

A Seroepidemiological Study of the Importance in Cow-Calf Pairs of Respiratory and Enteric Viruses in Beef Operations from Northwestern Quebec

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ABSTRACT

Serum antibody analyses for bovine herpesvirus type 1 (BHV-1), bovine viral diarrhoea virus (BVDV), bovine respiratory syncytial virus (BRSV), bovine coronavirus (BCV), and bovine rotavirus (BRV) were performed on 527 randomly selected cows, before calving, and on 407 three-week-old calves. In cows and calves, BCV and BRV were the most seroprevalent viruses (80% to 100% according to virus and vaccination status). Bovine respiratory syncytial virus was the least seroprevalent in the cows, independent of the vaccination status. In nonvaccinated cows the seroprevalence to BRSV was 36.7%, and 53.5% in cows vaccinated less than two weeks prior to collecting blood, and 67.6% in cows vaccinated two weeks or more prior to blood collection. In their calves, BHV-1 was the least seroprevalent, independent of the vaccination status. The serological status and antibody titers in calves were generally associated with those of the dam. The occurrence of respiratory diseases in the calves was associated with cow and calf serological profiles (BHV-1, BRSV and BCV in the nonvaccinated group, BHV-1, BVDV and BCV in the vaccinated group). The occurrence of diarrhoea was not associated with cow and calf serological profiles but was negatively associated with high level calf serum IgG in the nonvaccinated group (odds ratio = 0.73). Bovine coronavirus and BRV were shed by

1.4% and 4.9% of calves in the non-vaccinated group, and by 0% and 9.9% of calves in the vaccinated group, respectively. Bovine rotavirus shedding was associated with fecal diarrhoeic consistency at the moment of fecal sampling but not with previous occurrence of diarrhoea.

RÉSUMÉ

Les sérums de 527 vaches en gestation choisies de façon aléatoire et de 407 veaux de 3-4 semaines d'âge nés de ces vaches ont été analysés pour la présence d'anticorps contre le virus herpès de type 1 bovin (VHB-1), le virus de la diarrhée virale bovine (VDVB), le virus respiratoire syncytial bovin (VRSB), le coronavirus bovin (CVB) et le rotavirus bovin (RVB). Chez les vaches et les veaux, les CVB et RVB avaient les séroprévalences les plus élevées (80 à 100 % selon le virus et le statut vaccinal). Chez les vaches, le VRSB était le moins séroprévalent, quel que soit le statut vaccinal. La prévalence en anticorps contre le VRSB était de 36,7 % chez les non-vaccinées, de 53,5 % chez les vaches vaccinées moins de deux semaines avant la prise de sang, et de 67,6 % chez celles vaccinées deux semaines ou plus avant la prise de sang. Chez les veaux, c'est le VHB-1 qui était le moins séroprévalent. Une association était généralement observée entre les statuts sérologiques des veaux et ceux des mères et entre les titres en anticorps

des veaux et ceux des mères. La présence de problèmes respiratoires chez le veau était associée au profil sérologique de la vache et à celui du veau (VHB-1, VRSB et CVB dans le groupe non-vacciné, VHB-1, VDVB et CVB dans le groupe vacciné). Le développement d'un état diarrhéique par contre n'était pas associé aux profils sérologiques mais était négativement associé à un niveau élevé d'IgG chez le veau (odds ratio = 0,73). L'excrétion fécale de CVB et de RVB chez les veaux était respectivement de 1,4 % et 4,9 % chez les non-vaccinés et de 0 % et 9,9 % chez les vaccinés. La mise en évidence de RVB dans les fèces n'était pas associée à un état diarrhéique antérieur, mais était associée à la consistance diarrhéique des matières fécales au moment de leur prélèvement.

INTRODUCTION

Enteric and respiratory diseases are main health problems for northwestern Quebec beef calves (1). In their personal interview surveys, Larochelle *et al* (personal communication) and Couture and Major (1) reported a mortality rate of 9% between birth and weaning time, for calves seen alive around birth. More specifically, 80% to 89% of the reported mortalities occurred in the first month of life. According to the farmers, about 1/3 of these deaths were due to diarrhoea and respiratory diseases (1).

Bovine herpesvirus type 1 (BHV-1) and bovine respiratory syncytial virus

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(BRSV) are associated with respiratory diseases in young calves (2,3). Although the pathogenesis of bovine viral diarrhoea virus (BVDV) is complex, immunosuppression is an important component of BVDV pathogenicity (4,5). Bovine viral diarrhoea virus has been reported in association with other viral and/or bacterial agents to cause enteric or respiratory diseases in young calves (2,4,6,7). Bovine rotavirus (BRV) and bovine coronavirus (BCV) are causative agents of neonatal calf diarrhoea (8). In addition, BCV may also be responsible for respiratory problems in young calves (9). Thus, we decided to assess the importance of these viruses in northwestern Quebec cow-calf operations.

At birth, the newborn calf is almost agammaglobulinemic (10). Although immunologically competent, antibody production is low and it is unable to produce measurable immunological response to various infectious agents (11). Low antibody levels in calves predispose to infectious enteric or respiratory diseases in calves (3,12). Therefore, disease protection during the first weeks of life is dependent on the success of maternal antibody transfer. The half-life of maternal antibodies to BHV-1 and BVDV in the calf is about three weeks (13,14). Thus, at that age an agent specific association between calf and dam immunity should still be expected. The strength of that association is unknown, however.

The aims of this study were: 1) to describe cow and calf serological status for BHV-1, BVDV, BRSV, BCV and BRV and to test associations (odds ratio and linear correlation) between calf and cow serological profiles; 2) to study the association of cow and calf serological profiles with respiratory diseases and diarrhoea in three to four week old calves; 3) to determine the prevalence of BCV and BRV in calf feces and to test the association (odds ratio) with diarrhoea in three to four week old calves.

MATERIALS AND METHODS

SOURCE OF THE DATA

A random sample of 26 beef herds (25 cross-bred and one Simmental purebred) was selected from cow-calf herds with 25 or more breeding

females, which calved between January and May 1991, and located in western Abitibi. The participation rate was 71%. On each farm, from mid-November to December 1990, a systematic sampling of 50% of the assumed pregnant females was done and nonheparinized blood samples were collected. To limit the number of samples submitted to the laboratory, not more than 24 randomly selected dam sera per herd were used, except in herd 3 where 25 cows were sampled. The main unit of interest was the cow-calf pair, but some attention was given to the herd.

Data on cow vaccination schedule during 1989–1990 and 1990–1991 production periods, calf vaccination schedule, occurrence of enteric and respiratory diseases in calves during the first three to four weeks of life were recorded by the farmer. Additional information was collected by the senior author during fortnightly visits, from January to May 1991, at which time fecal samples and nonheparinized blood samples were collected from calves aged three to four weeks. Fecal consistency (normal vs diarrheic) was recorded.

The list of the agents against which the animals were vaccinated was validated by checking the vaccine packaging. If packaging was not available, a copy of the bill was obtained or the veterinarian was asked for the trademark of the vaccine. The list of the vaccine agents was obtained by consulting the compendium of veterinary products (15) at the given trademark.

Serum and fecal specimens were stored at -20° for a month and sent to the diagnostic laboratories where they were stored at -70° until analyzed, between March and July 1991.

ANTIBODY DETERMINATION

Serum antibody titers against BHV-1, BVDV, BRSV, BCV, and BRV were analyzed at the Institut Armand-Frappier (Laval-des-Rapides, Québec). Antibodies to BHV-1, BVDV, BRSV, and BRV were determined by the virus neutralization test (16). The presence of antibodies to BCV were determined by the hemagglutination inhibition test (17). Thresholds for seropositive samples were set at titers higher than 1/8 for BHV-1, BVDV, and BRSV, and 1/20 for BCV and BRV. Titers were expressed as loga-

rithm, base 2, of the reciprocal of the highest dilution with positive reaction. The serological profile, for a cow or a calf, was its serological status (positive or negative) to the given viruses.

To ascertain the reliability of antibody determination, we performed a quality control on serological tests. Twenty of the serum samples were each divided into three to five aliquots and tested independent of each other.

DETERMINATION OF SERUM IgG CONCENTRATION

Calf serum IgG concentration was determined by radial immunodiffusion using the Bovine Vet-Rid kit (Bethyl, Montgomery, Texas). Eight sera were each divided into four aliquots and tested independent of each other, in order to perform a quality control. A concentration less than 1000 mg/dL in a newborn calf, indicated hypogammaglobulinemia (18).

DETECTION OF VIRAL ANTIGENS IN FECAL SAMPLES

Feces from 375 calves were analyzed by a double sandwich indirect enzyme-linked immunosorbent assay (ELISA) test, to determine the presence of BCV and BRV type A (19). Equal volumes of feces and phosphate buffered saline (PBS pH 7.0) were mixed and incubated in duplicate in the antibody coated wells of microtiter plates. After one hour at room temperature, the microplates were washed three times with 200 μ L/well of PBS. Following incubation for one hour in the presence of specific rabbit anti-BCV or anti-BRV serum, the immune reaction was revealed by using HRP-conjugated goat antirabbit IgG serum and hydrogen peroxide and tetramethylbenzidine (TMB) as enzyme substrate and indicator. Optical density was determined at 450 nm using a Titertek Multiscan spectrophotometer (Flow Laboratories, McLean, Virginia). Optical densities classifying a sample as positive were ≥ 0.220 and ≥ 0.260 , for BCV and BRV, respectively.

DEFINITIONS

Vaccination-1 group was composed of cows which did not receive vaccine against any of the viruses within one year of blood sampling. Vaccination-2

TABLE I. Variance components due to the measurement error and sample total variance of serum IgG and antibodies to BHV-1, BVDV, BRSV, BCV, BRV in cows and calves in north-western Quebec beef operations

Serological analysis for	Variance component of the measurement error	Variance of the samples	
		in the cows	in the calves
BHV-1	0.533	2.543	1.28
BVDV	1.095	7.283	6.126
BRSV	1.866	3.293	2.727
BCV	2	3.174	4.409
BRV	0.716	1.78	1.516
IgG	15295.83	Not applicable	506303.4

group was cows vaccinated less than two weeks prior to blood sampling. Vaccination-3 group was the group of cows vaccinated between two weeks and less than one year prior to blood sampling.

STATISTICAL PROCEDURES

The quality control results were analyzed by the variance components estimation procedure (using the type I estimation method) (20). For each virus, the model for the estimation of the variance due to error measure was: $V = V_i + V_e$; where V was the total variance; V_i was the variance between sera; V_e was the variance due

to error. The results of the quality control are presented in Table I.

The statistical analyses were performed according to the vaccination status of the cow. Estimates of prevalences, by herd and by time interval between dam vaccination date and blood collection, were used to summarize the serological status of the cows and calves. Ninety-five percent confidence intervals (95% CI) were computed using the continuity corrected version of the score interval (21). The means of seroprevalence arcsine transformations in vaccination-2 and vaccination-3 were compared by one way analysis of variance

(22). The arcsine transformation was used in order to stabilize the variance as suggested for proportions (22). Because statistically significant difference was not observed between vaccination-2 and vaccination-3 groups, they have been combined in subsequent analyses (vaccination-4 group). An inference was claimed statistically significant when the probability to make a type 1 error was 0.05 or less.

The investigation of the cow-calf association was made in two steps: 1) by maximum likelihood logistic regression (23) using the serological status: $\ln[p_{ij}/(1 - p_{ij})] = a + b_i \cdot \text{herd}_i + c_j \cdot (\text{dam serological status})_j$; where p_{ij} = probability of a calf to be seropositive; 2) by linear correlation computation (22), to assess the associations between log-titers of seropositive cow-calf pairs.

Associations between serological profiles with previous occurrence of diarrhea or with previous occurrence of respiratory diseases were investigated by maximum likelihood logistic regression using a backward elimination procedure. The logistic model was:

TABLE II. Herd prevalences (%) of seropositive cows and calves to BHV-1, BVDV, BRSV and BCV in the northwestern Quebec beef cow-calf operations

No. ^a	Cows				Herd	No. ^a	Calves			
	BHV-1	BVDV	BRSV	BCV			BHV-1	BVDV	BRSV	BCV
23	30.43	4.35	56.52	91.30	1	28	21.43	0.00	42.86	92.86
24	33.33	4.17	29.17	79.17	2	19	31.58	5.26	42.10	100
25	30.77	0.00	4.00	84.62	3	20	10.00	0.00	5.00	70.00
24	33.33	75.00	45.83	95.83	4	9	22.22	55.56	55.56	88.89 ^d
24	41.67	87.50	37.50	83.33	5	10	20.00	90.00	50.00	100
24	75.00	100	12.50	95.83	6	10	30.00	90.00	10.00	80.00
19	21.05	15.79	0.00	100	7	15	20.00	20.00	0.00	86.67
24	53.85	65.39	70.83	100	8	23	34.78	43.48	26.09	91.30
20	60.00	90.00	35.00	90.00	9	18	27.78	77.78	22.22	83.33
18	27.78	50.00	16.67	100	10	11	27.27	36.36	18.18	100
19	26.32	47.37	26.32	100	11	16	18.75	43.75	31.25	100
12	33.33	8.33	8.33	91.67	12	6	50.00	0.00	0.00	83.33
17	5.88	100	5.88	94.12	13	14	21.43	85.71	28.57	92.86
15	26.67	86.67	80.00	100	14	5	0.00	100	80.00	100
23	39.13	56.52	52.17	100	15	15	20.00	60.00	33.33	86.67
24	58.33 ^b	95.83 ^b	50.00 ^b	79.17	16	30	36.67 ^b	66.67 ^b	33.33 ^b	90.00
15	86.67 ^b	93.33 ^b	73.33 ^b	100 ^b	17	5	40.00 ^b	80.00 ^b	40.00 ^b	80.00 ^b
24	75.00 ^b	87.50 ^b	66.67 ^b	87.50 ^b	18	25	60.00 ^b	88.00 ^b	60.00 ^b	80.00 ^b
23	41.67 ^b	50.00 ^b	56.52 ^b	83.33	19	22	22.73 ^b	81.82 ^b	40.91 ^b	68.18
13	23.08 ^b	15.39 ^b	7.69 ^b	92.31	20	10	30.00 ^b	20.00 ^b	10.00 ^b	90.00
24	95.83 ^c	91.67 ^c	75.00 ^c	95.83 ^c	21	16	25.00 ^c	50.00 ^c	25.00 ^c	100 ^c
24	79.17 ^c	83.33 ^c	33.33	95.83	22	17	29.41 ^c	64.71 ^c	35.29	100 ^d
23	88.00 ^c	84.00 ^c	56.52 ^c	100	23	29	34.48 ^c	62.07 ^c	58.62 ^c	100
23	43.48 ^c	69.57 ^c	86.96	65.22	24	22	31.82 ^c	77.27 ^c	63.64	90.91 ^d
12	75.00 ^c	83.33 ^c	58.33 ^c	100 ^c	25	7	42.86 ^c	57.14 ^c	42.86 ^c	100 ^c
11	72.73 ^c	54.55 ^c	90.91 ^c	90.91	26	5	60.00 ^c	20.00 ^c	80.00 ^c	100

^aNumber sampled

^bVaccination-2 group

^cVaccination-3 group

^dHerd where calves were vaccinated at birth, against BCV and BRV, with CALF-GUARD™ vaccine

TABLE III. Seroprevalences (%) to BHV-1, BVDV, BRSV, and BCV in cows and calves, by cow vaccination status, in the northwestern Quebec beef cow-calf operations

	Virus	Vaccination-1	Vaccination-2	Vaccination-3
Cows	BHV-1	37.4	58.6	75.7
	(%)	(32.0–43.0) ^a	(48.2–68.3)	(67.1–83.3)
	BVDV	52.7	72.7	80.0
	(%)	(47.0–58.4)	(62.7–81.0)	(71.8–86.9)
	BRSV	36.7	53.5	67.6
	(%)	(31.6–41.8)	(43.3–63.5)	(54.8–77.6)
	BCV	91.2	92.3	97.1
	(%)	(88.0–93.5)	(78.0–98.0)	(83.8–99.9)
Calves	BHV-1	23.7	39.1	33.3
	(%)	(18.4–30.0)	(29.3–49.9)	(24.2–43.8)
	BVDV	40.2	71.7	61.5
	(%)	(33.7–47.0)	(61.2–80.4)	(50.9–71.0)
	BRSV	31.9	40.2	49.1
	(%)	(26.2–37.9)	(30.3–51.0)	(35.8–62.6)
	BCV	89.4	80.0	100
	(%)	(85.4–92.6)	(60.9–91.6)	(82.2–100)

^a () 95% confidence interval

TABLE IV. Odds ratio of logistic regression of calf serological status on cow serological status for BHV-1, BVDV, BRSV and BCV, controlling for vaccination status, in northwestern Quebec cow-calf operations

Virus	Vaccination-1			Vaccination-4		
	No. ^a	Odds ratio	95% CI ^b	No. ^a	Odds ratio	95% CI ^b
BHV-1	187	2.25	1.59–3.19	140	1.19	0.83–1.7
BVDV	187	2.64	1.48–4.69	140	1.81	1.19–2.76
BRSV	216	1.91	1.35–2.69	110	1.54	1.03–2.3
BCV	255	3.15	2.12–4.67	46	8.3	2.98–23.08

^aNumber of cow-calf pairs used for the analysis

^b95% confidence interval for the odds ratio

$$\ln[p_{ijklm}/(1 - p_{ijklm})] = a + b_i \text{herd}_i + c_j * S_{bhv-1(j)} + c_k * S_{bvdv(k)} + c_l * S_{brsv(l)} + c_m * S_{bcv(m)}$$

where p_{ijklm} = probability of a calf to have developed diarrhea (or respiratory disease); $S_{bhv-1(j)}$ was the serological status to BHV-1. Variable removing criteria was set at a p value > 0.10. Models were built separately for cows and calves because the number of records for cow-calf pairs (327 for vaccination-1 and vaccination-4 groups) was not sufficient to put all cows and calves variables ($n = 8$) in the same model. For this analysis some cow-calf pairs in the vaccination-4 group were not specifically vaccinated against BRSV and/or BCV.

To account for herd effect the variable herd was forced into all the logistic models as fixed effect. The assessment of the fit of the logistic models was performed by analysis of the residuals by checking whether lack of adjustment did not occur in more than 5% of the cells.

Results of BCV and BRV antigen detection studies are presented in rel-

ative frequencies. Association between viral antigen shedding and diarrhea was assessed by crude odds ratio.

Confidence intervals around the odds ratio were computed using estimated asymptotic standard errors (23). All the analyses were performed using the Statistical Analysis System (20).

RESULTS

SEROLOGICAL PROFILES AND COW-HALF ASSOCIATIONS

Results from 527 cows, 407 calves, and 327 cow-calf pairs were obtained. Large variations of herd prevalence of seropositive cows to BVDV were observed among herds of vaccination-1 group and among herds of vaccination-2 group (Table II). Large variation of herd seroprevalence to BVDV was also observed in calves among herds of vaccination-1 group (Table II). For BHV-1, BRSV, and the other cases of BVDV the variations of herd seroprevalences were moderate,

TABLE V. Pearson correlation coefficients between seropositive calves log-titer to BHV-1, BVDV, BRSV, and BCV and those of seropositive cows in the northwestern Quebec beef cow-calf operations

Virus	Vaccination-1	Vaccination-4
BHV-1	0.317	0.323
	$p = 0.056^a$ (37) ^b	$p = 0.042$ (40)
BVDV	0.446	0.623
	$p < 0.001$ (85)	$p < 0.001$ (82)
BRSV	-0.187	0.403
	$p = 0.192$ (50)	$p = 0.016$ (35)
BCV	0.272	0.384
	$p < 0.001$ (211)	$p = 0.003$ (59)

^a p value

^b() number of cow-calf pairs used for the analysis

in cows and calves. Less variation was noted for herd seroprevalences to BCV and BRV in vaccination-1 group. Vaccines against BCV and/or BRV were used in few herds, therefore variation of herd seroprevalence was not estimated for vaccination-2 and vaccination-3 groups. Because almost all the animals were seropositive to BRV, seroprevalences to BRV are not used in subsequent analyses.

Seroprevalences according to cow vaccination status, regardless of the herd, are summarized in Table III. Cows seroprevalences for all viruses were lower in vaccination-1 group and were higher in vaccination-3 group. In calves, except for BCV, seroprevalences in vaccination-2 and vaccination-3 groups were higher than the one in vaccination-1 group (Table III). No difference between means of arcsine transformations of herd seroprevalences to each virus was observed between vaccination-2 and vaccination-3 groups ($p > 0.05$). Thereafter, vaccination-2 and vaccination-3 have been combined to form vaccination-4 for subsequent analyses.

For BHV-1, in the vaccination-1 group, calves born from seropositive dams were 2.25 times more likely to be seropositive than calves born from seronegative dams (Table IV) but there was no linear correlation between the log-titers of seropositive cow-calf pairs (Tables V). In the vaccination-4 group, on the other hand, calves born from seropositive cows were at similar probability to be seropositive as calves born from

TABLE VI. Associations between calf clinical respiratory diseases with cow and calf serological profiles to BHV-1, BVDV, BRSV, and BCV in the northwestern Quebec cow-calf operations (odds ratio and 95% CI)

	Vaccination-1				Vaccination-4			
	No. ^a	Virus	Odds ratio	95% CI ^b	No. ^a	Virus	Odds ratio	95% CI ^b
Dams	239	BHV-1	1.26	0.96–1.656 ^c	190	BHV-1	0.57	0.42–0.78
		BRSV	1.46	1.09–1.95		BVDV	0.62	0.43–0.88
		BCV	0.55	0.34–0.89		BCV	0.44	0.28–0.67
Calves	217	BHV-1	1.83	1.34–2.51	188	BHV-1	1.74	1.28–2.37
		BRSV	1.44	1.05–1.97		BVDV	0.64	0.47–0.87
		BCV	0.44	0.28–0.71		BCV	0.56	0.34–0.91

^aNumber of animals used to assess the model

^b95% confidence interval for the odds ratio

^cConfidence interval for BHV-1 in the model of vaccination-1 cows include 1 because the p value was 0.093; BHV-1 was kept in the model because its exclusion from the model led to lack of adjustment in a cell containing 13 observations

seronegative dams (Table IV). There was a linear correlation between log-titers of seropositive cows and seropositive calves (Table V).

For BVDV, calves of seropositive dams were 2.64 times (in the vaccination-1 group) and 1.81 times (in the vaccination-4 group) more likely to be seropositive than calves of seronegative dams (Table IV). Log-titers of seropositive cow-calf pairs were linearly correlated (Table V). The highest correlation coefficients in vaccination-1 and vaccination-4 groups were observed with BVDV (Table V).

For BRSV, calves born from seropositive dams were 1.91 times (in the vaccination-1 group) and 1.54 times (in the vaccination-4 group) more likely to have seropositive status than calves born from seronegative dams (Table IV). In the vaccination-1 group log-titers of seropositive cows were not correlated to that of seropositive calves but in the vaccination-4 group they were correlated (Table V).

For BCV, in the vaccination-1 group, calves of seropositive dams were 3.15 times more likely to be seropositive than calves of seronegative dams. In the vaccination-4 group, calves of seropositive cows were 8.30 times more likely to be seropositive than calves of seronegative dams (Table IV). In the vaccination-1 and vaccination-4 groups, log-titers of seropositive calves were correlated to log-titers of seropositive dams (Table V).

RESPIRATORY DISEASES AND DIARRHEA

The prevalences of diarrhea and respiratory diseases (recorded by the farmer), and diarrheic feces at

sampling (observed by the senior author), were 38.38%, 13.43%, 11.22% in the vaccination-1 and 36.65%, 6.37%, 10.59% in the vaccination-4 groups, respectively. During the first week of life, 61% and 50% of diarrhea and respiratory diseases occurred, respectively.

In the vaccination-1 group, the presence of antibodies to BHV-1 and BRSV in the dam was associated with an increased risk of respiratory diseases in the calf but the presence of antibodies to BCV was associated with a decreased risk (Table VI). In the model with calf antibodies, the occurrence of respiratory disease was accompanied by a seropositive status to BHV-1 and BRSV but by a seronegative status to BCV ($p < 0.001$, $p = 0.023$ and $p < 0.001$, respectively) (Table VI). In the vaccination-4 group, the model with dam serology indicated that the presence of antibodies to BHV-1, BVDV, and BCV was associated with a decreased risk of respiratory disease in the calf (Table VI). Based on calf serology, the occurrence of respiratory disease was associated with seropositive status to BHV-1 but with seronegative status to BVDV and BCV (Table VI).

In the vaccination-1 and vaccination-4 groups, no association was found between cow and calf serological profiles and previous occurrence of diarrhea. In absence of an association with specific antibodies, diarrhea was regressed on calf serum IgG level (in vaccination-1 and vaccination-4 groups). In the vaccination-1 group, the presence of high levels of IgG (>1000 mg/dL) was associated with decreased risk of previous occurrence of diarrhea in calves (odds ratio = 0.73 with 95% CI of (0.53, 0.99)). In the vaccination-4 group, no associa-

tion was observed between calf serum IgG level and previous occurrence of diarrhea.

VIRUS SHEDDING

Bovine rotavirus and BCV antigens were shed by 4.9% and 1.4% ($n = 284$) of calves from vaccination-1 herds and by 9.9% and 0% ($n = 91$) of calves from vaccination-4 ones, although BRV and BCV antibodies were found in almost all the calves. No calf was a joint shedder of BRV and BCV. The presence of BRV in the feces was not associated with previous occurrence of diarrhea. However, it was associated with the diarrheic consistency of the feces; the odds ratio (95% CI) was 4.8 (1.5, 15.3) in the vaccination-1 group and 7.7 (1.5, 40.3) in the vaccination-4 group.

DISCUSSION

SEROLOGICAL PROFILES AND COW-CALF ASSOCIATIONS

The large variations in cows and calves seroprevalences to BVDV within the vaccination-1 group might be due to differences in the degree and/or time of exposure to the virus. On the other hand, the lower variations observed for BCV and BRV and their high seroprevalences was perhaps an indication of a great level of exposure of the herds to these two viruses. The magnitude of seroprevalences estimates of the present study (Table III) are not unusual relative to other studies in Quebec (7,24).

The magnitude of the linear correlation between seropositive cow-calf pairs antibody log-titers varied from weak to moderate (Table V), except for BHV-1 where no correlation existed. The measurement error in

antibody determination (Table I) contributed probably to erase or to reduce the correlation strength (25). However, other factors such as failure of maternal immunoglobulin transfer, passive antibody decay in the calves, increase of antibody titers in cows between blood sampling and calving, or active antibody formation in the calves before they were bled might have contributed also to that lack of meaningful association.

In the vaccination-4 group, for BVDV, the presence of a moderate correlation coefficient ($r = 0.623$), despite the fact that about 46% of the cows sampled had just been vaccinated, indicated that an important increase of BVDV antibody level did not occur in these cows between cow and calf blood sampling. Such observations could be explained by the long persistence (years) of BVDV antibodies (5).

RESPIRATORY DISEASES AND DIARRHEA

In the dams' models (Table VI), the negative association between cow serological status to BHV-1 in vaccination-4 group and previous occurrence of respiratory diseases in calves, whereas positive in vaccination-1 group, seemed to indicate that the risk associated with a potentially infected dam decreased following vaccination. Indeed, it is well-established that infection by BHV-1 persists by a phenomenon of latency and that virus reexcretion can be prevented by a high level of specific immunity induced by vaccination (26). This result, and the finding that antibodies to BVDV in the cow or in the calf were negatively associated with respiratory disease status in the vaccination-4 group, suggested a protective effect of dam vaccination. With regard to the associations between calf serological status to BHV-1 and BRSV and the previous occurrence of respiratory diseases, these agents could be potentially responsible for calf respiratory diseases. However, due to the nature of the study, an evidence of a causal relationship cannot be assessed.

Seropositive status to BCV in the cow or calf was associated with a decreased risk for the calf to develop respiratory disease (Table VI).

According to the literature, BCV seemed to play a secondary role for respiratory disease occurrence since only a few cases of respiratory diseases in calves have been attributed to coronavirus (9); however, it has been frequently isolated from the calf respiratory tract (27,28). Therefore, that negative association was not interpreted as a protective effect of the presence of antibodies against BCV. It was speculated that a debilitating factor, responsible for the absence of seroreaction to BCV, could be suspected to be a potential risk factor associated with the occurrence of respiratory diseases.

Considering the high prevalence of seropositive animals, most of them should have been in contact with the agent. It is therefore possible that this lack of seroreaction in some calves could be an indicator of their incapacity to respond immunologically as efficaciously as other calves, due to other causes (i.e. nutrition). Griebel *et al* (29) found that the immunological response to K99 pili antigen was slower in protein/energy malnutrition calves.

For diarrhea, the lack of association found with cow and calf serological status to BCV might be indicative of noninvolvement of BCV in the occurrence of diarrhea. In vaccination-1 group, we found an association between calf serum IgG and previous occurrence of diarrhea (95% CI of (0.53, 0.99)). However, considering the weakness of that association, a clear evidence of a possible role of low level of immunity in the occurrence of diarrhea was not shown. An active immunological response to BCV in calves with failure of colostrum antibody transfer (30,31) and/or a greater BCV challenge, which overcame the passive antibody protection, were other reasons that could be put forward to explain the lack of association between cow-calf serological profiles and previous occurrence of diarrhea in calves. However, with regard to the higher seroprevalences to BCV and the age of the calf at blood sampling, i.e. after the occurrence of diarrhea, the presence of an active immunological response seemed to be the most plausible reason. Thus, if antibodies to BCV were measured under one week of age an

association with diarrhea might have occurred. In addition, an evaluation of the enteric local immunity, rather than the systemic immunity, might have been more informative.

Establishing a threshold for IgG concentration in order to define an hypogammaglobulinemic calf of three weeks of age raised concern. The IgG of maternal origin, in a three week old calf, was expected to be half of what it received during its first 48 hours of life (13,14). However, at three weeks the antibodies are not necessarily of maternal origin (30,31). Only 4.18% of the calves had serum IgG concentration less or equal to 500 mg/dL; theoretically should be the IgG concentration normally present in three week old calves if they had 1000 mg/dL at 48 hours of age (13,14,31). Prevalence of hypogammaglobulinemic calves reported by other authors were higher or equal to 20% for the winter season (32,33).

VIRUS SHEDDING

The prevalence of BCV fecal antigens were about the same order of magnitude as that reported in nondiarrheic calves in previous studies (8,28). It was lower than prevalences reported in diarrheic calves by Snodgrass *et al* (8) and Reynolds *et al* (28). Calf age reported by Snodgrass *et al* (8) ranged from 1 to 28 days but in the study of Reynolds *et al* (28) age was not specified. The age of the viral shedders reported in experimental infections was under two weeks (27,28) whereas Heckert *et al* (9) and Langpap *et al* (34), reported BCV shedding in older calves. However, Langpap *et al* (34) studying coronaviral enteritis in one day to three month old calves found much more BCV shedders in one to seven day old calves. Therefore, if fecal samples were taken within one week of age, where 61% of the diarrhea occurred, prevalence of BCV antigen shedders could probably be higher.

The prevalence of BRV antigen shedding in vaccination-1 and vaccination-4 groups was lower than the ones reported by Lucchelli *et al* (35) and even in nondiarrheic calves by Snodgrass *et al* (8). However, the mean age of the shedders reported in these studies was under two weeks. It is possible that the proportion of the

detected viral antigen shedders would have been higher if feces had been sampled around one week of age, when most enteric problems occurred. The fact that the presence of BRV antigen in the stools was associated with the diarrheic consistency of the feces supported this assumption. Another possible reason for low presence of BRV in feces was that the ELISA test used in this study detected only type A rotaviruses despite the fact that other BRV types (i.e. group B) are known to infect calves (36,37).

The fact that BRV shedding seemed to be more frequent in the vaccination-4 group than in vaccination-1 raised the following questions: Did farmers use vaccines because they had problems? or did they neglect sanitary management because they vaccinated? or finally, were vaccines or vaccination management ineffective in controlling BRV, since marketed vaccines are against type A BRV, only?

In conclusion, seroprevalences to BHV-1, BVDV and BRSV varied between herds and indicated a high presence of BCV and BRV in the northwestern Quebec beef cows and calves. Calf serological status and calf antibody titers were associated with those of the dam. The occurrence of respiratory diseases in the calf was associated with cow-calf serological profiles. Such associations were not observed with diarrhea. Prevalences of viral antigen shedding were very low at three to four weeks of age, but may have been higher if samples were taken one to two weeks sooner. The investigation of these cow-calf operations indicated the existence of a complex set of interactions between cows, calves, and infectious agents.

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