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# Survival and life history differentiation in Sympatric European Grayling populations in Lesjaskogsvatnet

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## Abstract

The present study entitled "Survival and life history differentiation in Sympatric European Grayling populations in Lesjaskogsvatnet" deals with individual growth trajectories and adult mortality differences among grayling populations in Lesjaskogsvatnet.

During this study 731 grayling scale samples from five different streams Steinbekken, Sandbekken, Søre Skotte, Valåe and Søre Hyrjon Lesjaskogsvatnet were taken. These five streams are the tributaries of Lake Lesjaskogsvatnet which is the feeding site for all the mature fish from different streams. Steinbekken, Sandbekken, Søre Skotte are warm streams and small whereas Valåe and Søre Hyrjon are cold and large.

The grayling from warm streams grow rapidly up to 58 mm in the first winter and the slow growing grayling is up to 47 mm, which is less by 19 percent. The present studies show that life spans of warm stream fishes are shorter than the fish in the cold streams.

Among the five streams, highest growth rate occurs in Sandbekken which is 31%, 28% and 29 % in male female and both sexes respectively. The lowest growth rate seems to occur in Shyrjon for male i.e. 21% and for female and both sexes in Sørskottåe i.e. 22 %.

The annual mortality rate of female is higher in Sandbekken i.e. 88.24 and male in Valåe i.e. 74.78. The lowest mortality rate occurs in Shyrjon i.e. 60.78 for female and for male, in Steinbekken i.e. 50.86.

In this study the warm water fishes has high growth rate and high mortalities in female and in male the mortality rate does not depends upon the water temperature.


Keywords: catch curve, parameter, mortality, spawning, growth trajectories

# Contents

# 1. Introduction

Populations within species are often separated from each other geographically in varying degrees which may result in reproductive isolation (Mooi, 2009). Variations in local - environmental conditions may result in adaptive variation of morphological characteristics, life history strategies, physiology and behaviour (Taylor, 1991). Fish are exothermal in nature which changes their body temperature according to the surrounding environment. The overall growth and development of the fish is determined by various factors among which the temperature of water is the most important one (Taylor, 1991).

The salmonid fish grayling (*Thymallus thymallus*) needs well-oxygenated water with minimum requirement of 5-7 ppm. They can tolerate the temperature maximum up to 25°C and prefers 18°C. In a study of local grayling populations in the Lesja region of Norway, the local European grayling populations living in warm streams survived increasingly better under increasing temperatures and individuals from cold-water populations survived less well under decrease in temperature (T. Haugen & Vøllestad, 2000).

This study involves data from European grayling populations that have been studied in detail over the last decades (Barson, Haugen, Vøllestad, & Primmer, 2009; Bass, Haugen, & Vøllestad, 2014; Junge et al., 2011; Kavanagh, Haugen, Gregersen, Jernvall, & Vøllestad, 2010; Koskinen, Haugen, & Primmer, 2002; TO, 2000). Due to natural as well as human introduction in intensive fisheries, the grayling population is flourishing (Northcote, 1995). The grayling populations in the Lesjaskogsvatnet immigrated from the river Gudbrandsdalslågen before last century (Øygarden, 2013) and has colonized all inhabitable tributaries since. These tributaries vary a lot in many environmental conditions, but, importantly, to a large extent in spring water temperatures (Figure 1).

## 1.1 Life Cycle

Grayling are spring spawners that migrate to streams and rivers after the melting of ice..  The larvae about 12-18 mm of length after hatching swims freely in the shallow water of the streams. The grayling can attain growth up to maximum 45 cm length (Leith, 2007). The fish becomes adult in the feeding area from where their parents come.

## 1.2 Spawning behavior:

The grayling fish during their spawning time migrate from slow to fast flowing river. The lake population migrates towards the streams during the spawning period (Northcote, 1995). In clear water, low fluctuation of water level and low water level the movement of fish is higher than in turbid water, high fluctuation and higher water level respectively. Due to increase in temperature of the surrounding, the spawning time of grayling increases by 3-4 weeks earlier on the basis of analysis of collected data from 1948-2009. The shifting of spawning was due to the increase of water temperature. The study result indicates the declining of the population abundance with the increase in the temperature (Wedekind & KUeNG, 2010). As many other salmonids, the life history of grayling is similar having specific spawning site. The feeding site of grayling fish is larger water body which moves with member of own species as well as other fish species (Kristiansen & Dølving, 1996) .

## 1.3 Age Determination in fish

The age determination helps to know the biology of fish and different activities of fish population. The age determination of fish was demonstrated for the first time in 1898 in Carp fish, *Cyprinus carpio*. Then after for 30 years the studied were limited only in the marine fishes. In North America the scales are used to determine the age of fish in 1930 for fresh water.(Carlander, 1987). The daily growth rings in the scales of fishes are the important discovery in age of fish from 1970 (Beamish, 1987). The estimation of age in fish helps in the fisheries management which helps to increase the fish population. The accurate estimation of age declines with increase in age. The concentric rings can be easily observed in the young fish but in older fish the rings are densely packed. In fisheries management the incorrect method should be considered (Horká, Ibbotson, Jones, Cove, & Scott, 2010).

The annulus formation in the scales helps to determine the age of fish .When the fish attains the size of 33.5-37 mm, the scales are formed. When the age of fish reaches the age of 5 the annuli are difficult to read as they are closely packed together. With increasing the age the annuli are tightly packed. The fish growth is rapid until they are sexually mature at the age of 3 years. Fish growth occurs in spring to autumn and very less in winter. During winter season the growth is very less and hence the annuli are densely packed. The maximum growth of the fish takes place in summer and $1/10^{th}$ part in other seasons (Ingram, Ibbotson, & Gallagher, 1999).

## 1.4. Growth and mortality:

In the studies performed in fresh water fish brown trout (Paulsen, 2014), the warmer temperature increases the metabolic rate. Thus, temperature plays an important role in the life history traits. In this study the four populations of brown trout in hatchery with same temperature was performed in controlled condition. The result shows that the fish from cold climate has the highest growth rates.
The growth and mortality are used in fisheries population dynamics to know the status of fish. The size of the fish and its length is the growth rate whereas the death of fish in particular population termed as mortality rate. The mortality of fish could be natural or by the fishing activities. The studies conducted on the five different populations on growth and survival effects on maturation in Norway shows that the mortality rate is higher in the population which mature early. The extensive fishing affects the fish which mature early leading high mortality. The mortality of the immature fish is largely due to fishing pressure (T. O. Haugen, 2000).

The growth rate of grayling fish is variable and male grows slightly faster than the female. The male have better feeding than the females due to its dominant nature which helps them for better growth. When the flow of water is less, the growth rate decreases in fish. The mortality rate is higher in large fish due to fishing activities. The studies show that, when the water flow is less the fish did not spawn in its traditional spawning area. The fish lives up to 20 years but the studies in Beaverlodge River shows that the fish were only from 2 years old to nine years old. The declining survival rate of grayling population is due to over fishing and natural mortality of adult fish (Carl, Walty, & Rimmer, 1992).The stress in the organisms results in the  slow growth which affects the maturity which leads to less fitness (Stearns & Crandall, 1984). In this study I expect the temperature plays an important role in the growth of fish. I expect the fish in warm streams to have highest growth rate and short life span as compared to cold streams. The adult mortality is expected to be higher in the grayling having highest growth rate and less in slow growing fish.

In this study I focus on the sympatric grayling fish of five different streams of which Steinbekken, Sandbekken, Søre Skotte are small and warm whereas Valåe and Shyrjon are large and warm. In this study I will look the first year growth of fish from different streams and the adult mortalities of fish with respect to temperature.

## 2. Objectives

i. To determine and compare individual age and growth trajectories from scale readings in populations of grayling in Lesjaskogsvatnet.

ii. To estimate and compare adult mortality differences among grayling populations in Lesjaskogsvatnet with respect to thermal regimes.

# 3. Materials and Methods

## 3.1 Study species: The European grayling (*Thymallus thymallus)*

The European grayling is a freshwater fish species that belongs to the family Salmonidae. The body of the fish is streamlined with large dorsal fin. Males and females can be easily distinguished, as dorsal fin is larger in males than of females. The maximum size is up to 60 cm in length, but varies largely among populations (Northcote 1995).

Naturally the European grayling is native to Europe where it occurs in most parts like France, England, Norway, Finland and Sweden, northern parts of Ural mountain and also in the northern part of Russia (Northcote, 1995). In Norway, grayling are distributed in south-eastern part and in the river system of south eastern part and in eastern parts of Finnmark County (Kvinge, 2014).

Naturally, the spawning of grayling takes place in the spring season shortly after ice out at water temperature 4-5 °C and during early summer. The spawning activities are mostly active during day time when the temperature is at maximum and if the temperature is suitable during night, the spawning activities could take place.  Light plays an important role in the courtship and territorial behavior as the grayling does not spawn in the turbid water (Parkinson, Philippart, & Baras, 1999).

The mature grayling migrates from the lakes to the streams for spawning. The males are aggressive during the spawning time fighting over access to the better spawning habitat. The eggs are deposited few centimeters under the gravel during the spawning period. Hatching of eggs takes place at 140-200 degree days (accumulated temperature sum) during day time. The larvae, after emerging from the gravel, fill the air bladder with air for short time and remain in a mid-water position. The free-swimming juvenile fish immediately swim downstream or they may remain for 1-1.5 months in the streams (Haddeland, Junge, Serbezov, & Vøllestad, 2015).

Spawning takes place in shallow water of depth 10-110 cm with high velocities of water 40-70 cm $s^{-1}$and gravel in the substratum 16-32 mm (Mari Nykänen, 2004). The larvae stay in the gravel until yolk sac is resorbed after which they emerged from the gravel. The free-swimming larvae prefer the dead zone close to the rivers during the early summer period. The habitat preference changes during ontogeny of the grayling fry and is different in summer and winter. In summer season, the fry prefer the shallow water with high velocity and coarse substratum but not in winter. In summer season, the fish needs high energy. The fast water current helps the fish to reach the large number of prey. During winter season the energy should be minimized. The deep water habitat of the fish helps to protect the fish from the frozen ice as well as predators  (M Nykänen, Huusko, & Lahti, 2004). For grayling populations that inhabit lakes, the juveniles leave the nursery streams during/by end of first summer (Haugen 2000) – leading to a mere 2-3 month of environmental segregation during the life in sympatric lake-dwelling grayling populations.

## 3.2 Study area

This study was conducted in the lake Lesjaskogsvatnet (Figure 1) which is situated 611 metre above the sea level in western parts of central Norway. The maximum depth of the lake is 24 metre with an area of 4.52 $km^2$ (http://vann-nett.no/). The lake is shallow sub-alpine lake with an average depth of 10 metres. The grayling species colonized the lake at the end of $19^{th}$ century  during a period where construction work gave access to the lake for down-stream grayling in the river Gudbrandsdalslågen (T. O. Haugen & Vøllestad, 2001).

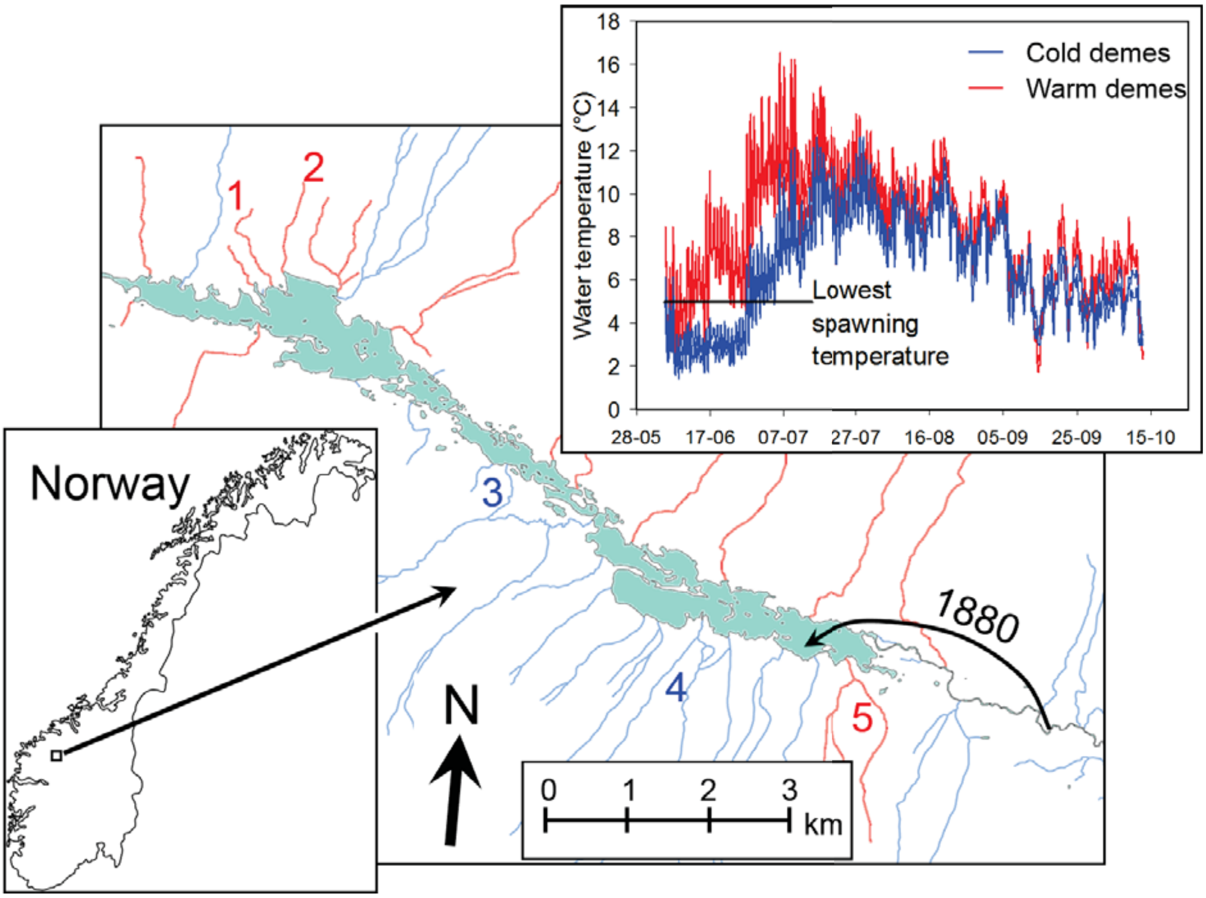



**Figure 1**. Study location map with warm and cold tributaries shown in red and blue colors, respectively. Daily average water temperature trajectories for warm and cold streams are show up right. Study populations are given as: 1- Søre Skotte,2- Steinbekken, 3- Søre Hyrjon , 4-Valåe, 5 - Sandbekken. (Source figure (Bærum, 2008))

There are many streams that empty into Lesjaskogsvatnet. The grayling use most of these streams as spawning- and nursery habitats. The northern part of the lakes receives more sunlight than the southern slopes. As a result the streams are classified as large and cold (LC) and small and warm (SW). Steinbekken (STE), Sandbekken (SAN), Søre Skotte (SSKO), Valåe(VAL) and Søre Hyrjon (SHYR) are the tributaries of the lake from where the fish were sampled in this study. Steinbekken and Sandbekken are classified as small and warm (SW) and similarly Søre Hyrjon and Valåe are classified as large and cold (LC) (Gregersen, Haugen, & Vøllestad, 2008).

**Table 1**: Environmental characteristics of five spawning tributaries (Gregersen, Haugen, & Vøllestad, 2008)

| Tributary | Tributary width (m) | Tributary type | Mean June temperature |
|---|---|---|---|

| | | | (°C) |
|---|---|---|---|
| Steinbekken | 0.75 | Small/ warm | 6.05 |
| Sandbekken | 1.0 | Small/ warm | 6.05 |
| Søre Skotte | 1.25 | Small/ warm | 6.0 |
| Valåe | 6.0 | Large/ cold | 2.9 |
| Søre Hyrjon | 5.0 | Large/ cold | 3.4 |

## 3.3 Sample collection

The mature grayling specimens were captured in 2010 from the five study tributaries of Lesjaskogsvatnet. The samples were collected using fyke nets (Figure 2) that were emptied several times per day. The fish were anesthetised using 5ml of benzocaine in 10 litres of water. The fork lengths of each individual were measured and distinguish their sexes. 4-5 scales of each fish were retrieved and kept in paper envelopes with sample ID (Figure 3). During the present study 731 individuals is studied. The captured fish were tagged (using PIT tags) and released right upstream from the respective fish trap.

Figure 2. Fyke nets used to sample mature grayling during the spawning run.

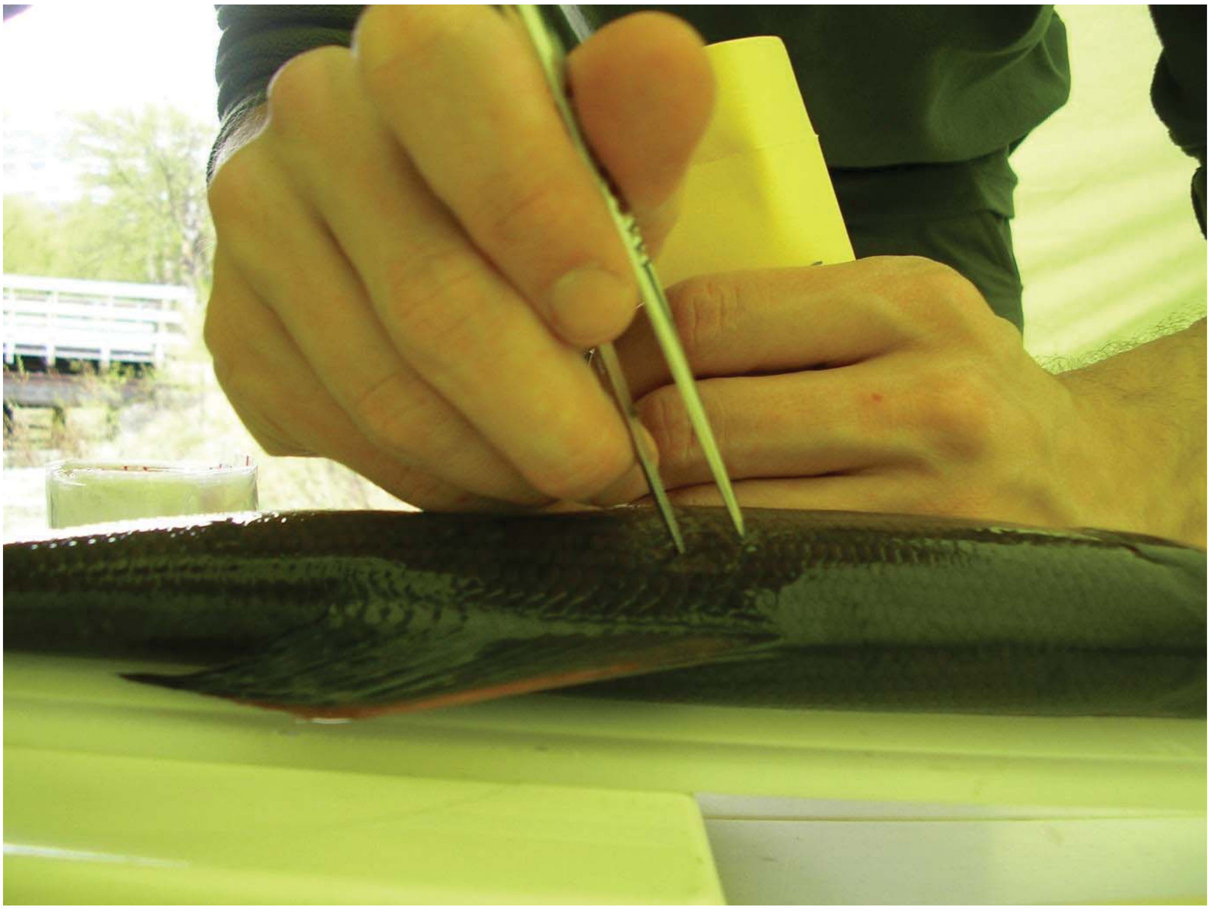


Figure 3. Scale sampling from mature grayling. The fish is anesthetized and released after sampling scales and application of PIT-tag.

### 3.4 Scale analysis

The scales samples were analyzed at the image centre and the MINA Ecology lab of NMBU. In the image centre, stereo microscope was used for scale reading. The scales having small central plates are used in analysis and the replacement scales are not used. The digital camera Leica DFC 320 was used in the microscope to take the photographs of the scales.


Figure4. Digital camera Leica DFC 320 used in the microscope

The photographs of the scales were analysed by using the software Image Pro Express in the laboratory. The total length of the scale from the centre was noted and marked as `Y` and each winter edge as ´V´. After marking the data were exported to an excel sheet and the snapshot of the scales were taken.

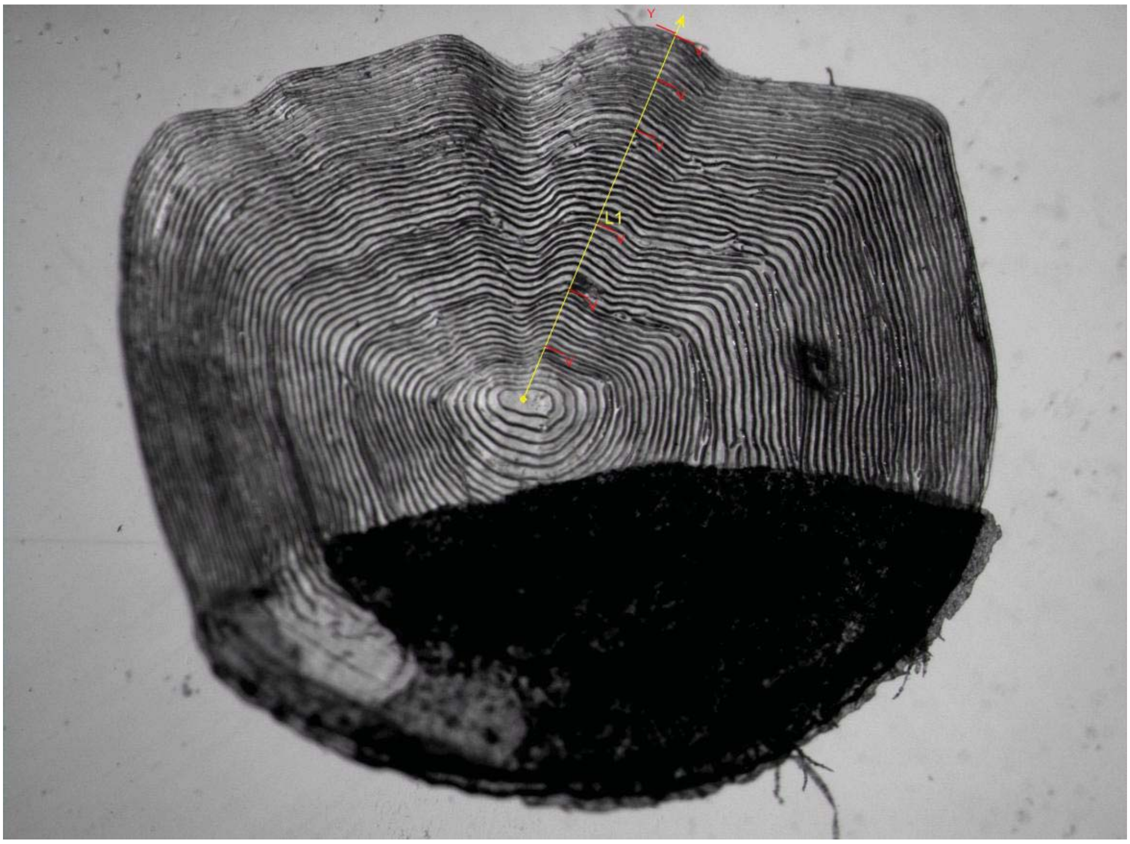


Figure 5: Individual STE10053 scale showing the V (winter edge) and Y (total length of scale)

The dark areas, where the circuli are close together, represent the winter and the light areas represents the summer where the circuli are far apart. On the basis of winter and summer growth zones, the age of the fish and the scale growth of every year were estimated. On the basis of the growth zones, the growth size of the fish were back calculated for individual fish assuming that the ratio between body size and scale radius is constant throughout entire life (Jonsson & Stenseth, 1976; Nygård, 2012; Spannhof, 1989).

## 3.5 Statistical Analyses

The data of the sample analyzed with the help of image pro express in the excel sheet is analyzed with the help of R software (R Core Team 2016). The measurement error was assessed from estimates on within-individual variation in randomized repeated measurements of 10 individual scales. The variance was scaled on the mean value of these repeated measurements - i.e. the variance coefficient. Also mention that these CV's were analyse in a linear mixed effects model (Zuur, Ieno, Walker, Saveliev, & Smith, 2009) with intercept as fixed effect and repeat samples and random effect.

. The data is analyzed by using different statistical methods.

i. Linear mixed effects models
ii. Generalized linear models
iii. Log likelihood ratio test
iv. Chi square test

**Linear mixed effects models**: This model approach combines fixed effects and random effects to simultaneously model both mean and variance components of the data (Zuur et al 2009). The difference in back-calculated length at end of first winter (i.e., during the stream-dwelling phase) and measurement error in scale reading was analysed using this modelling approach. To select among candidate models AICc was used as a criteria (Burnham,K.P K. P. & Anderson, D. R. Model selection and multimodel inference: a practical information-theoretic approach Springer, 2002)

Back-calculations were based on the Lea-Dahl method assuming a proportional growth of scales and body length(Ricker, 1992) .

The relationship between body and scale with the length of fish at capture $\mathbf{L_c}$, age of fish **c** and distance from the centre of the scale to edge termed as $\mathbf{S_c}$. The relation is assumed to be linear where $L_i$ of fish at age i where i≤ c and the equation can be given as

Li=Si/Sc*Lcatch , Lcatch=body length at capture (Weisberg & Frie, 1987).

This formula is used to estimate back-calculated lengths.

**von Bertalanffy growth model**: The individual back-calculated growth pattern of grayling will be fitted using the von Bertalanffy model. This model will be useful in the studies of length-growth studies of grayling population. The three parameters $L_\infty$, K and $t_0$ are used in the von Bertalanffy equation.

$$L = L_\infty [1 - \exp(-K(t - t_0))]$$

Where $L_\infty$ = maximum length

K = growth rate

$t_0$ = theoretical age at zero length.

To test and quantify differences in age structure among sexes and populations, I fitted candidate generalized linear models based on the age count data under a Poisson distribution (using a log-link). Model selection among the candidate models was undertaken by means of AICc (Burnham and Anderson 2002).

**Catch curves:** Catch curve represents the catch of the fish by age and length. In this study catch curve will helps in estimating the mortality of fish. The age distribution in fish population assumed to be in geometrical ratio. The yearly survival rate of the fish can be calculated with the help of catch curves (Chapman & Robson, 1960).Most statistics will be conducted using the FSA package in R (Ogle 2017). In addition glm in the base package was used for generalized linear models and lme4-library for linear mixed effects models. Model selection was performed using the AICcmodavg-package

# 4. Results

## 4.1 Measurement error

There was a large variation both between and within individuals for the repeated R1-readings (Figure 6). On average the CV for R1 was 0.081±0.067 (SD).

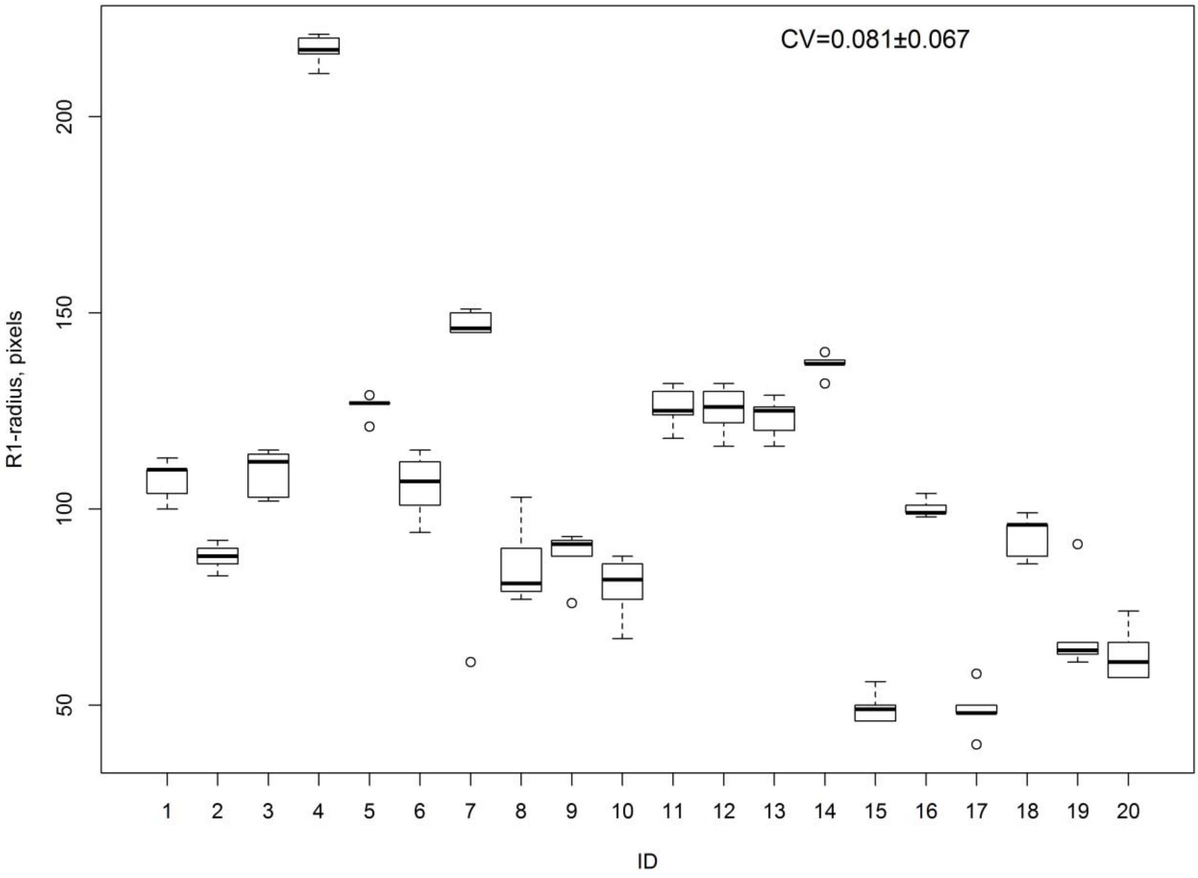



Figure 6. Boxplot of randomized repeated measurements of the R1-radius from 20 randomly selected individuals. The thick horizontal line within the boxes represent the median values. 50% of the observations are within the boxes and 90% between the whiskers.

There was a large variation both between and within individuals for the repeated R2-readings (Figure 7). On average the CV for R1 was 0.041±0.037 (SD)

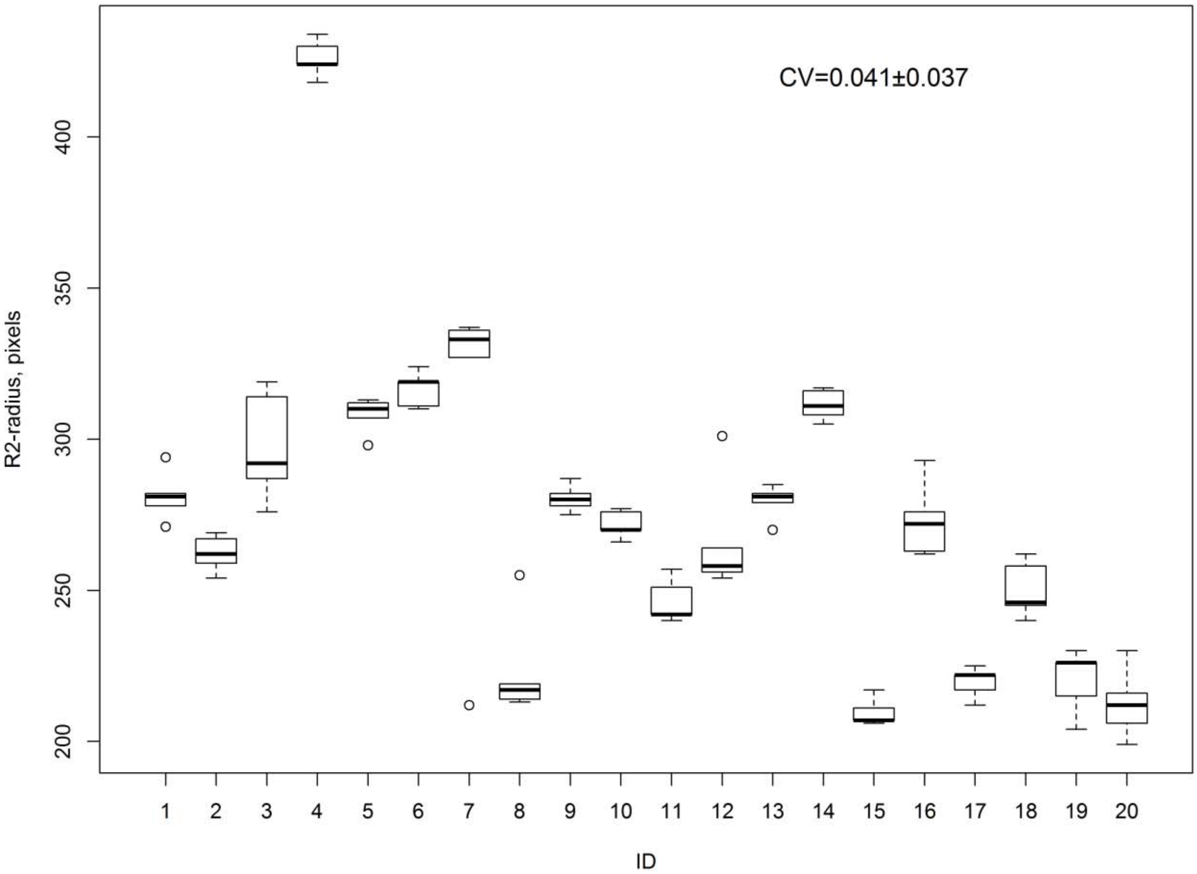



Figure 7. Boxplot of repeated measurements of the R2-radius from 20 randomly selected individuals. The thick horizontal line within the boxes represent the median values. 50% of the observbations are within the boxes and 90% between the whiskers.

There was a large variation both between and within individuals for the repeated R1-readings (Figure 8). On average the CV for R3 was 0.025±0.024 (SD)

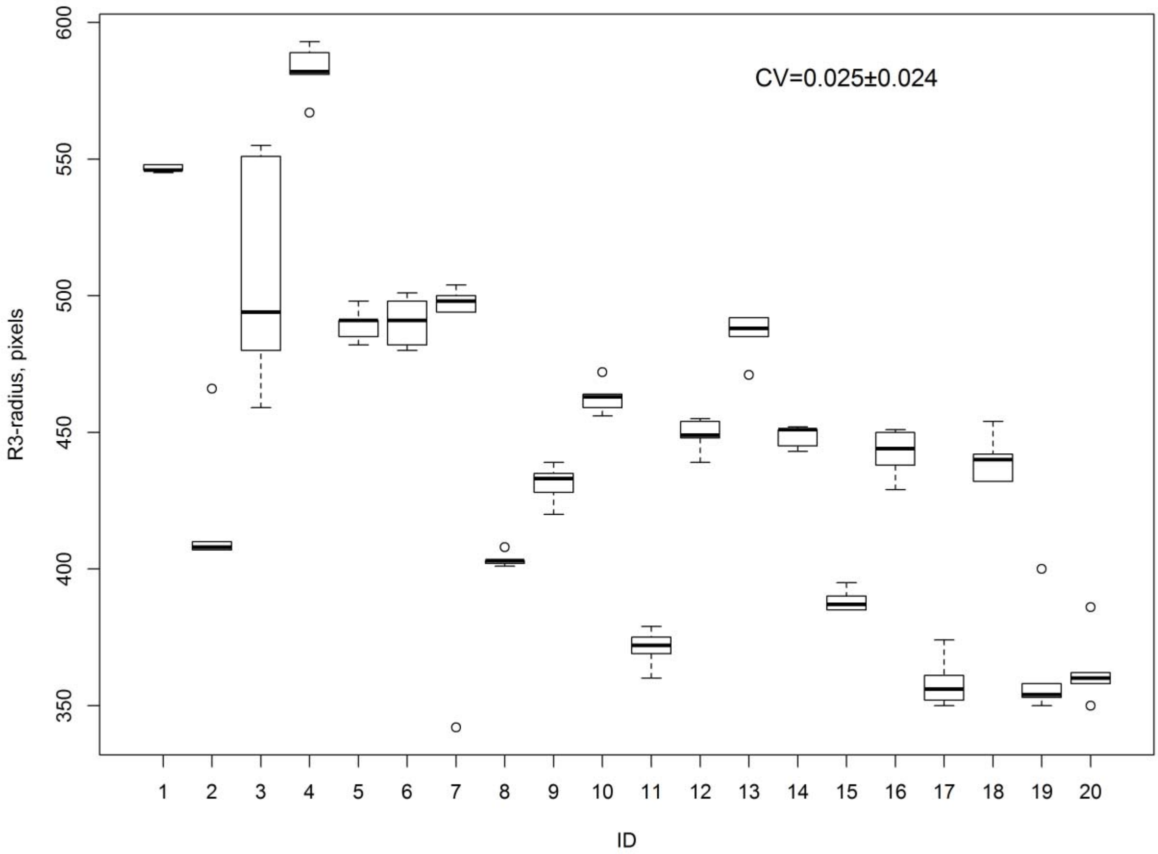



Figure 8. Boxplot of repeated measurements of the R3-radius from 20 randomly selected individuals. The thick horizontal line within the boxes represent the median values. 50% of the observbations are within the boxes and 90% between the whiskers.

There was a large variation both between and within individuals for the repeated Rtot-readings (Figure 9). On average the CV for Rtot was 0.015±0.014 (SD).

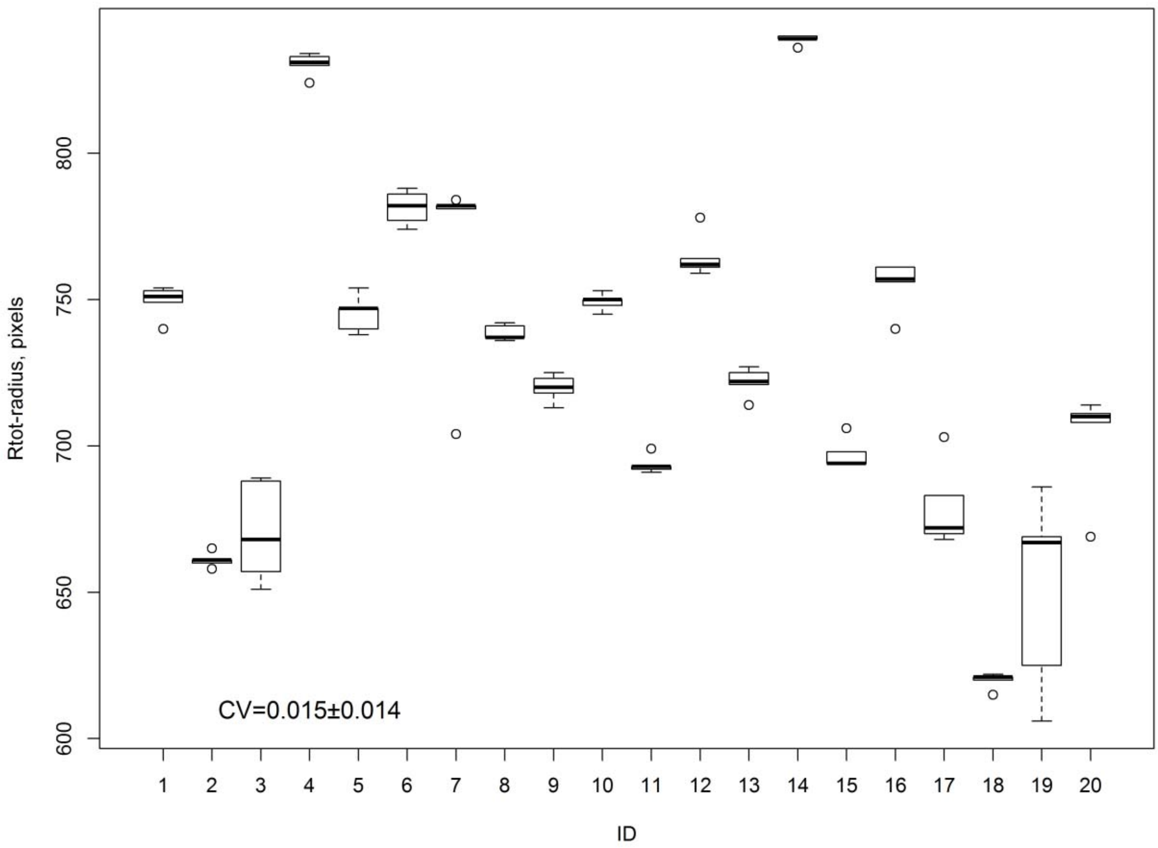



Figure 9. Boxplot of repeated measurements of the Rtot-radius from 20 randomly selected individuals. The thick horizontal line within the boxes represent the median values. 50% of the observbations are within the boxes and 90% between the whiskers

## 4.2 Measurement error analysis

As for the manually calculated measurment errors the CVs declined with increasing age of radius, but the statistically estimated CVs were generally larger than the manually calculated ones (Table 2).

Table 2. Parameter estimates for the linear mixed effects models fitted the repeated measuremten scale radius data.

| | Fixed effect | | Variances | | | |
|---|---|---|---|---|---|---|
| Response | Mean | SE | within-ID | Among-ID | Est CV | Manually calculated CV |
| R1 | 103.62 | 8.41 | 10.81 | 37.29 | 0.104 | 0.081 |
| R2 | 273.28 | 11.15 | 15.65 | 49.35 | 0.057 | 0.041 |
| R3 | 445.53 | 13.75 | 21.07 | 60.76 | 0.047 | 0.025 |
| Rtot | 726.77 | 12.69 | 13.4 | 56.45 | 0.018 | 0.015 |

## 4.3Back-calculated growth differences

The model selection performed to find the most supported candidate model for predicting size at end of first winter

Table 3. Model selection table (based on AICc) showing AIC rank of 12 candidate models fitted to estimate L1 in 5 Lesjaskogsvatn grayling populations. K= number of parameters,

| Model | K | AICc | ΔAICc | AICcWt | Cum.Wt | LL |
|---|---|---|---|---|---|---|
| Stream+Age | 7 | 162.81 | 0.00 | 0.28 | 0.28 | -74.32 |
| Temp+Age | 4 | 163.06 | 0.24 | 0.25 | 0.53 | -77.50 |
| Temp*Age | 5 | 163.14 | 0.33 | 0.24 | 0.77 | -76.52 |
| Type+Age | 4 | 165.19 | 2.38 | 0.09 | 0.85 | -78.57 |
| Type*Age | 5 | 165.52 | 2.71 | 0.07 | 0.93 | -77.72 |
| Temp | 3 | 167.54 | 4.73 | 0.03 | 0.95 | -80.75 |
| Stream | 6 | 168.64 | 5.83 | 0.02 | 0.97 | -78.26 |
| Stream+Sex | 7 | 168.74 | 5.93 | 0.01 | 0.98 | -77.29 |
| Stream*Age | 11 | 169.07 | 6.26 | 0.01 | 1.00 | -73.34 |
| Stream*Age*Sex | 21 | 171.87 | 9.06 | 0.00 | 1.00 | -64.23 |
| Stream*Sex | 11 | 174.65 | 11.84 | 0.00 | 1.00 | -76.12 |
| Age | 3 | 234.64 | 71.83 | 0.00 | 1.00 | -114.30 |

The stream Steinbekken, Valåe and age is statistically significant (Table 4).The age of fish depends upon the water temperature of the streams and streams as well.

Table 4. Parameter estimates for the most supported model (Table 3) predicting back-calculated length at end of first winter as function of age and stream.

| | Estimate | Std. Error | t value | Pr(>\|t\|) | |
|---|---|---|---|---|---|
| (Intercept) | 4.02724 | 0.07411 | 54.340 | < 2e-16 | *** |
| StreamSHyrjon | -0.07738 | 0.05095 | -1.519 | 0.129284 | |
| StreamSteinbekken | 0.07868 | 0.03940 | 1.997 | 0.046238 | * |
| StreamSørskottåe | 0.01873 | 0.04750 | 0.394 | 0.693502 | |
| StreamValåe | -0.14627 | 0.03881 | -3.769 | 0.000178 | *** |
| Age | -0.03718 | 0.01328 | -2.801 | 0.005245 | ** |

The back-calculated length at end of first winter decreases with catch age. The growth of fish in Valåe is poor and less which is about 47mm whereas the growth of fish in Steinbekken is good as compare to others which is about 58mm (fig 10).

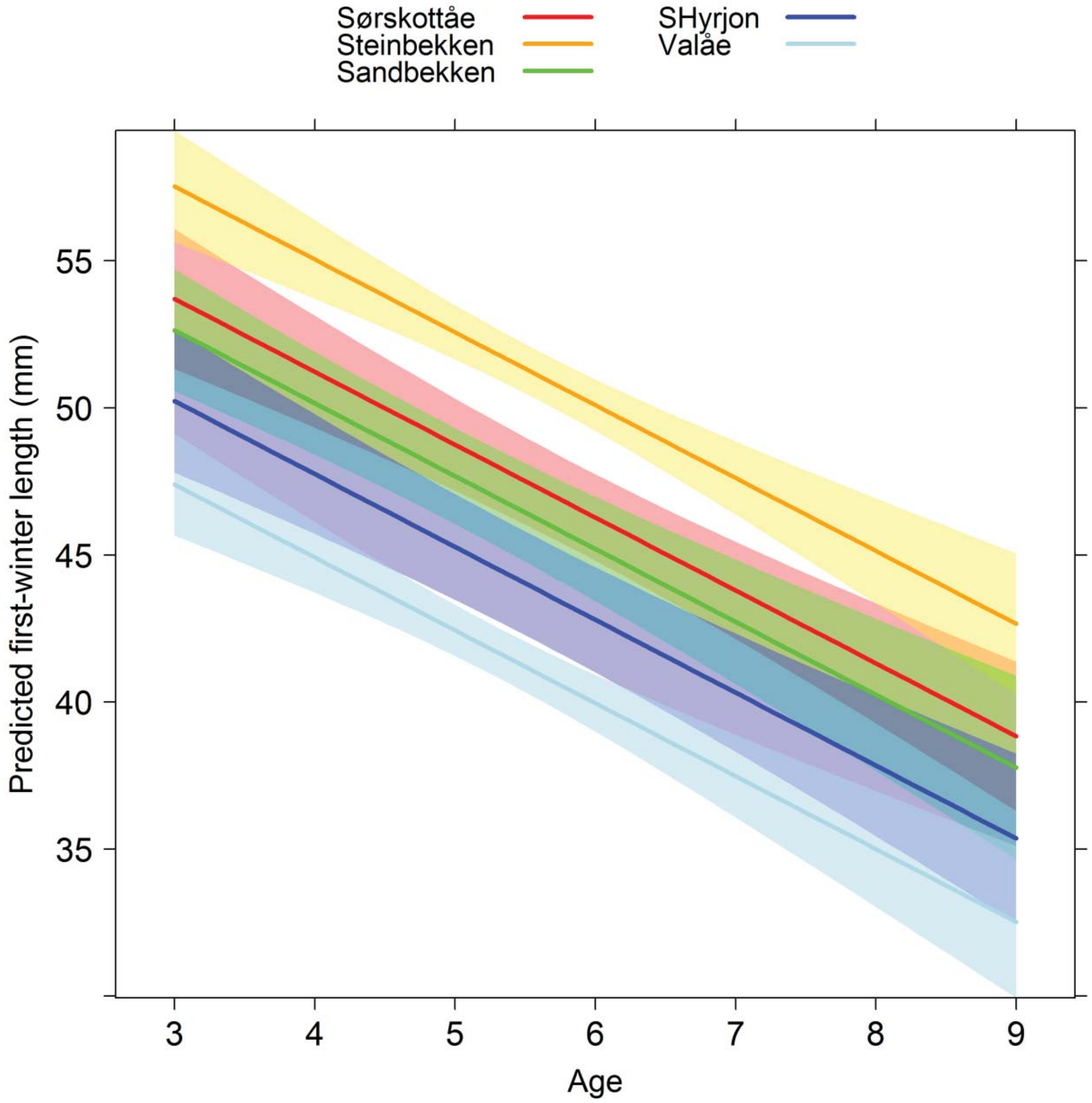



Figure10. First year winter length as function of age and stream

The temperature and age are statistically significant (table 5). Higher the temperature more will be the age.

Table 5. Parameter estmates for the 2nd-most supported model predicting back-calculated length at end of first winter as function of temperature and age.

```
              Estimate Std. Error t value Pr(>|t|)
(Intercept)  3.684333   0.074162  49.680   <2e-16 ***
Temp         0.061656   0.007006   8.800   <2e-16 ***
Age         -0.032706   0.012821  -2.551    0.011 *
```

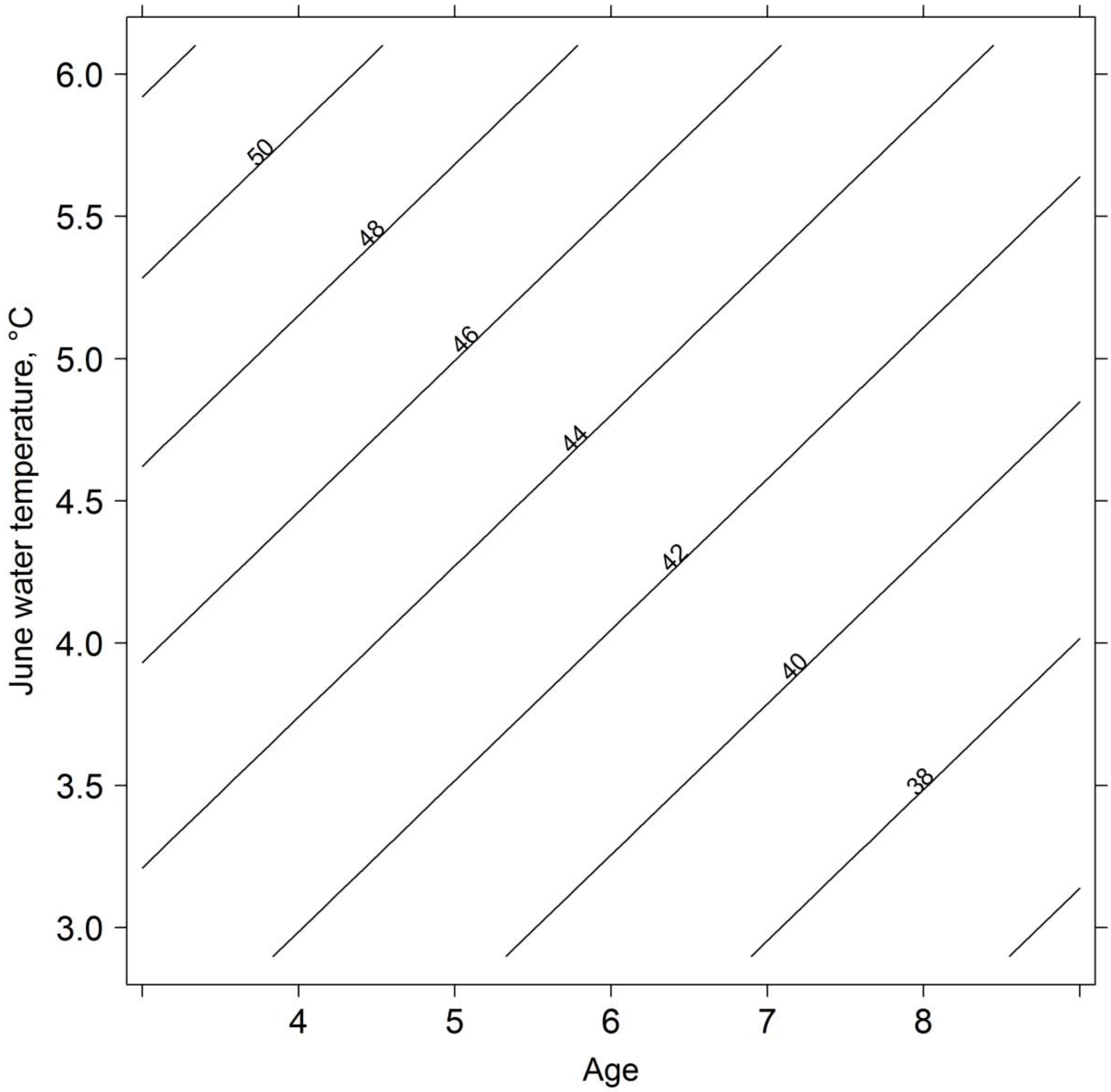



Figure 11. Contourplot of predicted L1-values based on estimates retrieved from the 2nd-most supported model (Table 5) where parameter estimates are shown in Table 5.

With increase of age the number of individual decreases. The maximum number of individual is 55 at age 4 when the June water temperature is about 5.7 °C and minimum at age 8 with 3.9°C June water temperature. The number gradually decreases with decrease in temperature and increase in age.

## 4.4 Differences in growth trajectories

In general, there are no vast differences between the male and female growth trajectories. Males are slightly larger than females in all the streams (fig 13). In male, the highest growth rate is in Sandbekken i.e 31 % and lowest in SHyrjon i.e 21 %. Similaly, the growth rate in female is also highest in Sandbekken i.e 28 % and lowest in Sørskottåe 22%. Sandbekken has highest rate of growth in both male and female i.e 29% and lowest in Sørskottåe i.e 22% (table 6)

The maximum potential length ($L_\infty$) in male, female and both sex are highest in Sørskottåe i.e. 456,445 and 456 respectively. In female, lowest in SHyrjon i.e 376 and in male and both sexes Sandbekken has the lowest potential length i.e 390 and 393 respectively (table 6).

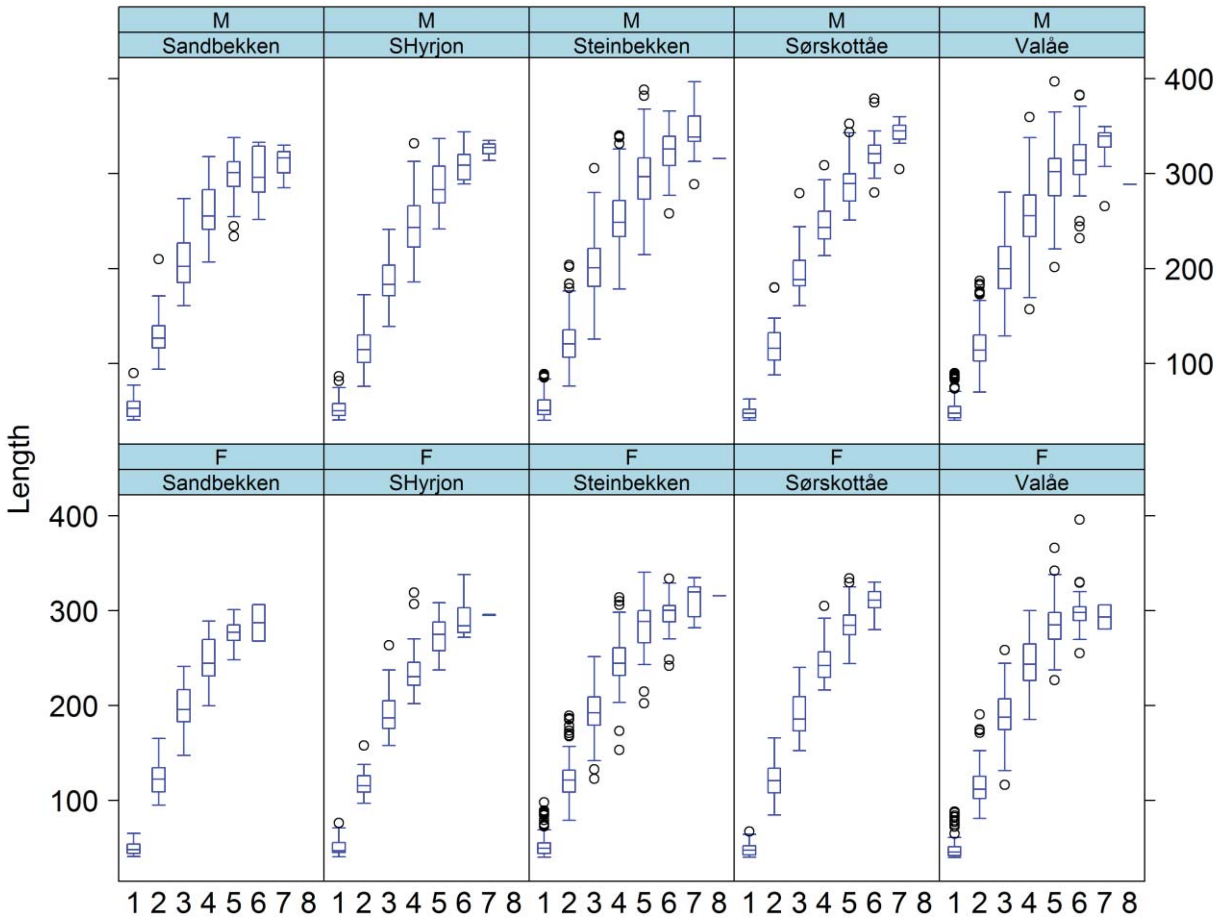
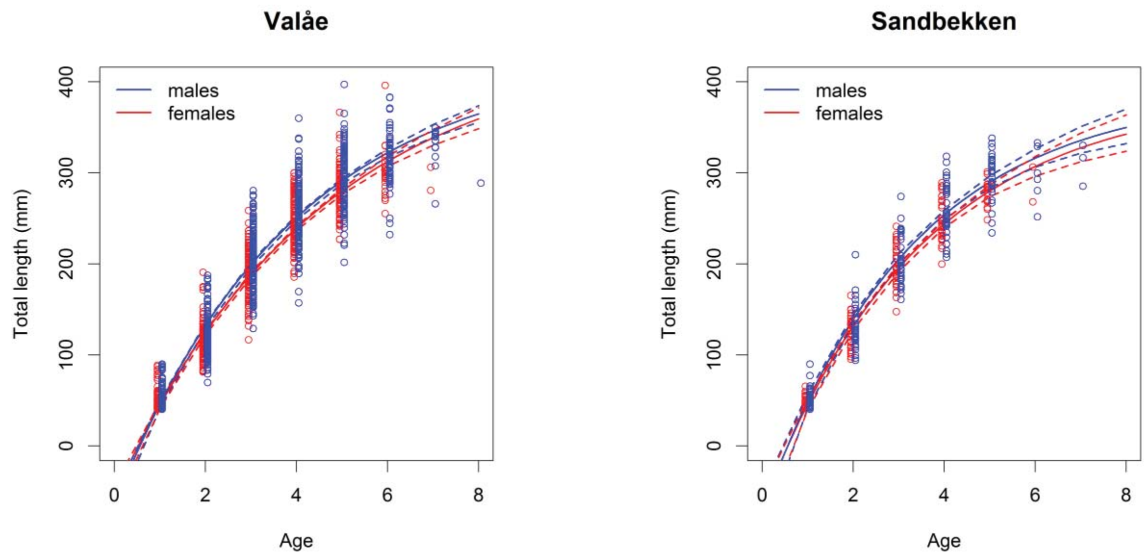



Figure 12. Boxplot of back-calculated age-specific lengths for males (M) and females (F) from the five grayling study populations in Lesjaskogsvatn. The horizontal lines located within the boxes represent the median value. 50% of the observations (at a given age) are

covered by the boxes and 90% of the observations are covered by the whiskers. Dots represent outliers.

Table 6. Parameter estimate table for Von Bertalanffy model

| Population | Gender | $L_\infty$ | K | $T_0$ |
|---|---|---|---|---|
| Valåe | Male | 428.4±13.2 | 0.256±0.015 | 0.560±0.032 |
| Valåe | Female | 441.8±18.5 | 0.224±0.016 | 0.53±0.033 |
| Valåe | Both | 437.8±11.2 | 0.238±0.011 | 0.54±0.273 |
| Steinbekken | Male | 455.03±13.80 | 0.230±0.013 | 0.48±0.031 |
| Steinbekken | Female | 399.04±13.19 | 0.267±0.017 | 0.504±0.035 |
| Steinbekken | Both | 438.8±10.25 | 0.238±0.010 | 0.488±0.024 |
| Sandbekken | Male | 389.7±20.50 | 0.306±0.033 | 0.558±0.058 |
| Sandbekken | Female | 388.1±23.54 | 0.287±0.032 | 0.562±0.050 |
| Sandbekken | Both | 393.33±15.93 | 0.292±0.023 | 0.557±0.04 |
| Shyrjon | Male | 453.6±34.07 | 0.214±0.028 | 0.457±0.071 |
| Shyrjon | Female | 375.60±20.45 | 0.284±0.031 | 0.536±0.061 |
| Shyrjon | Both | 418.12±19.85 | 0.241±0.0216 | 0.490±0.048 |
| Sørskottåe | Male | 456.8±18.18 | 0.22±0.016 | 0.540±0.042 |
| Sørskottåe | Female | 444.62±23.04 | 0.22±0.021 | 0.521±0.046 |
| Sørskottåe | Both | 456.41±14.50 | 0.22±0.012 | 0.52±0.031 |
| | | | | |



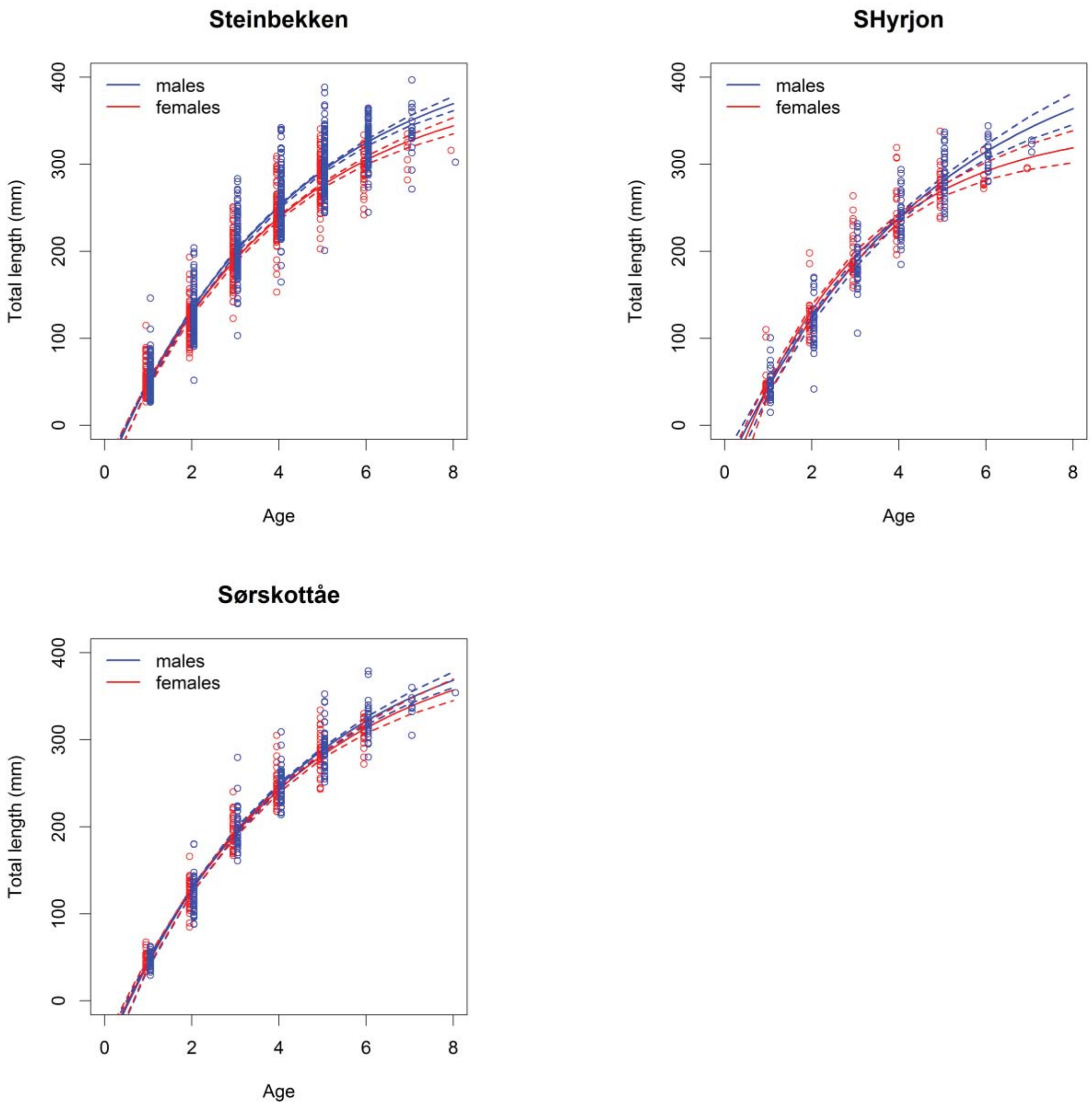



Figure 13. Estimated sex-specific von Bertalanffy growth curves (solid lines) with corresponding 95% confidence bounds (dashed lines) for the five study populations. Estimates were retrieved from the fitted models provided in Table 6. Dots represent raw back-caluculated length-at-age data.

## 4.4 Age distribution differences

The sex- and age distribution is shown in Figure 11. A generalized linear model (Poisson link) fitted to to explore differences in the age and sex distribution revealed a significant age distribution among streams but no difference in sex distribution (Table 7).

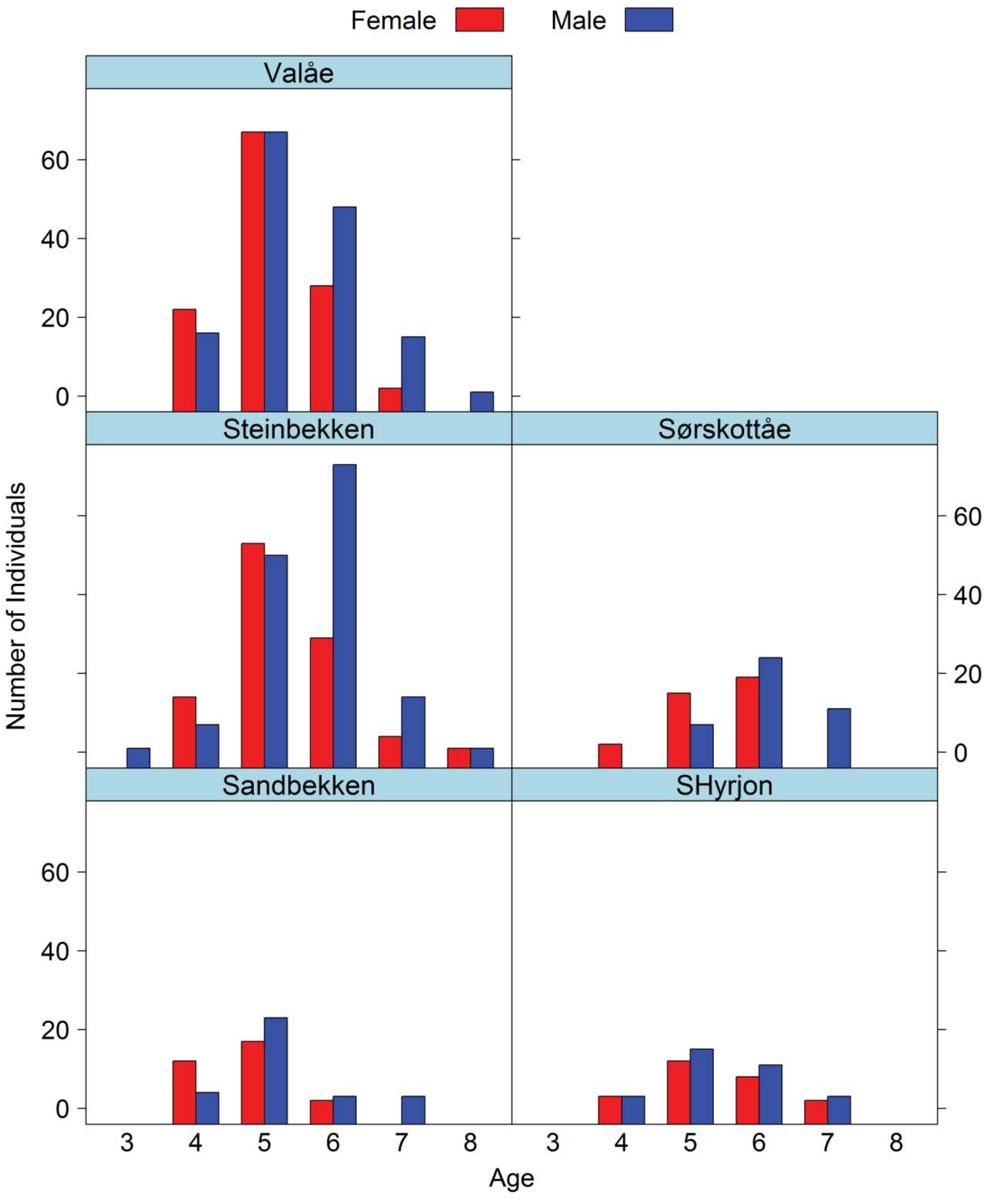



Fig 14. Sex and Age distribution of grayling among streams

As in figure 14, the number of males and females are equal at the age of 5 which is 65 in number of each sex in Valåe. At the same age the number of female is greater in Steinbekken and Sørskottåe but less in Sandbekken and SHyrjon. With the increase of age, the number of males is more as compare to females in all streams. The male and female of all the streams shows the unimodel relation except for the female of Sørskottae. Up to the age of 6, the female linearly increases in Sørskottae.

The age, stream, sex ,age*stream, age*sex are statistically significant but stream*sex, age*stream*sex are not significant (table 7)

Table 7. Analysis of variance table for the fitted GLM-model testing for differences in age and sex distribution among streams. The model was fitted using a Poisson distribution (log-link).

| Effect term | DF | Deviance | ResidDF | Chisq | P |
|---|---|---|---|---|---|
| Age | 5 | 300.381 | 39 | 556.12 | <0.00001 |
| Stream | 4 | 227.534 | 35 | 328.58 | <0.00001 |
| Sex | 2 | 212.037 | 33 | 116.54 | <0.00001 |
| Age*Stream | 13 | 67.904 | 20 | 48.64 | <0.00001 |
| Age*Sex | 6 | 35.626 | 14 | 13.01 | <0.00001 |
| Stream*Sex | 5 | 5.346 | 9 | 7.67 | 0.3751 |
| Age*Stream*Sex | 9 | 7.669 | 0 | 0 | 0.5678 |

Table 8. Catch curve based instantaneous mortality rates (Z) and corresponding annual mortality (A) and survival (S) rates for both sexes in the five study populations (appart from Søre Skotte).

| Population | Gender | Z | SE(Z) | A | S |
|---|---|---|---|---|---|
| Valåe | Male | 1.38 | 0.38 | 74.78 | 25.22 |
| | Female | 1.76 | 0.51 | 82.72 | 17.28 |
| Sandbekken | Male | 1.02 | 0.59 | 63.88 | 36.12 |
| | Female | 2.14 | NA | 88.24 | 11.77 |
| Steinbekken | Male | 0.71 | 0.13 | 50.86 | 49.14 |
| | Female | 1.37 | 0.24 | 74.54 | 25.46 |
| Shyrjon | Male | 0.81 | 0.42 | 55.28 | 44.72 |
| | Female | 0.94 | 0.18 | 60.78 | 39.22 |
| Sørskottåe | Male | Not estimable | | | |
| | Female | Not estimable | | | |

Generally in all the streams the mortality rate in adults is high. The mortality rate in Sørskottåe is high and thus it is not estimable. The highest annual mortality rate in female i.e. 88.24 in Sandbekken and lowest is in SHyrjon i.e. 60.78. In male the annual mortality rate is high in Valåe i.e 74.78 and lowest in Steinbekken 50.86 (Table 8)

## 5. Discussion

The back-calculated growth differences as shown in the figure 10 shows that the fish in the cold streams shows slow growth. The growth range of the fish in first winter is between 47-58 mm. The grayling in Valåe and SHyrjon are poor growers as compared to fish in warm streams by 19 percent. The studies performed in the Atlantic salmon, *Salmo salar* (Handeland, Imsland, & Stefansson, 2008) shows that at warmer temperature at 14 °C, the growth of the fish is highest and lowest at the temperature of 6°C. As the fish size increases , the optimal temperature for growth also increases whereas the feed conversion efficiency decreases with increase in age (Handeland et al., 2008). A study performed on the lump fish (*Cyclopterus lumpus*) shows that the warmer temperature of 10-13°C shows positive effect in the growth rate of fish to 31-35 %. The fish shows the increase in weight up to 48-53°C as compared to the fish growing at 4 °C. The temperature does not have effect on feed conversion efficiency but increase in temperature helps to increase in feed intake in lump fish (Nytrø et al., 2014). The studies performed in the Arctic charr shows that the growth of the fish is influenced by the life history characteristics of fishes rather than the temperature (Larsson et al., 2005). Similarly, the study on the brown trout (*Salmo trutta*) shows that temperature and growth of the fish is not significantly correlated (Forseth et al., 2009).

Most of the grayling in warm streams grow rapidly and have short life spans whereas the fish in the cold streams grow slowly and have longer life spans as in figure 11. The number of individuals decreases with increase in age and warmer the temperature more the number of individual. This supports the life history theory. According to life history theory mortality plays a vital role in determent the life history of an organism. The organisms having longer life span matures early than late maturing organism (Promislow & Harvey, 1990). In this study fish from Sandbekken have shorter life span. The study performed on effect of growth and survivals on maturation of grayling shows that the fish which mature early have higher adult mortalities. The mortalities in immature fish might be due to fishing activities (T. O. Haugen, 2000). The(Bass et al., 2014) study show that individuals from different streams use different parts of the lake, making them exposed to diffential fishing pressure.

Biologists used the scales to determine the age of fishes. The changes occurs in the growth of the fish depends upon the growth of the scales. As the fish grows, the scale deposition can be observed which makes the discontinuation with the circuli (Ottaway, 1978). To know how much to rely on our data comparing the different streams, the first three radius and total length of the scale is studied. The average measurement error is found to be 8 % in first radius. A lot of variation within and between individual is observed (fig 6). The second radius is easier to read as compared to first one (fig 7) in which measurement error reduced by half i.e. 4 %. In third radius R3, the measurement error is 2.5 % which is very low (fig 8). The measurement error in the total radius is significantly low i.e. 1.5% (fig 9). The finding of this study shows that the measurement error decreases with increase of age. The highest measurement occurs in 1st year and decreases in the following years. The repeating of the measurement helps to reduce the error. In this study the measurement error helps to reduce the random and systematic error (Atkinson & Nevill, 1998).

In this study I used von Bertalanffy model to predict the length- at-age of the grayling population (table 6). The parameters like maximum potential length ( $\mathbf{L_\infty}$ **),** growth rate of the fish (K) and theoretical age at zero length can be predicted by this model. I use Von Bertalanffy growth curves by using table 6 to estimate the total length on the basis of sex (fig 13). By using this model we found that overall sizes of males are slightly larger than that of females. In both sexes we found the highest growth rate in Sandbekken i.e. 29% and lowest in Sørskottåe i.e. 22%. The fish of the stream having the slowest growth rate seems to have maximum potential length, $L_\infty$ and vice versa. The growth rate of Sandbekken is highest in both sexes whereas the potential length is minimum i.e. 393. The fish of Sørskottåe have slowest growth rate in male, female and both sexes but highest in the potential length, $L_\infty$ **i.e.** 456,445 and 456 respectively. The studies performed in the life history traits of iteroparous fresh water fishes shows that the reproductive effort is equal to the adult mortality rate and the adult mortality rate is inversely proportional to the optimal age at maturity. Von Bertalanffy equation is good for the somatic growth after the fish matures. To study the life history traits, Von Bertalanffy equations are simple functions at the time when the fish mature (Lester, Shuter, & Abrams, 2004). In grayling, the fluctuation of water temperature and its effect on the growth and survival is studied by using Von Bertalanffy model. The variability in the growth of grayling occurs in the river where the temperature of water rises to the maximum tolerance level. Due to the fluctuation of water level the mortalities rate was higher and growth rate reduced. The variation in the growth of fish annually due to variation in temperature can be explained by combining daily water temperature with the Von Bertalanffy model (Mallet, Charles, Persat, & Auger, 1999).

To determine the age distribution differences among five different streams, I use Poisson GLM model (fig 14). During this study I found that there are significant differences among the age and streams but not with sex (Table 7). I found the number of female decreases with increase of age in all the streams. The number of males and females are equal in Valåe at the age of five but not in other streams. The findings of this result shows that the length decreases with the increase of the catch age (fig 10). In this study I use the scale reading to determine the age of fish. The studies in the brown trout, *Salmo trutta* shows that the error rate using scale reading of 375 fish to determine age is below 6.6 % .The result shows that the age calculated by using scale reading and the true age of fish has no significant difference (Rifflart, Marchand, Rivot, & Bagliniere, 2006).

During this study I have found the instantaneous mortality rate on the basis of catch curve, annual mortality rate and Survival of grayling in all the streams except Søre Skotte. The mortality rate of Søre Skotte is very high so that it is not estimable. In this study I found the highest mortality in female is 88.24 % in Sandbekken and lowest mortality in male grayling in Steinbekken. Our finding shows that the mortality rate among female in all the streams are higher than that of males. The studies performed in the brook trout (*Salvelinus fontinalis*) shows that the high temperature variability causes high mortality in fish. The experiment was common garden experiment where four treatments were used i.e. constant temperature, periodic variability, low variability and high variability of temperature. The result shows that in constant temperature the length of the fish is higher (Pisano, 2017).

The findings in this study clearly shows that the growth rate of warm water fishes are higher than the fishes in cold water streams whereas the life span is less in the fishes inhabiting in the warm streams than in the cold streams. The mortality rate seems to be higher in female than in male but the growth rate of male is higher than that of female. In both sexes, i find that the fish having fastest growth have highest mortality rate.

## 6. Conclusion

The findings from the present studies can be summarized as the fish which grows in the cold streams grow slowly than in the warm water. The fish in the cold waters mature late than in warm water. As a result the life span of the fish in cold streams is long. The decrease in the life span in early mature fish is due to extensive fishing and predators.

The result of the present studies shows that the annual mortality rate in adult females are higher in small warm water stream and lowest in large cold water stream whereas in male the mortality rate is highest in large cold water stream and lowest in warm water stream. The highest annual mortality rate for female is in Sandbekken i.e. 88.24 which is small warm water stream whereas in male Valåe has the highest annual mortality rate i.e. 74.78 which is large cold water stream. For female, the lowest annual mortality rate observed in Shyrjon i.e. 60.78 which is large cold water streams and for male is in Steinbekken i.e 50.86 which is small warm water streams.

Growth rate seems to be higher in small warm streams and lower in large cold water streams in male whereas in females the growth rate is highest and lowest both in small water streams. Generally, the males are slightly larger than females. In Sandbekken which warm water stream the growth of male and female seems to be highest i.e. 31% and 28% respectively. The lowest growth rate seems to occur in Large cold water stream Shyrjon i.e 21 % for male and for female warm water stream Søre Skottæe i.e 22 %.

The highest and lowest maximum potential length seems in warm water streams in all sexes except in female in Shyrjon for lowest $L_\infty$ .In Sørskottåe the maximum length; $L_\infty$ is highest which warm water stream is. The lowest maximum potential length is also seen in warm water stream, Sandbekken in male and both sexes.

Among all the streams, the annual mortality rate is more in female than the males. In general, the four streams, Sandbekken, Valåe, Steinbekken and Shyrjon it seems that higher the growth rate higher is the annual adult mortality rate. Regarding sex, the growth rate of males are slightly higher than that of female but mortality rate seems to be higher at females. In Sørskottåe the growth rate is slowest among all streams but the adult mortality seems to be higher and thus it is not estimable. It is clear that from the present finding, that the adult annual mortality does not depends upon the water temperature only.

## References

Atkinson, G., & Nevill, A. M. (1998). Statistical methods for assessing measurement error (reliability) in variables relevant to sports medicine. *Sports medicine, 26*(4), 217-238.

Bærum, K. M. (2008). Morphological divergence among sympatric demes of European grayling (Thymallus thymallus).

Barson, N. J., Haugen, T. O., Vøllestad, L. A., & Primmer, C. R. (2009). CONTEMPORARY ISOLATION-BY-DISTANCE, BUT NOT ISOLATION-BY-TIME, AMONG DEMES OF EUROPEAN GRAYLING (THYMALLUS THYMALLUS, LINNAEUS) WITH RECENT COMMON ANCESTORS. *Evolution, 63*(2), 549-556.

Bass, A. L., Haugen, T. O., & Vøllestad, L. A. (2014). Distribution and movement of European grayling in a subarctic lake revealed by acoustic telemetry. *Ecology of freshwater fish, 23*(2), 149-160.

Beamish, R. (1987). Current trends in age determination methodology. *Age and growth of fish*, 15-42.

Carl, L. M., Walty, D., & Rimmer, D. M. (1992). Demography of spawning grayling (Thymallus arcticus) in the Beaverlodge River, Alberta. *Hydrobiologia, 243*(1), 237-247.

Carlander, K. (1987). A history of scale age and growth studies of North American freshwater fish. *Age and growth of fish. Iowa State University Press, Ames*, 3-14.

Chapman, D., & Robson, D. (1960). The analysis of a catch curve. *Biometrics*, 354-368.

Forseth, T., Larsson, S., Jensen, A. J., Jonsson, B., Näslund, I., & Berglund, I. (2009). Thermal growth performance of juvenile brown trout Salmo trutta: no support for thermal adaptation hypotheses. *Journal of Fish Biology, 74*(1), 133-149.

Gregersen, F., Haugen, T., & Vøllestad, L. (2008). Contemporary egg size divergence among sympatric grayling demes with common ancestors. *Ecology of freshwater fish, 17*(1), 110-118.

Haddeland, P. J., Junge, C., Serbezov, D., & Vøllestad, L. A. (2015). Genetic parentage analysis confirms a polygynandrous breeding system in the European grayling (Thymallus thymallus). *PloS one, 10*(3), e0122032.

Handeland, S. O., Imsland, A. K., & Stefansson, S. O. (2008). The effect of temperature and fish size on growth, feed intake, food conversion efficiency and stomach evacuation rate of Atlantic salmon post-smolts. *Aquaculture, 283*(1), 36-42.

Haugen, T., & Vøllestad, L. (2000). Population differences in early life-history traits in grayling. *Journal of evolutionary biology, 13*(6), 897-905.

Haugen, T. O. (2000). Growth and survival effects on maturation pattern in populations of grayling with recent common ancestors. *Oikos, 90*(1), 107-118.

Haugen, T. O., & Vøllestad, L. A. (2001). A century of life-history evolution in grayling *Microevolution Rate, Pattern, Process* (pp. 475-491): Springer.

Horká, P., Ibbotson, A., Jones, J., Cove, R., & Scott, L. (2010). Validation of scale-age determination in European grayling Thymallus thymallus using tag-recapture analysis. *Journal of Fish Biology, 77*(1), 153-161.

Ingram, A., Ibbotson, A., & Gallagher, M. (1999). The Ecology and management of European Grayling [Thymallus thymallus (Linnaeus)] Interim Report.

Jonsson, B., & Stenseth, N. (1976). Regression of Body Length on Scale Size of Brown Trout, Salmo-Trutta-L. *Norwegian Journal of Zoology, 24*(4), 331-340.

Junge, C., Vøllestad, L., Barson, N., Haugen, T., Otero, J., Sætre, G., . . . Primmer, C. (2011). Strong gene flow and lack of stable population structure in the face of rapid adaptation to local temperature in a spring-spawning salmonid, the European grayling (Thymallus thymallus). *Heredity, 106*(3), 460-471.

Kavanagh, K. D., Haugen, T. O., Gregersen, F., Jernvall, J., & Vøllestad, L. A. (2010). Contemporary temperature-driven divergence in a Nordic freshwater fish under conditions commonly thought to hinder adaptation. *BMC evolutionary biology, 10*(1), 350.

Koskinen, M. T., Haugen, T. O., & Primmer, C. R. (2002). Contemporary fisherian life-history evolution in small salmonid populations. *Nature, 419*(6909), 826-830.

Kristiansen, H., & Dølving, K. B. (1996). The migration of spawning stocks of graylingThymallus thymallus, in Lake Mjøsa, Norway. *Environmental biology of fishes, 47*(1), 43-50.

Kvinge, A. S. (2014). Gene Expression in the Early Development stages in Populations of European Grayling (Thymallus thymallus).

Larsson, S., Forseth, T., Berglund, I., Jensen, A., Näslund, I., Elliott, J., & Jonsson, B. (2005). Thermal adaptation of Arctic charr: experimental studies of growth in eleven charr populations from Sweden, Norway and Britain. *Freshwater Biology, 50*(2), 353-368.

Leith, A. E. (2007). Life history.

Lester, N., Shuter, B., & Abrams, P. (2004). Interpreting the von Bertalanffy model of somatic growth in fishes: the cost of reproduction. *Proceedings of the Royal Society of London B: Biological Sciences, 271*(1548), 1625-1631.

Mallet, J., Charles, S., Persat, H., & Auger, P. (1999). Growth modelling in accordance with daily water temperature in European grayling (Thymallus thymallus L.). *Canadian Journal of Fisheries and Aquatic Sciences, 56*(6), 994-1000.

Mooi, R. (2009). Evolution, Douglas J. Futuyma: Soc Integ Comp Biol.

Northcote, T. G. (1995). Comparative biology and management of Arctic and European grayling (Salmonidae, Thymallus). *Reviews in fish biology and fisheries, 5*(2), 141-194.

Nygård, K. (2012). *Movement and growth of European grayling Thymallus thymallus in two Norwegian rivers.* Institutt for biologi.

Nykänen, M. (2004). *Habitat selection by riverine grayling, Thymallus thymallus L*: University of Jyväskylä.

Nykänen, M., Huusko, A., & Lahti, M. (2004). Changes in movement, range and habitat preferences of adult grayling from late summer to early winter. *Journal of Fish Biology, 64*(5), 1386-1398.

Nytrø, A. V., Vikingstad, E., Foss, A., Hangstad, T. A., Reynolds, P., Eliassen, G., . . . Imsland, A. K. (2014). The effect of temperature and fish size on growth of juvenile lumpfish (Cyclopterus lumpus L.). *Aquaculture, 434*, 296-302.

Ottaway, E. (1978). Rhythmic growth activity in fish scales. *Journal of Fish Biology, 12*(6), 615-623.

Øygarden, A. (2013). *Variation in early life history traits among populations of grayling (Thymallus thymallus); different temperature reaction norms.*

Parkinson, D., Philippart, J. C., & Baras, E. (1999). A preliminary investigation of spawning migrations of grayling in a small stream as determined by radio-tracking. *Journal of Fish Biology, 55*(1), 172-182.

Paulsen, S. M. (2014). *Population variation in early growth of brown trout inhabiting different climates: Evidence for countergradient growth?*

Pisano, O. (2017). The Influence of Temperature Variability on Metrics of Life History in Brook Trout (Salvelinus fontinalis).

Promislow, D. E., & Harvey, P. H. (1990). Living fast and dying young: A comparative analysis of life-history variation among mammals. *Journal of Zoology, 220*(3), 417-437.

Ricker, W. E. (1992). Back-calculation of fish lengths based on proportionality between scale and length increments. *Canadian Journal of Fisheries and Aquatic Sciences, 49*(5), 1018-1026.

Rifflart, R., Marchand, F., Rivot, E., & Bagliniere, J.-L. (2006). Scale reading validation for estimating age from tagged fish recapture in a brown trout (Salmo trutta) population. *Fisheries Research, 78*(2), 380-384.

Spannhof, L. (1989). Age and growth of fish: Robert C. Summerfelt (Managing Editor) and Gordon E. Hall (Technical Editor). Iowa State University Press, Ames, IA, USA, 1987. xvi+ 544 pp., figs. and tables, ISBN 0-81380-733-6: Elsevier.

Stearns, S. C., & Crandall, R. E. (1984). Plasticity for age and size at sexual maturity: a life-history response to unavoidable stress. *Fish reproduction: strategies and tactics. Academic Press, London*, 13-33.

Taylor, E. B. (1991). A review of local adaptation in Salmonidac, with particular reference to Pacific and Atlantic salmon. *Aquaculture, 98*(1-3), 185-207.

TO, H. (2000). Early survival and growth in populations of grayling with recent common ancestors—field experiments. *Journal of Fish Biology, 56*, 1173-1191.

Wedekind, C., & KUeNG, C. (2010). Shift of spawning season and effects of climate warming on developmental stages of a grayling (Salmonidae). *Conservation Biology, 24*(5), 1418-1423.

Weisberg, S., & Frie, R. (1987). Linear models for the growth of fish. *Age and Growth of Fish. Iowa State University Press, Ames, IA*, 127-143.

Zuur, A., Ieno, E., Walker, N., Saveliev, A., & Smith, G. (2009). Mixed Effects Models and Extensions in Ecology with R. Springer Science+: Business Media New York.

# Appendix

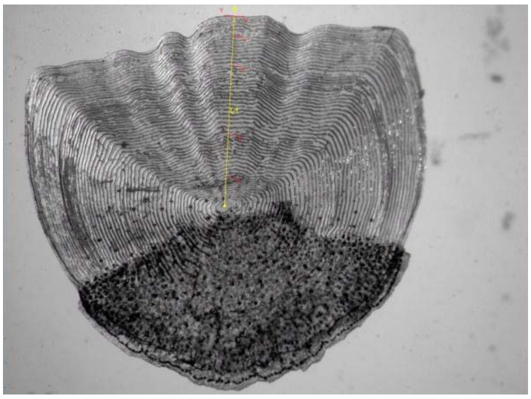
Appendix1. Data to study the measurement error while scale reading of randomly selected 100 samples.

| ID | ID2 | Rtot | R1 | R2 | R3 |
|---|---|---|---|---|---|
| 1 | SAN10001.TIF | 740 | 104 | 282 | 545 |
| 2 | SAN10048.TIF | 665 | 90 | 267 | 407 |
| 3 | STE10242.TIF | 714 | 116 | 270 | 471 |
| 4 | SSKO10026.TIF | 774 | 94 | 310 | 482 |
| 5 | SHYR10038.TIF | 830 | 217 | 424 | 582 |
| 6 | VAL10167.TIF | 668 | 40 | 212 | 352 |
| 7 | SAN10001.TIF | 751 | 110 | 294 | 548 |
| 8 | STE10006.TIF 2 | 737 | 90 | 255 | 403 |
| 9 | STE10015.TIF | 745 | 77 | 276 | 464 |
| 10 | SHYR10003.TIF | 651 | 102 | 287 | 459 |
| 11 | VAL10129.TIF | 694 | 46 | 207 | 385 |
| 12 | STE10033.TIF | 699 | 124 | 257 | 375 |
| 13 | SAN10001.TIF | 754 | 113 | 281 | 546 |
| 14 | SSKO10036.TIF | 782 | 151 | 333 | 498 |
| 15 | VAL10216.TIF | 625 | 91 | 204 | 350 |
| 16 | SHYR10054.TIF 2 | 738 | 121 | 298 | 482 |
| 17 | VAL10151.TIF | 756 | 99 | 272 | 438 |
| 18 | STE10012.TIF | 718 | 88 | 278 | 435 |
| 19 | SHYR10003.TIF | 657 | 103 | 292 | 480 |
| 20 | STE10114.TIF | 762 | 122 | 256 | 455 |
| 21 | SSKO10026.TIF | 782 | 107 | 319 | 498 |
| 22 | STE10015.TIF | 753 | 88 | 277 | 472 |
| 23 | STE10242.TIF | 725 | 125 | 282 | 488 |
| 24 | SAN10048.TIF | 658 | 83 | 262 | 410 |
| 25 | STE10263.TIF | 840 | 137 | 317 | 451 |
| 26 | SHYR10003.TIF | 668 | 112 | 276 | 494 |
| 27 | STE10114.TIF | 761 | 126 | 264 | 448 |
| 28 | STE10012.TIF | 723 | 92 | 280 | 433 |
| 29 | STE10006.TIF 2 | 741 | 79 | 213 | 403 |
| 30 | SHYR10054.TIF 2 | 747 | 127 | 307 | 491 |
| 31 | STE10033.TIF | 693 | 130 | 251 | 379 |
| 32 | VAL10129.TIF | 706 | 56 | 217 | 395 |
| 33 | SHYR10038.TIF | 831 | 216 | 424 | 581 |
| 34 | STE10015.TIF | 748 | 67 | 270 | 463 |
| 35 | VAL10167.TIF | 703 | 58 | 225 | 374 |
| 36 | SSKO10026.TIF | 788 | 115 | 324 | 501 |
| 37 | VAL10212.TIF | 621 | 99 | 245 | 454 |
| 38 | SAN10001.TIF | 749 | 100 | 271 | 546 |
| 39 | STE10242.TIF | 727 | 120 | 285 | 492 |
| 40 | VAL10151.TIF | 761 | 101 | 276 | 450 |
| 41 | STE10012.TIF | 713 | 76 | 275 | 428 |
| 42 | SHYR10054.TIF 2 | 740 | 127 | 310 | 485 |
| 43 | SSKO10036.TIF | 782 | 150 | 337 | 504 |
| 44 | VAL10216.TIF | 606 | 61 | 215 | 354 |
| 45 | SAN10048.TIF | 661 | 88 | 259 | 407 |

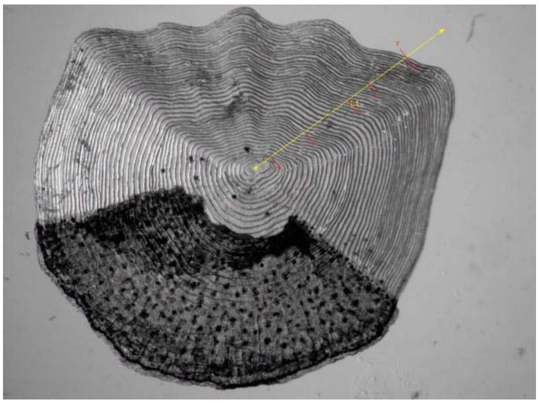
| | | | | | |
|---|---|---|---|---|---|
| 46 | STE10263.TIF | 836 | 140 | 308 | 445 |
| 47 | STE10114.TIF | 764 | 116 | 254 | 439 |
| 48 | STE10006.TIF 2 | 737 | 77 | 214 | 402 |
| 49 | STE10033.TIF | 693 | 125 | 242 | 360 |
| 50 | SAN10048.TIF | 660 | 92 | 254 | 466 |
| 51 | VAL10258.TIF | 669 | 66 | 230 | 358 |
| 52 | STE10114.TIF | 778 | 130 | 301 | 454 |
| 53 | SSKO10036.TIF | 784 | 146 | 336 | 500 |
| 54 | VAL10212.TIF | 615 | 86 | 240 | 432 |
| 55 | SHYR10038.TIF | 834 | 221 | 430 | 593 |
| 56 | VAL10216.TIF | 667 | 64 | 226 | 353 |
| 57 | STE10015.TIF | 750 | 86 | 270 | 459 |
| 58 | STE10012.TIF | 720 | 91 | 282 | 420 |
| 59 | STE10006.TIF 2 | 736 | 103 | 219 | 408 |
| 60 | VAL10129.TIF | 698 | 50 | 211 | 390 |
| 61 | SSKO10026.TIF | 786 | 112 | 319 | 491 |
| 62 | VAL10151.TIF | 761 | 104 | 293 | 451 |
| 63 | STE10242.TIF | 721 | 126 | 279 | 485 |
| 64 | SHYR10054.TIF 2 | 747 | 127 | 313 | 491 |
| 65 | VAL10258.TIF | 710 | 57 | 206 | 360 |
| 66 | STE10114.TIF | 759 | 132 | 258 | 449 |
| 67 | VAL10258.TIF | 711 | 61 | 212 | 362 |
| 68 | STE10033.TIF | 692 | 118 | 240 | 372 |
| 69 | VAL10212.TIF | 621 | 96 | 262 | 442 |
| 70 | SHYR10038.TIF | 824 | 211 | 418 | 567 |
| 71 | VAL10216.TIF | 686 | 63 | 226 | 400 |
| 72 | VAL10129.TIF | 694 | 49 | 207 | 385 |
| 73 | STE10015.TIF | 750 | 82 | 266 | 456 |
| 74 | STE10263.TIF | 840 | 137 | 311 | 452 |
| 75 | VAL10258.TIF | 714 | 74 | 216 | 386 |
| 76 | SHYR10003.TIF | 689 | 114 | 319 | 555 |
| 77 | VAL10151.TIF | 740 | 98 | 262 | 429 |
| 78 | VAL10212.TIF | 622 | 96 | 246 | 432 |
| 79 | STE10012.TIF | 725 | 93 | 287 | 439 |
| 80 | SSKO10036.TIF | 704 | 61 | 212 | 342 |
| 81 | VAL10258.TIF | 708 | 57 | 199 | 350 |
| 82 | STE10242.TIF | 722 | 129 | 281 | 492 |
| 83 | SSKO10026.TIF | 777 | 101 | 311 | 480 |
| 84 | VAL10216.TIF | 669 | 66 | 230 | 358 |
| 85 | SAN10001.TIF | 753 | 110 | 278 | 548 |
| 86 | VAL10167.TIF | 683 | 50 | 217 | 361 |
| 87 | VAL10129.TIF | 694 | 46 | 206 | 387 |
| 88 | VAL10212.TIF | 620 | 88 | 258 | 440 |
| 89 | SHYR10054.TIF 2 | 754 | 129 | 312 | 498 |
| 90 | VAL10167.TIF | 672 | 48 | 222 | 356 |
| 91 | STE10006.TIF 2 | 742 | 81 | 217 | 401 |
| 92 | STE10263.TIF | 839 | 132 | 305 | 443 |
| 93 | SHYR10003.TIF | 688 | 115 | 314 | 551 |
| 94 | VAL10167.TIF | 670 | 48 | 222 | 350 |
| 95 | SAN10048.TIF | 661 | 86 | 269 | 408 |
| 96 | VAL10151.TIF | 757 | 99 | 263 | 444 |
| 97 | SSKO10036.TIF | 781 | 145 | 327 | 494 |
| 98 | STE10263.TIF | 839 | 138 | 316 | 451 |

| | | | | | |
|---|---|---|---|---|---|
| 99 | SHYR10038.TIF | 833 | 220 | 434 | 589 |
| 100 | STE10033.TIF | 691 | 132 | 242 | 369 |

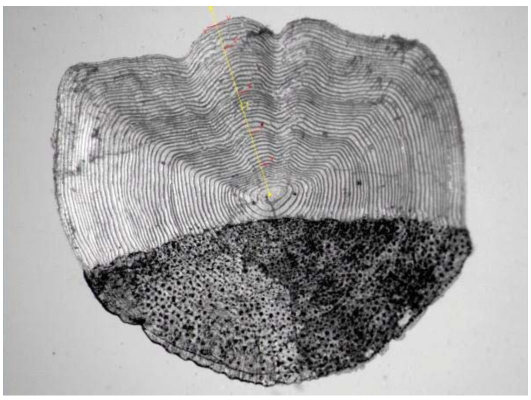
Appendix 2. Scale reading figures used in analyzing the data. During the present studies 731 scales were analyzed from the fish of five different streams.



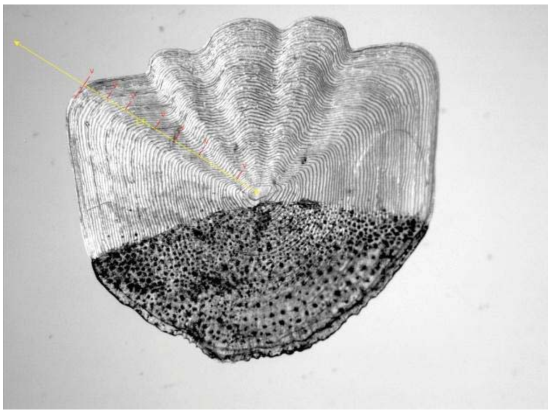
SAN10001



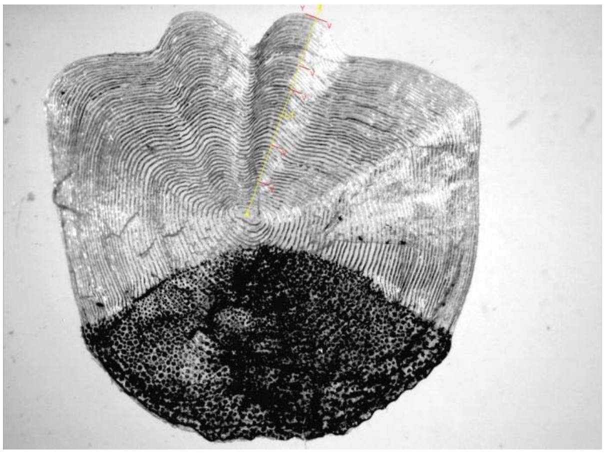
SHYR10005



SSKO10040



STE10005



VAL10021

Appendix 3: In image centre the 731 scale samples were photographed by this miscroscope


Microscope used to take the photographs

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