Effects of live *Brucella abortus* strain 19 vaccine on reindeer later challenge exposed with *Brucella suis* type 4.

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Abstract: Twelve reindeer *(Rangifer tarandus)* were vaccinated with *Brucella abortus* strain 19 vaccine and challenge exposed with *B. suis* type 4 two and one-half months later during mid-gestation. An additional 10 reindeer served as non-vaccinated controls. A sharp serologic titer response was observed in both vaccinates and controls. *Brucella suis* type 4 was isolated from tissues and blood from most controls (8 of 10, and 7 of 10 respectively). Seven of 11 vaccinated cows aborted, gave birth to weak fawns that died, or were not pregnant at the completion of the experiment. *Brucella suis* type 4 was isolated from the tissue of 4 of 12 vaccinates at necropsy. It was concluded that, under the conditions of this experiment, *B. abortus* strain 19 vaccine in reindeer did not provide adequate protection against challenge exposure with virulent *B. suis* type 4 organisms.

Key words: Reindeer, brucellosis, vaccine

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Introduction

Reindeer *(Rangifer tarandus)* herding in Alaska has been a viable industry since 1892 when herds were first introduced from Siberia for use by the Eskimos. The Alaska Native Claims Settlement Act has stimulated growth of commercial reindeer production in the past few years with approximately 30,000 animals now being herded. Brucellosis caused by *Brucella suis* type 4 has historically been a problem in these herds (Meyer, 1966, Dieterich, 1981). Because vaccination against brucellosis can potentially reduce herd production losses, investigations are continuing to determine the most efficient vaccine for use against *B. suis* type 4 infection in reindeer. Observations on the efficacy of *B. melitensis* strain H-38 and *B. abortus* strain 45/20 vaccines have been reported previously (Dieterich et al, 1980; 1981). Because *B. abortus* strain 19 vaccine is widely employed to protect domestic cattle against *B. abortus* infections, the efficacy of this vaccine against *B. suis* type 4 infections in reindeer was examined.

Materials and methods

Facilities - During the study, all reindeer were maintained in isolation rooms (10.5 m^2), 2 or 3 reindeer in a room; bedding material was wood shavings. The light-dark cycles in the animal room were adjusted to simulate natural conditions. Reduced air pressure was maintained in all animal and work rooms, and exhaust air was passed through absolute filters. Personnel entered through shower-in-shower-out air locks and wore protective clothing. Pass-through autoclaves and sewage kill tanks were used to prevent environmental contamination. Serologic and bacteriologic studies were performed in isolation suites.

Experimental procedure - Twenty-two reindeer, including 12 (11 mature females and 1 mature male) in the vaccinated groups and 10 (all mature females) in the non-vaccinated control-groups, were involved in two similar investigations (1978-79, 1979-80). All females had been with males during the September-October rutting season and were assumed to be pregnant. Prior to the investigation, all 22 reindeer were seronegative for brucellosis by the standard plate (SP), rivanol (Riv), complement fixation (CF), and buffered *Brucella* antigen (BBA) rapid card tests. The 1978-79 vaccinates and controls were also seronegative by the standard tube (ST), mercaptoethanol (ME) and antiglobulin (AG) test. All were negative on hemoculture, and their hematologic values (WBC, PCV, differential WBC) were within normal values. During the study a commercial grain and pellet mixture (Quality Texture, Fisher Mills Inc., Seattle, Washington, USA) was fed ad libitum.

Each vaccinated deer received a 1-ml dose of approximately $3x10^8$ colony forming units (CFU) of *B. abortus* strain 19 vaccine subcutaneously in the right shoulder area in November. Approximately two and one-half months later at mid-gestation, both the vaccinated and the control reindeer were challenge exposed conjunctivally with approximately $1x10^7$ CFU of *B. suis* type 4 organisms in 0.1 ml. of saline. Post-vaccination and post-challenge blood samples were obtained weekly or biweekly for serologic testing and bacteriologic culture. Serologic and bacteriologic examinations were carried out on blood and body tissues using the methods previously described (Dieterich, 1981). Reindeer from both groups and their viable offspring were necropsied approximately 4 months after challenge exposure. Tissues examined bacteriologically included: retropharyngeal, mandibular, suprapharyngeal, parotid, prescapular, prefemoral, popliteal, external and internal illiac, mesenteric, mediastinal, supramammary, and superficial inguinal lymph nodes, and portions of spleen, liver, biceps femoris muscle, uterus, cervix, and udder or testes. Tissues or fluids were collected from other areas if lesions were observed.

Results

Results of all serologic tests followed a similar pattern (Fig. 1). One to 3 weeks after vaccination (or challenge in the control group), there was a sharp increase in antibody titer which peaked at approximately 1 month. Positive serologic responses were first detected in the SP, BBA, CF and Riv tests followd by the ST, AG and ME tests. A slight anamnestic response was seen in the vaccinates 2 to 4 weeks after challenge exposure. Titers in both groups dropped to moderate levels by the end of the experiment. Serologic titers were detected in 1 of 6 fawns in the vaccinated group in 7 of 9 fawns in the control group.

Brucella abortus strain 19 was cultured from the blood of 4 of the 12 vaccinates within 1 month of vaccination. *Brucella suis* type 4 was isolated from hemocultures of 2 other vaccinates after challenge. Hemocultures from 7 of the 10 control animals were culture positiv for *B. suis* type 4 after challenge. *Brucella suis* type 4 was cultured from vaginal swabs from one of 6 vaccinates and 6 of 9 controls.

All 21 of the adult females (11 vaccinates and 10 controls) were presumed to be pregnant at the start of the study. One of the 11 vaccinates aborted, and 2 gave birth to weak fawns that died a short time later. Four vaccinates did not produce surviving fawns. The remaining 4 vaccinates did not produce fawns, and no external signs of abortion were observed. Two of the control reindeer aborted culture-positive fetuses, 2 delivered weak fawns that died (both culture positive), and the remaining 6 gave birth to live surviving fawns.

Brucella abortus strain 19 was isolated from the tissue of 1 of 12 vaccinates cultured at necropsy. *Brucella suis* type 4 was isolated from tissues of 4 vaccinated adults and from the 2 healthy fawns born in that group. *Brucella suis* type 4 was isolated from tissues from 8 of the 10 controls and from 9 of their fawns. One milk sample from a control reindeer was culture positive, but no milk samples from the vaccinates were culture positive for *B. suis* type 4.

Discussion

The challenge inoculum used in these studies was sufficient to cause infection in 8 of 10 controls. In addition, 7 of 10 were positive on hemoculture, all sero-converted, and 4 of 10 aborted or had weak fawns that died. These data indicate an adequate challenge procedure and dose were used on the eontrols and strain 19 vaccinates.

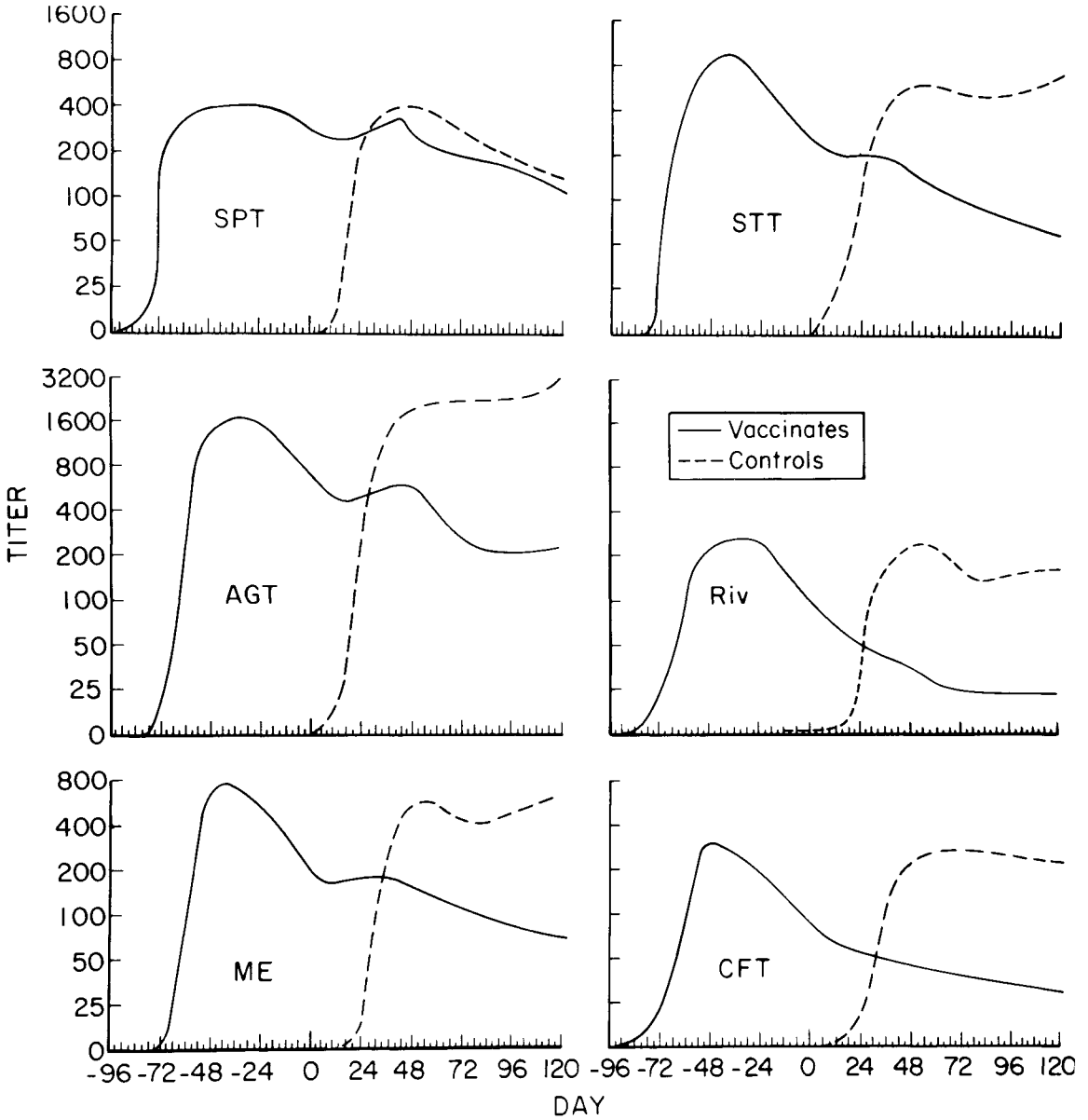

Fig. 1. Typical geometric mean titers of reindeer vaccinated with strain 19 and later challenge exposed with *B. suis* type 4.

The efficacy of the strain 19 vaccine against reproductive loss was unsatisfactory since 7 of 11 vaccinated cows aborted, gave birth to weak fawns that died, or were barren. While it was not possible to confirm early pregnancy in the test reindeer, it was unusual that all the controls were pregnant while 4 of the 11 vaccinates were barren. In 2 previous studies, only 1 of 20 reindeer was not pregnant when bred and held under similar conditions. In addition, tissues from 1 of the barren vaccinates was culture positive for strain 19 as well as *B. suis* type 4. Questions concerning the pathogenicity of strain 19 were addressed in a subsequent study (Dieterich, in Prep.).

Isolation of *B. suis* type 4 from tissues from 4 of the 12 vaccinates and from 2 of 7 of their fawns was further evidence of vaccine failure. Increased resistance of reindeer to brucellosis was successfully demonstrated in a previous similar experiment using *B. abortus* 45/20 vaccine and indicated reindeer are capable of producing

immunity to infection (Dieterich et al, 1981).

It was concluded that, under the conditions of this experiment, *B. abortus* strain 19 vaccine in reindeer did not provide adequate proteciton against challenge with virulent *B. suis* type 4 organisms.

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