The use of a live vaccine against this virus seems appropriate when vaccination is very close to weaning, transport and commingling because of the rapid immunity response.

In conclusion, vaccination with Rispoval 4® reduced morbidity and mortality associated with BRD in calves recently weaned and commingled in a feed-lot.

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