

Supplementary Material

Table S1. Results of all univariable analyses. The outcome measure used in ordinal logistic regression was the age group in which BLV was first identified. For variables that met the proportional-odds assumption, the P-value displayed is the P-value for each age group comparison.

Variable	Description	Odds Ratio	95% CI	P-value
Colostrum source fed to calves*	Fresh colostrum			
	Frozen, pasteurized, or commercial colostrum	1.064	0.303, 3.735	0.923
Amount of colostrum fed (L)		0.881	0.685, 1.132	0.321
Timing of first colostrum feeding	<6 hours after birth			
	>6 hours after birth	1.727	0.579, 5.151	0.327
Timing of second colostrum feeding	<12 hours after birth			
	>12 hours after birth	1.282	0.469, 3.505	0.629
Milk type fed to calves	Milk replacer			
	Milk from farm (any type)	0.723	0.265, 1.970	0.526
Treatment of milk fed to calves	Treated (pasteurized or acidified)			
	Not treated	3.299	0.845, 12.880	0.086
How milk is fed to calves	Individual bottle/bucket per calf			
	Communal bottles/buckets/feeders	0.592	0.172, 2.033	0.405
Is feeding equipment cleaned	After every feeding			
	Daily	2.855	0.684, 11.923	0.150
	Less often	1.726	0.505, 5.892	0.384
	Overall P-value			0.304
Is feeding equipment disinfected	At least every 2 days			
	Weekly	1.045	0.250, 4.361	0.952

	Never	1.040	0.260, 4.170	0.955
	Overall P-value			0.998
Type of calf housing	Individual pens or hutches			
	Group pens or hutches	2.051	0.705, 5.965	0.187
Number of calves per pen/hutch		1.126	0.926, 1.369	0.235
Are calves grouped before weaning	No			
	Yes	1.239	0.462, 3.326	0.670
Calf housing related to adults	>200m away			
	<200m away	1.115	0.184, 6.743	0.906
	In same building	0.669	0.103, 4.328	0.673
	Overall P-value			0.633
Number of people caring for calves		0.997	0.662, 1.500	0.987
Order that age groups are handled	Calves first			
	Heifers/adults first	0.717	0.259, 1.990	0.523
Needles reused, pre-weaned calves	No			
	Yes	0.750	0.239, 2.350	0.622
Syringes reused, pre-weaned calves	No			
	Yes	1.939	0.471, 7.982	0.359
Tube feeding performed, pre-weaned calves	No			
	Yes	0.842	0.209, 3.389	0.809
Minor surgery performed in pre-weaned calves	No			
	Yes	0.850	0.249, 2.898	0.795
Farm staff give IV injections, pre-weaned calves	No			
	Yes	1.533	0.539, 4.360	0.423
Fly control in pre-weaned calves	None			
	Environmental or topical	3.327	0.935, 11.841	0.063
	Environmental and topical	5.287	0.654, 42.711	0.118
	Overall P-value			0.133
Feed equipment	No			

shared with weaned heifers and older animals				
	Yes	1.522	0.550, 4.214	0.419
Group size for young weaned heifers		0.984	0.859, 1.128	0.820
Group size for older weaned heifers		1.006	0.962, 1.052	0.791
Age range within group of young weaned heifers		0.878	0.713, 1.082	0.223
Age range within group of older weaned heifers		1.006	0.800, 1.265	0.959
Weaned heifer housing related to adults	>200m away			
	<200m away	1.813	0.454, 7.249	0.400
	Same building	1.715	0.399, 7.374	0.468
	Overall P-value			0.670
Number of times weaned heifers are regrouped	Not regrouped			
	Regrouped once	2.791	0.502, 15.529	0.241
	Regrouped twice	9.644	1.791, 51.931	0.008
	Regrouped three times	2.146	0.384, 12.001	0.384
	Overall P-value			0.033
Weaned heifer access to pasture	Pastured >200m from adults			
	Pastured <200m from adults	0.785	0.181, 3.407	0.747
Hot iron dehorner use in weaned heifers	No			
	Yes	1.256	0.444, 3.556	0.667
Needles reused, weaned heifers	No			
	Yes	0.835	0.261, 2.676	0.762
Syringes reused, weaned heifers	No			
	Yes	1.517	0.445, 5.170	0.506
Foot trimming performed in weaned heifers	No			
	Yes	0.731	0.207, 2.589	0.628
Minor surgery performed in weaned heifers	No			
	Yes	1.012	0.344, 2.978	0.983
Fly control in	None			

weaned heifers	Environmental or topical	3.431	1.207, 9.760	0.021
	Environmental and topical	5.286	0.386, 72.452	0.213
	Overall p-value			0.051
Feed equipment shared with breeding-age heifers and adult cows	No			
	Yes	1.243	0.455, 3.396	0.671
Water source for breeding-age heifers	Only for breeding-age heifers			
	Shared with adult cows	2.452	0.505, 11.918	0.266
Group size for breeding-age heifers		1.003	0.972, 1.036	0.844
Age range within group of breeding-age heifers		0.980	0.843, 1.140	0.796
Age when heifers enter the breeding group		0.809	0.590, 1.110	0.190
Breeding-age heifer housing related to adults	>200m away			
	<200m away	1.420	0.372, 5.416	0.608
	Same building	3.181	0.836, 12.105	0.090
	Overall P-value			0.177
When are heifers moved into adult herd	At close-up			
	>3 months prior to calving	0.878	0.293, 2.634	0.816
Breeding-age heifer access to pasture	Pastured >200m from adults			
	Pastured <200m from adults	1.137	0.425, 3.042	0.798
Ear tagger use, breeding-age heifers	No			
	Yes	0.529	0.075, 3.707	0.521
Needles reused, breeding-age heifers	No			
	Yes	0.672	0.214, 2.110	0.496
Syringes reused, breeding-age heifers	No			
	Yes	1.681	0.455, 6.208	0.436
Foot trimming	No			

performed in breeding-age heifers**	Yes	2.753	0.808, 9.381	0.105
	No			
Minor surgery performed in breeding-age heifers	Yes	1.445	0.408, 5.118	0.569
	No			
Fly control in breeding-age heifers	None			
	Environmental or topical	4.645	1.1493, 14.454	0.008
	Environmental and topical	4.439	0.660, 29.877	0.125
	Overall P-value			0.019
Farm buys replacement animals	Yes	1.464	0.544, 3.945	0.451
	No			
Farm buys animals with unknown BLV status	Yes	1.135	0.407, 3.166	0.809
	No			
Farm buys from sale barns	Yes	1.415	0.274, 7.319	0.679
	No			
Farm buys open heifers	Yes	1.415	0.274, 7.319	0.679
	No			
Farm buys bred heifers	Yes	3.169	0.810, 12.403	0.098
	No			
Farm buys first-lactation cows	Yes	1.373	0.444, 4.243	0.582
	No			
Farm buys mature cows	Yes	2.142	0.680, 6.749	0.193
	No			
Cows leave the farm and return	Yes	0.855	0.319, 2.297	0.757
	No			
Fenceline contact with other herds	Yes	0.079	0.008, 0.741	0.026
	No			
Any other method of contact with other herds	Yes	0.315	0.080, 1.239	0.098
	No			

* Herds using fresh pooled colostrum: 44; herds using fresh colostrum from a single cow: 1; herds using frozen colostrum from a single cow: 6; herds using commercial colostrum: 2.

** For the variable that did not meet the proportional-odds assumption, the estimate and P-value displayed is the one for the relevant age group comparison – odds of BLV infection being first identified in breeding-age heifers versus BLV infection being first identified in the milking herd.

Appendix 1

QUESTIONNAIRE ADMINISTERED FOR DATA COLLECTION

A copy of the questionnaire administered to producers for data collection follows on the next pages. Due to margin constraints, the questionnaire has been modified slightly to fit.

BLV Temporal Patterns of Infection – Atlantic Healthy Herds

General Information Survey

Complete the following information for ALL animals of each age group, not just the individuals being sampled.

Farm Name: _____

Owner Name: _____

Veterinarian: _____

Date of sample collection (D/M/Y): _____

Dairy Board #: _____

Barn Type: _____

Total number of pre-weaned heifer calves: _____

Total number of weaned heifers, pre-breeding age: _____

Total number of breeding-age heifers: _____

Total number of mature cows, dry: _____

Total number of mature cows, lactating: _____

Part 1: Pre-weaned heifer calves

Complete the questions below taking into account ALL pre-weaned heifer calves, not just those sampled. Answer as accurately as possible, providing supporting comments as needed.

Colostrum management

APPROXIMATELY what percent of heifer calves receive the following types of colostrum?

	Fresh pooled colostrum
	Fresh colostrum from one cow
	Frozen pooled colostrum
	Frozen colostrum from one cow
	Commercial colostrum
	Pasteurised colostrum

List any other type(s) of colostrum also fed to heifer calves: _____

How much colostrum does each heifer calf receive (litres or quarts)? _____

What percent of heifer calves receive colostrum within the following time periods?

	First feeding	Second feeding
Within 2 hours		
Within 6 hours		
Within 12 hours		
Within 24 hours		

Comments, colostrum management:

Part 1: Pre-weaned heifer calves

Feeding

What percent of the time do heifer calves receive the following types of milk?

	Waste milk
	Surplus bulk tank milk
	Milk from individual cow (no pooling)
	Commercial milk replacer

Is milk treated prior to feeding it to calves?

	No treatment performed
	Milk is pasteurised
	Milk is acidified
	Milk is fermented

How is milk fed to calves?

	Bottle or bucket; dedicated bottle/bucket for each calf
	Bottle or bucket; communal equipment
	Free choice, individual nipple/feeder for each calf
	Free choice, multiple calves use the same nipple/feeder

How often are bottles/buckets/equipment cleaned and/or disinfected?

C	D	
		After every feeding
		Daily
		Every other day
		Other; please specify

Part 1: Pre-weaned heifer calves

Housing

What percentage of calves are housed in the following housing types?

	Individual calf hutches
	Group calf hutches; # of calves per hutch:
	Individual calf pens
	Group calf pens; average # of calves per pen:

Are calves moved to group housing BEFORE weaning? _____

If yes, how old are calves when they are grouped? _____

Where is the calf housing in relation to the adult cows?

	In the same building
	In a separate building/calf hutches, <200 meters from adult cows
	In a separate building/calf hutches, >200 meters from adult cows
	At a different location; please specify:

How many people are caring for (feeding, handling, cleaning) the preweaned heifer calves?

Do the people caring for the calves also care for the adult milking herd? _____

If yes, in what order are the different age groups handled? _____

Are there separate boots/coveralls used when caring for the calves? _____

Comments, milk feeding and housing:

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Part 2: Weaned heifer calves

Complete the questions below taking into account ALL pre-weaned heifer calves, not just those sampled. Answer as accurately as possible, providing supporting comments as needed.

Feeding

Describe the typical diet for weaned heifer calves:

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Are calves fed using the same equipment (mixing bins, mixing utensils, buckets) as adult cows?

Which of the following BEST describes the water source for the calves?

	Buckets or troughs used ONLY by weaned calves
	Buckets or troughs used by weaned calves and older heifers
	Buckets or troughs used by weaned calves and adult cows
	Other; please specify:

Comments, feeding:

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Part 2: Weaned heifer calves

Housing

What is the AVERAGE group size for weaned heifers? _____

What is the AVERAGE age range within one group of weaned heifers? _____

Where is the weaned calf housing in relation to adult cows?

	In the same building
	In a separate building, <200 meters from adult cows
	In a separate building, >200 meters from adult cows
	At a different location; please specify:

For the AVERAGE calf, how often is she re-grouped after weaning? Do NOT include the initial movement into a group from individual housing, or movement into the group of breeding-age heifers.

	>3 movements
	2-3 movements
	1 movement
	Not re-grouped after weaning

What BEST describes pasturing for weaned heifer calves?

	Pastured in the same field as adult cows
	Pastured separately with <200 meter separation from adult cows
	Pastured separately with >200 meter separation from adult cows
	Pastured at a different location
	Not pastured

Comments, housing:

Part 3: Breeding-age heifers

Complete the questions below taking into account ALL breeding-age heifers, not just those sampled. Answer questions based on heifer management BEFORE they join the adult herd, unless otherwise stated. Answer as accurately as possible, providing supporting comments as needed.

Feeding

Describe the typical diet for breeding-age heifers:

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Are heifers fed using the same equipment (mixing bins, mixing utensils, buckets) as adult cows?

Which of the following BEST describes the water source for the heifers?

	Buckets or troughs used ONLY by breeding-age heifers
	Buckets or troughs used by heifers and adult cows
	Other; please specify:

Comments, feeding:

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Part 3: Breeding-age heifers

Housing

What is the AVERAGE group size for breeding-age heifers? _____

What is the AVERAGE age range within one group of breeding-age heifers? _____

What is the AVERAGE age when a heifer enters the breeding group? _____

Where are the heifers housed in relation to adult cows?

	In the same building
	In a separate building, <200 meters from adult cows
	In a separate building, >200 meters from adult cows
	At a different location; please specify:

When are heifers moved into the main adult herd?

	During the close-up period/at calving
	Within 3 months of calving
	Within 6 months of calving
	At the time of successful breeding

What type of grouping is used for breeding-age heifers?

	All in-all out
	Continuous movement (when heifers are confirmed pregnant they are moved into the adult herd and replaced with younger heifers)
	Other; specify

How often are heifers moved into/out of the breeding group? _____

If heifers are pastured, choose the BEST description below.

	Pastured in the same field as adult cows
	Pastured separately with <200 meter separation from adult cows
	Pastured separately with >200 meter separation from adult cows
	Pastured at a different location
	Not pastured

Comments, housing:

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Part 3: Breeding-age heifers

Equipment use and fly control

Check ALL of the following equipment that is used on breeding-age heifers:

	Gouge or spoon-type dehorers
	Hot iron or chemical dehorers
	Ear taggers; specify if self-piercing (i.e. tag has sharp metal part) or not
	Reused needles
	Reused syringes
	Foot trimming equipment
	Equipment for minor surgery (e.g. extra teat removal, lancing abscesses, ear notchers)
	Reproductive equipment and palpation (e.g. rectal sleeves, ultrasound, AI equipment)

What type(s) of fly control are used for breeding-age heifers?

	Environmental – fly paper, fumigation, sprays/powders – used in the heifer environment
	Topical sprays/powders used specifically for heifers
	Both environmental and topical used specifically for heifers
	No fly control specifically for heifers

Comments, equipment and fly control:

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Part 4: Cow movement

Complete the questions below using information from the last TWO years. Answer as accurately as possible, providing supporting comments as needed.

Do you buy replacement animals? _____

What source(s) do you use when purchasing animals?

	Herds with KNOWN negative status for BLV
	Herds with KNOWN positive status for BLV
	Herds with UNKNOWN BLV status
	Sale barns/auctions
	Custom rearing operations
	Other; please specify:

Indicate ALL ages of cows that are purchased

	Pre-weaned calves
	Weaned calves
	Open heifers
	Bred heifers
	First-lactation cows
	Mature cows

During show season, how many instances are there of animals leaving the farm?

	Often (>8)
	Sometimes (4-8)
	Rare (<3)
	Never

Comments, cow movement:

Animal Sampling

Select 6 animals AT RANDOM from the following three age groups. If there are 6 or fewer animals in a specific age group, sample all of the animals available. Write the tag/ID number of the animal on the sheet as well as on the blood tube. Also include the tag/ID number of the mother.

Pre-weaned calves

Age group	Tag/ID number	Dam tag/ID number
Pre-weaned heifer calves		
Weaned heifer calves		
Bred heifers		