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Effectiveness of a commercial leptospiral vaccine on urinary shedding in naturally exposed sheep in New Zealand

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ABSTRACT

L. borgpetersenii serovar Hardjo and *L. interrogans* serovar Pomona are endemic in New Zealand sheep. An effective vaccine and vaccination strategy would protect both humans and livestock.

Four to 12 lambs were selected from each of eight farms (total = 84, vaccinated group), while four to 16 lambs (total = 98) served as unvaccinated controls. A commercial Hardjo/Pomona vaccine was given at 1–6 weeks of age, 5–11 weeks later and 33–67 weeks later on seven farms and at 18 weeks of age and 5 weeks later on the eighth farm. Vaccinates and controls were grazed together. Blood was regularly collected from the control group to assess flock exposure. Urine was collected from both groups 26–82 weeks after the second vaccination and tested by quantitative PCR.

Seroprevalence in controls at the time of urine sampling ranged from 2.7 to 98.2% for Hardjo and from 0 to 54.1% for Pomona with seroconversion occurring 13 to 67 weeks after the second vaccination in all but one farm where exposure had happened by the time of vaccination. The shedding prevalence adjusted for clustering in farms was 45.1% [95% CI 17.6–72.7] (for an observed number of 50/98) in the control animals and 1.8% [95% CI 0.0–10.1] (for an observed number of 5/84) in the vaccinated animals. The vaccine was 100% effective on five farms where animals were vaccinated before 12 weeks of age and before natural exposure occurred, but the effectiveness was 80% [0–97] on one farm where the lambs were exposed before vaccination and 65% [9–87] to 80% [0–97] on one farm where the animals were fully vaccinated by 24 weeks of age. The overall vaccine effectiveness was 86.3% [63.6–94.8%] despite maternal antibodies in some flocks at first vaccination. Vaccination timing seemed to be crucial in achieving optimum reduction in shedding in urine in vaccinated sheep.

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1. Introduction

Leptospirosis is widespread in New Zealand, with 97% of sheep flocks having one or more animals seropositive for serovars Hardjo or Pomona [1]. Sheep are reservoir hosts for Hardjo [2]. They are able to shed live leptospires persistently or intermittently for at least 11 months in urine after natural Hardjo infection [3] or, for Pomona, at least 102 days after artificial challenge and up to 9 months after natural challenge [4].

Leptospiral infection in sheep can have adverse economic effects by causing clinical disease manifested by fever, jaundice, hepatic and renal dysfunction, haemoglobinuria, anaemia and lamb mortality [5–8]. Subclinical disease is also reported, with suspected impaired reproductive efficiency, abortion and agalactia [9,10]. Additionally, seropositive sheep can be shedding without clinical signs, and are thus exposing humans to the risk of infection.

In 2012, 113 human cases of leptospirosis were notified in New Zealand [11], a figure likely to be underestimated by 40 (95% CI 16–56) times [12]. Meat workers are particularly at risk when processing sheep [13–16]. Workers in one sheep abattoir were exposed to up to 54 kidney culture positive carcasses per day

Abbreviations: GEE, Generalized estimating equation; MAT, Microscopic agglutination test; RR, Risk ratio; VE, Vaccine effectiveness.

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during high risk periods [15], and in four other abattoirs had an average annual infection risk of 11.1% [12].

Some reports suggest that human acute severe leptospirosis can be successfully treated with a combination of corticosteroids and antibiotics if diagnosed early [17,18]. However, the disease often remains undiagnosed or is diagnosed too late due to its non-specific clinical signs. Therefore prevention of infection is the most efficient control measure. The use of personal protective gear has been shown not to reduce the risk of infection in meat workers [16]. No human vaccine is available in New Zealand, and protection therefore relies on avoiding exposure to infected urine. The most effective way to protect humans in New Zealand would thus be reducing or eliminating *Leptospira* shedding in farm animals by vaccination [19].

Leptospiral vaccines currently registered for livestock in New Zealand are inactivated whole-cell vaccines with an adjuvant. No cross-protection exists between serogroups, so vaccination schemes must include the serogroups that are endemic in the area [20]. One of 161 New Zealand sheep farmers surveyed reported vaccinating their sheep against leptospirosis [12]. Vaccines have shown variable efficacy in reducing shedding or renal carriage in cattle after artificial challenge with serovars Hardjo or Pomona using various challenge routes, strains and protocols: some reports showed vaccines were efficacious [21,22], while others showed poor efficacy [21,23,24], for an overall range of 0–100%. The reported efficacy in the face of natural challenge was 71 to 100% reduction in urine shedding up to one year after vaccination [25–27]. A vaccination campaign started in 1983 in New Zealand dairy cattle was associated with a >80% reduction in the incidence of disease among dairy farmers and dairy workers [28].

Marshall et al. [29] artificially challenged sheep with serovar Hardjo 6 weeks after vaccination and found 2/9 vaccinated and 10/10 control sheep positive at kidney culture. Early studies with serovar Pomona showed a vaccine efficacy of 100% when challenge occurred 20 days, 40 days or 49 weeks after vaccination [4], but only dark field microscopy was used, a method which lacks sensitivity and specificity especially for low shedding rates. An experimental challenge trial on 9–11-week-old lambs [30] challenged four months after vaccination with Hardjo or Pomona showed an efficacy of 100% as measured by urine and kidney culture. Hence there is a dearth of data on the efficacy of either vaccines *per se* or vaccination programmes in a natural challenge, commercial sheep farming context. Studies on vaccine effectiveness for preventing shedding after natural challenge in a commercial farming environment are therefore needed as an essential step to validate the use of vaccine by sheep farmers.

This study evaluated the effectiveness of *Leptospira* vaccination in sheep in reducing shedding in urine in naturally infected commercial sheep flocks.

2. Materials and methods

2.1. Animals

From September to November 2010 (farms A-G) and in January 2013 (farm H), between 170 and 327 ewe lambs per farm from eight commercial sheep and beef farms in the North and South Islands of New Zealand (Table 1) were enrolled for a study of production effects of leptospirosis [31]. One third of the enrolled lambs were randomly selected to receive a bivalent commercial vaccine against *L. borgpetersenii* serovar Hardjo type Hardjo-bovis and *L. interrogans* serovar Pomona. This constituted no more than 5.2% of sheep on any farm, and no more than 1/3 of any management group at enrolment, with the intention to minimise the disturbance of natural exposure dynamics. On some farms the group composition

Table 1
Farm location, breed, median age and Hardjo and Pomona seroprevalence (MAT titre ≥48) of sheep at the first vaccination ("Leptavoid-2", MSD Animal Health), weeks between vaccination 1 and mid-study sampling, Hardjo and Pomona seroprevalence (MAT titre ≥48) of control sheep at mid-study, and vaccination schedule.

Farm	District	Sheep breed	Median age at first vaccination (weeks)	Seroprevalence (%) in vaccinated lambs at first vaccination		Seroprevalence (%) in control lambs at mid-study			Date of first vaccination	Weeks between vaccination 1 and 2	Weeks between vaccination 2 and 3
				Hardjo	Pomona	Weeks since vaccination 1	Hardjo	Pomona			
A	Manawatu	Romney composite	1	57.7 (56/97)	4.2 (4/96)	11	4.9 (9/182)	0 (0/182)	1/10/2011	11	33
B	Tararua	Perendale composite	6	15.9 (17/107)	1.9 (2/107)	27	5.1 (9/176)	13.1 (23/176)	22/11/2011	8	46
C	Wairoa	Romney Coopworth cross	3	67.0 (67/100)	10.1 (10/99)	27	0 (0/188)	0.5 (1/188)	17/10/2011	6	NA*
D	Central Hawke's Bay	Romney texel cross	2	6.3 (5/79)	2.5 (2/79)	25	1.3 (2/159)	1.9 (3/161)	30/09/2011	8	NA*
E	Hurunui	Romney	4	76.9 (83/108)	0 (0/110)	27	1.0 (2/197)	0 (0/197)	25/10/2011	8	67
F	Hurunui	Lincoln merino cross	4	25.8 (24/93)	0 (0/94)	27	20.2 (33/163)	4.3 (7/163)	28/10/2011	8	NA*
G	Manawatu	Romney	2	18.5 (10/54)	1.9 (1/54)	23	0 (0/111)	0.9 (1/111)	19/10/2011	8	33
H	Waikato	Romney Finn coopworth composite	18	1.1 (1/94)	1.1 (1/94)	13	3.0 (5/169)	2.9 (5/170)	8/01/2013	5	NA*

* *Not applicable*: the third injection was given on the day of urine sampling, after or not given at all, so it did not contribute in the evaluation of vaccine effectiveness.
** *Not applicable*: no blood sample.

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