Aquaculture 596 (2025) 741729

Contents lists available at ScienceDirect

Aquaculture

journal homepage: www.elsevier.com/locate/aquaculture

Vertebral deformities in cultured big size Rainbow Trout: Radiological analysis from juvenile to harvest size

Per Gunnar Fjelldal[a,*], Sankar Murugesan[a,b,c], Tone Vågseth[a], Audun Østby Pedersen[a], Angelico Madaro[e], Samantha Bui[d], Harald Kryvi[b], Lars Helge Stien[d], Jonatan Nilsson[e]

[a] Reproduction and Developmental Biology Group, Institute of Marine Research (IMR), Matre Aquaculture Research station, Matre 5, 5984 Matredal, Norway
[b] Department of Biosciences, University of Bergen, Norway
[c] ICAR-Central Marine Fisheries Research Institute, Kochi, India
[d] Animal Welfare, Institute of Marine Research (IMR), Institute of Marine Research (IMR), Matre Aquaculture Research station, Matre 5, 5984 Matredal, Norway
[e] Animal Welfare, Institute of Marine Research (IMR), PO Box 1870, Nordnes, 5817 Bergen, Norway

ARTICLE INFO

Keywords:
Rainbow Trout
Deformity
Vertebrae
Welfare
Tail fin

ABSTRACT

Vertebral deformities are a big challenge in Rainbow Trout seawater aquaculture. However, supportive scientific literature is missing. The present study used radiology to follow the development of vertebral deformities in a population of farmed Rainbow Trout as they grew from 36 g to 5.5 kg. In addition, separately collected deformity screening data from three other farmed populations were included for comparison.

The fish developed deformities in different vertebral regions over time, eventuating to affect almost the entire vertebral column. At this point, the fish were ~ 5.5 kg and 93 % of the fish had one or more deformed vertebrae. A negative relationship between severity of deformity (number of deformed vertebrae per individual), and fish length and weight strongly suggest a negative impact on fish welfare. The most frequently affected area was the ural region of the vertebral column. This region is a part of the caudal fin complex which also suffered from degradation of the fin rays. Ural region deformities were also frequent in the separately investigated populations.

The current results indicate that Rainbow Trout are not able to maintain normal bone development under current farming conditions, consequently, jeopardizing the welfare of the fish.

1. Introduction

Rainbow Trout (*Oncorhynchus mykiss*) has been introduced to all continents except Antarctica and is today one of the world's most important aquaculture species (Gall and Crandell, 1992; FAO, 2022). Several different production methods and marked sizes are applied, from 250 to 500 g 'table fish' produced in ponds (Woynarovich et al., 2011) to 5 kg fish produced in net pens (sea-cages) (Aas et al., 2022). In Norway, one of the world leading producers of Rainbow Trout the production consists of two phases. First an initial phase in on-land freshwater tanks, whereafter the fish are transferred to sea-cages for grow-out to harvest size at 2–6 kg (FAO, 2024).

Recently, a workshop on R&D challenges in Rainbow Trout farming in Norway listed vertebral deformities as a significant problem (FHF workshop March 21, 2023). There are indeed many publications on vertebral deformities in cultured Rainbow Trout (e.g. Madsen and Dalsgaard, 1999; Madsen et al., 2000, 2001; Kacem et al., 2004; Deschamps et al., 2008; Gislason et al., 2010; Yadegari et al., 2011; Boglione et al., 2014). The literature on deformities in farmed Rainbow Trout are however almost exclusively on fish raised on-land in freshwater tanks, mostly on <300 g fish (Madsen and Dalsgaard, 1999; Madsen et al., 2000, 2001; Kacem et al., 2004; Deschamps et al., 2008) and maximum 2.7 kg (Kause et al., 2005; Gislason et al., 2010; Kacem et al., 2004; Poirier Stewart et al., 2014). To our knowledge, there is only one study on deformities in Rainbow Trout raised in sea-cages, and this study only followed the fish to 1 kg (Kause et al., 2005). The deformity assessment in that study was done by visual examination of the external phenotype without radiological examination. Thus, there is a need for literature on vertebral deformity development in Rainbow Trout raised in seawater using radiology, and not only up to 1 kg, but all the way to harvest weight at 5–6 kg.

Vertebral deformities pose problems in modern aquaculture. Naturalists have long shown a fascination for anomalies in fish (Dawson, 1964), initially manifested as descriptions of wild fish specimens (e.g.

* Corresponding author at: Institute of Marine Research (IMR), Matre Research Station, Matre 5, 5984 Matredal, Norway.
E-mail address: pergf@hi.no (P.G. Fjelldal).

https://doi.org/10.1016/j.aquaculture.2024.741729
Received 2 July 2024; Received in revised form 1 October 2024; Accepted 4 October 2024
Available online 5 October 2024
0044-8486/© 2024 The Authors. Published by Elsevier B.V. This is an open access article under the CC BY license (http://creativecommons.org/licenses/by/4.0/).

Barrington, 1767; Ford, 1930) and later evolving into research prompted by problematic deformities occurring in farmed fish (Boglione et al., 2001; Fjelldal et al., 2012; Boglione et al., 2013a, 2013b; Eissa et al., 2021; Chandra et al., 2024; Aydın et al., 2024). Specialized for an aquatic mode of life, fish rely on specific locomotor adaptions involving a specialized vertebral column (Webb, 1982). Deformities in the vertebral column of farmed fish therefore have a wide range of unwanted negative effects in aquaculture, including reduced growth, swimming performance, product quality, and welfare (Michie, 2001; Branson and Turnbull, 2008; Powell et al., 2009; Hansen et al., 2010; Noble et al., 2018). Vertebral deformities have indeed been addressed as an individual-based welfare indicator in a welfare indicator handbook for Rainbow Trout (Noble et al., 2020).

Fish can develop a wide range of vertebral deformity phenotypes. Possible deformities that can develop in the vertebral column of fish are spinal curvature and/or shortening (Witten et al., 2005, 2009; Munday et al., 2016; Perrott et al., 2018; Lovett et al., 2020). Spinal curvatures can be categorized into lordosis, scoliosis, and kyphosis, or combinations of those, and may affect different regions of the vertebral column (Fjelldal et al., 2012; Chatzakis et al., 2024; Mariasingarayan et al., 2024). Spinal shortening involves vertebral body deformities, in which Witten et al. (2009) identified 20 different types and developed a classification system. Subsequently, identifying deformity type and location has become an important diagnostic tool in ichthyological research and veterinary science. The type of deformity reflects the stage of pathogenesis (i.e. 'age' of deformity), and location reflects the onset time (life stage) of the deformity. As classical examples, change in the intervertebral regions or mild compressions of the vertebra endplates are early deformity stages, while fusion centra with several fused vertebrae is a final deformity stage (Witten et al., 2009). With regards to vertebral deformity location, research on Ballan Wrasse (*Labrus bergylta*) (Fjelldal et al., 2021) and Atlantic Salmon (*Salmo salar*) (Sullivan et al., 2007; Grini et al., 2011) indicate that deformities in the abdominal region of the vertebral column develop earlier in life than those in the tail region.

Various methods are applied for vertebral column regionalization. Several authors have addressed the location of vertebral deformities in farmed Rainbow Trout (Aubin et al., 2005; Deschamps et al., 2008; Deschamps et al., 2009; Deschamps et al., 2014; Fontagné et al., 2009; Boglione et al., 2014; Gislason et al., 2010). Some authors have used anatomy to separate between truncal and caudal vertebra and morphometrics for a further subdivision into 4 vertebral column regions (Ramzu and Meunier, 1999; Kacem et al., 2004; Meunier and Ramzu, 2006; Deschamps et al., 2008), while others have used vertebra number without emphasizing region (Madsen and Dalsgaard, 1999). De Clercq et al. (2017) developed a refined regionalization system for the vertebral column of Chinook Salmon (*Oncorhynchus tshawytscha*) based on histology and anatomy. Later, Sankar et al. (2024) used that system to define specific radiographic hallmarks for vertebral column regionalization in Atlantic Salmon, separating the vertebral column into 5 regions: postcranial, abdominal, transitional, caudal and ural. This method is tested to be applicable for Rainbow Trout, Brown Trout (*Salmo trutta*), Arctic Char (*Salvelinus alpinus*), Pink Salmon (*Oncorhynchus gorbuscha*), and Chinook Salmon (Sankar et al., 2024), and should hence be used in further research on vertebral deformities in farmed Rainbow Trout.

The main aim of the present study was to describe the development of vertebral deformities and their location in farmed Rainbow Trout, from the freshwater stage and throughout the entire grow-out phase in seawater.

2. Material and methods

This study followed growth and clinical radiology in a cohort of Rainbow Trout from 36 g in freshwater (April 2022) until harvest at 5.5 kg in seawater (May 2023). The study showed that the common most deformity type in this population was vertebral deformities in the ural region of the vertebral column, the region supporting the caudal fin complex. To pursue this further, the condition of the caudal fin was monitored during the final seawater phase. After assessing the results of the study, it was decided to examine some extra fish groups to evaluate the transferability of the main results. The extra groups were: (*i*) one group reared in seawater tanks at 9 °C up to 4 kg at the Institute of Marine Research; (*ii*) one group reared in freshwater tanks up to 0.2 kg at the Institute of Marine Research; (*iii*) one group sampled from a separate commercial sea farm at 0.3 kg.

Table 1
Main study. Fish length, weight and condition factor (mean ± SE) throughout the one-year study period from April 2022 to May 2023. Different lower-case letters indicate significant difference (one-way ANOVA, $P < 0.05$).

	April 2022 (n = 90)	June 2022 (n = 100)	December 2022 (n = 80)	May 2023 (n = 90)
Length (cm)	13.8 ± 0.1	22.0 ± 0.1	47.6 ± 0.5	62.6 ± 0.5
Weight (g)	36.0 ± 0.6	161.2 ± 2.7	2155.8 ± 58.6	5485.7 ± 115.4
Condition factor	1.33 ± 0.01a	1.50 ± 0.01b	1.94 ± 0.02c	2.21 ± 0.03d

2.1. Main study

2.1.1. Fish stock and rearing condition

On 15 December 2021, all-female Rainbow Trout eyed eggs from the Aquagen strain arrived at the Institute of Marine Research (IMR), Matre research station. The eggs were incubated at 8 °C and hatched 8–10 days later. First feeding started on 01 February 2022 and was done in 3 m tanks at 13 °C and continuous light. On 23–25 March 2022, the fish (7.5 ± 1.4 g) were distributed between six 3 m tanks (5.6 m^3, 2000 fish per tank) supplied with 13 °C freshwater and at continuous light. The trout were vaccinated on 20 June (Alpha Ject 5–3, PHARMAQ, AS, Overhalla, Norway), and later transferred to a sea cage (12 m × 12 m wide, 15 m deep) at the research station's facility in Solheim, Masfjorden, on 12 August 2022, and kept there for the remainder of the study. Mortality rates in freshwater and seawater were 1.2 and 3.3 %, respectively.

2.1.2. Sampling

Fish were sampled on 27 April ($n = 90$) and 08 June ($n = 100$) 2022 in freshwater, and on 14 December 2022 ($n = 80$) and 25 May 2023 (n = 90) in seawater. Random fish were netted out and euthanized by an overdose of anesthetics (0.5 g L^{-1} of tricaine methanesulfonate, Finquel, MS222). Body weight was measured to the nearest 0.1 g and fork length to the nearest 0.1 cm. Subsequently, the fish were radiographed the same day as sampled. Biometric data are displayed in Table 1.

2.1.3. Radiology and regionalization of the vertebral column

Fish were radiographed with a Direct Radiology System (Canon CXDI410C Wireless, CANON INC., Kawasaki, Japan) using a portable X-ray unit (Portable X-ray Unit Hiray Plus, Model Porta 100 HF, JOB Corporation, Yokohama, Japan) at 88 cm distance with 40 kV and 4 mAs. Fish sampled in April, June and December 2022 were radiographed as whole fish, while those sampled in May 2023 were carefully dissected and had their vertebral columns radiographed. Each fish was evaluated for different types of vertebral deformities (Witten et al., 2009), and if present, type and location were recorded.

Ninety fish sampled on 08 June 2022 were used for region specific and total vertebrae counts. Region specific vertebrae counts were performed according to the method recently described by Sankar et al. (2024). This method uses 4 specific radiographic hallmarks to define 5 different vertebra regions: Postcranial, Abdominal, Transitional, Caudal and Ural regions. The hallmarks are as follow:

1. The lack of ribs in the postcranial region.

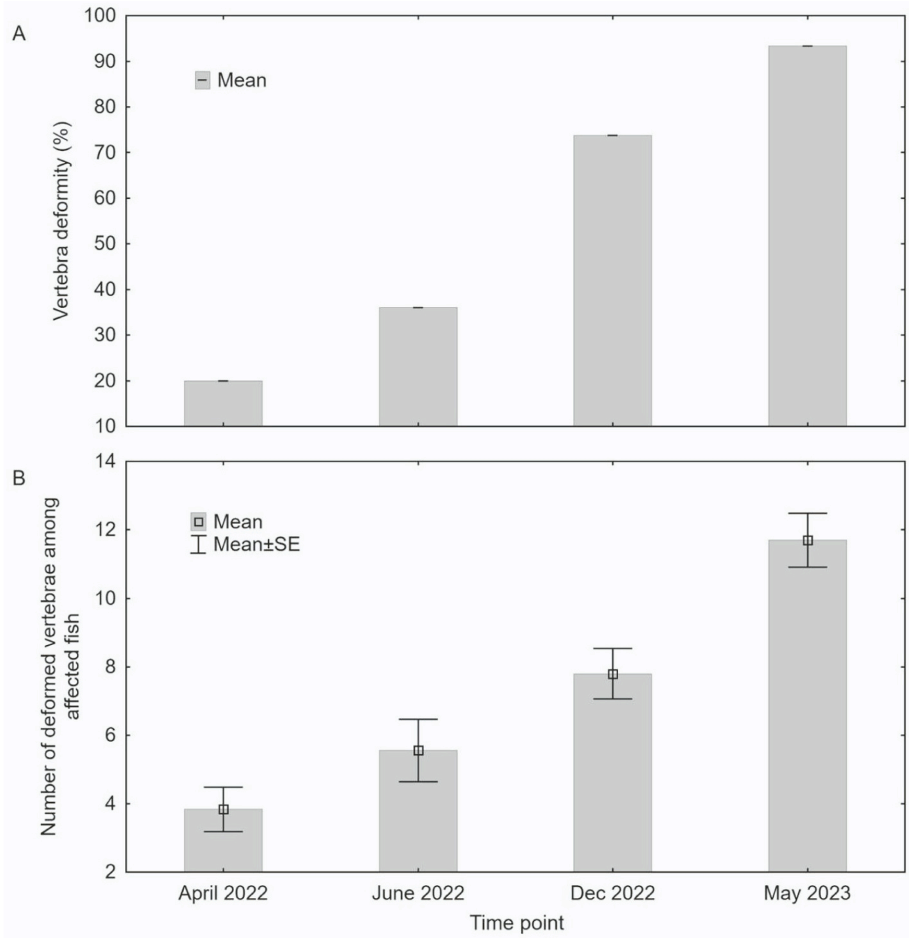

Fig. 1. Main study - vertebral deformity occurrence and severity over time from April 2022 (36 g) to May 2023 (5.5 kg). The fish were reared in freshwater tanks until August 2022 when they were transferred to sea-cages. (A) Occurrence (percentage) of fish with one or more deformed vertebrae. (B) Number of deformed vertebrae (mean ± SE) among deformed fish only, reflecting severity. Number of fish examined: April 2022 ($n = 90$), June 2022 ($n = 100$), December 2022 ($n = 80$), May 2023 (n = 90).

2. The modified parapophysis of the first vertebra of the transitional region.
3. The prominent haemal spine of the first vertebra of caudal region.
4. The separated haemal spine of the most cranial pre-ural vertebra of the ural region.

Six fish without radiological detectable vertebra deformities sampled in June 2022 and May 2023 were analysed for vertebra cranio-caudal lengths and dorso-ventral diameter to study vertebra morphometrics in different vertebra regions. Vertebra lengths and diameters were measured on radiographs by means of an image analysing software (ImageJ, 1.53 k, Wayne Rasband and contributors, National Institutes of Health, USA).

2.1.4. Fin condition

During the study it was discovered that the majority of the fish developed vertebra deformities in the ural region of the vertebral column. Since this vertebral region is a part of the caudal fin complex, it was decided to assess the condition of the caudal fin during the second half of the study. For this purpose, all fish investigated by radiology in December 2022 and May 2023 were also photographed. The euthanized fish were placed individually in a custom-made semi-closed photobox (900 × 400 × 700 mm, L × B × H) with an Olympus Tough TG-6 camera mounted on top, and the fish were photographed on the left side. Fish were measured in ImageJ (1.53 k, Wayne Rasband and contributors, National Institutes of Health, USA) for fork length, standard length, length of the central caudal fin lepidotrichs (i.e., the difference between fork length and standard length) and maximum dorso-ventral height of the caudal fin.

2.2. Extra fish groups

After finalizing the analysis of the data from the main study it was decided to screen some additional groups of farmed Rainbow Trout that were available to explore transferability of the results. These extra groups were euthanized, radiographed, and assessed for vertebrae deformities in the same way as described for the main fish group above.

The first extra group (Extra 1) consisted of all-female Rainbow Trout from the Aquagen strain purchased from a local farm. On 02 November 2022, about 800 fish of around 300 g were transported to IMR, Matre research station. The fish were subsequently reared under LD12:12 in a 3 m diameter tank (5.6 m^3 volume) supplied with 6 °C freshwater. On 26 June 2023 when the fish were approximately 1.9 kg, they were transferred to a 5 m outdoor tank supplied with 25 ppt seawater. Following, on 14 September 2023, 23 fish were transferred to 2 m tanks supplied with 34 ppt and 9 °C seawater. On 07 November 2023, these 23 fish (W: 4042 ± 92 g, L: 58.4 ± 0.4 cm, mean ± S.E.) were euthanized and radiographed.

The second extra group (Extra 2) consisted of mixed sex Rainbow Trout from the Osland strain, transferred from a local hatchery to IMR, Matre research station in June 2023 at 20 g. The fish were stocked in two 3 m tanks (5.6 m^3) at continuous light and supplied with ambient

Table 2
Simple linear regressions between total number of deformed vertebrae (V) and length, weight and condition factor in December 2022 (n = 80) and May 2023 (n = 90).

Time point	Y-axis	X-axis	R	R^2	*P*-value
December 2022	Length	Number of deformed V	−0.35	0.12	0.0016
	Weight	Number of deformed V	−0.27	0.07	0.0158
	Condition factor	Number of deformed V	0.42	0.17	0.0001
May 2023	Length	Number of deformed V	−0.59	0.34	<0.00001
	Weight	Number of deformed V	−0.35	0.12	0.0008
	Condition factor	Number of deformed V	0.52	0.27	<0.00001

temperature freshwater; 1500–1600 trout per tank. On 4 September 2023, 105 (52 and 53 from each tank) randomly sampled fish (W: 230 ± 5.3 g, L: 24 ± 0.2 cm, mean ± S.E.) were euthanized and radiographed.

The third extra group (Extra 3) were fish of the same cohort as Extra 2. However, these were collected from the sea-cage of a commercial farm on 4 October 2023 (W: 327 ± 7.6 g, L: 28.7 ± 0.2 cm, mean ± S.E.) and transferred to IMR, Matre research station where they were stocked in 1 m tanks supplied with 34 ppt seawater under continuous light. Five days later, 74 randomly sampled fish were euthanized and radiographed.

2.3. Statistics and calculations

The condition factor (CF) was calculated as: $CF = 100 \times BW/L^3$, where BW was the body weight (g) and L was the fork length (cm).

The data were analysed using Statistica version 12 (StatSoft, Inc., 2300 East 14th Street, Tulsa, Oklahoma, USA). Results are given as means and standard errors.

Data were tested for homogeneity in variance (Levene's F test) and normality (Kolmogorov Smirnov test). Significant differences between time points in CF were tested by one-way ANOVAs with an alfa level of 0.05. Significant differences between time points in occurrence of deformed fish were tested by Chi-square tests with a Bonferroni corrected alfa level of 0.0083. Significant differences between time points in deformity severity (number of deformed vertebrae among deformed fish) were tested by Kruskal-Wallis ANOVA with an alfa level of 0.05. Significant differences between time points in vertebra morphometrics (length / diameter) within vertebra regions were tested by one-way ANOVAs with an alfa level of 0.05. Possible significant relationships between measured parameters were tested by simple linear regression.

3. Results

3.1. Main study

3.1.1. Fish growth and deformities

The fish grew from 36 g to 5486 g throughout the one-year study period (Table 1). There was a significant and close to two-fold increase in CF from 1.33 at start to 2.21 at termination (Table 1).

There was a continuous and significant increase over time with respect to the occurrence of fish with one or more deformed vertebrae vs. non-deformed fish (Fig. 1A). The level increased from 20 % at 36 g body size (April 2022) to 93 % at 5.5 kg (May 2023), and there was a significant increase (Chi-square test, Bonferroni corrected alfa level; $P < 0.0008$) between all timepoints except between April and June 2022 (36

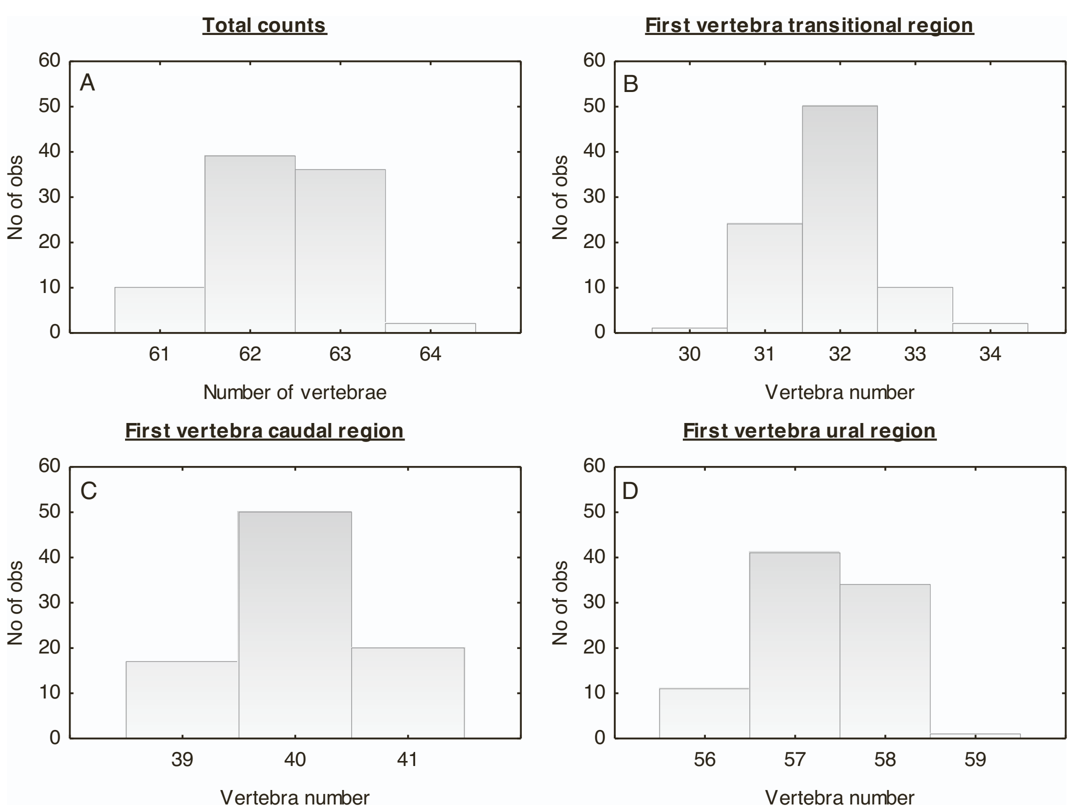

Fig. 2. Main study - vertebra meristics June 2022 (n = 90). (A) Total counts, same as the number of the last vertebra in the ural region, includes the most caudal vertebra which is fused to the urostyle (U2, Schultze and Arratia, 1989). (B) The number of the most cranial vertebra of the transitional region. (C) Number of the most cranial vertebra of the caudal region. (D) Number of the most cranial vertebra of the ural region. The regions were defined based on specific radiographic hallmarks outlined by Sankar et al. (2024).

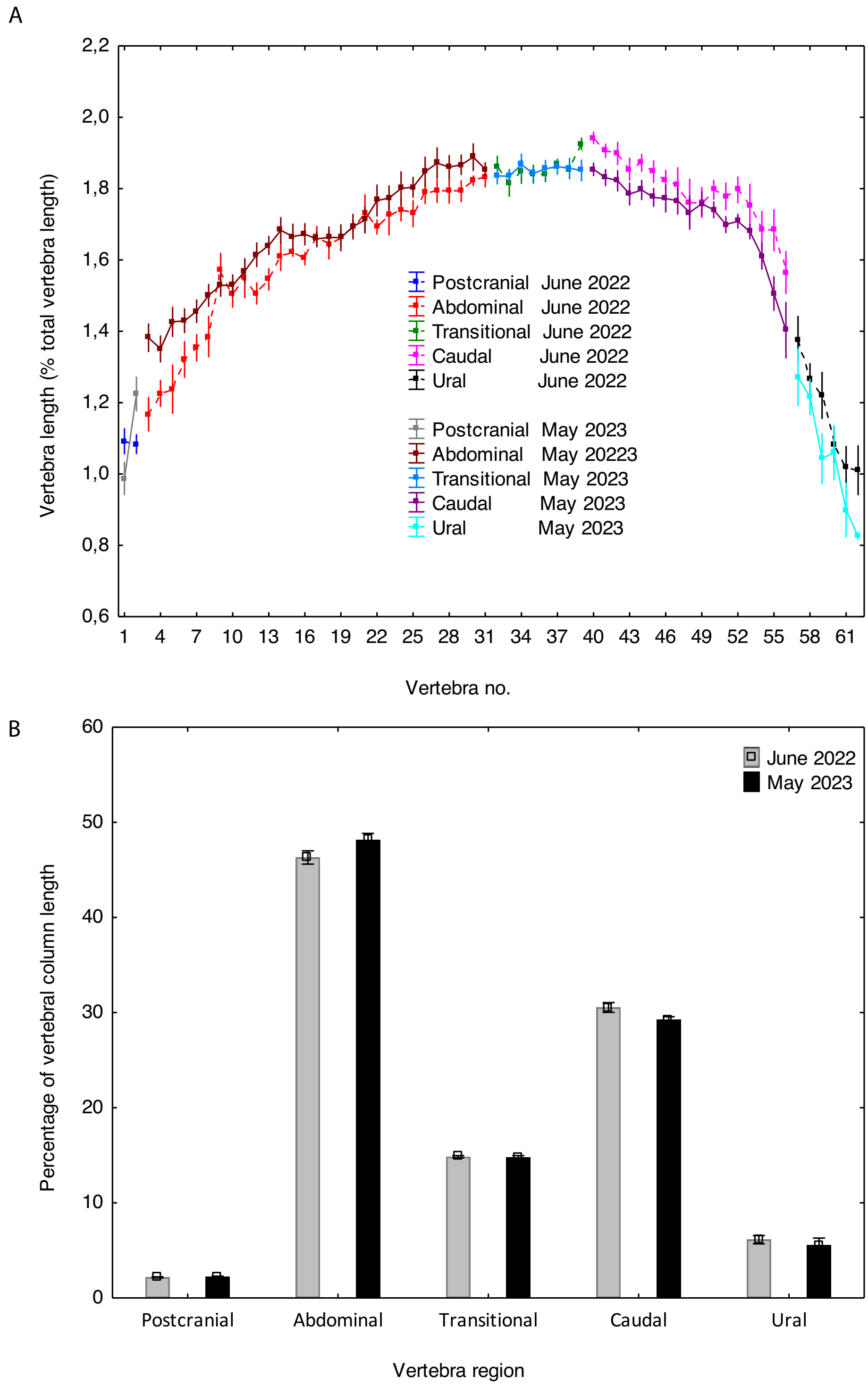

Fig. 3. Main study - standardized vertebra cranio-caudal lengths (mean ± SE). Relative lengths of single vertebra calculated as percentage of the sum of all vertebra lengths per individual at two different time points, June 2022 ($n = 6$, 160 g) and May 2023 (n = 6, 5.5 kg). Different vertebra regions are indicated; the postcranial, abdominal, transitional, caudal and ural regions. (A) Data on single vertebra numbers. (B) Data on the sum of relative lengths per region to calculate the relative size (length) contribution of each region.

%).

With respect to deformity severity, data analyses showed the same significant (Kruskal-Wallis ANOVA, $P < 0.0001$) trend as for the deformity occurrence (Fig. 1B). There was a three-fold increase in the mean number of deformed vertebrae among the deformed fish from 36 g in April 2022 (4 def. vertebrae) to 5.5 kg in May 2023 (12 def. vertebrae). Number of deformed vertebrae among deformed fish was significantly higher in May 2023 compared to all earlier timepoints, and significantly higher in December 2022 compared to the initial sampling in April 2022 (Kruskal-Wallis multiple comparison post hoc test, $P < 0.05$).

With respect to possible impact of deformity severity on growth and CF, there were significant linear regressions between number of deformed vertebrae and length, weight and CF in both December 2022 and May 2023 (Table 2). However, the relationships were all stronger in

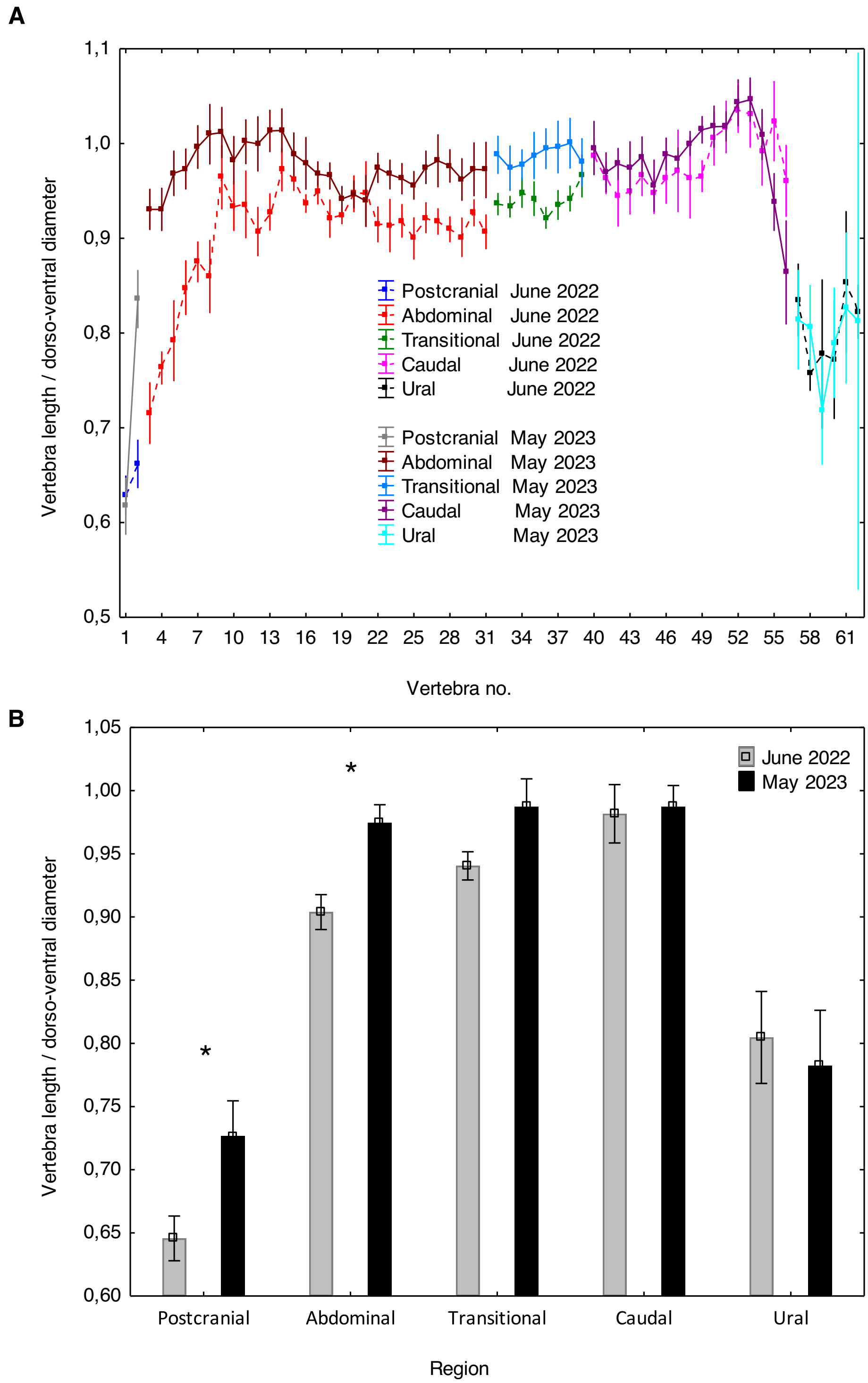

Fig. 4. Main study - vertebra dimensions (mean ± SE). The ratio between vertebra cranio-caudal length and dorso-ventral diameter in the postcranial, abdominal, transitional, caudal and ural regions of the vertebral column at two different time points, June 2022 (n = 6, 160 g) and May 2023 (n = 6, 5.5 kg). (A) Data on single vertebra numbers. (B) Data on average values per region. Black asterisk indicates significant differences within regions (one-way ANOVA, $P < 0.05$).

May 2023 (5.5 kg) compared to December 2022 (2.2 kg), and there were negative relationships between deformity severity and length and weight, and positive relationships between deformity severity and CF (Table 2).

3.1.2. Regionalization of the vertebral column

The total vertebrae counts varied between 61 and 64, with 62 and 63 as the most common total counts (Fig. 2A). The postcranial region comprised of vertebra 1 and 2, the most common last vertebra of the abdominal region was vertebra 31 (min: 29, max: 33), the transitional region started at vertebra 32 (min: 30, max 34, Fig. 2B) and ended at vertebra 39 (min: 38, max 40), the caudal region typically started at vertebra 40 (min: 38, max: 41, Fig. 2C) and ended at vertebra 56 (min: 55, max: 58), while the ural region typically started at vertebra 57 (min: 56, max: 59, Fig. 2D) and ended at vertebra 62 (min: 61, max: 64).

The size and dimensions of the individual vertebrae varied between

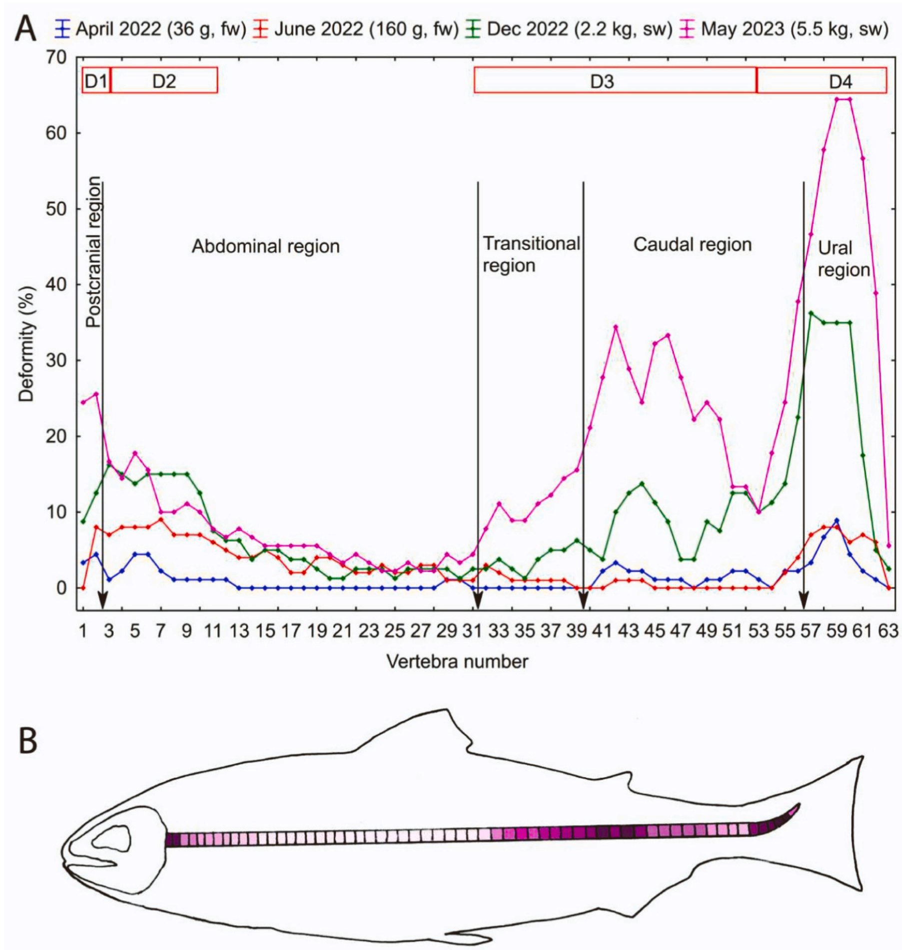

Fig. 5. Main study – vertebral deformity location. (A) Occurrence (%) of deformed vertebra per vertebra number along the vertebral column at four different timepoints: April 2022 (36 g, freshwater, $n = 90$), June 2022 (160 g, freshwater, $n = 100$), December (2.2 kg, seawater, $n = 80$), May 2023 (5.5 kg, seawater, n = 90). The fish were reared in freshwater tanks until August 2022 when they were transferred to sea-cages. The vertebral column is regionalized into 5 regions according to Sankar et al. (2024). Different deformity centra (D) indicated with D1–4. (B) Schematic drawing illustrating the location and level of deformity occurrence at termination in May 2023. White vertebrae illustrate low occurrence, increasing colour intensity illustrates increasing deformity rate.

the different vertebral regions. The cranio-caudal length of the single vertebra centra increased until the beginning of the caudal region (V40) and then decreased gradually towards the tail in June 2022 (160 g), while in the May 2023 sampling (5.5 kg), the vertebra length increased until the end of the abdominal region (V30) followed by a gradually decrease towards the tail (Fig. 3A). Vertebra cranio-caudal length / dorso-ventral diameter ratio was significantly higher (more elongated vertebrae; one-way ANOVA, $P < 0.05$) in the postcranial and abdominal region in May 2023 compared to June 2022 (Fig. 4B).

3.1.3. Vertebral deformity location

There was a development of distinct patterns in deformity prevalence along the vertebral column during the one-year study period where the fish grew from 36 g to 5.5 kg. Four deformity centres were evident:

Deformity centre no. 1 was in the postcranial region. There was an increased deformity rate in V1–2 in seawater, with the highest increase between 2.2 and 5.5 kg body weight (Fig. 5A,B). At 5.5 kg (May 2023), these deformities were mostly compressions and fusions (type 6, Witten et al., 2009) (Fig. 6A,B).

Deformity centre no. 2 was in the anterior part of the abdominal region. The most severe deformities developed between V3 and V10 (Fig. 5A), and these deformities gave the affected fish a distinct 'humpback' external phenotype. The deformities that developed in this specific area were already frequent when the fish were 160 g (June 2022) but there were already some pathologies at 36 g (April 2022) (Fig. 5A). At 36 g body weight, 75 % of the deformities in V3–10 were a reduction of the intervertebral space (type 1, Witten et al., 2009) (Fig. 6C,D). This deformity type reflects early pathological changes (Fjelldal et al., 2007, 2009a) and its prevalence fell with time: 38 % June 2022, 3 % December 2022 and 7 % May 2023. On the contrary, there was 0 % prevalence of fusion centre vertebrae (type 8, Witten et al., 2009) (Fig. 6E) at the first sampling, and its occurrence rose with time: 18 % June 2022, 78 % December 2022, and 56 % May 2023. Fusion centre is a terminal deformity stage (Fjelldal et al., 2007, 2009a) and was the predominate deformity type in V3–10 in December 2022 and May 2023. In fish with fusion centre in V3–10, the fusion centre often also involved one or both postcranial vertebrae (V1–2) (Fig. 6E). The remaining of the abdominal region – vertebra 11 to 31 – also had an increased deformity rate over time but it was lower compared to the remaining of the vertebral column (Fig. 5A,B).

Deformity centre no. 3 covered the transitional and caudal regions. There was an increase in deformity rate in this area from 160 g in freshwater to 2.2 kg in seawater, which further increased up to 5.5 kg (May 2023) (Fig. 5A,B). The most common changes observed in this deformity centre were early pathologies: one-sided compressions (45 %, type 5, Witten et al., 2009), two-sided compressions (38 %, type 2 and 3), and reduced intervertebral space (15 %, type 1) (Fig. 7A-D).

Deformity centre no. 4 covered the ural region. At 36 g body size the deformity rate was 9 % in V59. The deformity rate in V59 did not

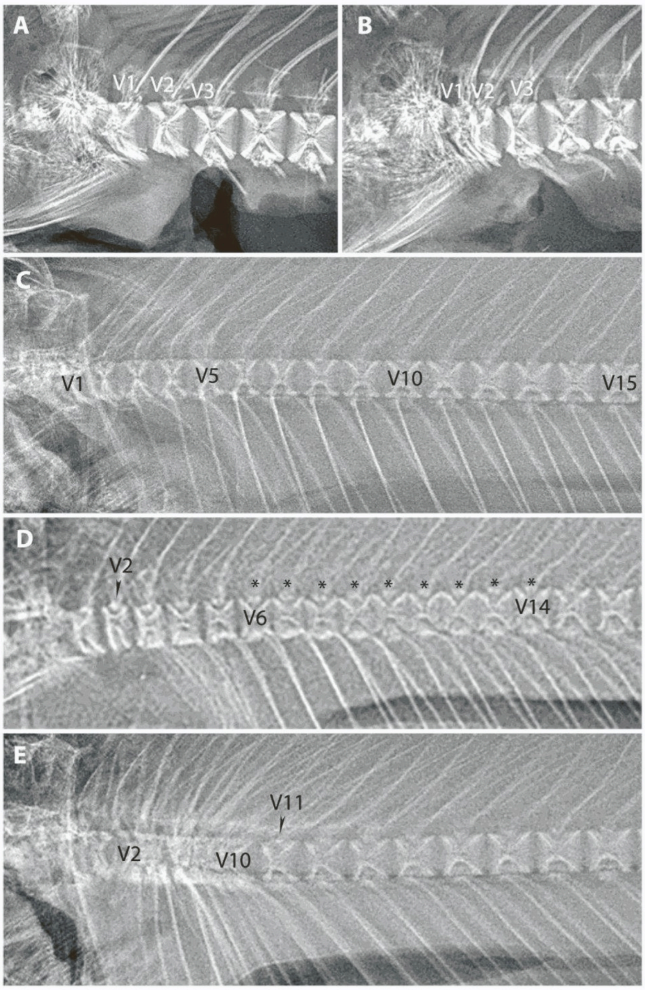

Fig. 6. Main study - lateral radiology postcranial and abdominal region deformities (Deformity centre 1 and 2). (A) Normal phenotype in May 2023 (5.5 kg). (B) Fusion of V1 and 2 in May 2023. (C) Normal phenotype in December 2022 (2.2 kg). (D) One-sided compression of V2 (black arrowhead) and lack of intervertebral space V6 to V14 (black asterisk) in June 2022 (160 g). (E) Fusion centre V2 to V10 and one-sided compression V11 (black arrowhead) in December 2022. Deformity types classified according to Witten et al. (2009).

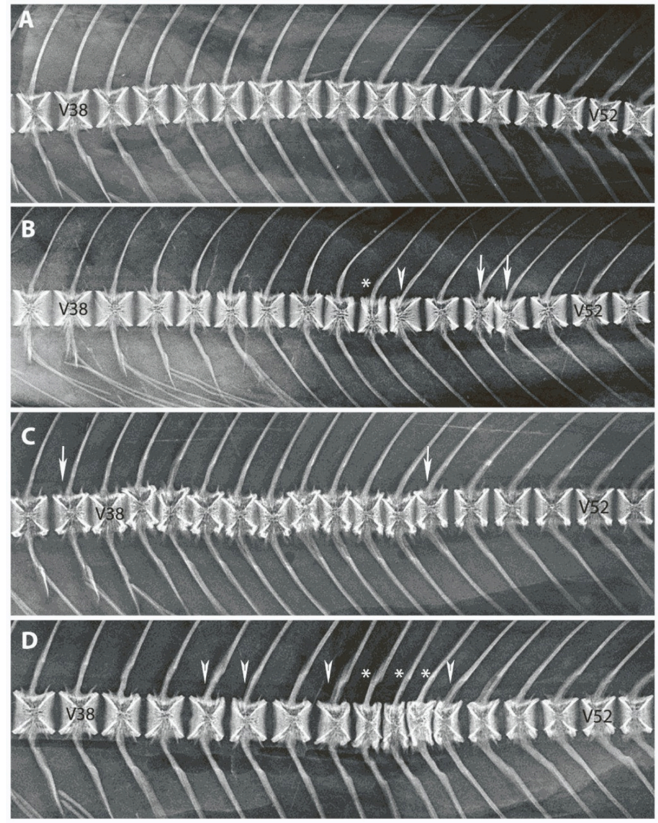

Fig. 7. Main study - lateral radiology caudal region deformities (Deformity centre 3) in May 2023 (5.5 kg). (A) Normal phenotype. (B–D) Different examples of caudal region deformities. Examples of vertebra with different deformity types are indicated: one-sided compressions (white arrowhead), two-sided compressions (white asterisk), lack of intervertebral space (white arrow). Deformity types classified according to Witten et al. (2009).

increase between 36 and 160 g (8 %) in freshwater but showed a clear increase in seawater up to 5.5 kg (2.2 kg: 36 %, 5.5 kg: 65 %) (Fig. 5A,B). The deformities in the ural region spread into the most posterior vertebrae of the caudal region. One-sided compressions dominated among ural deformities at 36 g (45 %), while compression and fusion (type 6) were the predominate types at the later samplings (46–61 %) (Fig. 8A-F). In concert with the increasing prevalence of ural deformities over time, there was an obvious change in the condition of the caudal fin between 2.2 and 5.5 kg in seawater (Fig. 9). There was a significant reduction in the ratio between caudal fin length and fork length over time, while the morphology of the caudal fin measured as the ratio between the length and the height of the fin did not change (Fig. 9).

3.2. Extra fish groups

All extra groups had a high occurrence (Extra 1: 83 %, Extra 2: 74 %, Extra 3: 81 %) of fish with one or more deformed vertebrae (Fig. 10A), and the mean number of deformed vertebrae in affected fish ranged between 7.6 and 10.1 (Fig. 10B). The location of the deformities throughout the vertebral column is shown Fig. 11. The Extra 1 group had highest occurrence of deformed postcranial vertebrae. The Extra 2 and 3 groups had highest occurrence of deformed transitional and caudal vertebrae. All groups had a high occurrence of deformed ural vertebrae, and the Extra 1 and 3 groups were higher compared to the Extra 2 group. The deformity rate in V58 was 65 % in Extra 1, 39 % in Extra 2, and 61 % in Extra 3. Among the Extra 2 and 3 groups, 3.3 % of the fish had kyphosis and/or lordosis.

4. Discussion

The present study show that Rainbow Trout developed deformities in different regions of the vertebral column as they grew from 36 g in freshwater up to 5.5 kg in seawater. The resulting deformities, covering most of the vertebral column, had a negative effect on fish length and weight in seawater (>2 kg) a strong indication of reduced fish welfare. Most of the fish developed deformities in the ural region of the vertebral column in concert with a degradation of the lepidotrichs of the caudal fin.

4.1. Vertebral deformities reduced growth

As the fish grew more and more fish were affected by vertebral deformities. The increase in deformity rate was caused by a steady increase of deformities in different regions of the vertebral column. On the final sampling when the fish were on average 5.5 kg, 93 % of the fish had one or more deformed vertebrae, and these fish had on average 12 deformed vertebrae each. Hansen et al. (2010) found that in Atlantic Salmon, which have a comparable number of total vertebrae as Rainbow Trout, fish with more than 10 deformed vertebrae showed reduced growth rate

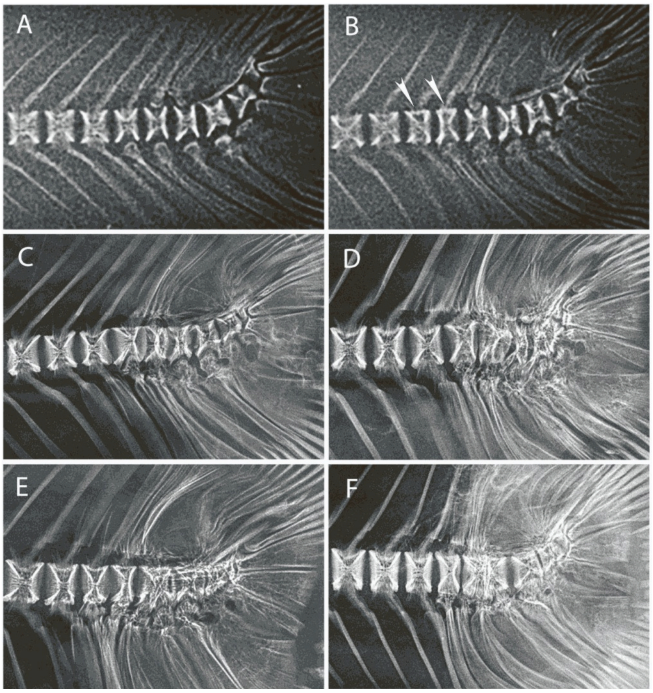

Fig. 8. Main study - lateral radiology of ural region deformities (Deformity centre 4). (A) Normal phenotype in June 2022 (160 g). (B) Individual with mild one-sided compressions (white arrowheads) in June 2022. (C–F) Various degrees of vertebra compression and fusion in May 2023 (5.5 kg).

up to harvest size in seawater. Likewise, the present study showed a significant negative relationship between number of deformed vertebrae and harvest length and weight in Rainbow Trout. Hence, the present study supports vertebral deformities as an important welfare indicator for Rainbow Trout grown out to large size in seawater.

4.2. High foraging activity as a possible explanation for deformity development

In the present study, most fish had 62 or 63 vertebrae which agrees with Kacem et al. (2004) who reported 61 and 62 vertebrae as most common counts. Also, similar to Kacem et al. (2004), the present study showed 4 deformity centres in the vertebral column of Rainbow Trout. The first centre was in the postcranial region (V1–2), where vertebra fusion dominated over time. This is also common in adult Atlantic salmon and may be an age-related skeletal change (Fjelldal et al., 2009b). However, the occurrence reported herein is much higher than that reported in Atlantic Salmon and may indicate a pathological condition. Deformities in postcranial vertebrae have not been reported in wild Rainbow Trout (McCrimmon and Bidgood, 1965; Van Velson, 1978), which supports this notion. The second centre was between V3 and V10 which is the anterior part of the abdominal region. Interestingly, V10 is at border of the postcranial region defined by Ramzu and Meunier (1999) which based their regionalization on vertebral morphometrics. Thus, the vertebral deformities developing between V3 and V10 may be linked to vertebral morphometrics and/or functional demands. Indeed, recent research has revealed that Rainbow Trout bends the cranial portion of the vertebral column dorsally and ventrally during feeding (Camp, 2021; Jimenez and Camp, 2023). This illustrates that vertebral column regionalization systems based on anatomy (Sankar et al., 2024) and morphometrics (Ramzu and Meunier, 1999) can be combined in deformity research on salmonids. The presently studied population developed the most severe deformities between V3 and V10, which gave a clear external phenotype. Domesticated Rainbow Trout have an extreme appetite resulting in high foraging activity. This may make domesticated Rainbow Trout susceptible for deformities in the cranial part of the vertebral column which may explain the development of the first (V1–2) and second (V3–10) deformity centres found in this study. Studies on smaller Rainbow Trout in freshwater have also pointed out deformities in the cranial portion of the vertebral column (Kacem et al., 2004; Deschamps et al., 2014). This agrees with the results of the present study where these deformities were existing already in freshwater as early developing pathologies (one-sided compression, reduced intervertebral space) which changed to terminal stage pathologies (fusion) towards the end of the study. Similar results have also been described in Atlantic Salmon (Fjelldal et al., 2007).

4.3. Caudal fin degradation as a possible explanation for deformity development

The third deformity centre covered the transitional and caudal regions. These deformities increased in occurrence as the fish grew bigger in seawater, and the pathologies were mostly early stages at the final sampling. Similar observations have been recorded in Atlantic Salmon (*Salmo salar*) (Fjelldal et al., 2009c; Grini et al., 2011) and Rainbow Trout (Deschamps et al., 2009), where the authors have suggested a link to high mechanical loading imposed by the lateral swimming musculature in this region. However, in Rainbow Trout, these deformities may also be coupled to the deformities developing in the ural region, the fourth deformity centre addressed herein, and/or to the degradation of the caudal fin lepidotrichs. Sixty-five percent of the fish had a deformity in vertebra nos. 59 and 60 at the final sampling. These vertebrae belong to the caudal fin complex, which also suffered from a degradation of the fin lepidotrichs (rays). The latter condition was severe at the final sampling and must have had an absolute negative effect on swimming capabilities. In turn, this handicap has most probably challenged the vertebral column through its effect on swimming muscular activity. Vertebra cranio-caudal lengths increased from the head until vertebra no. 40 and then decreased further towards the tail at 160 g size in freshwater, while this shift point had moved towards the head to vertebra no. 30 at 5.5 kg size in seawater. A cranial shift in vertebra maximum length with age/size has not been observed in Atlantic Salmon (Fjelldal et al., 2009c), but in Atlantic Cod (*Gadus morhua*) (Fjelldal et al., 2013). Whether the present observation in Rainbow Trout reflects a compensatory mechanism related to the degradation of the caudal fin lepidotrichs or a natural ontogenetic change is at present unknown. Nevertheless, the observed age/size related change in vertebra maximum length was associated with proportional changes of the vertebral bodies in the postcranial and abdominal regions, shown by an increase in vertebra length relative to diameter – more elongated vertebrae – in these regions as the fish grew bigger.

4.4. Comparative evidence from farmed Atlantic Salmon

Based on the literature on deformity development in farmed Atlantic Salmon, possible deformity inducing factors can be suggested. The early pathologies currently observed in the main study population were one-sided compressions and reduced intervertebral spaces. The latter often combined with dorso-ventral shifts between adjacent vertebrae (Witten et al., 2009). In Atlantic salmon, reduced intervertebral space and dorso-ventral shifts can develop in the cranial portion of the abdominal region in freshwater and change into fusions over time (Fjelldal et al., 2007), but may also develop in the caudal region in seawater as a negative response to intra-abdominal vaccination (Thorarinsson et al., 2024). In the latter context more recently referred to as 'cross stich vertebrae'. Studies on this condition have reported cartilage deposition between the endplates of adjacent vertebrae and inside the intertrabecular spaces (Holm et al., 2020), and evidence of gas inside the notochord, along with perforations in the vertebral endplates with either notochordal tissue prolapsing into the vertebral body or vascularized fibrochondroid proliferations extending into the notochord (Trangerud et al., 2020). The

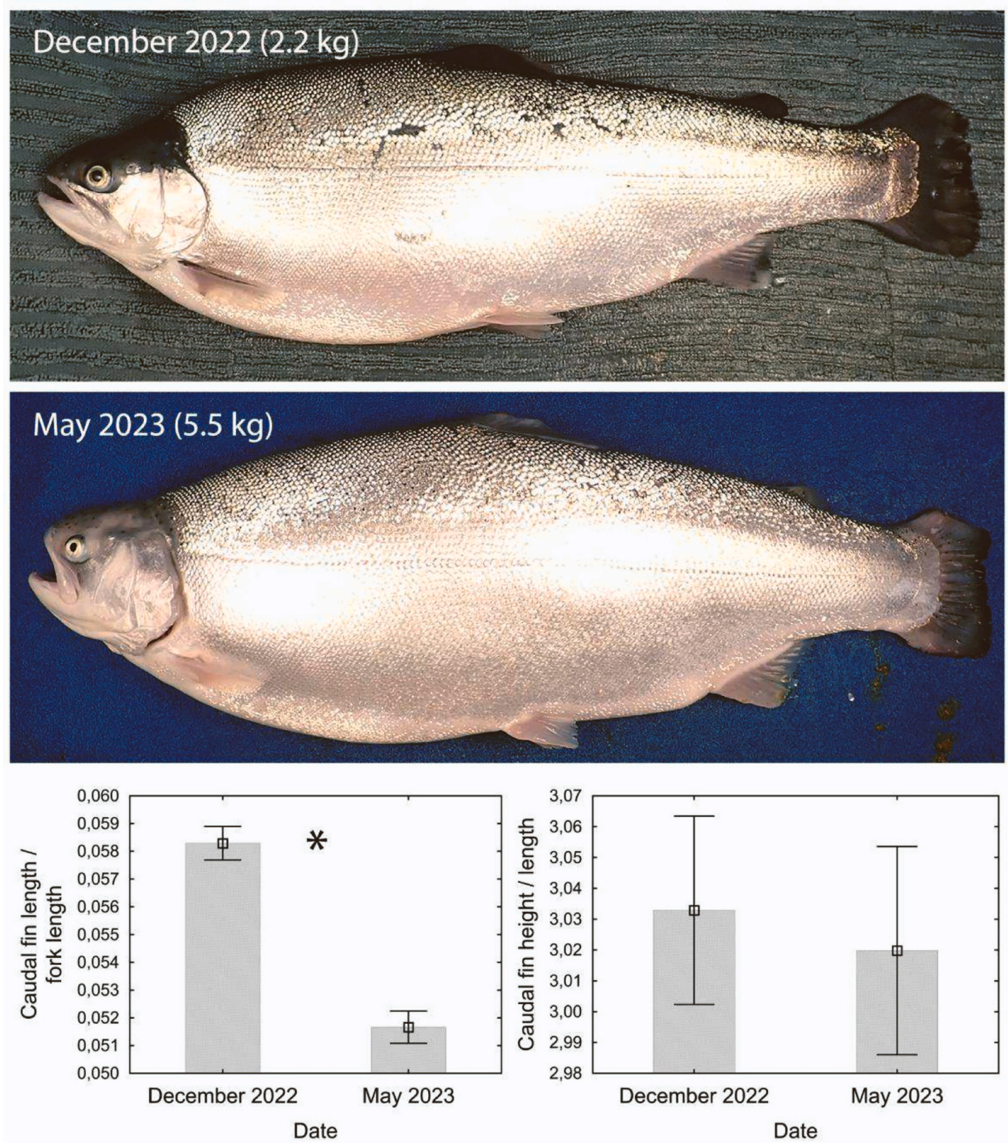

Fig. 9. Main study - caudal fin condition in seawater. The upper two panels show pictures of fish from December 2022 and May 2023. Note the small and degraded tail fins. There was a significant reduction in the ratio between caudal fin length and fork length between December 2022 ($n = 80$) and May 2023 ($n = 90$) (lower left panel). The shape of the caudal fin measured as the ratio between caudal fin length height and length did not change over time (lower right panel). Data are show as mean ± standard error. Black asterisk indicates significant difference (one-way ANOVA, $P < 0.05$).

pioneer work by Kvellestad et al. (2000) reported an infiltration by inflammatory cells around deformed vertebrae and suggested that vertebral deformities in farmed Atlantic salmon starts in intervertebral tissues. Signs of inflammation has later been reported in ‘cross stich vertebrae’ (Holm et al., 2020; Trangerud et al., 2020). The currently studied trout showed ‘cross stich vertebrae’ both in the cranial part of the abdominal and in the caudal regions, developing before and after vaccination, respectively. Vertebra compression has been associated with reduced mechanical strength of the vertebral bodies (Baeverfjord et al., 1998), and a changed growth pattern of the vertebral body endplates along with a replacement of notochordal tissue by cartilage (Witten et al., 2005). Limitations in dietary phosphorus (Baeverfjord et al., 1998; Fjelldal et al., 2009c) and elevated temperature (Ytteborg et al., 2010; Grini et al., 2011) can result in vertebral compression in Atlantic Salmon. Vertebral deformity development in Rainbow Trout may indeed involve the combined forces of the possible inducing factors earlier defined for Atlantic salmon, and the new factors proposed in this study for trout (i.e. foraging activity, caudal fin degradation).

4.5. A widespread problem in Rainbow Trout in seawater

The results from the main study and the extra groups together suggest that ural region vertebral deformities are a wide-spread problem in seawater farming of Rainbow Trout, and that this most probably links to the external phenotype of the fish. It may be that the early domestication and the subsequent selective breeding of Rainbow Trout has changed the phenotype of the fish to an extent so that it challenges normal bone development and bone health. As a parallel example, rapid growth imposes a high risk for leg bone disorders in broiler chicken (Liu et al., 2023), where genetic selection has been suggested as a possible redemption for disease prevention and improved leg health. Earlier studies on wild Rainbow Trout support the notion that ural region vertebral deformities is a wide-spread problem among cultured Rainbow Trout, and do not represent ‘normal variation’. McCrimmon and Bidgood (1965) radiographed 291 wild Rainbow Trout captured in five different Great Lakes watersheds and 80 hatchery reared Rainbow Trout of the Normandale stock and found 7.6 and 3.8 % fish with vertebral

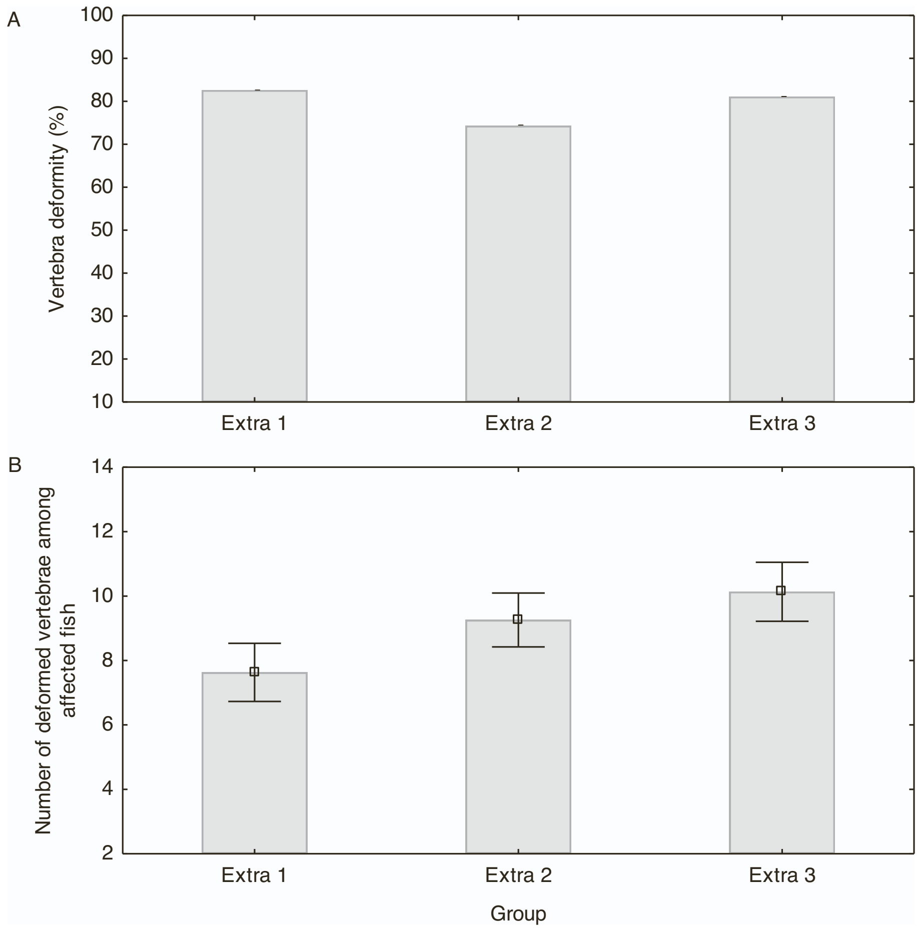

Fig. 10. Extra groups - vertebral deformity occurrence and severity. (A) Occurrence (percentage) of fish with one or more deformed vertebrae. (B) Number of deformed vertebrae (mean ± SE) among deformed fish only, reflecting severity. Number of fish examined: Extra 1 (n = 23), Extra 2 (n = 105), Extra 3 (n = 74).

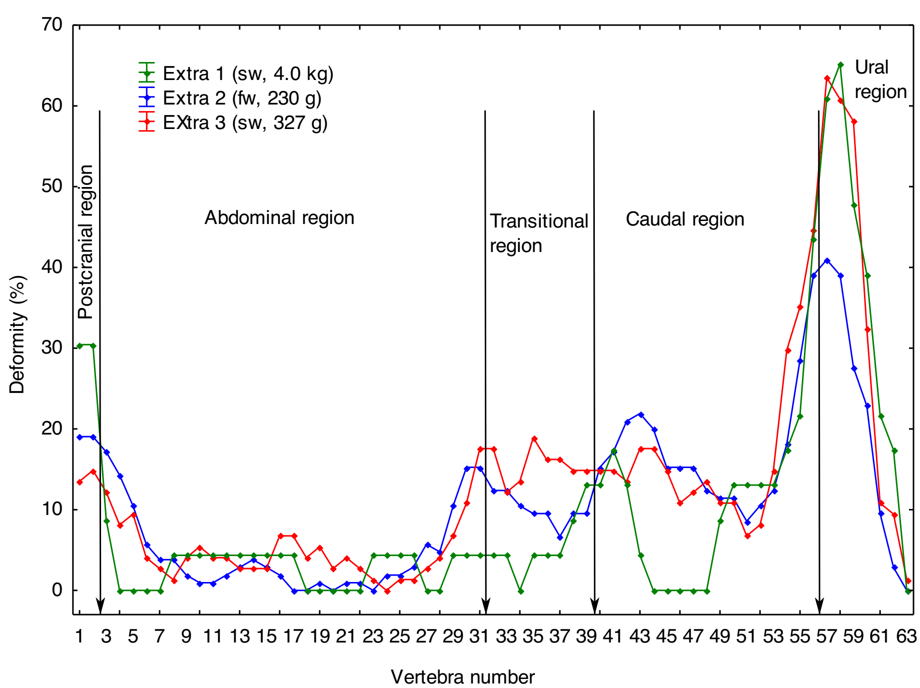

Fig. 11. Extra groups - vertebral deformity location. Occurrence (%) of deformed vertebra per vertebra number along the vertebral column. The vertebral column is regionalized into 5 regions according to Sankar et al. (2024). Number of fish examined: Extra 1 (n = 23), Extra 2 (n = 105), Extra 3 (n = 74).

deformities, respectively. Of the totally 25 deformed trout (wild and reared pooled), 23 had deformed vertebrae located in the abdominal region between the dorsal and pelvic fins (6.2 %), 1 had deformities in vertebrae number 5 to 9 (0.3 %), and 1 had deformities in vertebrae number 54 and 55 (0.3 %) (McCrimmon and Bidgood, 1965). The average number of deformed vertebrae among deformed fish was 6.2 in that study (wild and reared pooled). Van Velson (1978) examined 4944 adult wild Rainbow Trout trapped in the upper North Platte River drainage in Nebraska and found 4.6 % fish with vertebral deformities, which were categorized into two types; vertebral column curvature (lordosis or scoliosis), and deformity in the caudal peduncle with vertebra 'smashed or jammed' together giving the fish a 'stubby' appearance. The examined fish in the latter study may have contained some hatchery reared fish (likely low) and may have been exposed to electricity early in life. Van Velson (1978) concluded that if deformities approach 8 to 10 %, they should no longer be considered natural, and investigations should be made to determine the cause. Like the studies on wild Rainbow Trout, a radiological study on wild Sockeye (*Oncorhynchus nerka*), Pink (*Oncorhynchus gorbuscha*), and Chum Salmon (*Oncorhynchus keta*) showed a low occurrence of vertebral deformities (2.8–3.3 %, Gill and Fisk, 1966).

5. Conclusions

Rainbow Trout farmed to a large size in seawater have a high risk of developing vertebral deformities. The most severe deformities start to develop early and affect the anterior part of the abdominal region of the vertebral column. The most abundant deformities develop in the ural region of the vertebral column and may be linked to degradation of the caudal fin lepidotrichs. The latest developing deformities affect the postcranial, transitional, and caudal regions of the vertebral column. Together, these deformities cover a large portion of the vertebral column and depress fish growth. Vertebral deformities are thus an important individual based welfare indicator for farmed Rainbow Trout. Based on experience from farmed Atlantic Salmon relevant research topics for finding a solution may be mineral nutrition, rearing temperatures, and vaccine side effects. In addition, improvement of caudal fin health should be a top priority.

CRediT authorship contribution statement

Per Gunnar Fjelldal: Writing – original draft, Visualization, Validation, Supervision, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **Sankar Murugesan:** Writing – original draft, Methodology, Formal analysis, Data curation. **Tone Vågseth:** Writing – review & editing, Software, Methodology, Formal analysis, Data curation. **Audun Østby Pedersen:** Writing – review & editing, Software, Methodology, Investigation, Data curation. **Angelico Madaro:** Writing – review & editing, Validation, Project administration, Investigation, Formal analysis. **Samantha Bui:** Writing – review & editing, Validation, Resources, Project administration. **Harald Kryvi:** Writing – review & editing, Visualization, Methodology, Formal analysis, Conceptualization. **Lars Helge Stien:** Writing – review & editing, Validation, Resources, Project administration, Investigation, Conceptualization. **Jonatan Nilsson:** Writing – original draft, Visualization, Software, Project administration, Methodology, Investigation, Formal analysis, Data curation, Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

Acknowledgements

This study was funded by the Institute of Marine Research (internal project 'Overvåking velferd', no. 1493
0, project leader Lars Helge Stien). The authors want to thank the staff at Matre Research Station, especially Trond Asheim, for taking care of the fish and helping out during sampling. The authors want to thank the two anonymous reviewers for their valuable insight and helpful comments.

References

Aas, T.S., Åsgård, T., Ytrestøyl, T., 2022. Chemical composition of whole body and fillet of slaughter sized Atlantic salmon (*Salmo salar*) and Rainbow Trout (*Oncorhynchus mykiss*) farmed in Norway in 2020. Aquacult. Rep. 25, 101252.

Aubin, J., Gatesoupe, F.J., Labbé, L., Lebrun, L., 2005. Trial of probiotics to prevent the vertebral column compression syndrome in Rainbow Trout (*Oncorhynchus mykiss* Walbaum). Aquac. Res. 36, 758–767.

Aydın, İ., Alemdağ, M., Terzi, Y., Öztürk, R.Ç., Küçük, E., Polat, H., Çoban, D., 2024. Ontogeny of vertebral column and fin development, and detection of deformations in hatchery-reared European flounder (*Platichthys flesus*) during early life stage. Aquaculture 582, 740498.

Baeverfjord, G., Åsgård, T., Shearer, K.D., 1998. Development and detection of phosphorus deficiency in Atlantic salmon, *Salmo salar* L., parr and post-smolts. Aquac. Nutr. 4, 1–11.

Barrington, D., 1767. XXIII. A letter to Dr. William Watson, FRS from the Hon. Daines Barrington, FRS on some particular fish found in Wales. Philos. Trans. R. Soc. Lond. 57, 204–214.

Boglione, C., Gagliardi, F., Scardi, M., Cataudella, S., 2001. Skeletal descriptors and quality assessment in larvae and post-larvae of wild-caught and hatchery-reared gilthead sea bream (*Sparus aurata* L. 1758). Aquaculture 192, 1–22.

Boglione, C., Gavaia, P., Koumoundouros, G., Gisbert, E., Moren, M., Fontagné, S., Witten, P.E., 2013a. Skeletal anomalies in reared European fish larvae and juveniles. Part 1: Normal and anomalous skeletogenic processes. Rev. Aquac. 5, S99–S120.

Boglione, C., Gisbert, E., Gavaia, P., Witten, P.E., Moren, M., Fontagné, S., Koumoundouros, G., 2013b. Skeletal anomalies in reared European fish larvae and juveniles. Part 2: main typologies, occurrences and causative factors. Rev. Aquac. 5, S121–S167.

Boglione, C., Pulcini, D., Scardi, M., Palamara, E., Russo, T., Cataudella, S., 2014. Skeletal anomaly monitoring in Rainbow Trout (*Oncorhynchus mykiss*, Walbaum 1792) reared under different conditions. PLoS One 9, e96983.

Branson, E.J., Turnbull, T., 2008. Welfare and deformities in fish. Fish welfare 202–216.

Camp, A.L., 2021. A neck-like vertebral motion in fish. Proc. R. Soc. B 288, 20211091.

Chandra, G., Saini, V.P., Kumar, S., Fopp-Bayat, D., 2024. Deformities in fish: a barrier for responsible aquaculture and sustainable fisheries. Rev. Aquac. 16, 872–891.

Chatzakis, D., Al Belbeisi, R., Karagianni, S., Karagiannakidis, E., Koumoundouros, G., 2024. Prevalence and typology of skeletal abnormalities in fishes of the eastern Mediterranean. J. Fish Dis. e13992.

Dawson, C.E., 1964. A bibliography of anomalies of fishes. Gulf Carib. Res. 1, 308–399.

De Clercq, A., Perrott, M.R., Davie, P.S., Preece, M.A., Wybourne, B., Ruff, N., Huysseune, A., Witten, P.E., 2017. Vertebral column regionalisation in Chinook salmon, *Oncorhynchus tshawytscha*. J. Anat. 231, 500–514.

Deschamps, M.H., Kacem, A., Ventura, R., Courty, G., Haffray, P., Meunier, F.J., Sire, J. Y., 2008. Assessment of "discreet" vertebral abnormalities, bone mineralization and bone compactness in farmed rainbow trout. Aquaculture 279, 11–17.

Deschamps, M.H., Labbé, L., Baloche, S., Fouchereau-Péron, M., Dufour, S., Sire, J.Y., 2009. Sustained exercise improves vertebral histomorphometry and modulates hormonal levels in rainbow trout. Aquaculture 296, 337–346.

Deschamps, M.H., Poirier Stewart, N., Demanche, A., Vandenberg, G.W., 2014. Preliminary study for phenotypic description of vertebral abnormalities in triploid trout subjected to prolonged deficiency in phosphorus. J. Appl. Ichthyol. 30, 833–839.

Eissa, A.E., Abu-Seida, A.M., Ismail, M.M., Abu-Elala, N.M., Abdelsalam, M., 2021. A comprehensive overview of the most common skeletal deformities in fish. Aquac. Res. 52, 2391–2402.

FAO, 2022. The State of World Fisheries and Aquaculture 2022. Towards Blue Transformation. FAO, Rome. https://doi.org/10.4060/cc0461en.

FAO, 2024. *Oncorhynchus mykiss*. Cultured Aquatic Species Information Programme. Text by Cowx, I. G. Fisheries and Aquaculture, Rome. https://www.fao.org/fishery/en/culturedspecies/oncorhynchus_mykiss/en.

FHF workshop, 2023. Arbeidsmøte om FoU-utfordringer ved oppdrett av regnbueørret, 21 March 2023. Clarion Hotel Bergen Airport, Flyplassvegen, 551, p. 5869. Bergen. https://www.fhf.no/arrangementer/arrangementer/arbeidsmoete-om-fou-utfordringer-ved-oppdrett-av-regnbueoerret/.

Fjelldal, P.G., Hansen, T.J., Berg, A.E., 2007. A radiological study on the development of vertebral deformities in cultured Atlantic salmon (*Salmo salar* L.). Aquaculture 273, 721–728.

Fjelldal, P.G., van der Meeren, T., Jørstad, K.E., Hansen, T.J., 2009a. A radiological study on vertebral deformities in cultured and wild Atlantic cod (*Gadus morhua*, L.). Aquaculture 289, 6–12.

Fjelldal, P.G., Glover, K.A., Skaala, Ø., Imsland, A., Hansen, T.J., 2009b. Vertebral body mineralization and deformities in cultured Atlantic salmon (*Salmo salar* L.): effects of genetics and off-season smolt production. Aquaculture 296, 36–44.

Fjelldal, P.G., Hansen, T., Breck, O., Sandvik, R., Waagbø, R., Berg, A., Ørnsrud, R., 2009c. Supplementation of dietary minerals during the early seawater phase increase vertebral strength and reduce the prevalence of vertebral deformities in fast-growing under-yearling Atlantic salmon (*Salmo salar* L.) smolt. Aquac. Nutr. 15, 366–378.

Fjelldal, P.G., Hansen, T., Breck, O., Ørnsrud, R., Lock, E.J., Waagbø, R., Wargelius, A., Witten, P.E., 2012. Vertebral deformities in farmed Atlantic salmon (*Salmo salar* L.) – etiology and pathology. J. Appl. Ichthyol. 28, 433–440.

Fjelldal, P.G., Totland, G.K., Hansen, T., Kryvi, H., Wang, X., Søndergaard, J.L., Grotmol, S., 2013. Regional changes in vertebra morphology during ontogeny reflect the life history of Atlantic cod (*Gadus morhua* L.). J. Anat. 222, 615–624.

Fjelldal, P.G., Madaro, A., Hvas, M., Stien, L.H., Oppedal, F., Fraser, T.W., 2021. Skeletal deformities in wild and farmed cleaner fish species used in Atlantic salmon *Salmo salar* aquaculture. J. Fish Biol. 98, 1049–1058.

Fontagné, S., Silva, N., Bazin, D., Ramos, A., Aguirre, P., Surget, A., Abrantes, A., Kaushik, S.J., Power, D.M., 2009. Effects of dietary phosphorus and calcium level on growth and skeletal development in Rainbow Trout (*Oncorhynchus mykiss*) fry. Aquaculture 297, 141–150.

Ford, E., 1930. Some abnormal fishes received at the Plymouth laboratory. J. Mar. Biol. Assoc. U. K. 17, 53–64.

Gall, G.A., Crandell, P.A., 1992. The rainbow trout. Aquaculture 100, 1–10.

Gill, C.D., Fisk, D.M., 1966. Vertebral abnormalities in sockeye, pink, and chum salmon. Trans. Am. Fish. Soc. 95, 177–182.

Gislason, H., Karstensen, H., Christiansen, D., Hjelde, K., Helland, S., Bæverfjord, G., 2010. Rib and vertebral deformities in rainbow trout (*Oncorhynchus mykiss*) explained by a dominant-mutation mechanism. Aquaculture 309, 86–95.

Grini, A., Hansen, T., Berg, A., Wargelius, A., Fjelldal, P.G., 2011. The effect of water temperature on vertebral deformities and vaccine-induced abdominal lesions in Atlantic salmon, *Salmo salar* L. J. Fish Dis. 34, 531–546.

Hansen, T., Fjelldal, P.G., Yurtseva, A., Berg, A., 2010. A possible relation between growth and number of deformed vertebrae in Atlantic salmon (*Salmo salar* L.). J. Appl. Ichthyol. 26, 355–359.

Holm, H., Ytteborg, E., Høst, V., Reed, A.K., Dalum, A.S., Bæverfjord, G., 2020. A pathomorphological description of cross-stitch vertebrae in farmed Atlantic salmon (*Salmo salar* L.). Aquaculture 526, 735382.

Jimenez, Y.E., Camp, A.L., 2023. Beam theory predicts muscle deformation and vertebral curvature during feeding in Rainbow Trout (*Oncorhynchus mykiss*). J. Exp. Biol. 226 (20).

Kacem, A., Meunier, F.J., Aubin, J., Haffray, P., 2004. Caractérisation histo-morphologique des malformations du squelette vertébral chez la truite arc-en-ciel (*Oncorhynchus mykiss*) après différents traitements de triploïdisation. Cybium 28, 15–23.

Kause, A., Ritola, O., Paananen, T., Wahlroos, H., Mäntysaari, E.A., 2005. Genetic trends in growth, sexual maturity and skeletal deformations, and rate of inbreeding in a breeding programme for rainbow trout (*Oncorhynchus mykiss*). Aquaculture 247, 177–187.

Kvellestad, A., Høie, S., Thorud, K., Tørud, B., Lyngøy, A., 2000. Platyspondyly and shortness of vertebral column in farmed Atlantic salmon *Salmo salar* in Norway-description and interpretation of pathologic changes. Dis. Aquat. Org. 39, 97–108.

Liu, K.L., He, Y.F., Xu, B.W., Lin, L.X., Chen, P., Iqbal, M.K., Mehmood, K., Huang, S.C., 2023. Leg disorders in broiler chickens: a review of current knowledge. Anim. Biotechnol. 34, 5124–5138.

Lovett, B.A., Firth, E.C., Tuck, I.D., Symonds, J.E., Walker, S.P., Perrott, M.R., Davie, P.S., Munday, J.S., Preece, M.A., Herbert, N.A., 2020. Radiographic characterisation of spinal curvature development in farmed New Zealand Chinook salmon *Oncorhynchus tshawytscha* throughout seawater production. Sci. Rep. 10, 20039.

Madsen, L., Dalsgaard, I., 1999. Vertebral column deformities in farmed Rainbow Trout (*Oncorhynchus mykiss*). Aquaculture 171, 41–48.

Madsen, L., Arnbjerg, J., Dalsgaard, I., 2000. Spinal deformities in triploid all-female rainbow trout (*Oncorhynchus mykiss*). Bull. Eur. Assoc. Fish Pathol. 20, 206–208.

Madsen, L., Arnbjerg, J., Dalsgaard, I., 2001. Radiological examination of the spinal column in farmed rainbow trout *Oncorhynchus mykiss* (Walbaum): experiments with *Flavobacterium psychrophilum* and oxytetracycline. Aquac. Res. 32, 235–241.

Mariasingarayan, Y., Danaraj, J., Veeraiyan, B., Fjelldal, P.G., Karuppiah, K., Narayanasamy, R., 2024. Vertebral column deformity in six species of wild fish at the Coromandel coast, Bay of Bengal India. Aquacult. Fish. 9, 635–641.

McCrimmon, H.R., Bidgood, B., 1965. Abnormal vertebrae in the rainbow trout with particular reference to electrofishing. Trans. Am. Fish. Soc. 94, 84–88.

Meunier, F.J., Ramzu, M.Y., 2006. La régionalisation morphofonctionnelle de l'axe vertébral chez les Téléostéens en relation avec le mode de nage. Comptes Rendus Palevol. 5, 499–507.

Michie, I., 2001. Causes of downgrading in the salmon industry. In: Kestin, S.C., Warris, P.D. (Eds.), Farmed Fish Quality. Fishing News Books, Oxford, pp. 129–136.

Munday, J.S., Perrott, M.R., Symonds, J.E., Walker, S.P., Lovett, B., Preece, M.A., Davie, P.S., 2016. Unilateral perivertebral fibrosis associated with lordosis, kyphosis and scoliosis (LKS) in farmed Chinook salmon in New Zealand. Dis. Aquat. Org. 121, 211–221.

Noble, C., Gismervik, K., Iversen, M.H., Kolarevic, J., Nilsson, J., Stien, L.H., Turnbull, J. F. (Eds.), 2018. Welfare Indicators for farmed Atlantic salmon: tools for assessing fish welfare, 351pp.

Noble, C., Gismervik, K., Iversen, M.H., Kolarevic, J., Nilsson, J., Stien, L.H., Turnbull, J. F. (Eds.), 2020. Welfare Indicators for farmed Rainbow Trout: tools for assessing fish welfare, 310 pp.

Perrott, M.R., Symonds, J.E., Walker, S.P., Hely, F.S., Wybourne, B., Preece, M.A., Davie, P.S., 2018. Spinal curvatures and onset of vertebral deformities in farmed Chinook salmon, *Oncorhynchus tshawytscha* (Walbaum, 1792) in New Zealand. J. Appl. Ichthyol. 34, 501–511.

Poirier Stewart, N., Deschamps, M.H., Witten, P.E., Le Luyer, J., Proulx, E., Huysseune, A., Bureau, D.P., Vandenberg, G.W., 2014. X-ray-based morphometrics: an approach to diagnose vertebral abnormalities in under-mineralized vertebrae of juvenile triploid all-female rainbow trout (*Oncorhynchus mykiss*) fed with a phosphorus deficient diet. J. Appl. Ichthyol. 30, 796–803.

Powell, M.D., Jones, M.A., Lijalad, M., 2009. Effects of skeletal deformities on swimming performance and recovery from exhaustive exercise in triploid Atlantic salmon. Dis. Aquat. Org. 85, 59–66.

Ramzu, M., Meunier, F.J., 1999. Morphological parameters of the vertebral column in the Rainbow Trout (*Oncorhynchus mykiss*) (Teleostei, Salmoniform). Ann. Sci. Nat. 3, 87–97.

Sankar, M., Kryvi, H., Fraser, T.W.K., Philip, A.J.P., Remø, S., Hansen, T.J., Witten, P.E., Fjelldal, P.G., 2024. A new method for regionalization of the vertebral column in salmonids based on radiographic hallmarks. J. Fish Biol. https://doi.org/10.1111/jfb.15873 accepted for publication.

Schultze, H.P., Arratia, G., 1989. The composition of the caudal skeleton of teleosts (Actinopterygil: Osteichthyes). Zool. J. Linnean Soc. 97, 189–231.

Sullivan, M., Hammond, G., Roberts, R.J., Manchester, N.J., 2007. Spinal deformation in commercially cultured Atlantic salmon, *Salmo salar* L.: a clinical and radiological study. J. Fish Dis. 30, 745–752.

Thorarinsson, R., Negaard, P., Baeverfjord, G., Peña, P., Skjerve, E., 2024. The effect of vaccine-associated cross-stitch vertebrae pathology on growth of farmed Atlantic salmon. J. World Aquacult. Soc. 55, 240–256.

Trangerud, C., Bjørgen, H., Koppang, E.O., Grøntvedt, R.N., Skogmo, H.K., Ottesen, N., Kvellestad, A., 2020. Vertebral column deformity with curved cross-stitch vertebrae in Norwegian seawater-farmed Atlantic salmon, *Salmo salar* L. J. Fish Dis. 43, 379–389.

Van Velson, R.C., 1978. The McConaughy Rainbow …Life History and a Management Plan for the North Platte River Valley. Nebraska Game and Parks Commission – Staff Research Publications, 52. https://digitalcommons.unl.edu/nebgamestaff/52.

Webb, P.W., 1982. Locomotor patterns in the evolution of actinopterygian fishes. Am. Zool. 22, 329–342.

Witten, P.E., Gil-Martens, L., Hall, B.K., Huysseune, A., Obach, A., 2005. Compressed vertebrae in Atlantic salmon *Salmo salar*: evidence for metaplastic chondrogenesis as a skeletogenic response late in ontogeny. Dis. Aquat. Org. 64, 237–246.

Witten, P.E., Gil-Martens, L., Huysseune, A., Takle, H., Hjelde, K., 2009. Towards a classification and an understanding of developmental relationships of vertebral body malformations in Atlantic salmon (*Salmo salar* L.). Aquaculture 295, 6–14.

Woynarovich, A., Hoitsy, G., Moth-Poulsen, T., 2011. Small-scale rainbow trout farming. In: FAO Fisheries and Aquaculture Technical Paper, 561, p. I.

Yadegari, M., Raissy, M., Ansari, M., 2011. A radiographical study on skeletal deformities in cultured Rainbow Trout (*Oncorhynchus mykiss*) in Iran. Global Veterinaria 7, 601–604.

Ytteborg, E., Baeverfjord, G., Torgersen, J., Hjelde, K., Takle, H., 2010. Molecular pathology of vertebral deformities in hyperthermic Atlantic salmon (*Salmo salar*). BMC Physiol. 10, 1–16.