



Quantitative genetic variation of resistance against *Piscirickettsia salmonis* in Atlantic salmon (*Salmo salar*)



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ABSTRACT

Piscirickettsiosis (*Piscirickettsia salmonis*) is one of the diseases that cause large economic losses in Chilean salmon industry. Genetic improvement of disease resistance represents one strategy for controlling infectious diseases in farmed fish. However, knowledge of whether genetic variation exists for piscirickettsiosis resistance is needed in order to determine the feasibility of including this trait into the breeding goal. Using data from a challenge test performed on 2601 Atlantic salmon (*Salmo salar*) from 118 full-sib groups (40 half-sib groups) we found significant genetic variation for resistance to piscirickettsiosis. We used a cross-sectional linear model (CSL) and a binary threshold (probit) model (THR) to analyze the test-period survival, a linear model (LIN), Cox (COX) and Weibull (WB) frailty proportional hazard models to analyse the day at death, and a survival score (SS) model with a logit link to analyze the test-day survival. The estimated heritabilities for the different models ranged from 0.11 (SS) to 0.41 (COX). The Pearson and Spearman correlation coefficients between full-sib families estimated breeding values (EBVs) from the six statistical models were above 0.96 and 0.97, respectively. We used different data subsets, splitting the entire dataset both at random and by tank, in order to predict the accuracy of selection for each model. In both cases COX (0.8 and 0.79) and CSL (0.76 and 0.71) models showed the highest and the lowest accuracy of selection, respectively. These results indicate that resistance against *P. salmonis* in Atlantic salmon might be genetically improved more efficiently by means of using models which take both time to death and data censoring into account in the genetic evaluations.

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1. Introduction

Chile is one of the leading producers of farmed Atlantic salmon (*Salmo salar*) in the world. One of the key factors affecting the profitability and sustainability of this industry is the sanitary status of farmed fish. The infectious disease piscirickettsiosis, caused by *Piscirickettsia salmonis*, is one of the main causes of significant losses in the Chilean salmon industry. This intracellular pathogen was initially isolated from individuals of coho salmon (*Oncorhynchus kisutch*) from an outbreak that caused high mortality in fish farms in Chile (Cvitanich et al., 1991; Fryer and Hedrick, 2003). Moreover, it has been reported that *P. salmonis* is now covering a wider geographic range and can affect other salmonids species, such as rainbow trout (*Oncorhynchus mykiss*), cherry salmon (*Oncorhynchus masou*), chinook salmon (*Oncorhynchus tshawytscha*), pink salmon (*Oncorhynchus gorbuscha*) and Atlantic salmon (Fryer and Hedrick, 2003).

It has been proposed that the main routes of the initial infection in natural conditions are injured skin and/or gills. In fact, subcutaneous injection of the bacteria can lead to similar mortality rates as those seen

with intraperitoneal injection (IP) and also can double the mortality rates caused by a skin patch infection, indicating that skin damage is crucial in the manifestation of the disease (Smith et al., 1999). In the later stages of infection, the pathological signs observed in fish infected via different routes become similar, most likely because of the septic nature of the disease and several tissues and organs can be compromised (Cvitanich et al., 1991; Fryer and Hedrick, 2003).

In addition to vaccines and antibiotics, genetic improvement for disease resistance may represent a feasible and sustainable approach for the control of infectious diseases (Stear et al., 2001). Thus, it has been proposed that resistance to infectious diseases should be included in the breeding goal of genetic improvement schemes in salmonids, considering both economically and sustainability issues (Gjedrem, 2000; Gjedrem et al., 2012; Ødegård et al., 2011). However, including resistance to piscirickettsiosis into the breeding objective would require an understanding of the level of additive genetic variation for this trait. To date, no studies aiming to determine additive genetic variation for piscirickettsiosis resistance in Chilean commercial populations of Atlantic salmon have been published.

A considerable number of studies have shown genetic variation that would account for resistance against different infectious diseases in salmonids. In these studies, resistance is typically measured as the survival

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rate of infected individuals using data from field outbreaks and experimental challenges (Yáñez and Martínez, 2010). Different models have been used for analyzing survival data, which can be basically classified as cross-sectional and longitudinal models, such as threshold and proportional hazard frailty models, respectively. These models differ in terms of how they use the available information, and this may have an impact on the accuracy in estimating genetic variance and the breeding values for the selection candidates (Ødegård et al., 2011).

In this work we aim to investigate levels of genetic variation and estimate breeding values for resistance against piscirickettsiosis in Atlantic salmon using survival data obtained from an experimental challenge using cross-sectional and longitudinal models, and then to compare the accuracy of the predictions obtained between them. This information will be useful for planning breeding strategies for controlling this disease by genetic improvement.

2. Materials and methods

2.1. Fish

Survival data were obtained from a total of 2601 Atlantic salmon smolts belonging to 118 full-sibs families (the progeny of 40 sires and 118 dams) from the breeding population of Salmones Chaicas, which were hatched in May 2010. Each individual was tagged with a PIT-tag (Passive Integrated Transponder) inserted into the abdominal cavity, at an average weight of 13.1 (SD = 3.4) g, in order to retain pedigree information during the challenge test. Tagged fish were reared in a single communal tank about 14 months before the transfer to Aquainnovo's Research Station located in Lenca River, Xth Region, Chile. After a 29 day acclimation period in salt water (31 ppt), an average of 22 (ranging between 9 and 24) fish per family was challenged as described below. The average weight of these fish at the moment of inoculation was 274.8 (SD = 90.6) g and they were negative for the presence of Infectious Salmon Anemia virus, Infectious Pancreatic Necrosis virus, *Renibacterium salmoninarum* and *Flavobacterium* spp.

2.2. Challenge test

The challenge test was carried out with a virulent strain of *P. salmonis* which had been isolated in May 2011 from Chiloe and provided by SGS Aquatic Health Chile S.A. Briefly, the LD₅₀ dose was determined using three different dilutions (1:10, 1:100, 1:1000) from the original inoculum in a preliminary IP challenge, which was carried out in a random sample of 75 fish from the same group (25 fish per dilution). This preliminary challenge spanned 35 days and based on the mortality at the end of the period a dilution of 1:50 was determined as the LD₅₀. In the definitive challenge test, infection was induced by means of IP injection with 0.2 ml of the previously determined LD₅₀ of *P. salmonis*. After inoculation, infected families were split and kept in three different tanks with salt water (31 ppt) during the test period. Thus, the challenge was carried out using a three-replicate design in which a similar number of fish from each family were separated in three communal tanks (see Table 1 for summary statistics by tank replicate).

Table 1

Summary statistics for each replicated tank of the experimental challenge test against *Piscirickettsia salmonis* in Atlantic salmon (standard deviation in parenthesis).

| Tank | 1 | 2 | 3 |
|-----------------------------------|---------------|---------------|---------------|
| Total number of fish | 860 | 867 | 874 |
| Average number of fish per family | 7.3 (1.2) | 7.3 (1.1) | 7.4 (1.1) |
| Average body weight | 323.2 (141.2) | 321.1 (133.1) | 322.6 (137.8) |
| Total number of dead fish | 356 | 298 | 342 |
| Final survival rate | 0.59 | 0.66 | 0.61 |

2.3. Records, trait definitions

Mortalities were recorded daily until day 40 post-inoculation. The Kaplan–Meier curves of the survival function (Kaplan and Meier, 1958) by family and tank were plotted for the test period in order to show the cumulative mortality across the challenge (Fig. 1). Body weight was recorded at the time of death for every dead fish or at the end of the assay for survivors. Necropsy examination was performed on each dead fish and molecular diagnostic in a sample of dead fish was carried out in order to both confirm *P. salmonis* and discard other pathogens (mentioned in Section 2.1) as the cause of death.

Resistance has been assessed as the challenge-test survival using the following trait definition:

1. Test survival (TS), which was scored as 0 if the fish died in the challenge test period and scored 1 if the fish survived at the end of the experiment. This trait was analyzed using a cross-sectional linear model (CSL) and a binary threshold (probit) model (THR), in order to take the binary nature of the trait into account.
2. Day of death (DD), which had values ranging between 1 and 40 depending on the day of the event. If fish survived at the end of the testing period the record was assumed censored. This trait was analyzed using a linear model (LIN), without accounting for censored data, and both COX and Weibull frailty proportional hazard models, which take censored data into account.
3. Test-day survival (TDS), in which the survival time (days) was transformed to binary survival records with one record per day the

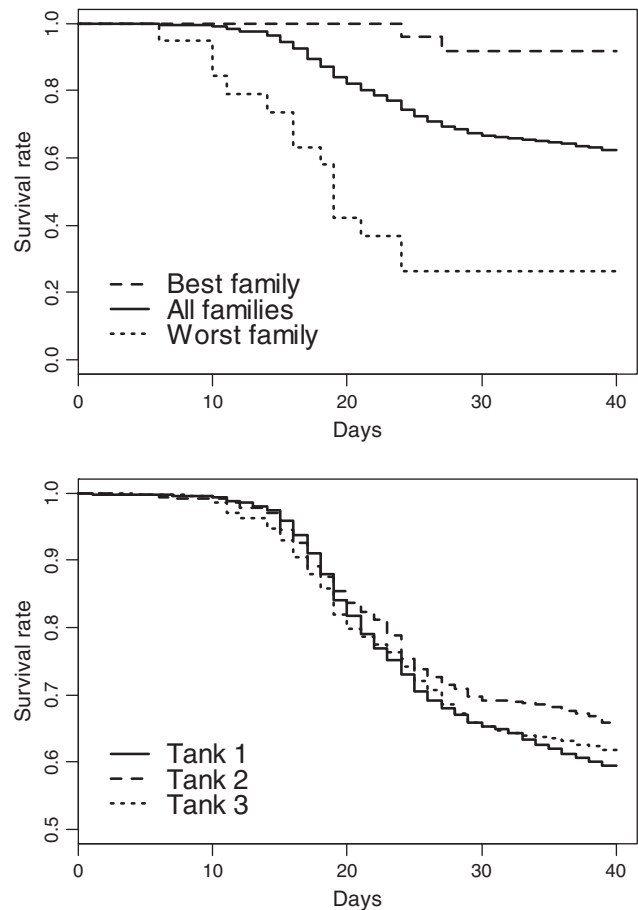


Fig. 1. Kaplan–Meier mortality curves of the best and the worst family and an average of the 118 full-sib families (above) and replicated tanks (below) during 40 days of a challenge with *Piscirickettsia salmonis* in Atlantic salmon.

fish stayed in the test and the number of records per fish equals the number of days (the test-day of the first mortality due to disease was set as a starting point) until death or censoring. For each period, an observation was scored as 0 if the fish was still alive on the actual test-day and scored 1 if the fish died on the actual-test day, e.g., a fish dying at day 6 had survival scores of [0 0 0 0 0 1] (Gitterle et al., 2006; Ødegård et al., 2006, 2007). This trait was analyzed using a survival score (SS) model with a logit link function in order to take the binary nature of the trait into account.

2.4. Models

Data was analyzed with different univariate sire–dam models as defined below:

CSL and LIN: A linear model was used to fit the binary variable of TS and the continuous variable of DD:

$$y_{ijkl} = \mu + t_i + d_j + s_k + e_{ijkl} \quad (1)$$

Where, y_{ijkl} is the phenotype (TS for CSL and DD for LIN) for the fish l , in full-sib family jk , in tank i ; μ is the fixed effect of the overall mean; t_i is the fixed effect of tank i ; d_j is the random genetic effect of the dam j ; s_k is the random genetic effect of the sire k , and e_{ijkl} is the random residual for fish l .

THR: A binary threshold (probit) model was used for analyzing TS:

$$\Pr(Y_{ijkl}) = \Phi(\mu + t_i + d_j + s_k) \quad (2)$$

where, Y_{ijkl} is the phenotype (TS) for the fish l ; $\Phi(\cdot)$ is the cumulative standard normal distribution and the other parameters as described above.

COX and WB: Cox and Weibull frailty proportional hazard models were fitted for analyzing DD:

$$\lambda_{ijkl}(d) = \lambda_0(d) \exp(t_i + d_j + s_k) \quad (3)$$

where, $\lambda_{ijkl}(d)$ is the hazard function at day d for fish l from dam j and sire k ; $\lambda_0(d)$ is the baseline hazard function at day d , which can be totally arbitrary (COX) or can have a Weibull (WB) distribution (i.e. $\lambda\rho(\lambda d)^{\rho-1}$) with parameters λ and ρ ($\lambda > 0$ and $\rho > 0$), and the other parameters are as defined above.

SS: A survival score (SS) model with a logit link function was used for analyzing TDS:

$$\Pr(Y_{ijkl} = 1) = \frac{\exp\left(\sum_{p=0}^4 \beta_{ip} Z(d)_p + \mu + t_i + d_j + s_k + e_{ijkl}\right)}{1 + \exp\left(\sum_{p=0}^4 \beta_{ip} Z(d)_p + \mu + t_i + d_j + s_k + e_{ijkl}\right)} \quad (4)$$

where, Y_{ijkl} is the phenotype (TDS) for the fish l ; β_{ip} = p th order regression coefficient of the i th tank, $Z(d)_p$ = p th order orthogonal polynomial of day d , and the other parameters are as defined above.

CSL, THR, LIN and SS models were fitted using ASREML 3.0 package (Gilmour et al., 2009) whereas COX and WB models were analyzed using the Survival Kit (Ducrocq et al., 2010). Initially, we run models including sire–dam additive genetic effect plus random common environmental of full-sibs effect (full model). For all models, the proportion of variance explained by common environmental of full-sibs effect was negligible (close to zero). This may be due to the fact that common environmental of full-sibs effect is confounded with additive genetic effect, which is difficult to separate given the mating design (Gjerde,

2005). Moreover, the difference in variation due to environment was expected to be negligible as fish were kept in a common environment (one single tank) for about 14 months before they were challenged. Thus, our final model included only sire–dam additive genetic effect (reduced model). In addition, reduced model was compared with full model using likelihood ratio (LR) tests (Lynch and Walsh, 1998) and random common environmental of full-sibs effect was not significant ($P < 0.05$). For all the models, sire and dam effects assumed $\sim \mathbf{N}(0, \mathbf{A}\sigma_{sd}^2)$ where $\sigma_{sd}^2 = \sigma_s^2 = \sigma_d^2$ and \mathbf{A} is the additive genetic relationship matrix among the animals (including parents). Residuals (if included in the model) assumed $\sim \mathbf{N}(0, \mathbf{I}\sigma_e^2)$ or calculation of heritability, the implicit residual variance on the underlying scale for THR model (probit link) was set to 1 and for SS model (logit link) was set to $\pi^2/3 \sim 3.3$. Similarly, for COX and WB models the residual variance on the log-scale was set to $\pi^2/6$ (from an extreme value distribution) (Ducrocq and Casella, 1996).

2.5. Heritabilities

The sire–dam variances are expected to contain $1/4$ of the total additive genetic variance (as $1/4$ of the additive genetic variance is explained by each of the parents). Hence, for all the models heritability was calculated as:

$$h^2 = \frac{4\sigma_{sd}^2}{2\sigma_{sd}^2 + \sigma_e^2}$$

where σ_{sd}^2 is the additive genetic sire–dam variance and σ_e^2 is the residual variance.

2.6. Model comparison

Due to the fact that the models presented in Section 2.4 use different trait definitions, we cannot directly compare them based on heritability estimates or fit statistics. Therefore, the models were compared based on their accuracy of selection, which was determined through the correlation among full-sib family estimated breeding values (EBV), based on subsets of data from tank replicates and also from two random data subsets. In both cases we used the variance components that were estimated using the entire dataset. We calculated the full-sib family EBVs as $(\mathbf{a}_d + \mathbf{a}_s)$ where \mathbf{a}_d and \mathbf{a}_s represent the EBV of the dam and sire, estimated from the models, respectively. The accuracy of selection (r_T) is related to the product-moment (Pearson) correlation coefficients among the full-sib family EBV (r_{EBV}) calculated using different data subsets. From selection index theory, it has been proven that $r_{EBV} \approx r_T^2$ if there is a similar amount of information per family in the different data subsets and lack of bias of the predicted breeding value (Gitterle et al., 2006). Thus, the square root of the r_{EBV} can be used for evaluating models on the basis of their accuracy of selection. Furthermore, the rank correlation (Spearman) coefficient among the full-sib family EBVs from each model was calculated to determine the agreement among genetic predictions of the different statistical methods.

3. Results

3.1. Challenge test

Fish inoculated with *P. salmonis* showed typical clinical signs and pathological lesions of piscirickettsiosis. The total cumulative mortality across all the families reached 38.5% at the end of the test period. Kaplan–Meier mortality curves showed that mortalities increased rapidly around day 11 and also demonstrated differences in survival rates between families (Fig. 1). Thus, cumulative mortality rates ranged from 8.3% to 73.7% between different families, thus indicating a substantial genetic variation in susceptibility for piscirickettsiosis.

Table 2

Estimates of additive genetic sire–dam variance (σ_{sd}^2) and heritabilities (h^2) with their standard errors (\pm SE) for resistance to *Piscirickettsia salmonis* in Atlantic salmon analyzed using six statistical models.

| Model ¹ | Estimates | |
|--------------------|------------------------|-----------------|
| | $\sigma_{sd}^2 \pm$ SE | $h^2 \pm$ SE |
| CSL | 0.036 \pm 0.007 | 0.15 \pm 0.03 |
| THR | 0.269 \pm 0.059 | 0.24 \pm 0.04 |
| LIN | 19.95 \pm 4.04 | 0.18 \pm 0.03 |
| SS | 0.507 \pm 0.107 | 0.11 \pm 0.02 |
| COX | 0.2108 ² | 0.41 |
| WB | 0.1146 ² | 0.25 |

¹ The models used were: CSL = binary cross sectional linear model, THR = binary threshold (probit) model, LIN = time until death linear model, SS = survival score (logit) model, COX = Cox proportional hazard frailty model and WB = Weibull proportional hazard frailty model.

² Mode of the posterior distribution.

3.2. Heritabilities and correlations

Significant additive-genetic variation was observed for all the trait definitions and models used with low to moderate heritability estimates ranging from 0.11 to 0.41 (Table 2). Estimated heritability values for *P. salmonis*' resistance were slightly variable, depending on trait definition and the model used. In general terms, considerably lower values of heritability for resistance were obtained when the trait was assessed as the TDS, analyzed using a SS model (0.11). Dividing the test period in many test-day survival scores may have diluted heritability in SS models. For both linear models, CSL and LIN, similar heritability values were obtained (0.15 and 0.18, respectively), independently that the trait definition used is different for both linear models. In addition, heritability was intermediate in THR model (0.24) in comparison with other models. Higher values of heritability were obtained when the continuous trait, day of death, was analyzed using WB and COX models (0.25 and 0.41, respectively), which takes the baseline hazard function and the right-censored data into account.

3.3. Model comparison

Table 3 shows the Pearson and Spearman correlation coefficients among the full-sib family EBVs from different models. Disregarding the sign of the correlation coefficients, which may be positive or negative depending on different trait definition between the models used, all correlations were higher than 0.96. This result indicates that an almost identical ranking of families is generated by using whichever of the models fitted. The highest correlation was obtained among CSL and THR (\sim 1), and the lowest one was achieved between THR and LIN (0.96 and 0.97 for Pearson and Spearman, respectively).

Table 3

Pearson (above diagonal) and Spearman (below diagonal) correlation coefficients between full-sib² family EBVs for six statistical models for survival to *Piscirickettsia salmonis* infection in a challenge test.

| Model ¹ | CSL | THR | LIN | SS | COX | WB |
|--------------------|--------|--------|--------|--------|--------|--------|
| CSL | | 0.999 | 0.966 | −0.994 | −0.986 | −0.992 |
| THR | 0.999 | | 0.964 | −0.994 | −0.988 | −0.993 |
| LIN | 0.971 | 0.970 | | −0.982 | −0.975 | −0.973 |
| SS | −0.994 | −0.994 | −0.986 | | 0.993 | 0.995 |
| COX | −0.988 | −0.988 | −0.980 | 0.991 | | 0.998 |
| WB | −0.993 | −0.993 | −0.979 | 0.994 | 0.997 | |

¹ The models used were: CSL = binary cross sectional linear model, THR = binary threshold (probit) model, LIN = time until death linear model, SS = survival score (logit) model, COX = Cox proportional hazard frailty model and WB = Weibull proportional hazard frailty model.

² A total of 118 families were included in the analysis.

Table 4

Estimated Pearson correlation coefficients¹ (r_{EBV}) between the predicted family² EBVs (r_{EBV}) from two test tanks and two subsets of data and accuracy of selection³ (r_T) using six statistical models.

| Models ⁴ | Tank | | Subset | |
|---------------------|-----------|-------|-----------|-------|
| | r_{EBV} | r_T | r_{EBV} | r_T |
| CSL | 0.508 | 0.712 | 0.571 | 0.755 |
| THR | 0.512 | 0.715 | 0.578 | 0.760 |
| LIN | 0.613 | 0.782 | 0.617 | 0.785 |
| SS | 0.557 | 0.746 | 0.608 | 0.779 |
| COX | 0.620 | 0.787 | 0.648 | 0.804 |
| WB | 0.588 | 0.766 | 0.624 | 0.789 |

¹ All correlations were significantly different from zero ($P < 0.0001$).

² A total of 118 families were included in the analysis.

³ Accuracy of selection (r_T) = $\sqrt{r_{EBV}}$.

⁴ The models used were: CSL = binary cross sectional linear model, THR = binary threshold (probit) model, LIN = time until death linear model, SS = survival score (logit) model, COX = Cox proportional hazard frailty model and WB = Weibull proportional hazard frailty model.

The Pearson correlation coefficients among full-sib family EBVs from two test tanks and two data subsets, and accuracy of selection are presented in Table 4. The highest correlations and accuracies of selection between two tank replicates data subsets and the two random data subsets were achieved with COX model ($r_T \approx 0.79$ and 0.8, respectively). Moreover, the latter behaved very similar to WB and LIN with respect to accuracy of selection (with r_T values ranging between 0.77 and 0.79). The SS model also showed a high accuracy of selection when calculated from the two random data subsets ($r_T \approx 0.75$ and 0.78, for the two tank replicates data subsets and the two random data subsets, respectively). The lowest correlations and accuracies of selection were obtained with the CSL and THR models. In addition, accuracies were very similar between these two models ($r_T \approx 0.71$ and 0.72 for CSL and THR and $r_T \approx 0.76$ for both, when using the two tank replicates data subsets and the two random data subsets, respectively).

4. Discussion

Injured skin and/or gills are the most likely routes of entry of *P. salmonis* to the host in a natural infection. Subcutaneous inoculation of the agent can cause similar levels of mortality as IP injection and when both ways are compared against a skin patch infection, the mortality can only reach around a half in the latter, suggesting that skin injuries are fundamental in the severity of the disease (Smith et al., 1999). Besides, in the final stages of the disease, the pathological signs become similar independently of the way of entry, most likely because of the systemic nature of this infection (Cvitanich et al., 1991). Therefore, we assume that although IP injection may not be the normal route of infection, this method may both imitate the effects of the natural way of entry of the pathogen, in which an injury is required for establishing the infection, and allows a standardization of the experiment in practical terms. However, more studies are necessary to elucidate if genetic resistance mechanisms involved in IP injection are similar to those implicated in natural infection.

The detection of significant genetic variation for resistance to *P. salmonis* and the estimation of moderate to high heritability values using different trait definitions and statistical models are in accordance with other studies that also have found significant genetic variation and similar intermediate values of heritabilities for resistance against other bacterial diseases in farmed salmonid populations (Gjedrem et al., 1991; Gjøen et al., 1997; Ødegård et al., 2006; Perry et al., 2004; Withler and Evelyn, 1990).

The differences between the estimated heritabilities for resistance are certainly due to differences in trait definition and analysis. The lower value of the heritability calculated for SS in comparison with other models was expected due to the split of information into several

days across the test period. This has also been shown by other studies for resistance to furunculosis in Atlantic salmon (Ødegård et al., 2006) and white spot syndrome virus in shrimp (Gitterle et al., 2006). Although, the cumulative rate of mortality did not approach 50%, level which maximizes the phenotypic variance of a binary trait, significant and moderated heritability values were found when TS was analyzed using both CSL and THR models. Furthermore, both models behave very similar in terms of ranking of families (correlation between family EBVs ~ 1) and the accuracy of selection, indicating a low gain by including a probit function for taking the binary nature of the trait into account when analyzing TS. A similar heritability value was obtained for both linear models LIN and CSL, in spite of the fact that they used a different trait definition (DD and TS, respectively). However, a slightly higher accuracy of selection was obtained with LIN and the ranking of families was somewhat different when compared with CSL. This result indicates a better use of the information when fitting a linear model, using day of death as trait definition compared to the overall test period survival, in this particular case.

The difference in the heritability estimates between COX and WB models was most likely due to differences in the distribution from the baseline hazard function and the parameters (i.e. mode, mean, and standard deviation) calculated from the marginal posterior distribution of these models (data not shown). However, these differences are considerably less marked than those reported by Gitterle et al. (2006), study in which, for example, the modes of the posterior distribution were 0.014 and 0.41 for COX and WB, respectively. In addition, differences in ranking of families were small between the use of COX, WB and SS, indicating a minor effect in the determination of genetically resistant families. When both time to death and censored data were taken into account using COX and WB proportional hazard frailty models, accuracy of selection was very marginally higher compared to LIN and SS models, indicating a minor gain of using COX and WB in this case.

In general terms, COX, WB, LIN and SS performed very similar and slightly better than THR and CSL regarding the accuracy of selection. These results indicate that models which take time to death into account are more precise than models which use a binary trait definition. Similar results have been shown by other studies for disease resistance in shrimp (Gitterle et al., 2006) and Atlantic salmon (Ødegård et al., 2006, 2007). Nevertheless, these findings using survival challenge test data of IP infected *P. salmonis* animals have to be validated with survival data from natural infection under field conditions.

In the present study we demonstrated that significant additive genetic variation for resistance against piscirickettsiosis exists in Atlantic salmon. The presence of low to moderate heritabilities for *P. salmonis* resistance indicates that selective breeding for these traits can be successfully applied in the studied population. Finally, more studies aiming to determine the epidemiological consequences of improving resistance are needed in order to establish the guidelines of the long-term control of infectious diseases in farmed salmon populations.

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References

- Cvitnich, J.O., Garate, N., Smith, C., 1991. The isolation of a rickettsia-like organism causing disease and mortality in Chilean salmonids and its confirmation by Koch's postulate. *J. Fish Dis.* 14, 121–145.
- Ducrocq, V., Casella, G., 1996. A Bayesian analysis of mixed survival models. *Genet. Sel. Evol.* 28, 505–529.
- Ducrocq, V., Sölkner, J., Mészáros, G., 2010. Survival kit v6 — a software package for survival analysis. Proceedings of the 9th World Congress on Genetics to Livestock Production, August 1–6, 2010, Leipzig, Germany.
- Fryer, J., Hedrick, R., 2003. *Piscirickettsia salmonis*: a Gram-negative intracellular bacterial pathogen of fish. *J. Fish Dis.* 26, 251–262.
- Gilmour, A., Gogel, B., Cullis, B., Thompson, R., Butler, D., Cherry, M., Collins, D., Dutkowski, G., Harding, S., Haskard, K., 2009. ASReml User Guide Release 3.0. VSN International Ltd., UK 275 (<http://www.vsnl.co.uk>).
- Gitterle, T., Ødegård, J., Gjerde, B., Rye, M., Salte, R., 2006. Genetic parameters and accuracy of selection for resistance to White Spot Syndrome Virus (WSSV) in *Penaeus (Litopenaeus) vannamei* using different statistical models. *Aquaculture* 251, 210–218.
- Gjedrem, T., 2000. Genetic improvement of cold-water fish species. *Aquac. Res.* 31, 25–33.
- Gjedrem, T., Gjoen, H., Gjerde, B., 1991. Genetic origin of Norwegian farmed Atlantic salmon. *Aquaculture* 98, 41–50.
- Gjedrem, T., Robinson, N., Rye, M., 2012. The importance of selective breeding in aquaculture to meet future demands for animal protein: a review. *Aquaculture* 350–353, 117–129.
- Gjerde, B., 2005. Mating design. In: Gjedrem, T. (Ed.), *Selection and Breeding Programs in Aquaculture*. Springer, Dordrecht, The Netherlands, pp. 178–182.
- Gjøen, H.M., Refstie, T., Ulla, O., Gjerde, B., 1997. Genetic correlations between survival of Atlantic salmon in challenge and field tests. *Aquaculture* 158, 277–288.
- Kaplan, E.L., Meier, P., 1958. Nonparametric estimation from incomplete observations. *J. Am. Stat. Assoc.* 53, 457–481.
- Lynch, M., Walsh, B., 1998. *Genetics and Analysis of Quantitative Traits*. Sinauer Associates Sunderland, MA.
- Ødegård, J., Olesen, I., Gjerde, B., Klemetsdal, G., 2006. Evaluation of statistical models for genetic analysis of challenge test data on furunculosis resistance in Atlantic salmon (*Salmo salar*): prediction of field survival. *Aquaculture* 259, 116–123.
- Ødegård, J., Olesen, I., Gjerde, B., Klemetsdal, G., 2007. Evaluation of statistical models for genetic analysis of challenge-test data on ISA resistance in Atlantic salmon (*Salmo salar*): prediction of progeny survival. *Aquaculture* 266, 70–76.
- Ødegård, J., Baranski, M., Gjerde, B., Gjedrem, T., 2011. Methodology for genetic evaluation of disease resistance in aquaculture species: challenges and future prospects. *Aquac. Res.* 42, 103–114.
- Perry, G.M.L., Tarte, P., Croisetiére, S., Belhumeur, P., Bernatchez, L., 2004. Genetic variance and covariance for 0+ brook charr (*Salvelinus fontinalis*) weight and survival time of furunculosis (*Aeromonas salmonicida*) exposure. *Aquaculture* 235, 263–271.
- Smith, P., Pizarro, P., Ojeda, P., Contreras, J., Oyanedel, S., Larenas, J., 1999. Routes of entry of *Piscirickettsia salmonis* in rainbow trout *Oncorhynchus mykiss*. *Dis. Aquat. Org.* 37, 165–172.
- Stear, M., Bishop, S., Mallard, B., Raadsma, H., 2001. The sustainability, feasibility and desirability of breeding livestock for disease resistance. *Res. Vet. Sci.* 71, 1–7.
- Withler, R., Evelyn, T., 1990. Genetic variation in resistance to bacterial kidney disease within and between two strains of coho salmon from British Columbia. *Trans. Am. Fish. Soc.* 119, 1003–1009.
- Yáñez, J.M., Martínez, V., 2010. Genetic factors involved in resistance to infectious diseases in salmonids and their application in breeding programmes. *Arch. Med. Vet.* 42, 1–13.