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A preliminary study of the production and financial impacts of clinical salmonellosis outbreaks in lactating cows in seasonal calving, pasture-based dairy herds in New Zealand, from which *Salmonella enterica* subsp. *enterica* were isolated

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ABSTRACT

Aims: To quantify the production effects of outbreaks of clinical salmonellosis in seasonal calving, pasture-based dairy herds in New Zealand, from which non-Brandenburg *Salmonella enterica* subsp. *enterica* were isolated, and to use these results to undertake a financial analysis of their consequences.

Methods: In this retrospective cohort study, we collated routinely available data for cow demographics, disease treatment, and accumulated whole of lactation milk solids (MS) yield from five spring-calving, pasture-based dairy herds that experienced naturally occurring clinical salmonellosis outbreaks in milking cows over the study period from 1 June 2021 until 31 May 2022. We used multivariable generalised linear models to estimate the effects of cow-level risk factors for clinical salmonellosis and the effects of disease on accumulated whole of lactation MS yield and risk of removal by culling or death. The financial consequences of salmonellosis were estimated with a partial budget model using the study results and publicly available information.

Results: The median herd incidence risk of clinical salmonellosis over the study period was 2.6% (min 0.5, max 7.0%). The adjusted marginal mean incidence risk for disease for cows of age 2, 3–4, 5–6 and ≥ 7 years of age were 0.93% (95% CI = 0.39–2.18%), 0.98% (95% CI = 0.49–1.93%), 3.5% (95% CI = 2.15–5.63%) and 4.8% (95% CI = 3.06–7.40%), respectively. In three of the four herds with milk production records, there was no measurable effect of disease on MS yield and in a single herd, the adjusted marginal mean effect was a reduction in affected cows of 150 (95% CI = 98–202) kg ($p < 0.001$) accumulated MS yield per cow (36%). In a study of median-sized hypothetical herds, under six different scenarios differing in the level of impact of salmonellosis on MS yield and with differing incidence risks of disease, we estimated a total herd-level increase in expenditure and decrease in income for the season in which the outbreak occurred of NZ\$1,873–NZ\$13,444.

Conclusions: This preliminary study suggests that the financial consequences of clinical salmonellosis in New Zealand dairy herds are highly variable and are driven mainly by the incidence risk of disease and its effect on milk production.

Clinical relevance: Our results provide a preliminary guide to the range of biological and financial impacts of salmonellosis outbreaks in New Zealand dairy herds, which can aid in the planning of future research and inform farmer decision-making on control measures.

Abbreviations: DAG: Directed acyclic graphs; MS: Milk solids; PSC: Planned start of calving.

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Introduction

Salmonella enterica subspecies *enterica* are Gram-negative bacteria of the family Enterobacteriaceae, of which more than 2,600 serovars are described worldwide (Jajere 2019), and many of which can cause disease in both animals and humans. In New Zealand, clinical salmonellosis caused by this subspecies (hereafter referred to as “salmonellosis”) is a sporadic, mainly enteric disease among pre-weaned calves and adult dairy cattle that adversely

affects their welfare, health and productivity, particularly in the early to middle period of the herd lactation (Teague 2011). Animal reservoirs of *Salmonella* spp. acting as sources for human infection in New Zealand (Baker *et al.* 2007) are a concern for veterinary and public health researchers, as is the risk of the development or spread of existing antimicrobial resistant strains within animals, including cattle, or from animals to humans (Broughton *et al.* 2010).

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A financial evaluation of control measures against a disease requires data on the biological impacts of the disease in a relevant population and its financial consequences. Teague (2011) provided herd-level estimates of costs and losses of serovar Typhimurium outbreaks using historic production records as control data, but no reports could be located by the authors on either salmonellosis associated with non-Dublin *Salmonella* serovars in milking cows or production and financial impacts measured at the cow level. Our main research aim was to describe the biological and financial impact of naturally occurring salmonellosis outbreaks in milking cows from a convenience sample of pasture-based spring-calving New Zealand dairy herds in the dairy season commencing in 2021. Our secondary aim was to investigate the associations between the occurrence of clinical salmonellosis and animal-level factors such as age, breed and when the individual cow calved relative to the planned start of the calving (PSC) for the herd, measured as the interval between the cow's calving date and the PSC. The objective of our research was to provide dairy farmers and their veterinary advisers with information that would inform programmes to control salmonellosis outbreaks.

Materials and methods

Study design

This was a retrospective cohort study among New Zealand dairy farms diagnosed with naturally occurring outbreaks of salmonellosis and was approved by the Massey University Human Ethics Committee (Ethics Application SOA 21/67, Palmerston North, NZ). Surveillance by the Ministry for Primary Industries (Wellington, NZ) had identified an increase in the number of laboratory submissions from outbreaks of salmonellosis on dairy farms and the emergence of novel serovars over the 2012–2020 period (Bingham and Buckle 2021). This situation prompted the initiation of the National Salmonellosis Case–Control Study over two dairy seasons between June 2021 and May 2023. That study sought to describe the patterns of disease and investigate potential management-associated risk factors with the objective of developing control programmes (Compton *et al.* 2024). Farmers who had registered as possible case farms for enrolment in that study and who had agreed to contact from the researchers were contacted by the first author of the current study (CC) by e-mail and telephone between May and July 2022 and invited to enrol in this new study. However, recruitment of case farms and their data from the national study was delayed because of Covid-19 pandemic-related disruptions to normal farm operations. Farms were eligible for the current study if they had one or

more lactating cows with clinical signs of acute onset of diarrhoea with symptoms of systemic illness, with or without cow deaths, and from which non-Brandenburg *Salmonella enterica* subsp. *enterica* were isolated from samples from one or more presumptive case cows during the study period of 1 June 2021–31 May 2022. No data were available on whether salmonellosis diagnoses were made by farmers or veterinarians or a combination of both, and therefore we considered the recorded diagnoses as presumptive. Samples for bacteriological testing were not collected from all presumptive cases. We requested farmers in the current study to provide access to data on cow demographics, total accumulated whole of lactation milk solids (MS) yield, animal treatments, pregnancy testing and animal removals for cows in their herds via their MINDA LIVE (Livestock Improvement Corporation, Hamilton, NZ) and Dairy Diary (Fonterra, Auckland, NZ) software applications for the study period.

Data management

To be included in the analysis, cows had to have a calving date between 1 June 2021 and 31 October 2021. Cows were excluded if they were removed from their herd before the first diagnosis of salmonellosis in their herd. Otherwise, cows were included in the analysis until their removal for any reason or until the end of the dairy season (31 May 2022), whichever came first.

We investigated three main outcome variables. First, cows were categorised by their salmonellosis status as non-cases (no) or cases (yes) from electronic farmer-reported treatment records. Second, we categorised cows by their fate at removal from the herd. We initially categorised the fate from farmer-recorded electronic records as “culled” (for slaughter), “sold” (for further productive use), or “died” (on farm). Non-pregnant cows are routinely sold or culled at the end of their lactation on seasonal supply dairy farms in New Zealand. However, on one study farm, the owner carried over cows that were not pregnant at the end of the breeding programme to the subsequent dairy season, by continuing to milk them over the winter period. Therefore, for our analysis, we considered “carried-over” cows to be the same as those that were culled or sold. This enabled us to include all study herds in the analysis using the same and equivalent outcome variable. Consequently, we created a binary variable: “culled, sold or carried over” for every cow that was culled or sold or carried over (yes) or that remained in the herd until 31 May 2022 (no). For the production of descriptive statistics, cow deaths for any reason were summarised separately from cows that were culled, carried over or sold. Hereafter, cows that were culled, sold or carried over are termed “removed.” Cow deaths were attributed to

salmonellosis if they occurred within 21 days of the diagnosis of salmonellosis in that individual cow or if salmonellosis was recorded as the cause of death. The third outcome variable was the accumulated whole of lactation milk solids yield (kg MS/cow) provided in electronic data via MINDA LIVE. The unit of analysis was the individual cow.

We investigated associations of each of the outcomes with several predictor variables. Herd was categorised as a factor variable. Cows were categorised by age as 2, 3–4, 5–6, or ≥ 7 years old, and by breed as Holstein-Friesian or Jersey if $\geq 12/16$ of that breed, or otherwise as Holstein-Friesian/Jersey crossbred. The interval from the PSC for cows within each herd was calculated as the interval in days between the PSC date for the herd and the calving date of an individual cow. This interval was centred for regression modelling. The herd PSC date is calculated as 282 days (the average gestation period for a dairy cow) following the start of the breeding programme in the previous season. Cow treatment data were collated and binary disease status (no/yes) variables were created for cows that had one or more treatments for mastitis, lameness, sick (undiagnosed), and other (all other diagnoses combined). The date of diagnosis of animal health conditions, including salmonellosis, was assumed to be the same as the first day of treatment for that diagnosis.

Statistical analyses

All data analysis was undertaken with R statistical software (R Core Team, R Foundation for Statistical Computing, Vienna, Austria). The enrolled herds and the frequency of cases of salmonellosis were described using standard statistical methods. We explored the relationship between the potential predictor variables and the three outcome variables of interest – the odds of a cow being diagnosed with salmonella, the odds of removal from the herd and the accumulated whole of lactation MS yield – using path diagrams or directed acyclic graphs (DAG) and the R packages *ggdag* and *ggplot2* (Wickham 2016). The predictor variables we considered for odds of being a salmonellosis case were categories of age and breed, disease status prior to the salmonellosis outbreak and interval between calving and median herd calving date. For the second outcome – removal from the herd – salmonella status was added to this list of candidate predictors. For the third outcome – accumulated whole of lactation MS yield – candidate predictors were the same as for the odds of being removed from a herd. A categorical variable for herd was forced into each model to account for the correlation of outcome variables within herds. The binary and continuous outcomes were modelled by logistic and linear models, respectively.

Having identified the minimal adjustment sets for each outcome, in the second step, we followed the

modelling approach described by Hosmer and Lemeshow (2013) to gain insight into useful predictor variables. We fitted regression models iteratively with a single predictor variable from each of the minimal adjustment sets. Variables with p-values < 0.25 and any others that were biologically plausible were retained for investigation with multivariable models. All the retained variables were then fitted in a multivariable model, and variables were removed sequentially that were clinically insignificant or with p-values ≥ 0.05 and with wide CI overlapping the null effect value. If the CI was narrow and only slightly overlapped the null effect, the variable was carried forward to the next phase of selection. Confounding variables were identified and subsequently retained if deletion of the variable in question led to a change of $> 20\%$ in one or more of the coefficients in the reduced model compared to the full model. Finally, any variables excluded from the initial single predictor screening process were added back into the model to determine if they were compatible with the data when other variables were included. Interaction terms between retained variables were added one by one and retained if clinically meaningful or likelihood ratio test p-values for their inclusion were < 0.05 . The linearity of any continuous predictors was determined by fitting a lowess-smoothed curve of the predicted log-odds for the variable against the predictor.

We evaluated the logistic models with Hosmer-Lemeshow tests and poorly fitted subjects by visualising plots of deviance residuals versus leverage. We used Cook's distance to identify potential outliers and the influence of these on the fit of the model with and without their inclusion, using bubble plots of change in Pearson χ^2 values versus estimated probability. We evaluated the continuous model by visualising raw residuals plots versus predicted values for homoscedasticity, linearity of a Q-Q plot, and plots of the square root of standardised residuals versus predicted values and standardised residuals versus leverage. Diagnostic plots of preliminary final models were created with the R package *ggResidPanel* and the results of final models or the final model marginal means were estimated and plotted using the R package *ggeffects* (Lüdtke 2018). Marginal means aid understanding of the relationship between predictor and outcome variables. The marginal mean estimates are predictions on the response scale from the final models (probability for logistic models and continuous for linear models) for each level of the focal predictor variable of interest (e.g. in this study, salmonellosis status), and the means of other continuous covariates or weighted averages over levels of factor covariates. No adjustments were made for Type 1 error inflation because the independent variables were identified *a priori*.

Financial analyses

The direct financial consequences of a salmonellosis outbreak in a herd were calculated using a simple partial budget model reflecting increased losses and costs from salmonellosis, and parameterised using data from our descriptive analysis, the regression modelling of the actual study herds, and publicly available sources. Three different levels of reporting – first, at the level of the individual diseased cow; second, for all diseased cows in the herd; and third, at the level of the herd – under two different hypothetical loss-incurring scenarios, were estimated. In the first scenario, it was postulated that there was no effect of salmonellosis on milk production, and losses were incurred only as a result of milk that had to be discarded during the mandatory withhold period for cows undergoing antimicrobial treatment. In the second scenario, it was postulated that salmonellosis did affect milk production, and the losses incurred included this effect, estimated from the regression model for the effects of salmonellosis on milk production from the current study. In both scenarios, the herds were the same median size (363 cows), and the herd incidence risk of salmonellosis over a lactation was defined for each scenario from the median, minimum and maximum of the study herds (0.5%, 2.6% or 7.0%, respectively) and other veterinary expenditures unrelated to the salmonellosis were assumed to be equal. No other changes in potential income or costs were recognised for our financial model.

The financial consequences at the diseased cow level were the sum of reduced income from unrealised milk sales and increased expenditure on veterinary treatments.

Reduced income at the individual diseased cow level

The reduced income from unrealised milk sales in a diseased cow in a hypothetical herd with no postulated effect of salmonellosis on MS production (first scenario) was estimated as the mean daily MS production of cows in those same study herds, multiplied by the number of days that milk was withdrawn from supply because of antimicrobial treatment and the milk price received by an owner-operator. The mean daily MS production per cow was calculated as the accumulated whole of lactation MS yield divided by the mean number of lactation days estimated in MINDA LIVE. The total number of cow-days that milk was withdrawn from supply was calculated as the sum of the mean milk withholding duration during and following treatment weighted by the frequency of those treatments. The MS price was calculated as the mean inflation-adjusted MS price paid to farmers

for the dairy seasons, where one dairy season is the period between June 1 and May 31 in the subsequent year, between 2017 and 2022, inclusive (NZ\$8.65; Reynish 2023).

The reduced income from milk sales in a diseased cow in a hypothetical herd with an effect of salmonellosis on MS production (second scenario) was estimated as the product of decreased MS production and the mean inflation-adjusted milk solids price. We estimated decreased MS production as the contrast of marginal means between cows with and without salmonellosis from the final regression model for that association, where the results were consistent with a reduction.

Increased expenditure at the individual diseased cow level

The increased expenditure on veterinary treatments for a diseased cow was estimated from electronic treatment records of cow identifiers, date of administration, and volume of treatment administered and treatment prices from indicative wholesale and retail prices (K Pearce,¹ pers. comm.). We summarised veterinary treatments for cows with salmonellosis by product and product class (antibiotic or other), calculated the retail price of each product per dose unit (mL), and multiplied these prices in turn by the units administered to calculate the cost for each product per cow treated. The mean retail costs of each product class were multiplied by the proportion of cows treated with each product class, and these weighted costs were finally summed to calculate a mean overall cost of veterinary treatments for a single cow with salmonellosis.

Financial consequences for all diseased cows in a herd

We calculated the financial consequences for all diseased cows in a hypothetical herd as the product of the total financial consequences for a single diseased cow, herd size, and salmonellosis incidence risk over a lactation.

Financial consequences at the herd level

We calculated the total additional herd-level expenditures regardless of salmonellosis incidence risk over a lactation only at the herd level. This total was calculated as the sum of veterinary fees for travel and professional time (NZ\$150), diagnostic laboratory fees for bacteriological testing of samples (NZ\$50), and vaccine costs calculated as the retail vaccine price (NZ\$1.20) per dose multiplied by hypothetical herd size and the mean number of doses used per cow used by all study herds (1 dose). We assumed that the farmer would administer the vaccine at no labour cost.

Finally, we calculated the total financial consequences of a salmonellosis outbreak in each

¹K. Pearce, Massey Farm Services Clinic, Palmerston North, NZ

hypothetical milking cow herd under each scenario and lactation incidence risk as the sum of the increased expenditure and decreased income for all affected cows and the increased herd-level expenditures. We divided this sum by the number of all cows, both diseased and non-diseased, in the hypothetical herd to report a total financial consequence per herd and per cow in the herd.

Sample size estimation

We estimated the required sample sizes to investigate the impact of salmonellosis on the two categorical outcomes and the effect of salmonellosis on the accumulated whole of lactation MS yield using the R package *epiR* and the *epi.sscohortc* function. For all outcomes, we used the same parameter values for power = 0.8 and confidence level = 0.95. The other parameter values for the categorical outcomes were: sided test = 2; expected incidence risk over a lactation of the outcome in the exposed and unexposed groups = 0.12 and 0.04, respectively; expected prevalence of exposure to the putative risk factor in the population = 0.2; ratio of number of cows in the exposed to the unexposed groups = 0.25; and the design effect = 5.39 based on an assumed mean cluster (herd) size of 440 and intraclass correlation coefficient of 0.01. A design effect was included in the power analyses to increase the sample size estimates because cows within each herd were not independent of each other due to shared management factors. The target number of herds and cows to enrol for the categorical outcomes was calculated to be 6 and 2,625, respectively.

For the estimation of the sample size required for the effect of salmonellosis on milk production, the *epi.sscmpc* function was used, and the additional parameter values were: mean accumulated whole of lactation MS yield = 397 kg MS/cow (DairyNZ and LIC 2021); SD of accumulated whole of lactation MS yield = 70 kg MS/cow (Bryant *et al.* 2021); effect of salmonellosis on MS production = -10%; the number in the exposed group divided by the number in the unexposed group = 0.05; and the design effect = 5.39 based on an assumed mean cluster size of 440 and intraclass correlation coefficient of 0.01. A one-sided test was applied for this outcome because it was assumed that salmonellosis would only have negative effects on the outcome variables. The target number of herds and cows to enrol for the effect of salmonellosis on milk production was 5 and 2,183, respectively.

Results

Description of study farms

The owners or managers of nine presumptive case herds identified from the National Salmonellosis

Case-Control Study database that had bacteriological results confirming their eligibility for this study were contacted and five farmers agreed to participate. These farmers provided access to their animal health records and herd management software, and subsequently, these five herds and 1,981 cows met the enrolment criteria. One of these herds did not undertake herd testing and therefore did not contribute MS production records to the analysis of that outcome (Table 1).

The range of incidence risk of salmonellosis in the herds over the lactation observed in the study period was 0.46–6.97% cases/herd/lactation, with salmonellosis diagnosed in 2–40 cows in each herd (Table 2). Salmonellosis cases occurred in early to mid-lactation of affected cows (median 76 (min -1, max 206) days post-calving) and herds (median days from herd's PSC to first treatment was 110 (min 27, max 199) days). The incidence risk of salmonellosis over the study period varied markedly between herds (Table 3), and there were differences in the fate of removed cows by salmonellosis status and by herd (Table 2). The incidence risk of culling, sale or carrying over during the study period was numerically greater in cows with positive salmonellosis status only in Herds 4 and 5. No deaths were directly attributed to salmonellosis. Crude means of accumulated whole of lactation MS yield and duration of lactation did not differ greatly between cows with different salmonellosis status, except in Herd 5, in which positive cows on

Table 1. Descriptive statistics of herd and cow-level data collected from seasonal calving pasture-based dairy herds in New Zealand as part of a preliminary study of the risk factors and financial impacts of clinical salmonellosis outbreaks in lactating cows from which *Salmonella enterica* subsp. *enterica* were isolated.

Variable	Value
Herd size (number of cows)	
Min, max	303, 574
Median	363
N (NNA)	5 (0)
Date of planned start of calving	
Min, max	6 Jul, 11 Aug
Median	21 Jul
N (NNA)	5 (0)
Date of start of outbreak	
Min, max	7 Sep, 11 Jan
Median	18 Sep
N (NNA)	5 (0)
Milk solids production (kg MS/cow/lactation)	
Min, max	326, 409
Median	364
N (NNA)	4 (1)
Age category (count (%))	
2 years	384 (19%)
3–4 years	684 (35%)
5–6 years	471 (24%)
≥ 7 years	442 (22%)
Breed category (count (%))	
H-F/J crossbred	797 (40%)
Holstein-Friesian	1,136 (57%)
Jersey	48 (2%)

H-F/J crossbred = Holstein-Friesian/Jersey crossbred; N = number of non-missing records; NNA = number of missing records

Table 2. Percentage incidence risk (and numerator/denominator) of clinical salmonellosis, common diseases and reasons for removals collected from seasonal calving pasture-based dairy herds in New Zealand as part of a preliminary study of the risk factors and financial impacts of clinical salmonellosis outbreaks in lactating cows from which *Salmonella enterica* subsp. *enterica* were isolated. Descriptive statistics for the planned start of calving (PSC) date^a and intervals to cases, grouped by herd, are also shown.

Variable or statistic	Herd 1	Herd 2	Herd 3	Herd 4	Herd 5
Disease					
Salmonellosis	6% (20/363)	2% (5/305)	0.5% (2/436)	7% (40/574)	3% (8/303)
Mastitis	4% (14/363)	12% (38/305)	15% (65/436)	11% (66/574)	18% (55/303)
Lame	1% (2/363)	0% (0/305)	10% (44/436)	1% (5/574)	3% (10/303)
Removals					
Culled or sold	19% (69/363)	7% (22/305)	14% (62/436)	20% (113/574)	19% (57/303)
Carried over	0% (0/363)	0% (0/305)	0% (0/436)	9% (50/574)	0% (0/303)
Culled or sold or carried over	19% (69/363)	7% (22/305)	14% (62/436)	28% (163/574)	19% (57/303)
Died	1% (3/363)	0.3% (1/305)	2% (8/436)	3% (16/574)	1% (4/303)
PSC date	2 August	21 July	11 August	21 July	6 July
Interval PSC to case diagnoses (days)					
Min, max	38, 50	59, 59	27, 33	110, 142	189, 199
Median	38	59	30	110	196
Interval calving to case diagnoses (days)					
Min, max	-1, 36	20, 61	5, 15	41, 128	122, 206
Median	4	49	10	97	183

^aDate 282 days (bovine gestation period) following the start of the breeding programme in the previous herd lactation

average produced 128 kg fewer MS per cow and milked 116 fewer days compared with negative cows. *Salmonella enterica* subsp. *enterica* serovars Typhimurium and Give were serotyped from three and two of the herds, respectively.

Use of registered veterinary medicines for treatment and prevention of salmonellosis

Veterinarians prescribed either of two antimicrobials for cows affected by salmonellosis: oxytetracycline in either long-acting (oxytetracycline dihydrate, Bivatop 200; Boehringer Ingelheim, Auckland, NZ) or both short- and long-acting dose rates (3–5 and 10 mg/kg, respectively, oxytetracycline hydrochloride, Engemycin; MSD Animal Health New Zealand, Upper Hutt, NZ), or a combination of trimethoprim and sulphadimethylpyrimidine (Amphoprim; Virbac NZ, Hamilton, NZ), with the latter

being the most frequently prescribed (68% of antimicrobial treatments). Additionally, some cows were treated with non-steroidal anti-inflammatory formulations of meloxicam (Metacam 20 mg/mL Solution for Injection; Boehringer Ingelheim) or ketoprofen (KetoMax 15%; AgriHealth NZ Limited, Auckland, NZ). One farmer used an anthelmintic, moxidectin (Cydectin Pour-On; Zoetis, Auckland, NZ), without veterinary consultation (Supplementary Table 1).

No herd had a preventive salmonellosis vaccination programme in place prior to its outbreak. Vaccination of the whole herd with Salvexin B (MSD AH) was undertaken in four herds, on average 1 (min 0, max 3) day after the first case was treated. A booster vaccination, as recommended by the manufacturer, was given to all the cows in one herd 31 days after the first vaccination, while the cows in the remaining herds did not receive a booster vaccination.

Table 3. Count (and incidence risk %) of cows removed for any reason from seasonal calving pasture-based dairy herds (n = 5) in New Zealand after the first diagnosis of clinical salmonellosis in a preliminary study of the risk factors and financial impacts of clinical salmonellosis outbreaks in lactating cows from which *Salmonella enterica* subsp. *enterica* were isolated, grouped by herd and clinical salmonellosis status. Statistics for lactation milk production (kg milk solids per cow) and duration (days) are also shown.

	Herd 1		Herd 2		Herd 3		Herd 4		Herd 5	
	Neg	Pos	Neg	Pos	Neg	Pos	Neg	Pos	Neg	Pos
Fate at removal										
Culled or sold	67 (20%)	2 (10%)	22 (7%)	0 (0%)	62 (14%)	0 (0%)	100 (19%)	13 (32%)	55 (19%)	2 (25%)
Carried over	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	44 (8%)	6 (15%)	0 (0%)	0 (0%)
Culled or sold or carried over	67 (20%)	2 (10%)	22 (7%)	0 (0%)	62 (14%)	0 (0%)	144 (27%)	19 (48%)	55 (19%)	2 (25%)
Died	3 (1%)	0 (0%)	1 (0.3%)	0 (0%)	8 (2%)	0 (0%)	15 (3%)	1 (2%)	4 (1%)	0 (0%)
Milk solids production										
Min, max	34, 738	58, 509	NA	NA	91, 631	416, 471	16, 584	140, 534	139, 619	127, 387
Mean (SD)	341 (96)	326 (100)	NA	NA	393 (85)	444 (39)	370 (76)	381 (75)	427 (78)	299 (80)
N (NNA)	326 (17)	20 (0)	0 (300)	0 (5)	418 (16)	2 (0)	525 (9)	40 (0)	293 (2)	8 (0)
Lactation duration										
Min, max	0, 305	106, 279	NA	NA	0, 236	209, 225	0, 305	141, 262	0, 294	75, 167
Mean (SD)	247 (60)	241 (46)	NA	NA	209 (43)	217 (11)	234 (45)	231 (34)	260 (42)	144 (30)
N (NNA)	336 (7)	20 (0)	0 (300)	0 (5)	427 (7)	2 (0)	532 (2)	40 (0)	294 (1)	8 (0)

N = number of non-missing records; NA = not available, Neg = negative; NNA: number of missing records; Pos = positive

Table 4. Point estimates of coefficients (with 95% CI and associated p-values) from the final linear regression model of associations between predictor variables and accumulated lactation milk solids yield (kg/cow) for data collected during a preliminary study of seasonal calving pasture-based dairy herds in New Zealand, assessing the risk factors and financial impacts of clinical salmonellosis outbreaks in lactating cows (n = 1,632) from which *Salmonella enterica* subsp. *enterica* were isolated.

Parameter	Coefficient	95% CI	p-value
Intercept	272.17	260.92–283.43	< 0.01
Clinical salmonellosis case			
No	REF		
Yes	−18.54	−51.83 to 14.75	0.28
Age			
2 years	REF		
3–4 years	80.55	70.39–90.71	< 0.01
5–6 years	115.67	104.67–126.68	< 0.01
≥ 7 years	80.29	69.16–91.42	< 0.01
Breed			
Holstein-Friesian	REF		
H-F/J crossbred	−6.14	−14.31 to 2.04	0.14
Jersey	−16.18	−50.08 to 17.72	0.35
Herd number			
1	REF		
3	49.96	39.05–60.88	< 0.01
4	28.53	18.22–38.85	< 0.01
5	82.78	70.84–94.72	< 0.01
Interaction: clinical salmonellosis case x herd number			
Case x Herd 1	REF		
Case x Herd 3	59.48	−48.19 to 167.16	0.28
Case x Herd 4	11.70	−29.33 to 52.73	0.58
Case x Herd 5	−131.25	−192.82 to −69.68	< 0.01

H-F/J crossbred = Holstein-Friesian/Jersey crossbred; REF = reference category

Associations with odds of diagnosis of clinical salmonellosis and odds of removal from the herd

The results of the final multivariable logistic regression model for salmonellosis status indicated that the odds of being a case were greatest among cows aged ≥ 7 years compared with 2-year-old cows, and varied between herds (Supplementary Table 2). The marginal mean of the final model estimated that the risk of being diagnosed with salmonellosis averaged over the herd effects of cows aged 2, 3–4, 5–6 and ≥ 7 years was 0.93 (95% CI = 0.39–2.18), 0.98 (95% CI = 0.49–1.93), 3.5 (95% CI = 2.15–5.63) and 4.78 (95% CI = 3.06–7.4)%, respectively.

The 95% CI for OR for the effect of salmonellosis on the odds of removal from a herd was wide (0.67–2.07), indicating that the data were compatible with a small decrease in the risk and a larger increase in the risk, so no clear association can be established (Supplementary Table 3). However, cows with relatively later calving dates, as indicated by increasing intervals from PSC to individual cow calving dates, and cows with one or more cases of clinical mastitis, had increased odds of removal from herds. No cow deaths met the definition of being attributed to salmonellosis.

Association between salmonellosis and milk solids production

There was a significant interaction between herd and the diagnosis of salmonellosis regarding the impact on MS production. Diagnosis of salmonellosis was consistent with reduced accumulated whole of lactation MS yield in Herd 5, but the CI for the effect of salmonellosis on whole of lactation MS yield was wide in the other herds, indicating that the data were consistent with both increased and decreased yields, so no clear association could be established (Table 4 and Figure 1). The difference in marginal means from the final model for cows with negative compared to positive salmonellosis status (negative minus positive) in Herd 5 was 150 (95% CI = 98–202) kg less accumulated whole of lactation MS yield per cow, a 36% reduction, whereas the differences between negative and positive salmonellosis cows in Herds 1, 3 and 4 were 19 (95% CI = −14.8 to 51.8), −41 (95% CI = −144 to 62) and 7 (95% CI = −17 to 31) kg MS/cow, respectively.

Financial analysis

We estimated that the largest single component and cause of the greatest variation of the financial impact of a salmonellosis outbreak in the hypothetical herds with the same salmonellosis incidence risk was from decreased income from the sale of milk solids. When this was from discarded milk with no measurably

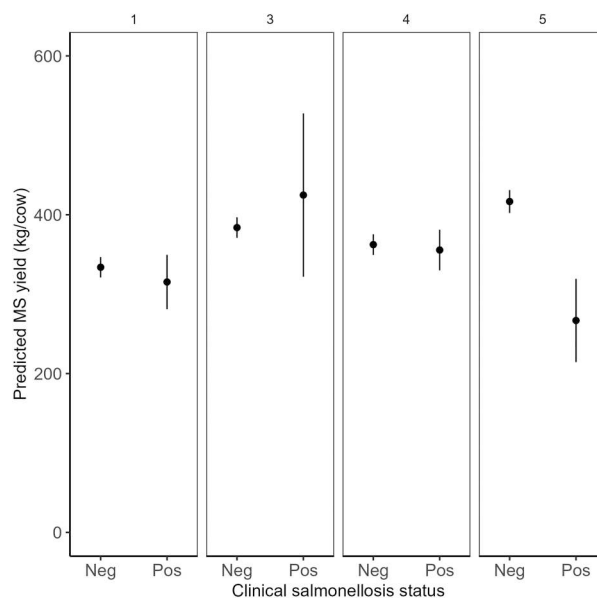


Figure 1. Estimated marginal mean predictions with 95% CI of accumulated whole of lactation milk solids yield from a final regression model of the association between clinical salmonellosis status (positive = Pos; negative = Neg) and accumulated whole of lactation milk solids yield (kg MS per cow) for herds 1 and 3–5 in a preliminary study assessing the risk factors and financial impacts of clinical salmonellosis outbreaks in lactating dairy cows from which *Salmonella enterica* subsp. *enterica* were isolated.

Table 5. Estimated financial consequences (NZ\$) expressed at three levels of analysis for hypothetical dairy herds of 363 cows in terms of decreased income, increased expenditure and their total, and for differing levels of production loss associated with infection and differing incidence risks over the lactation of clinical salmonellosis (median, min, max)^a. Data were collected during a preliminary study of dairy herds in New Zealand to assess the risk factors and financial impacts of clinical salmonellosis outbreaks in lactating cows from which *Salmonella enterica* subsp. *enterica* were isolated.

Level of analysis	Financial consequence	Item	Value in herd with no measurable production loss ^b			Value in herd with measurable production loss ^c		
			Median	Min	Max	Median	Min	Max
A. Diseased cow								
	Decreased income	Unrealised milk sales	72	72	72	1,298	1,298	1,298
	Increased expenditure	Veterinary medicine costs	59	59	59	59	59	59
		Subtotal	131	131	131	1,357	1,357	1,357
B. All diseased cows in herd								
	Decreased income	Unrealised milk sales	680	131	1,830	12,251	2,356	32,982
	Increased expenditure	Veterinary medicine costs	557	107	1,499	557	107	1,499
		Subtotal	1,237	238	3,329	12,807	2,463	34,481
C. Herd								
	Increased expenditure	Veterinary visit fees	150	150	150	150	150	150
	Increased expenditure	Diagnostic laboratory fees	50	50	50	50	50	50
	Increased expenditure	Vaccine costs	436	436	436	436	436	436
		Subtotal	636	636	636	636	636	636
		Total per herd ^d	1,873	874	3,965	13,444	3,099	35,117
		Total per cow in herd	5	2	11	37	9	97

^aIncidence risk over the lactation of clinical salmonellosis: median = 2.6%, min = 0.5%, max = 7%

^bHypothetical herd with no measurable effect of salmonellosis on lactation milk solids production of affected cows had unrealised milk income from discarded milk only and age-weighted mean daily production of 1.7 kg milk solids/cow/day.

^cHypothetical herd with reduced lactation milk solids production of 150 kg in diseased cows

^dCalculated from adding subtotals for B and C

reduced MS yield, the impact for a herd of 363 cows over a single lactation with a median incidence risk of affected cows of 2.6%, amounted to a loss of NZ\$680. When a measurable impact on MS yield of affected cows was included, the model predicted that for equivalent conditions of herd size and incidence risk, the impact amounted to a loss of NZ\$12,251 (Table 5). Table 5 also indicates the potential wide range of financial impacts of salmonellosis outbreaks for varying incidence risks in the hypothetical herds, which were amplified by the effect of disease on milk production.

Discussion

Previous reports of salmonellosis outbreaks on New Zealand dairy farms have mainly described overall herd incidence risk of clinical disease and death, and some have provided costs attributed to the outbreaks (Teague 2011). This is the first preliminary but detailed report of non-Brandenburg *Salmonella* outbreaks on a convenience sample of pasture-based New Zealand dairy farms, including the investigation of cow-level risk factors for clinical disease and the variability of cow and herd-level financial consequences of the outbreaks.

A novel finding in this study among pasture-based herds was that cows of 5–6 and ≥ 7 years of age were at greater risk of salmonellosis in herds with outbreaks compared with younger age-group cows. Scientists undertaking research in confinement herds in the United States reported contradictory results when investigating age as a risk factor for

salmonellosis. Cummings *et al.* (2009) identified among cows suspected of being salmonellosis cases, greater incidence rates of *Salmonella* isolation among multiparous compared with primiparous cows, whereas Fossler *et al.* (2005), working in herds selected without regard to salmonellosis history, reported no difference among parities. The differences between the findings of these international studies may be due to herd and animal selection criteria, but together with our findings, there is some evidence to suggest that milking cows of older age groups should be targeted for increased surveillance and biosecurity measures for salmonellosis control.

The choice of antimicrobials for treatment of presumptive cases of salmonellosis in cows followed the guidelines published by the New Zealand Veterinary Association (Chambers *et al.* 2018) for this disease and age group. However, the use of antimicrobials for the treatment of clinical salmonellosis cases is controversial (Holschbach and Peek 2018), and these authors state that the routine use of antimicrobials in mature animals is less justifiable than in calves, where there is a greater risk of systemic complications. We were unable to draw any conclusions about the effect of antimicrobial treatment compared with no antimicrobial treatment in this study, because treatment programmes were undertaken at the herd level, mixing the effects of treatment and herd, and there were too few herds to undertake an analysis of treatment effect at the herd level. However, beyond this controversy, Jajere (2019) highlighted the threat posed to human populations by multidrug-resistant *Salmonella*, and both the antimicrobial classes used

for treatment of salmonellosis in adult cattle in this study are categorised as “Highly Important Antimicrobials” by the World Health Organisation. Although authorised for use in both humans and animals (WHO 2024), veterinarians should consider their public health responsibilities when prescribing antimicrobials for this disease. Hence, we endorse efforts to protect both animal and public health by the use of management and non-antimicrobial measures to reduce the occurrence of outbreaks of salmonellosis in dairy herds, which would otherwise require intensive use of antimicrobials and increase the risk of development of antimicrobial resistance. The financial impact of a salmonellosis outbreak in an affected cow in this study was increased ten-fold in a hypothetical herd when the impact of the disease on milk production was modelled. This overall effect was likely mediated through both decreased daily milk yield and fewer lactation days of affected cows in one study herd, meaning that the cows were dried-off early. However, appropriate data were not retrieved to differentiate between these effects. Drying-off and culling decisions by farmers are affected by many factors, including the availability of sufficient pregnant replacement heifers, the number of discretionary culls and the stage of the herd lactation when the culling decision needs to be made, and are therefore highly variable between farms. However, using herd-level data for outbreaks of salmonellosis with incidence risk of clinical disease up to 100% and mortality up to 3%, Teague (2011) estimated that the per cow losses from unrealised milk income combined with increased veterinary expenses, ranged from NZ\$130–NZ\$2,200 per diseased cow, similar to the losses estimated in the current study where salmonellosis impacted MS production. We could find no cow-level reports of the effect of non-Dublin *Salmonella* serovars on milk production, but O’Doherty *et al.* (2015) estimated that MS production was reduced by 26 kg per cow lactation in herds exposed to *Salmonella* Dublin or Typhimurium as identified by bulk tank milk antibody status. A potentially important contributing factor towards the financial impact from the risk of removal of cows diagnosed with salmonellosis may be the proximity in time of the outbreak relative to the breeding programme. This was also reported by Taylor (2020) and was suggested in one of the current study herds, when the outbreak occurred during the breeding programme, 110–142 days after the PSC. In seasonal systems, the start of the breeding programme is usually 82 days after the PSC to maintain the same PSC in the next dairy season and hence the salmonellosis outbreak occurred in the second and third month of breeding. This likely disrupted normal reproductive cycling or conception of diseased cows and was possibly a factor in the relatively high incidence risk of cows culled, sold, or carried

over in this study herd. It should also be noted that this study only considered salmonellosis outbreaks in milking cows, whereas the disease may also manifest as diarrhoea or death in calves or dry cows or heifers, or abortion in dry cows or heifers, any of which would likely have different biological and financial impacts.

There were several limitations of this preliminary study. First, the study herds were a small convenience sample of herds and could not be considered representative of the target population of New Zealand dairy herds. The herds enrolled in this study were of smaller median size (363) and lower median milk production (364 kg MS/lactation) compared with the national averages for those measures (444 cows, 397 kg accumulated whole of season MS per cow per lactation, respectively; DairyNZ and LIC 2021), and the relative proportion of Holstein-Friesian and Holstein-Friesian/Jersey cross-bred cows was reversed in the study herds compared with national averages (33% and 50%, respectively; DairyNZ and LIC 2021). A larger number of herds, with more representative herd feeding systems and stages of herd lactation when outbreaks occurred, would be needed to generalise our findings to the population of New Zealand dairy herds.

We acknowledge that our preliminary study lacked statistical power to find effects that may have truly existed. Our estimates of sample size requirements were not met, but this was considered reasonable as the study was preliminary and still useful for estimating effect sizes and variance in the outcome measures for future research. Also, the incidence risk of salmonellosis over this study period was in most cases much lower than we had anticipated for the power calculations (20%) and lower than reported in a series of three herd outbreaks by Teague (incidence risk of clinical disease 8–100%, mortality < 1–3%, 2011) and also as provided by Stevenson *et al.* (2012) in a definition of acute salmonellosis outbreaks in dairy herds in New Zealand (incidence risk of clinical disease > 5% in 14 days). This lack of power potentially contributed to the wide CI for the effect of salmonellosis on the odds of removal from the herd (OR = 1.19; 95% CI = 0.67–2.07). Therefore, while the data were consistent with a relatively large increase in the odds of removal, they were also consistent with a smaller decrease, and so any potential effects of salmonellosis on the probability of removal from the herd as reported by Fossler *et al.* (2005) could not be factored into our financial model. A study in a larger number of herds in New Zealand might provide increased statistical power to identify whether such an association actually exists. Any impact of such an association would need to be included in a revised analysis of the financial impact of salmonellosis outbreaks. We also acknowledge

that the accumulated whole of lactation MS yield data included information from test records prior to the diagnosis of cases in some cows and may have biased the estimate of the effect of salmonellosis on production because of unmeasured confounding effects. However, insufficient data were available to exclude pre-case records and still include the accumulated whole of lactation milk solids yield as an outcome variable in this study. Additionally, our simple economic model did not consider a range of other potential effects at the cow and herd level, such as the sparing effects of salmonellosis on the feed demand of affected cows because of reduced appetite, or of affected cows that are dried off early, and further effects on cow body condition score, which would require a more complex bioeconomic model.

Despite these limitations, we provide preliminary and novel data for farmers and their advisers to consider when planning prevention or control programmes. We also provide data on the variability of biological and financial impacts of salmonellosis outbreaks and possible causes of that variation that can aid researchers to design future studies to increase knowledge about this disease in seasonal-calving pasture-based dairy systems.

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Disclosure statement

J Holter is an employee of the study funder and was involved only in the study design and manuscript review phases.

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