

## RESEARCH ARTICLE

# Analysing mortality patterns in salmon farming using daily cage registrations

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## Abstract

This study describes the patterns of mortality and investigates the sources of variation in mortality during the marine phase of commercial salmon farming. The study included daily mortality records from stocking to harvest of 21 million salmon from ten hatcheries in 136 fish-groups (fish in the same cage from the same hatchery). The fish was stocked in 2017–2018 at 21 marine farms within two Norwegian companies. The sources of variation in mortality were investigated using multilevel linear regression models with ‘fish-group’ nested within ‘farm’ as a random effect, cross-classified with ‘hatchery’. In the final model, ‘fish-group’ was the source of most variation (70%). Furthermore, the mortality categories ‘smolt-related mortality’, ‘infectious diseases’ and ‘handling and treatment’ were responsible for 10%, 17% and 29% of the total number of dead fish respectively. Overall, the study shows that smolt-related mortality is one of the major causes of death in the first part of the production, while handling and treatment was the dominating cause of mortality in total. Mortality varied by fish-group to a large extent. This means that targeted preventive strategies to decrease mortality for individual fish-groups might be more effective than overall measures at farm or hatchery level.

## KEYWORDS

cause-specific mortality, cross-classified multilevel regression model, fish health management, salmon farming, sources of variation

## 1 | INTRODUCTION

The life of a farmed salmon (*Salmo salar*) in Norway starts in the hatcheries, where the fish traditionally are hatched and raised to smolt in freshwater over a period of 6–12 months. The smolts are then transferred from the hatcheries into marine farms (marine phase), where they grow until harvested after another 12–18 months. Mortality during the production cycle will cause economic loss for the farmer but also has a cost in terms of reduced health and welfare for the fish. To prevent fish from dying, the causes and risk factors of mortality need to be investigated as a part of the fish health management at

the farm. Analysis of registered biological data from the production is crucial when making decisions aimed at reducing mortality and subsequently improving fish welfare.

Cause-specific mortality is routinely recorded in most Norwegian salmon farms where the farm staffs assign a cause of death to each fish every day. The causal categories can be based on the macroscopic assessment of the fish, knowledge of events likely to have caused the mortality at the farm the last day (lice treatment, handling of fish, other operations at the farm, etc.) or information from fish health personnel after clinical investigation, autopsy or samples analysed. However, the practice of classifying cause-specific mortality

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registration is currently not standardized, neither with a common list of causal categories nor with a common standard operating procedure. Cause-specific mortality is therefore not to be confused with a diagnosis, which is strictly given by fish health personnel.

The cause-specific mortality adds a qualitative attribute to the registrations compared with the sole crude mortality. Hence, the mortality data become both quantitative and qualitative and have a potential to help the farmer in health management and decision-making (Aunsmo et al., 2008; Nilsen et al., 2020).

Fish farmers report monthly mortality numbers from each farm to the Norwegian authorities, in accordance with national regulation (Akvakultudriftsforskriften, 2008). According to the 'fish health report' 2020, issued by the Norwegian Veterinary Institute, the median mortality of finished production cycles (fish transferred to sea at the same time to the same farm) at marine salmon farms in Norway 2020 was 17.9% (Sommerset et al., 2021). However, there is substantial variation in mortality between farms and production cycles, and mortality of more than 50% in single production cycles has been reported (Bang Jensen et al., 2020). The hatcheries also report mortality numbers to the authorities monthly. However, the information about hatchery mortality at a national level is scarce, since the reported numbers do not follow the fish throughout the production (Tørud et al., 2019). In addition, few standardized health measurements are performed when the salmon are transferred to sea from the hatcheries, and data on mortality causes at this stage are sparse at the national level. Hence, there are several knowledge gaps about the status of the smolt transferred to sea, mortality causes at both hatcheries and at sea, as well as how the early life of the salmon at the hatchery affects the later performance and survival in the marine phase (Bang Jensen et al., 2020; Gåsnes et al., 2021; Tørud et al., 2019).

Smoltification is a complex biological process where the physiology of the salmon transforms from life in a fresh water environment to saltwater (McCormick, 2012). This energy-demanding physiological transformation of the fish also affects the immune system negatively, making fish more vulnerable to stressful events and diseases during this period in life (Johansson et al., 2016). In salmon farming, the smoltification process is immediately followed by the transfer from hatchery to sea for the fish. The transfer involves several stressful events, including transportation, introduction to the saltwater environment and new pathogens (Iversen et al., 2005). These factors contribute to an increased risk of fish dying at the start of the sea phase. Bang Jensen, Qviller, et al. (2020) showed that there was an increase in mortality at the start of the sea phase compared with other periods in the production in Norway. Similar findings were also reported by Soares et al. (2011) from salmon production in Scotland. However, there is still limited information about the causes of 'smolt-related mortality' and what proportion of the overall mortality at cage level it constitutes in the marine phase.

Today, few scientific studies make use of production data retrieved directly from the production management systems (Fishtalk®, Akvagroup or Mercatus®, Scaleaqua) of the fish farmer (Bang Jensen, Mårtensson, et al., 2020). These production management systems were primarily built to help the farmer keep control of the inventory, i.e. number of fish, feeding and mortality. These

are the primary tools used by farmers for following fish throughout production and hence constitute the most detailed records of daily mortality from each production unit. However, since the systems were not built as a fish health management tool primarily, the data need to be adapted when used to study fish health challenges. Such secondary use of data is common in veterinary epidemiological research and is considered a cost-effective way to perform population studies as long as the appropriateness of the data are assessed for the intended use (Houe et al., 2011; Sørensen et al., 1996).

The aim of this study was to investigate mortality patterns in the marine phase, both early (during the first 180 days) and for the entire production. This was approached through two objectives: To describe mortality during production using cause-specific mortality classifications from production management systems, and to estimate the variance component proportion (VCP) of mortality at different organizational levels in salmon production.

## 2 | MATERIAL AND METHODS

### 2.1 | Descriptive statistics

The study unit was the 'fish-group', defined as fish from the same hatchery transferred to sea at the same time and to the same cage. The fish was followed retrospectively through the entire marine phase from the day of transfer to sea ('day 0').

The study population consisted of 20,716,314 salmon in 136 cages at 21 marine farms belonging to two salmon farming companies. The fish was transferred to sea from ten land-based hatcheries in four consecutive stocking periods between spring 2017 and autumn 2018.

### 2.2 | Data sources and data management

Data for this study were collected from two sources. Data were extracted from the production management database of the farmers (Fishtalk®, Akvagroup) and from the documentation that followed the fish to the sea farm ('smolt documentation sheet').

Daily registrations on cage level were extracted in March 2020; Table 1 describes the details of the variables. In addition, a graphic timeline from the production system with an overview of all movements of the fish between cages in the farms was retrieved to help trace the fish-groups.

Information about hatching date and temperature at smolt farm (at the day of transfer to sea) was gathered from the 'smolt documentation sheet'. If absent, the information was collected through direct contact with the site manager at the smolt production site.

#### 2.2.1 | Data management

Data from Fishtalk® were extracted as an Excel file (Microsoft Corporation), using a template made in Fishtalk® to ensure that

**TABLE 1** Descriptive statistics of variables in the study, presented for each of the subsets I and II. Continuous variables are presented with mean, min and max values. Categorical variables are displayed with the number of fish-groups in each category. For the variables 'number of hatcheries with FT' and 'number of self-owned hatcheries', the associated number of fish-groups is shown in addition to the number of hatcheries

Variable	Subset I	Subset II
	'Early mortality'	'Harvest mortality'
Number of fish-groups	121	74
Mortality in production [%], mean (min-max)	2.7 (0.3-21.2)	8.1 (2.7-23.9)
Ln-transformed outcome, mean (min-max)	0.58 (-1.17-3.05)	1.98 (0.99-3.18)
Number of hatcheries	10	9
Numbers of farms	20	16
Number of companies	2	2
Stocking period <sup>a</sup>		
Spring [number of fish-groups]	57	39
Autumn [number of fish-groups]	64	35
Year when transferred to sea		
2017 [number of fish-groups]	58	43
2018 [number of fish-groups]	63	31
Number of hatcheries with FT	7	7
Fish-group from hatcheries with FT	111	71
Number of self-owned hatcheries <sup>a</sup>	4	4
Fish-groups from self-owned hatcheries	90	55
Days in hatchery, mean (min-max)	350 (219-574)	361 (247-530)
Weight at transfer to sea [g], mean (min-max)	124 (74-250)	131 (74.3-250)
Temperature in sea at transfer [°C], mean (min-max) <sup>a</sup>	10.3 (4.9-16.0)	10 (5.0-16.0)
Delta temperature [°C], mean (min-max) <sup>b</sup>	0.8 (-5.3 - 4.5)	1.1 (-5.25-4.5)
Fish-groups treated against lice		
Yes [number of fish-groups]	7	60
No [number of fish-groups]	114	14
Fish-groups with 0 treatments against lice	-	14
Fish-groups with 1-4 treatments against lice <sup>b</sup>	-	42
Fish-groups with >4 treatments against lice <sup>b</sup>	-	18
Moved between cages		
Yes [number of fish-groups]	-	30
No [number of fish-groups]	-	44
Days in production, mean (min-max)	-	433 (352-517)

<sup>a</sup>Variable included in final model of 'early mortality'.

<sup>b</sup>Variable included in final model of 'harvest mortality'.

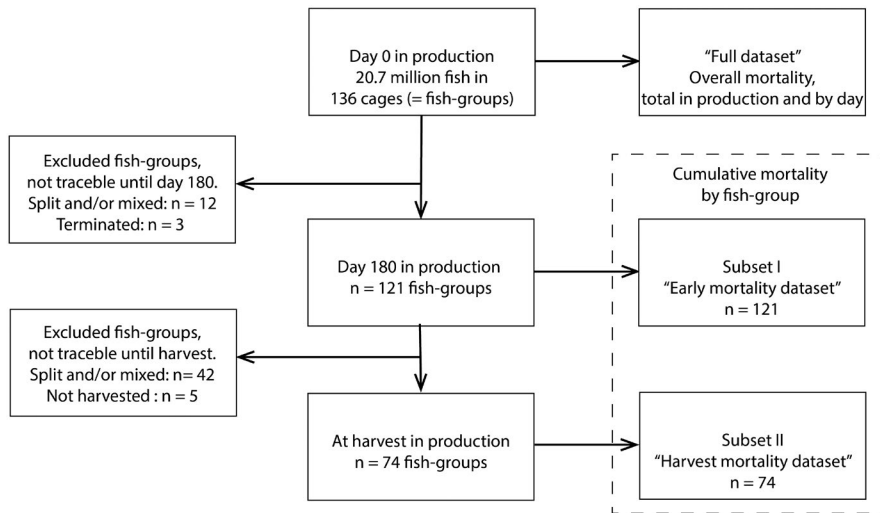
identical production information was retrieved from both companies. The information from the 'smolt documentation sheets' was plotted in an Excel file. All Excel files were imported to Stata (Stata SE/15; Stata Corp.) for further data management and statistical analysis.

One data set with two subsets were used in the study. The 'full data set' included all fish-groups transferred to sea and the registered daily mortality throughout the production. Two subsets of the 'full data set' were constructed in order to investigate the effect of smolt-related factors on cumulative mortality during the early marine phase (180 days post transfer, 'early mortality') and up until harvest ('harvest mortality') respectively. Fish-groups that had been either mixed, split or terminated were excluded. Five fish-groups that had not been harvested at the time of data extraction were also excluded. Only fish-groups that were traceable as one unit until

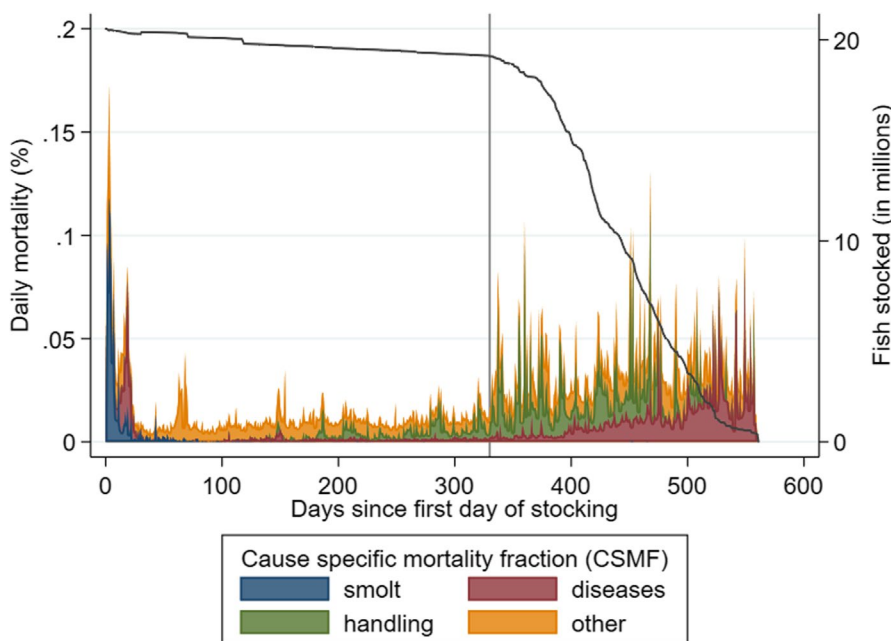
the timepoint of interest was kept for the analysis, this being either during the first 180 days (subset I, early mortality) or up until harvest (subset II, harvest mortality; Figure 1).

### 2.3 | Mortality and cause-specific mortality

The two companies recorded cause-specific mortality daily. The data set contained a total of 65 different categories of mortality causes, including one for unknown cause. Several were also pointing towards the same cause, but with different names. Mortality causes associated with either 'smolt-related mortality', 'handling or lice treatment' or 'infectious diseases' were grouped together in three different groups targeting main challenges in Norwegian salmon



**FIGURE 1** Flow chart describing the study population in the 'full data set' and the number of fish-groups in the two subsets 'early mortality' and 'harvest mortality'. Mortality measurements used are also indicated for each data set



**FIGURE 2** Graphic display of the daily mortality (%) from day of transfer to harvest in the 'full data set'. Coloured areas correspond to the different cause-specific mortality fractions (CSMF) within the mortality each day. The black line indicates the total number of fish in stock each day (right y-axis). The grey vertical line at day 330 is the first day with slaughter, meaning the population is, in addition to mortality, also decreasing because of harvest from this day and onwards

production today (Sommerset et al., 2021). The groups were named: 'smolt', 'handling' and 'diseases' respectively. All other causes were grouped in a fourth group 'other'. The group of 'smolt' included all mortality causes assessed to be related to the transition of fish from hatchery to the marine farm. Therefore, the mortality causes 'dead at arrival' and 'transportation', which indicate handling of fish, were included in the group of 'smolt' after a graphical assessment to when in the production mortality was recorded. Causes related to gill damages or gill diseases were first grouped in a separate group, but because of the low prevalence, they were later allocated to the group 'other'. An overview of all mortality classification categories with the grouping used for analysis is shown in Appendix 1.

Mortality in the full dataset was described as either 'daily mortality' (%) or 'total mortality' (%) across all fish-groups in the data set. 'Daily mortality' was the sum of dead fish each day divided by the total number of fish in all cages each day (Figure 2). 'Total mortality' was the sum of all dead fish, from transfer to harvest, divided by the sum of all fish

transferred to sea. Mortality in the subsets was calculated for each fish-group. Cumulated mortality, at 180 days ('early mortality', %) or at harvest ('harvest mortality', %), was divided by the number of fish transferred to sea in each fish-group (Figures 1 and 3). The proportion in different mortality causes were described as cause-specific mortality fractions (CSMF, %), meaning the different causes of mortality were expressed as proportions of either the total number of dead fish or of the mortality expressions above (Figures 2 and 3).

## 2.4 | Variables and statistical analyses

### 2.4.1 | Variables

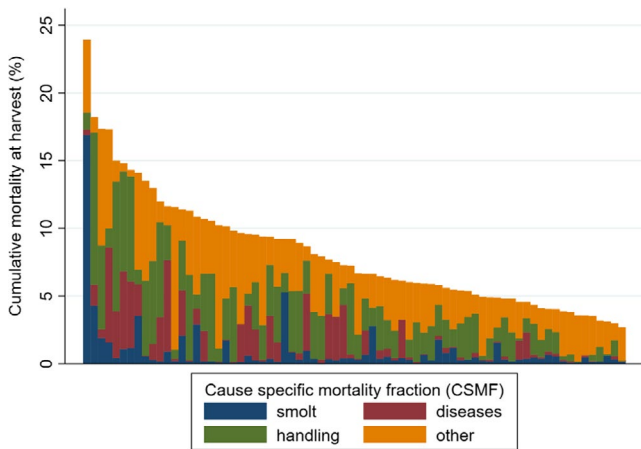
The exposures of interest for the statistical analyses were factors related to the transition of salmon from the hatchery to the sea site. This represents events in the production preceding the first period

in sea, which is identified as the time in production with highest risk of dying according to the earlier studies (Bang Jensen, Qviller, et al., 2020; Soares et al., 2011). In addition to those smolt-related variables, factors related to lice treatments and handling of fish were also included as this represents events known to cause extensive mortality throughout the production in sea (Sommerset et al., 2021). Variables tested in each model are shown in Table 1.

Eight variables associated with the sea transfer were included in the analysis (Table 1). Three were calculated from the raw data; 'days at smolt supplier' was a continuous variable based on the number of days from hatching to transfer to sea. The 'delta temperature' was a continuous variable based on the difference between the water temperature from the last day in the smolt facility to the daily average sea temperature at the sea farm the first week. 'Stocking period' was a dichotomous variable indicating whether the fish were transferred to sea within the first or last six months of the year. The other variables associated with sea transfer were as follows: fish weight at sea transfer ('weight at transfer to sea', continuous), whether the hatchery had flow through water system (FT); recirculation aquaculture systems or a combination of those, the two latter combined due to small numbers ('FT hatchery', dichotomous), if the hatchery

was owned by the owner of the sea site or not ('self-owned', dichotomous); sea temperature at transfer, calculated as the daily average during the first week for each group as stated earlier ('temperature in sea at transfer', continuous) and hatchery ('hatchery').

To control for lice treatments and handling during the marine phase, in addition to other events that potentially affected the outcomes, seven other variables were included in the analyses. To account for salmon lice treatments, two treatment variables were constructed. The first indicated whether the fish-group had been treated against sea lice ('treated', dichotomous) and the second quantified the total number of treatments, which were categorized into three groups (0, 1-4 and >4 treatments) and named 'number of treatments' (categorical). The company and farm at sea ('company' and 'farm', categorical), which year the fish were transferred to sea ('year', categorical) and if the fish-group was moved to another cage during the marine phase of production ('moved', dichotomous). The number of days in production at sea ('days in production', continuous) was the number of days from transfer to sea until harvest (slaughter). For fish-groups harvested over multiple days, the 'days in production' was calculated as an average between the first and last day harvest.

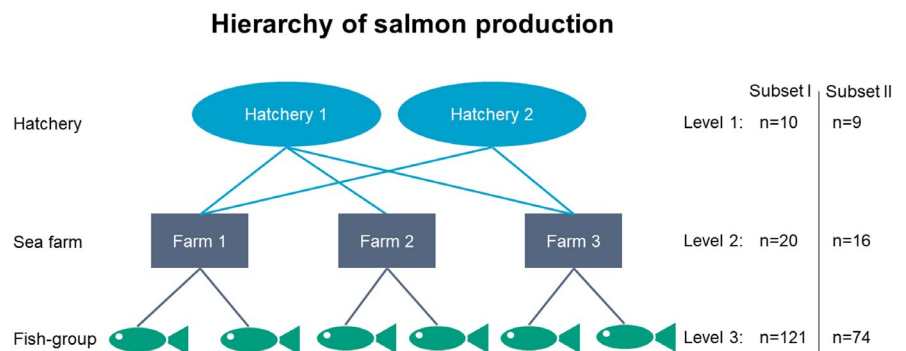


**FIGURE 3** Graphical display of the cumulative mortality in each fish-group at harvest (subset II,  $n = 74$  fish-groups), where the mortality is expressed as a proportion (%) of the fish transferred to sea. The different cause-specific mortality fractions (CSMF) are indicated for each fish-group with colours, and the fish-groups are sorted descending according to the total mortality

### 2.4.2 | Regression modelling

The outcome variables ('early mortality' and 'harvest mortality') had to be ln-transformed to reach the assumption of normally distributed residuals. All explanatory variables were tested with univariable linear regression for each of the two outcomes. Variables associated with the outcome at a level of  $p < .1$  were included in further analyses. Models were built as multi level cross-classified linear regression models, using 'farm' as level and cross-classified with 'hatchery' (Figure 4). For the cross-classification multilevel modelling, MLwiN (MLwiN Version 3.05, University of Bristol) was used within Stata with the stata command 'runmlwin' (Charlton et al., 2020; Leckie & Charlton, 2012). The approach when building the model followed the method described by Aunsmo et al. (2009). Briefly, the modelling was performed using Markov chain Monte Carlo estimation with Gibbs sampling for the posterior distribution (Browne, 2019) with a burn-in period of 1500 iterations and a final model run of 100,000 iterations. To establish the prior

**FIGURE 4** Illustration of the hierarchy of salmon production and the corresponding levels used in the cross-classified multilevel regression model for each of the subsets I ('early mortality') and II ('harvest mortality'). The number of units in each level for the two subsets is also indicated



distribution, the models were first run in iterative generalized least square. Models were built using forward selection and guided by a causal diagram. The improvement between models (for the same dependent variable) was evaluated using Bayesian deviance information criterion. Raftery-Lewis diagnostic and Brooks-Draper diagnostic were used as suggestion of the number of iterations, and the convergence of the models was assessed by kernel density plots (normality of the posterior estimates), plots for autocorrelation (AFC) and partial autocorrelation (PACF) (Aunsmo et al., 2009; Browne, 2019). The independent variables included in the final models were checked for collinearity using graphical assessments for the relationship between categorical and continuous variables. Residuals and trajectory plots were assessed for the final model for each outcome.

Sources of variation were investigated by comparing the proportion of the total variance explained by each random effect in the different models (Browne, 2019; Dohoo et al., 2001). The VCP was estimated for each level in the hierarchy for both the random intercept model and the final model. In addition, the variance component reduction (VCR) was estimated for each random effect, and overall, between the intercept model and the final model for each outcome (Aunsmo et al., 2009).

## 3 | RESULTS

### 3.1 | Descriptive statistics

The total mortality count of the full data set was 1,797,467 salmon, corresponding to an overall total mortality proportion of 8.7% in the marine phase. Mortality in the four stocking periods varied between 7.3% and 10%. Total mortality for the two companies was 7.3% and 11.2%. Daily mortality ranged from 0% to 0.17%, with a median of 0.017% (Figure 2).

In the 121 fish-groups in subset I (first 180 days), the mean mortality was 2.7% and the median was 1.7% with a range from 0.3% to 21% (Table 1). For the fish-groups followed until harvest (subset II,  $n = 74$ ), the mean mortality was 8.1% (median 7.0%) and varied from 2.7% to 23.9% between groups (Figure 4). Days in production at sea varied between the fish-groups from 352 to 517 days with a mean of 433 days from transfer to sea to harvest.

### 3.2 | Mortality classification

The grouped mortality causes of 'smolt', 'handling' and 'diseases' had a combined CSMF of nearly 60% of the registered dead fish in the full data set. Mortality due to 'handling' was the single most important group of causes (CSMF = 29.2%), followed by infectious diseases (CSMF = 17.3%) and smolt-related mortality (CSMF = 9.8%). During the first 180 days (subset I), 'smolt' mortality was the predominant cause identifying 31.7% of the registered dead fish at this point, followed by 'diseases' (17%) and 'handling' (1.5%). The differentiation

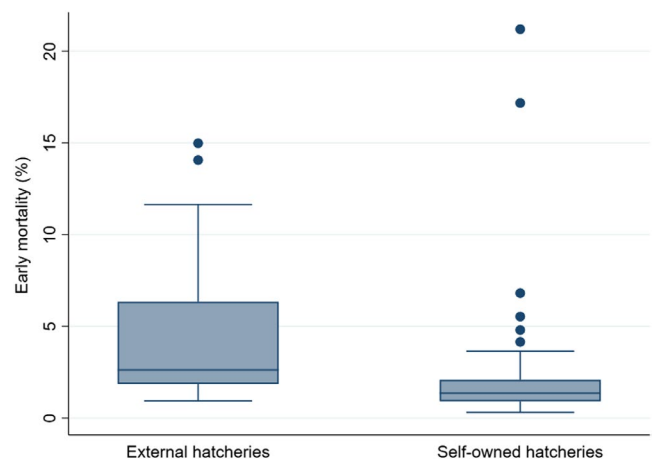
in time was further evident in Figure 2, which displays the causes of death day by day in production (full data set). The 'smolt' mortality dominated the period immediately after transfer, followed by a short period of mortality due to 'diseases'. 'Handling' was the most frequent cause of death from mid production and towards the end. At the very end, the 'disease' category was again a dominant cause. In Figure 3, the mortality causes of the 74 fish-groups in subset II (harvest mortality) were sorted by cumulative mortality and the bars further divided and stacked by the different mortality causes. Here, 'smolt' mortality and 'diseases' appear to dominate the cause of death in specific groups, whereas mortality due to 'handling' is more evenly spread between the fish-groups.

## 3.3 | Statistical analyses

### 3.3.1 | Variables

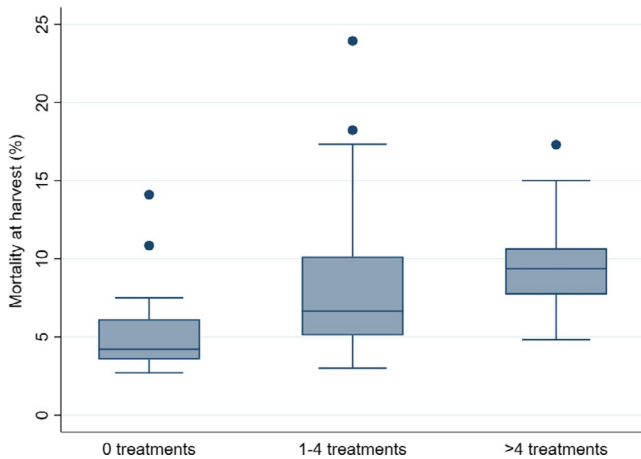
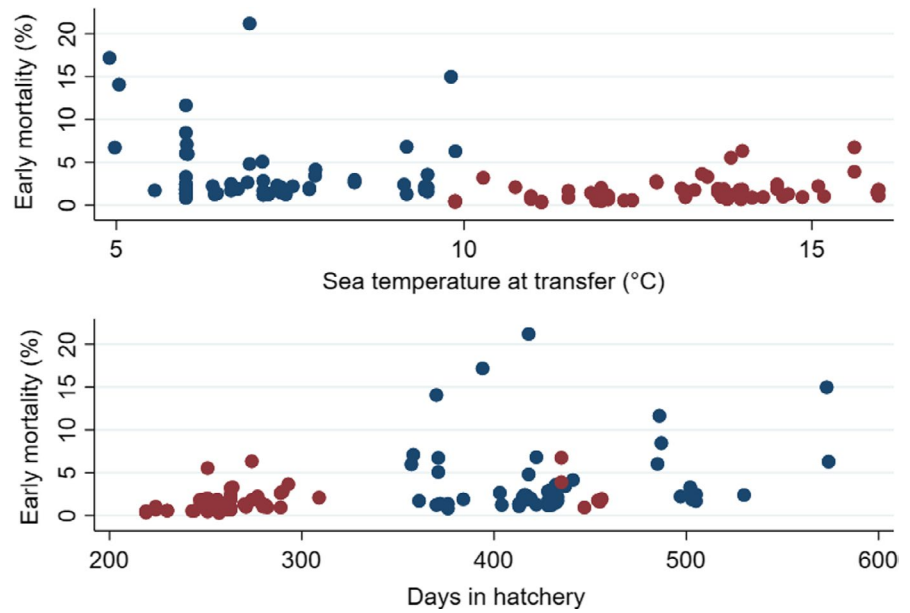
Descriptive statistics of the variables for each outcome are found in Table 1. Fish-groups from self-owned hatcheries showed a lower median and variation of mortality compared with fish-groups deriving from external hatcheries (Figure 5), when relations between the outcome and the exposures in the 'early mortality' model (subset I) were investigated. The relations between temperature, days in hatchery and stocking period displayed in Figure 6 indicate an increased mortality in fish-groups when sea temperature at transfer was below 10°C, which coincided with fish stocked in spring and exceeding 350 days in the hatchery.

Treatments against salmon lice were only performed by non-medical methods. For the 'harvest mortality' (subset II), there were



**FIGURE 5** Boxplot to display the relation between early mortality (subset I,  $n = 121$  fish-groups) and if the fish-group derived from a hatchery owned by the same company running the marine farm ( $n = 90$  fish-groups), or if the fish-groups were bought from an external hatchery ( $n = 31$  fish-groups). The line inside the box represents the median value, the box marks the values within the 25th to the 75th percentile of observations, and the upper and lower whiskers represents the respective adjacent values. Outliers are visualized as solid dots

**FIGURE 6** These scatterplots describe the relation between early mortality (%) and the variables (a) 'sea temperature at transfer' and (b) 'days in hatchery', in subset I ( $n = 121$  fish-groups). In both graphs, the fish-groups are coloured blue if they were stocked in the spring and red if they were stocked in autumn



**FIGURE 7** Boxplot to describe the cumulated mortality (%) at harvest (subset II, 'harvest mortality',  $n = 74$  fish-groups) by the number of treatments against lice. The fish-groups were grouped into three treatment categories: treated 0 ( $n = 14$  fish-groups), 1-4 ( $n = 42$  fish-groups) or  $>4$  ( $n = 18$  fish-groups) times during the production. Boxplot constructed as explained in Figure 5

14 fish-groups with no treatments registered, 42 groups with one to four treatments and 18 groups with more than four treatments. In the 'early mortality', seven fish-groups had registered treatments (Table 1). Mortality was affected by the treatments, where fish-groups treated had higher mean mortality. Figure 7 shows fish-groups with different number of treatments plotted against mortality at harvest.

### 3.3.2 | Cross-classified multilevel modelling

Results from the final cross-classified multilevel models (CCMM) are shown in Tables 2 and 3. The fixed effects included in the final model

of the ln-transformed early mortality outcome (subset I) were 'stocking period', 'temperature in sea at transfer' and 'self-owned hatchery'. For the ln-transformed harvest mortality outcome (subset II), the final model included 'delta temperature' and the categories of the number of treatments (Table 2). All fixed effects included in the final models were significant ( $p < .05$ ).

The chain length of 100,000 iterations was sufficient for both fixed and random effects of both final models, as assessed by the Raftery-Lewis and Brooks-Draper diagnostics. Autocorrelation for fixed effects was minimal, judged by ACF and PACF. For the random effects of farm and hatchery, the PACF plots were not reduced to zero after lag 1, indicating some degree of autocorrelation for these distributions. Residual plots showed no major shortcomings in general, apart for some outliers at the farm and hatchery level deviating from the linear relationship.

### 3.3.3 | Sources of variation and model explanation

For the outcome variable of 'early mortality', the VCR between the intercept and the final models indicated a model explanation of 46% (Table 3). In the final model, the hatchery and farm level accounted for 23% and 6% of the VCP respectively. The variance at the fish-group level remained almost unaffected by the fixed effect (VCR: 3%) and accounted for 70% of the VCP (Table 3).

A similar pattern was observed for the fish-group level in the 'harvest mortality' model; the VCP in the final model was 70%, and VCR indicated a low model explanation (VCR: 13%). However, the remaining VCP was split the opposite way between hatchery and farm level (10% and 20% respectively). Assessed by the VCR, the fixed effects accounted for 35% of the model explanation for this outcome (Table 3).

**TABLE 2** Results from the final models for the ln-transformed outcomes of 'early mortality' ( $n = 121$ ) and 'harvest mortality' ( $n = 74$ ). Fixed effects are displayed with  $\beta$ -values and SD. The categorized variable 'number of treatments' has the category of '0 treatments' as baseline

Model	'Early mortality'		'Harvest mortality'	
Number of fish-groups	121		74	
Number of farms	20		16	
Number of hatcheries	10		9	
Fixed effects	$\beta$	SD	$\beta$	SD
Stocking period	0.33	0.28		
Temperature in sea at transfer	0.1	0.04		
Self-owned hatchery	-0.77	0.26		
Delta temperature			-0.06	0.03
Number of treatments, 1-4			0.32	0.14
Number of treatments, >4			0.58	0.17
Intercept	-0.49	-0.56	1.73	0.15

**TABLE 3** Variance estimates of the sources of variation in the random part of the cross-classified multilevel models for the outcomes 'early mortality' ( $n = 121$ ) and 'harvest mortality' ( $n = 74$ ). Results from both the random intercept models and the final models, including the variance component proportion (VCP) within each model and variance component reduction (VCR) between the intercept and final models

Outcome	Sources of variation	Random intercept model		Final model		Model explanation
		Variance	VCP (%)	Variance	VCP (%)	VCR (%)
Early mortality	Hatchery	0.33	38	0.11	23.4	67
	Farm	0.19	22	0.03	6.4	84
	Fish-group	0.35	40	0.33	70.2	6
	Total	0.87	100	0.47	100	46
Harvest mortality	Hatchery	0.05	16	0.02	10	60
	Farm	0.1	32	0.04	20	60
	Fish-group	0.16	52	0.14	70	13
	Total	0.31	100	0.2	100	35

## 4 | DISCUSSION

The study population had low mortality compared with national statistics (Somerset et al., 2021). The origin of the fish affected mortality in the marine phase, and this was especially evident during the first 180 days in sea. The overall pattern seen was that smolt-related mortality dominated in the start of the marine phase, whereas mortalities related to handling or treatment were the main causes of death in total. The findings also indicate that fish-group was the most substantial source of variation in mortality, both in the early phase and when investigating the entire marine phase of production.

The grouped mortality causes ('smolt', 'handling' and 'infections') were assembled to target well-known health challenges in Norwegian salmon farming (Somerset et al., 2021). The original causes (listed in Appendix 1) were retrieved from the farmers' production management system, and the precision is thus unknown. However, episodes of increased daily mortality are often driven by events in the production (Aunsmo et al., 2008; Aunsmo et al., 2020; Nilsen et al., 2020), for example, handling of fish (e.g. lice treatment), environmental impact

(e.g. strong current) or diseases. It is reasonable to assume that mortality classification related to such known events has a high level of precision. Some diseases cannot be distinguished macroscopically (e.g. infectious pancreas necrosis and pancreas disease), and hence, it is difficult for the farmer to identify the cause of death. However, in Norway, fish health personnel must investigate cases of increased daily mortality in addition to mandatory monthly visits (Akvakultudriftsforskriften, 2008). The findings described by fish health personnel, including disease diagnoses in the population, will help guide the site staff and thus is assumed to increase the precision of daily cause-specific mortality registrations at the farm.

In human health management, cause-specific mortality registrations are used to identify health challenges and causes of mortality in the human population (Naghavi et al., 2015, 2017; WHO, 2021). Through a standardized system, including a list of mortality causes, each death is given one cause based on the underlying cause of death (WHO, 1979, 2018). Since the system is standardized, and the list of causes is hierarchical, the results can be summarized to any population (nationally and globally) and the mortality causes can further be grouped to a relevant



level of details (WHO, 2021). Farming of salmon is also in need of information about health challenges and mortality causes in the population in order to manage the production. However, there is no standard list of mortality causes, neither a common understanding of how the information should be processed and presented. This study has, in addition to the findings, explored some methods in how data from cause-specific mortality registrations can be presented and assist in identifying challenges within health management of salmon farming.

A summary of the total number of dead fish in each cause-specific mortality group will give the farmer an overview of the variation of causes of death throughout the production. In this study, the group of smolt-related causes was high in the start of the production. In addition, out of the 10 highest daily mortality registrations in the production period, eight were within the first 20 days after the fish had been transferred to sea. Mortality identified as smolt-related was the main cause of death during six of those days. The finding is in line with the study by Bang Jensen, Qviller, et al. (2020) where salmon had the highest risk of dying during the first period in sea (Bang Jensen, Qviller, et al., 2020). When all mortality was summarized, mortality caused by handling and treatment was the predominant cause of death. However, when mortality in each fish-group was assessed at harvest, certain fish-groups had smolt-related mortality or diseases as the most important overall cause of death. This information gives valuable insight and helps identify the farms' overall challenges when it comes to mortality and where to prioritize the resources to reduce mortality.

Median annual mortality of fish-groups harvested between 2016 and 2020 in Norway was 15%–18% (Sommerset et al., 2021). The median mortality was 7% in our study. However, the variation in mortality was 3%–24% among the fish-groups. This demonstrates both the better results and further potential of the two companies were followed in this study. The variation was also evident within 'early mortality' (subset I), where the 15 fish-groups (out of 121) with the highest cumulative mortality (mean 9.7%) constituted 46% of all dead fish at this point (data not shown). This is consistent with other studies that also reported high mortality in few groups, which increased the overall mean and produced a skewed mortality pattern (Aunsmo et al., 2008; Nilsen et al., 2020). This further emphasizes the importance of considering each fish-group within each farm when investigating causes of mortality.

A CCMM was built to further investigate events affecting mortality. Using CCMM as the structure of the regression model makes it possible to build in the production hierarchy into the model, where farms can receive smolt from several hatcheries. However, the number of observations in each level of the hierarchy decreases rapidly with this approach, with potential negative effects to the robustness of the model. This was probably the cause of the increased autocorrelation and partly nonlinear relationship between residuals and normal scores at the higher levels of the models, which results in some uncertainty regarding the estimates of the random effects in this study.

The choice of analysing 'early mortality' and 'harvest mortality' as two separate models was made to identify any differences in how the explanatory variables might affect the mortality differently

during the marine production. This contributes to the validity of the model setup, since the importance of the fish origin before sea transfer decreases gradually during the time in production. Looking at the random effects of 'farm' and 'hatchery' in the intercept models (no fixed effects included), the model with 'early mortality' as outcome placed most variance at the 'hatchery' level, whereas 'harvest mortality' outcome had most variance at the 'farm' level. This supports our theory that time in production (together with the production hierarchy) is important when building such models. In the final models, where the fixed effects are accounted for, the variance in the random part was reduced at both 'hatchery' and 'farm' level. However, the variance at the 'fish-group' level remained almost unchanged but with an increase in VCP to 70%. This indicates that the fixed effects of the models explained events occurring at the 'hatchery' and 'farm' level, but to a lesser degree have an impact to each individual fish-group, meaning the variables available had limited ability to identify the causes of mortality at the 'fish-group' level. Starting point for this study was one of the most detailed set of variables possible to obtain from the production system used in the salmon farming today. Hence, the study shows a need for more detailed knowledge to each fish-group in order to explain the 70% of variation in the model detected at this level with this approach.

Among the fixed effects, stocking period was the variable with most effect on mortality in the 'early mortality' model. Fish stocked in spring had increased mortality compared with fall-stocked fish. This is similar to findings from earlier studies (Bang Jensen, Qviller, et al., 2020; Nilsen et al., 2020; Pincinato et al., 2021). Sea temperature at transfer was also significant in the model, indicating an increase in mortality when the temperature rises. However, the temperature at transfer alone in the model was not significant, and graphical assessment of the relationship showed that mortality decreased with fish stocked during spring, along with rising temperatures, while fish stocked in autumn had the highest mortality at the highest temperatures. These variables are also related to 'days in hatchery' (Figure 6), where fish-groups transferred to sea at temperatures below 10°C and after more than 350 days at the hatcheries are stocked in spring. In addition, temperature will be an indicator of when in the stocking period the fish was transferred to sea, and low temperatures indicate stocking either early in the spring or late in autumn for example. Temperature, stocking period, season of year and days in hatchery all affect the mortality; however, causality is difficult to establish.

Four of the ten hatcheries in this study were owned by the companies who ran the marine farms. Fish-groups deriving from these four hatcheries ( $n = 90$ ) had a lower mean mortality at 180 days than the remaining 25% ( $n = 31$ ) of the fish-groups deriving from the other six hatcheries. The variable ('self-owned hatcheries') was included in the final model for early mortality outcome. This indicates a lower early mortality if the farming company owns both the hatchery and the marine farms. At large, this could be interpreted as if you are in control of the entire life of the fish; you are in a better position to control the production at the hatchery and reduce mortality in the early stage of the production in sea.

As described by others (Bang Jensen, Qviller, et al., 2020; Salama et al., 2016; Soares et al., 2011), this study also emphasizes the high risk of mortality in the first period after transferring the smolt to sea. A study by Pincinato et al. (2021) identified smolt-related mortality (or smolt quality) as important when analysing factors affecting losses in Norwegian aquaculture through a questionnaire-based survey performed in 2011, based on farm-level data (Bleie & Skrudland, 2014; Pincinato et al., 2021). They further associated these production losses to differences between hatcheries. The sources of variation in our study, based on cage-level data, identified the fish-group to be more important than the hatchery, where fish-groups with an increased early mortality in sea were spread between the hatcheries. This is important, since it requires a more detailed monitoring and recording of the fish health status of each fish-group in order to further reveal the causes of mortality. Measures taken across fish-groups at a hatchery or farm will not necessarily be sufficient.

Looking at the final model of 'harvest mortality', the number of salmon lice treatments drives the mortality and none of the significant fixed effects in the early mortality model remained significant at this stage. Treatments have been reported from several other studies to be the main cause of mortality in the Norwegian salmon farming during the last years (Bang Jensen, Qviller, et al., 2020; Overton et al., 2019; Sommerset et al., 2021) as well as a recent study showing increased mortality especially from non-medical treatments when investigating lice treatments in detail (Sviland Walde et al., 2021). Our study supports these findings of high mortality associated with lice treatments, also by identifying 'handling' as the most prevalent group of mortality causes in terms of number of dead fish. The CCMM indicates that mortality in the fish-group increases with the number treatments. However, future studies should also include details of which type of non-medical treatment method (preferably down to which vessel) used to further increase knowledge about mortality related to treatment. This information could also be integrated as a part of the mortality classification system, identifying not only 'lice treatment' as a cause of death, but the actual method or vessel used in the treatment. The farmer would then have access to detailed information, explaining causes of mortality related to lice treatments, as an integrated part of the health management.

Apart from the treatment variables, the 'delta temperature' was also significant in the final model of harvest mortality, meaning the difference in water temperature from the hatchery to the sea at transfer affected the mortality at the end of production. The measured effect on mortality was limited according to the model, but the effect was robust and stable throughout the modelling work. However, the causal pathway of the effect of delta temperature on harvest mortality remained unclear, and this result should be interpreted cautiously.

The study population consisted of fish-groups of salmon within two companies, making the number of study units relatively few when the salmon farming industry in total is considered the target population. Hence, the results should be interpreted with care and with considerations of the limitation in external validity. Internal validity (validity of the causal relationships presented from the models) is considered adequate. The study is based on the data possible to retrieve

retrospectively at this resolution today. However, the validity would have been improved further if more measurements (and traceability of the study unit) in the production of salmon were standardized in time and space. As of today, the authors regard this as a limitation in epidemiological studies comparing fish-groups across salmon farming companies. The structure of data in Norwegian fish farming favours 'farm' as the epidemiological unit (Bang Jensen, Mårtensson, et al., 2020; Bang Jensen, Qviller, et al., 2020); however, this study shows that cage to cage variation is important. Hence, it is difficult to produce studies of fish-groups with an increased validation and applicable results beyond the study population. Results from this study emphasize the importance (statistical and biological) of fish-groups within the farm, meaning improvement of structure and traceability of data are necessary to further investigate biological variation between fish-groups, not only between farms.

This study has shown that daily cause-specific mortality records can be used to effectively describe mortality patterns at a chosen unit and time in salmon farming. This has the potential to be an important tool within fish health management. Furthermore, sources of variation deriving from cross-classified multilevel modelling can be used when analysing causes of mortality and identify which part of the production hierarchy contributes the most. The fish-group attributed 70% of the variation in mortality in this study. This points towards the need for more detailed information from each fish-group to further investigate the causes of mortality in the sea phase. Overall in the production, the mortality due to handling of fish and treatment of salmon lice was the major cause of death. However, the cause-specific mortality classification registrations identified smolt-related causes to be the major cause of death during the first 180 days and for specific fish-groups also when mortality was summarized at harvest. This means that targeted preventive strategies against mortality at the fish-group level are important to increase survival, improve fish welfare and improve production of farmed salmon.

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## CONFLICT OF INTEREST

The authors declare that they have no competing interests.

## DATA AVAILABILITY STATEMENT

The data set generated in the study is not included but is available from the corresponding author on reasonable request.

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**APPENDIX 1**

Overview of all mortality causes included in the constructed groups from the 'full data set'. Displayed as both the actual number of dead fish for each cause, the cause-specific mortality fraction (CSMF) of the total number of dead fish (%) and the corresponding grouped CSMF used in this study. CSMF of less than 0.1% was for readability set to '<0.1%'. The mortality causes are sorted descending by the prevalence within each group. In the raw data (in Norwegian), some mortality causes occurred multiple times, only with differences in spelling or different acronyms used towards the same cause. When translated into English, those causes were merged. For traceability, merged causes have the number of original variables (2 or 3) in brackets behind the name. This also emphasize the need of a common standard of mortality causes in salmon farming.

Grouped mortality cause	Mortality cause	Number of dead fish	CSMF of total number of dead fish (%)	Grouped CSMF of total number of dead fish (%)
Smolt-related	Incomplete smoltification (2)	82,605	4.6 %	9.80%
	Transportation	46,625	2.6 %	
	Dead at arrival	25,323	1.4 %	
	Nephrocalcinosis	13,147	0.7 %	
	Fin damage	7,044	0.4 %	
	Haemorrhagic smolt syndrome (HSS) (2)	778	<0.1%	
Infectious diseases	Tenacibaculum	81,958	4.6 %	17.30%
	Cardiomyopathy syndrome (CMS) (2)	74,031	4.1 %	
	Heart and skeletal muscle inflammation (HSMI) (2)	73,636	4.1 %	
	Pancreas disease (PD) (2)	70,150	3.9 %	
	Infectious pancreatic necrosis (IPN) (2)	6,737	0.4 %	
	Yersinia infection (2)	3,512	0.2 %	
	Mouth rot	1,399	<0.1%	
	Fungal infection	413	<0.1%	
Handling and lice treatment	Handling (2)	243,264	13.5 %	29.20%
	Lice treatment (2)	238,728	13.3 %	
	Grading	34,317	1.9 %	
	Moving grading	7,714	0.4 %	
	Bath treatment	1	<0.1%	

Grouped mortality cause	Mortality cause	Number of dead fish	CSMF of total number of dead fish (%)	Grouped CSMF of total number of dead fish (%)
Other	Unknown (2)	376,206	20.9 %	43.70%
	Undefined	111,192	6.2 %	
	Runts (3)	81,664	4.5 %	
	Ulcer (3)	71,295	4.0 %	
	Sexual maturation (2)	31,948	1.8 %	
	Winter ulcer	30,388	1.7 %	
	Normal	23,261	1.3 %	
	Birds—cormorant	9,685	0.5 %	
	Proliferative gill infection (PGI)	8,093	0.5 %	
	Injuries (2)	7,612	0.4 %	
	Fin rot	7,522	0.4 %	
	Old	4,871	0.3 %	
	Amoebic gill disease (AGD)	4,870	0.3 %	
	Deformities (2)	4,531	0.3 %	
	Other gill problems	2,324	0.1 %	
	Dead due to incidents	2,084	0.1 %	
	Culled (2)	1,926	0.1 %	
	Gill infection	1,676	<0.1%	
	Predators (3)	1,396	<0.1%	
	Discarded	1,217	<0.1%	
	Suspected disease	960	<0.1%	
	Sampling	928	<0.1%	
	Egg not fertilized	284	<0.1%	
	Gill infection—other	89	<0.1%	
	Technical failure	52	<0.1%	
	Birds—heron	11	<0.1%	
<b>Total</b>	<b>Total number of dead fish</b>	<b>1,797,467</b>	<b>100%</b>	<b>100%</b>