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Linking PFOS to physiology and health in Baltic Sea salmon (Salmo salar)

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Abstract

Per- and polyfluoroalkyl substances (PFAS), including perfluorooctane sulfonic acid (PFOS), are persistent environmental contaminants widely distributed in aquatic ecosystems. The Baltic Sea, with its limited water exchange and high anthropogenic pressure, is particularly vulnerable to PFAS exposure, which has been documented across multiple trophic levels. Baltic salmon (*Salmo salar*), an ecologically and economically important species, serves as a valuable bioindicator for contaminant exposure in this region. However, the associations between PFAS concentrations and key biological variables in Baltic salmon remain poorly understood. Baltic salmon populations have also experienced various health problems, including reproductive disorders and decreased survival rates, which may be linked to environmental stressors such as contaminant exposure.

This study examines potential associations between PFOS blood serum concentrations and biological markers related to reproduction, metabolism, and immune function in Baltic salmon. Using random forest and decision tree modelling for exploratory analysis, coupled with PFOS-adjusted linear models for ecological interpretation, we identified relationships between PFOS and health biomarkers such as Gonadosomatic Index (GSI) and Liver Somatic Index (LSI). The inverse relationship between PFOS concentrations and GSI suggests that when females allocate resources into gonad development, PFOS is redistributed to the developing eggs. Additionally, immune-related variables (e.g. white blood cell count and monocytes) appeared to be associated with PFOS concentrations, suggesting potential links between contaminant exposure and immune status. A relationship between plasma chloride concentration and PFOS was found for some individuals and hemodilution caused by heavy Saprolegnia infection may have lowered PFOS concentrations in these individuals. Site-specific differences were observed, with stronger associations in wild salmon compared to salmon hatched in restocking farms. By focusing on general patterns rather than causal mechanisms, this study provides insights into how PFAS exposure correlates with physiological and ecological variables in Baltic salmon.

Introduction

Per- and polyfluoroalkyl substances (PFAS) represent a group of synthetic chemicals extensively utilized as surfactants in diverse applications such as textiles, cookware, and fire-fighting foams. These substances are characterized by their exceptional chemical stability, persistence in the environment, and lipophilic nature, which facilitates bioaccumulation in aquatic organisms (Banyoi et al. 2022). PFAS compounds, including perfluorooctane sulfonic acid (PFOS), are known to disrupt endocrine function, immune responses, and metabolic pathways, raising concerns about their long-term ecological and physiological effects (Fenton et al. 2021).

The Baltic Sea has a semi-enclosed nature, limited water exchange, and high anthropogenic pressures. PFAS are introduced into this ecosystem primarily through riverine discharge and atmospheric deposition (Filipovic et al. 2013), resulting in significant contamination across multiple trophic levels (Faxneld et al., 2016; Gebbink et al., 2016; Koponen et al., 2015; de Wit et al., 2020). Species such as

herring, cod, grey seals, and white-tailed eagle have demonstrated increasing PFAS concentrations over time, reflecting the pervasive nature of these contaminants (Faxneld et al., 2016; Schultes et al 2020). While much is known about the presence and bioaccumulation of PFAS, the broader patterns of association between PFAS concentrations and biological responses in aquatic organisms remain poorly characterized.

Baltic salmon (*Salmo salar L.*) occupies a vital ecological niche within the Baltic Sea ecosystem. This species is economically and ecologically significant, supporting fisheries and serving as an indicator for ecosystem health (Hindar et al., 2011, Kulmala et al., 2012). As an apex predator, Baltic salmon bioaccumulates contaminants present in lower trophic levels. Baltic salmon is known to depend on size-dependent foraging and food availability, and the majority of the different river populations to the Baltic Proper which constitutes their main growth and feeding ground (Jacobson et al. 2019). Their diet depends on size-specific prey availability and consists almost exclusively of sprat (*Sprattus sprattus* L.), herring (*Clupea harengus* L.) and three-spined stickleback, (*Gasterosteus aculeatus* L., Jacobson et al. 2018). This reliance on pelagic fish species, themselves known to bioaccumulate PFAS (Faxneld et al. 2016), raises concerns about contaminant transfer through the food web. However, understanding how PFAS concentrations are associated with physiological and ecological variables in Baltic salmon remains a critical knowledge gap.

Baltic salmon populations have experienced significant health issues for decades, including the M74 syndrome, which leads to high mortality rates among yolk-sac fry (Bengtsson et al., 1999). Additionally, these populations have faced declining numbers due to reduced sea phase survival rates, likely due to a combination of factors such as environmental changes, overfishing, and contaminant exposure. The International Council for the Exploration of the Sea (ICES) has reported fluctuations in Baltic salmon stocks, emphasizing the need for continued monitoring and effective management strategies to ensure their sustainability (ICES, 2025).

PFAS exposure is linked to reproductive, immunological, and metabolic disruptions in fish (Fenton et al., 2021; Dale et al., 2020; Kanerva et al., 2020). In salmonids, PFAS exposure has been associated with immunosuppression, physiological stress responses, and altered reproductive investment (Davis et al., 2008). Despite these findings, many knowledge gaps remain, particularly regarding the general patterns of association between PFAS concentrations and biological responses. Identifying such patterns is essential for interpretation of which physiological processes may be most sensitive to PFAS exposure, how these associations vary across ecological contexts, and whether geographic or environmental factors influence the observed relationships.

This study addresses these gaps by examining associations between PFOS concentrations and key physiological and ecological variables in Baltic salmon. Rather than trying to identify causal patterns, our analysis aims to uncover general patterns and potential relationships that may inform future hypothesis-driven studies. Using complementary statistical approaches, including Random Forest modelling for exploratory data analysis and PFOS-adjusted linear models for focused ecological interpretation, we aim

to identify priority variables that are strongly associated with PFOS concentrations. By integrating data mining with ecologically driven interpretations, this research provides insights into the multifaceted associations between PFAS exposure in free living individuals and biological processes in a species central to the Baltic ecosystem.

Materials and Method

Samples

Adult salmon returning to their spawning grounds were sampled during May to October 2018 in two Swedish rivers discharging into the Baltic Sea (Ume-/Vindelälven and Indalsälven) and one river (Lagan) discharging into the Atlantic Ocean (Kattegat). Details about sampling sites and procedures for sampling, biometrics and histopathology are described in Weichert et al. 2021.

Serum samples from both female and male salmon were used for PFOS analysis. Only female specimens were available from the river Lagan. The Neutrophil/Lymphocyte ratio (N/L), based on screening of cytological samples stained with Giemsa, was used as a marker of inflammatory immune response: typically, neutrophil counts increase and lymphocyte counts decrease under both stress and infection (reviewed in Davis et al., 2008). However, this pattern can vary, as lymphocytes may increase during certain types of infections, particularly in later stages when antibody production is initiated, since lymphocytes are responsible for this process.

All perfluorinated analytical standards were purchased from Wellington Laboratories (Guelph, ON, Canada), including: Sodium perfluoro-1-octane sulfonate (NaPFOS) and perfluoro-1-hexane sulfonate (NaPFHxS), perfluoro-n-octanoic (PFOA), -nonanoic (PFNA), -decanoic (PFDA), and -undecanoic acid (PFUnA), as well as $^{13}C_4$ labeled NaPFOS and PFOA. Acetonitrile, methanol, and water for liquid chromatography were purchased from Merck Millipore (Burlington, MA, USA). Formic acid and ammonium formate from Supelco (Bellefonte, PA, USA) were of LC-MS quality.

Extraction

Extractions of the serum samples were carried out in a similar manner as previously described (Reardon et al., 2019), with minor modifications. Labelled internal standards (^{13}C) of PFOS (50 ng) and PFOA (5 ng) in methanol (100 µL), were added to circa 0.5 g serum in a 10 mL polypropylene (PP) tubes. Protein precipitation was achieved using acetonitrile (4 mL) and 10 min of sonication. After centrifugation (2000*g*) the acetonitrile was isolated and transferred to a new PP test tube. The volume was reduced by evaporation at 60°C under a stream of nitrogen gas to 0.2–0.3 mL. Methanol (0.3 mL) was then added, and the samples were transferred to 1.5 mL conical Eppendorf tubes. The samples were centrifuged once more (7000*g*) for 10 min at 8°C to remove any residual particles. Aliquots of 250 µL of the samples were then transferred to vials with PP inserts for instrumental analysis using liquid chromatography tandem mass spectrometry (LC-MS-MS). Procedural blanks were run in parallel with the samples.

Liquid chromatography mass spectrometry

LC-MS-MS analyzes were conducted on a Shimadzu 20AD HPLC-system, with a SIL 20A autosampler (Shimadzu Corporation, Kyoto, Japan), coupled to a Sciex 3200 Q TRAP mass analyzer (Sciex, MA, USA), operated in negative electrospray ionization (ESI) mode. Transitions and collision energies are shown in Table 1 (left column). The HPLC column (Ascentis Express F5 PFP, 10 cm × 2.1 mm, 2.7 µm, Sigma Aldrich) and mobile phases were the same as those used by Reardon and coworkers (2019). Mobile phase A consisted of formic acid (5 mM) and ammonium formate (5 mM) in water/methanol (9:1), and mobile phase B was 100% methanol. The gradient was similar to that used by Reardon and coworkers (2019) (Table 1, right column), with a flow of 0.2 mL/min. ^{13}C PFOA was used as an internal standard for all fluorinated carboxylic acids (FCAs; PFOA, PFNA, PFDA, and PFUnA), while ^{13}C PFOS was used for all fluorinated sulfonic acids (FSAs; PFOS, PFOS isoforms and PFHxS). Quantification was conducted against external calibration curve and blank subtraction was only necessary to account for small amounts of PFOA in the blanks. Three peaks corresponding to isoforms of PFOS were quantified using a response factor compared to linear PFOS of 1. One of the three isomer peaks (designated PFOS iso1) was in turn a poorly resolved double peak, from the work of Reardon et al. (2019) it is known that the branched PFOS 3*m* and 4*m* are not resolved using this LC setup.

Table 1
MS transitions and collision energies for the different analytes as well as programming of the binary flow-system for the LC.

PFOS & iso	**Q1 (*m/z*)**	**Q3 (*m/z*)**	**DP (V)**	**CE (V)**	**Time (min)**	**Mob.B (%)**
	499	**80**	**-40**	**-90**	**Start**	**0**
PFOS $^{13}C_4$	503	80	-40	-90	0.1	0
PFOA	413	369	-20	-15	3.0	50
PFOA $^{13}C_4$	417	372	-20	-15	14	85
PFHxS	399	80	-40	-90	14.5	100
PFNA	463	419	-20	-15	15	100
PFDA	513	469	-20	-15	16	0
PFUnA	563	519	-20	-15	20	0

DP = Declustering potential.

CE = Collision energy.

Mob.B = Percentage of mobile phase B at the given time.

Statistical analyses

A total of 46 candidate variables were compiled from clinical chemistry, biometric, and histopathological analyses for which details are provided in Supplementary Table S1.

Serum concentrations of PFAS (ΣFCAs, ΣFSAs, and PFOS) were quantified and compared between salmon populations from the three rivers (Ume-/Vindelälven, Indalsälven, and Lagan). For rivers where both male and female specimens were available (Ume-/Vindelälven and Indalsälven), differences in PFAS levels between sexes were also assessed. Statistical analyses were conducted using two-tailed t-tests for general comparisons, and one-tailed t-tests where directional hypotheses were considered. Statistical significance was set at $p < 0.05$. Only female specimens were available from river Lagan.

To investigate the relationship between PFOS concentrations in the salmon and various biological variables we employed feature selection using random forest on (1) unadjusted PFOS values, and (2) PFOS concentrations adjusted based on individual blood chloride (Cl^-) concentrations

Prior to analyses an extensive screening and exploration of variables was carried out. The dataset included many candidate variables in relation to the number of individuals. Some of the candidate variables showed a high degree of correlation with one or several of the other candidate variables. Despite the fact that analysis using random forest is relatively robust to co-correlation there are no statistical tests that can differentiate between two or more highly correlated variables. Pearsons correlation coefficients were calculated between all candidate variables and at correlation ≥ 0.8, one of the variables was discarded. The decision on which variables to discard was based on species specific ecological and physiological knowledge, details are provided in Supplementary Table S2.

Feature selection was carried out on the remaining candidate variables using the Boruta package (Kursa and Rudnicki 2010) and PFOS concentration as the explanatory variable. The Boruta algorithm is a wrapper that uses random forest classification, implemented in the random Forest package (Liaw and Wiener 2002), to iteratively remove features less relevant than random probes (shadow attributes created by shuffling values of the original attribute across objects). Important variables are selected by comparing the candidate variables' *Z* score against the maximum *Z* score among shadow attributes (MZSA). Candidate variables with a statistically significant lower Z score (compared with the MZSA) are discarded and the analysis is rerun until all candidate variables are assigned (important or unimportant) or the set number of random forest runs is reached. The number of random forest runs was set to 300 whereafter unassigned candidate variables were assigned as important or not unimportant by comparing median Z scores (TentativeRoughFix command).

When screening the data it was noted that some individuals, primarily from Umeå/Vindelälven, had abnormally low blood chloride levels, indicative of osmoregulation failure. A strong relationship between chloride levels and PFOS was also apparent when carrying out feature selection. Haemodilution would result in diluted concentrations of PFOS measured in blood and a failure to establish relationships between PFOS and other candidate variables. Based on serum chloride levels in Atlantic salmon from available literature and the spread in concentrations in this study a threshold concentration was established. A linear model (PFOS ~ Cl) was carried out for fish under the threshold chloride value and

PFOS values in these individuals adjusted according to the regression coefficient and the mean chloride value for fish above the threshold value (primarily individuals from Lagan and Indal). Feature selection, as described above, was carried out again using the adjusted PFOS values and excluding the chloride concentration variable.

Results and discussion

Salmon from the two east coast rivers (Ume/Vindelälven and Indalsälven) displayed significantly higher serum concentrations of PFAS than salmon from the west coast river (Lagan) (Fig. 1). In fact, only PFOS was of high enough concentration to be quantified in salmon from west coast river Lagan. Serum levels of both ΣFCAs and ΣFSAs were significantly higher in salmon from Ume-/Vindelälven than in salmon from Indalsälven ($p < 0.001$ for ΣFCAs and $p < 0.0001$ for ΣFSAs, 2-tailed *t*-test). Male specimens from Indalsälven had higher serum levels than females, both for ΣFCAs ($p = 0.026$, 2-tailed *t*-test) and ΣFSAs ($p = 0.005$). In specimens from Ume-/Vindelälven ΣFCAs levels differed significantly between sexes ($p = 0.017$), while the ΣFSA levels did not show a significant difference using 2-tailed *t*-test ($p = 0.082$). However, a one-sided *t*-test showed a slight significant difference indicating higher levels in males ($p = 0.0408$). Only female specimens were available from river Lagan (see Fig. 1).

The pronounced relationships observed in salmon from the River Ume-/Vindelälven, in contrast to those from the Indalsälven system, emphasize the role of local environmental conditions in shaping PFOS dynamics. These site-specific associations likely reflect differences in contaminant sources, dietary exposure, and ecological factors such as prey contamination levels (Jacobson et al., 2019). While the associations identified in this study are not sufficient to establish causation, they underscore the value of examining regional variation in PFAS exposure to uncover patterns that could guide targeted conservation and regulatory efforts.

The pronounced relationships observed in salmon from the River Ume/Vindelälven should be interpreted in light of differences in stock origin and life history, rather than solely as effects of local freshwater exposure. Fish from the Ume/Vindelälven are of wild origin, whereas salmon from the Indalsälven system are predominantly hatchery-produced. Given that the vast majority of somatic growth and contaminant accumulation in Atlantic salmon occurs during the marine feeding phase—when individuals increase in mass from approximately 100–150 g at smoltification to several kilograms prior to spawning—it is unlikely that PFOS concentrations measured in returning adults primarily reflect exposure during the freshwater or early smolt stages. Instead, the observed differences are more plausibly linked to variation in marine phase duration, growth trajectories, or at-sea behaviour, such as differences in foraging areas, diet composition, or migration routes. Although fish from different river systems are generally considered to form mixed stocks at sea, persistent differences in PFOS concentrations between wild and hatchery-origin fish may indicate subtle but ecologically relevant divergence in marine habitat use or bioaccumulation dynamics. While the associations identified here do not establish causality, they highlight the importance of considering life-history variation and marine ecology when interpreting regional patterns of PFAS exposure in spawning salmon.

Data screening:

There were 46 candidate variables measured in 54 fish from the three rivers. Twenty-two of these candidate variables had a correlation coefficient ≥ 0.8 (**Figure S1**). The following were excluded prior to further analysis due to high correlations with one or more other variables: "T4total", "Site", "Date", "Weight", "Somaticweight", "Gonadalweight", "T3.T4Total", "muscle AChE activity", "T3/T4free", "T3total", "Condition factor", "nCA" and "T.Ca" (**Table S2**). Length was deemed an adequate proxy for fish size and less influenced by differences in gonad investment between sexes (i.e. differing allometric growth relationships) and sites, thus the correlated variable weight was excluded. Gonadal investment is an overriding physiological feature and gonadosomatic index (GSI) was retained in favour of the correlated variables site, date, condition factor, muscle AChE activity and gonadal weight because it was deemed to be the biologically most informative variable. GSI correlated with site, which was due to differences in sampling time between locations with sampling from Lagan and Indalsälven occurring later in the season than Ume-/Vindelälven.

Baltic salmon are treated as a mixed stock at sea, and it is thought that populations from different rivers overlap in their foraging areas and behaviour at sea. Atlantic salmon on the west coast forage and grow in the north Atlantic and no overlap in foraging areas with Baltic fish are known. This could explain differences in PFOS concentrations measured in salmon from Lagan and the east coast rivers. Measurements of thyroid hormones were correlated and the variables T3 free and T4 free were used as proxy for these in subsequent feature selection analysis. The variable WBC neutrophils was removed from the dataset because it is the continuous counterpart of neu_cut, a categorized version of the same measure, and including both would result in redundant information. After screening 29 candidate variables remained for feature selection using random forest models.

Random forest

In the random forest analysis, 9 variables were identified as important predictors of PFOS concentrations, while 22 were classified as unimportant (**see Table S3**, Fig. 2). This provides a comprehensive view of which variables are most strongly associated with PFOS levels.

The Random Forest analysis identified the most important predictors of PFOS concentrations in Baltic salmon, ranked by their mean importance scores (Fig. 2); GSI (mean importance: 9.97) was the most important variable related to PFOS concentrations suggesting a link between PFOS and reproductive investment. Chloride (6.86) also emerged as a key variable indicating a relationship between osmoregulation and PFOS concentrations. Liver-related variables, LSI (6.70) and EROD activity (5.85), highlighted the central role of hepatic physiology in structuring PFOS-related patterns. While PFAS are not known to directly activate the aryl hydrocarbon receptor and induce EROD activity, the observed association may reflect co-exposure to other planar organic contaminants, such as polycyclic aromatic hydrocarbons, which are known EROD inducers (Fent & Bätscher 2000). Thus, EROD activity likely serves as an indicator of overall contaminant burden and hepatic response rather than a direct mechanistic

effect of PFOS. Moderately important variables included thio-complement activity (3.83) and fish length (3.79), reflecting physiological and growth-related factors. The marker N:L ratio (3.44) suggested PFOS-related effects on immune function, while kidney damage (3.02) indicated potential renal impacts. Gender (2.54) was less influential but may reflect sex-specific differences in PFOS dynamics. These findings underscore the multifaceted impacts of PFOS exposure, and potential coupling to reproductive, metabolic, and immune systems in Baltic salmon. The results provide valuable insights into the biological variables most closely associated with PFOS concentrations, laying the groundwork for further research into underlying mechanisms.

In this analysis, Chloride, emerged as the second most important variable associated with PFOS concentrations, following GSI. A notable finding was that salmon from the river Ume-/Vindelälven exhibited significantly lower Cl^- plasma levels compared to individuals from other sites. One possible explanation is that the health status of the fish may influence Cl^- levels (Weichert et al 2021). Diseased or stressed fish often exhibit diluted blood plasma, which can lead to reduced Cl^- concentrations. The health of the sampled salmon from river Ume-/Vindelälven was indeed worse than the salmon from Indalsälven or Lagan, with several fish suffering from massive oomycete infection (Saprolegniosis). Saprolegniosis disrupts the skin barrier, allowing leakage of fresh water into the body at a rate the fish cannot handle. This leads to hemodilution. Saprolegniosis commonly occur secondary to other health issues, and is thus potentially linked to environmental, physiological or pathogen stressors during migration.

Adjustment of PFOS concentrations based on chloride levels

To examine the relationship between PFOS concentrations and chloride (Cl- levels more closely, we adjusted PFOS values to account for variations in Cl- concentrations among the fish samples. We performed a linear regression analysis (PFOS ~ Cl-) on fish with Cl concentrations below 125 mmol/L, after excluding two outliers that could skew the results. The threshold of 125 mmol/L was selected based on the distribution of Cl- levels considered 'normal,' primarily observed in fish from the rivers Lagan and Indalsälven. This threshold is further supported by data from Rozas-Serri et al. (2022), who reported plasma chloride concentrations in adult Atlantic salmon in seawater environments averaging around 155 mmol/L, indicating that values significantly lower than this suggest disrupted osmoregulation. The overall regression was statistically significant ($R^2 = 0.31$, $F(1, 16) = 8.66$, $p = 0.01$) with a regression coefficient of 1.9. PFOS concentrations in fish under the threshold Cl concentration were adjusted using the regression coefficient and the mean Cl concentration measured in fish above the threshold Cl concentration. This adjustment accounts for the variability in PFOS levels due to factors such as osmoregulatory changes or health status (Fig. 3).

Random forest using adjusted PFOS values

Feature selection was carried out as outlined above but using adjusted PFOS concentrations and after removing Cl concentrations as a candidate variable. 11 variables were identified as important and 19 as unimportant (Fig. 4).

GSI remained the most important variable but with a much higher score (14.58) than for unadjusted PFOS values. The majority of fish with lower Cl values also had a lower GSI. After adjusting for osmotic haemodilution these individuals had an even higher estimate for PFOS concentrations making possible decontamination through gonadal investment more pronounced. Liver related variables (LSI and EROD activity; 5.38 and 6.66 respectively) retained a relatively high importance. Thio complement activity (3.91), kidney damage (5.34), fish length (3.01) and gender (2.50) also remained as variables of importance. Variables selected using adjusted PFOS concentrations that were not selected when using unadjusted PFOS concentrations included the thyroid hormones thyroxine (T4free; 3.51), potassium (Kcorrected; 2.50), carotinoids (2.24) and most notably monocyte/macrophage leucocytes (WBC.monocytesandmacrophages; 4.93)

The results from the variable importance analysis consistently highlight several biologically and ecologically significant factors associated with PFOS concentrations in Baltic salmon (see **Table S3**). Among these, Gonadosomatic Index (GSI) emerged as the most critical predictor. This underscores the strong association between PFOS and reproductive investment, likely reflecting maternal transfer of contaminants to eggs. Unlike legacy lipophilic contaminants that partition into lipids, maternal transfer of PFAS is primarily driven by their high affinity for proteins such as vitellogenin and albumin. Indeed, recent wildlife studies have confirmed that maternal-embryonic transfer efficiency of PFOS correlates directly with protein-water partition coefficients rather than lipid content (Mehdi et al. 2025). Such a process can alleviate the contaminant burden on spawning females but may have long-term implications for offspring fitness and population dynamics. For instance, female zebrafish (*Danio rerio*) exposed to PFOS at 0.05 and 0.25 mg/L for 70 days showed a significant reduction in GSI (Du et al., 2009). Further evidence of maternal transfer comes from a study by Conard et al. (2022), which investigated Lake Michigan salmonids, including Chinook salmon, coho salmon, and steelhead trout. The study found higher PFOS concentrations in eggs compared to maternal muscle tissue, consistent with maternal offloading of PFOS to eggs demonstrated here (**Figure S2**). Crucially, recent experimental work on rainbow trout (*Oncorhynchus mykiss*) eggs has demonstrated that such simulated maternal transfer of PFOS induces oxidative stress and biochemical alterations during early embryonic development (Haché et al., 2026).

Maternal transfer of PFOS from female salmon to eggs may have important ecological and population-level implications for Baltic salmon. PFAS compounds are known to be transferred to developing oocytes (Wang et al. 2011), resulting in exposure during early life stages. Baltic salmon populations already face multiple health challenges, including poor smolt performance and reduced marine survival of adults (ICES 2025)). Early-life exposure to PFOS may therefore add to existing stressors by affecting immune development, physiological performance, or survival in juveniles. Such effects could reduce recruitment and, over time, exacerbate population-level impacts in already vulnerable salmon stocks.

Beyond direct effects on salmon populations, maternal transfer of PFOS may also introduce these contaminants into freshwater food webs, as salmon eggs are highly nutritious and constitute an important food source for fish, birds, and other wildlife. This pathway could be particularly relevant in northern river systems, where PFAS contamination has historically been lower compared to legacy pollutants from, for example, the pulp and paper industry. Maternal transfer of PFOS to eggs may therefore represent a mechanism by which PFAS are redistributed into riverine food webs that would otherwise experience limited direct exposure.

Implications for Baltic Salmon Health and Resilience

The observed associations between PFOS concentrations and key physiological variables suggest that contamination may influence the health and resilience of Baltic salmon. Although causality cannot be inferred, patterns such as the inverse relationship between GSI and PFOS concentrations raise important questions about how PFAS exposure may interact with reproductive processes. Similarly, the associations with immune and metabolic markers highlight potential areas of vulnerability for salmon populations facing additional environmental stressors, such as climate change and overfishing. These findings provide a foundation for future studies aimed at disentangling the complex interactions between PFAS exposure and fish health.

Future directions

This study lays the groundwork for understanding how PFAS exposure is associated with key physiological and ecological processes in Baltic salmon. Building on these findings, future research is needed to study possible variations in PFAS burden in wild vs compensatory hatched salmon in the Baltic Sea as well as investigate extent to which PFAS are transferred to eggs and their subsequent effects on offspring fitness, focusing on validating observed associations as well as explore the mechanisms linking PFAS exposure to immune and stress-related biomarkers in salmonids to deepen understanding of observed patterns.

Conclusion

By focusing on general associations rather than causation, this study provides valuable insights into the relationships between PFOS concentrations and biological variables in Baltic salmon. The identified patterns highlight areas for further investigation and contribute to a growing understanding of PFAS impacts in aquatic ecosystems. Continued exploration of these associations will be critical for advancing conservation strategies and mitigating the risks posed by PFAS contamination in the Baltic Sea.

Declarations

Ethics approval and consent to participate:

All procedures involving the handling and sampling of Atlantic salmon (*Salmo salar*) in this study were approved by the local animal ethics committee in Gothenburg, Sweden (regionala djurförsöksetiska nämnden i Göteborg), under permit number 5.8.18–02260/2018 (identification number 001380).

Consent for publication:

Not applicable.

Competing interests:

The authors declare that they have no competing interests.

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Author contributions:

ED and DJ conceptualized and initiated the study. ED served as the project leader, coordinated the study, and drafted the manuscript. DJ performed the statistical analyses, generated the figures, and contributed to the interpretation and writing of the paper. DL conducted the chemical analyses, provided input on the study design, and assisted in manuscript preparation. CA contributed to the study design, performed the fish health examinations and sampling, and participated in interpreting results and writing. All authors read and approved the final manuscript.

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Availability of data and materials:

The datasets supporting the findings of this study are available within the article and its supplementary materials, or from the corresponding author upon reasonable request.

References

1. Banyoi SM, Porseryd T, Larsson J, Grahn M, Dinnétz P (2022) The effects of exposure to environmentally relevant PFAS concentrations for aquatic organisms at different consumer trophic levels: Systematic review and meta-analyses. Environ Pollut 315:120422. https://doi.org/10.1016/j.envpol.2022.120422
2. Bengtsson BE, Hill C, Bergman A, Brandt I, Johansson N, Magnhagen C, Södergren A, Thulin J (1999) Reproductive disturbances in Baltic fish: a synopsis of the FiRe project. Ambio-Journal Hum Environ Res Manage 28(1):2–8
3. Conard WM, Whitehead HD, Harris KJ, Lamberti GA, Peaslee GF, Rand AA (2022) Maternal offloading of per-and polyfluoroalkyl substances to eggs by Lake Michigan salmonids. Environ Sci Technol Lett 9(11):937–942. https://doi.org/10.1021/acs.estlett.2c00627
4. Dale K, Yadetie F, Müller MB, Pampanin DM, Gilabert A, Zhang X, Tairova Z, Haar A, Lille-Langøy., Lyche J, Porte C, Karlsen A, Goksøyr A (2020) Proteomics and lipidomics analyses reveal modulation of lipid metabolism by perfluoroalkyl substances in liver of Atlantic cod (Gadus morhua). Aquat Toxicol 227:105590. https://doi.org/10.1016/j.aquatox.2020.105590
5. Davis A, Maney D, Maerz JC (2008) The use of leukocyte profiles to measure stress in vertebrates: a review for ecologists. Funct Ecol 22:760–772. https://doi.org/10.1111/j.1365-2435.2008.01467.x
6. de Wit CA, Bossi R, Dietz R, Dreyer A, Faxneld S, Garbus SE, Hellström P, Koschorreck J, Lohmann N, Roos A, Sellström U, Sonne C, Treu G, Vorkamp K, Yuan B, Eulaers I (2020) Organohalogen compounds of emerging concern in Baltic Sea biota: Levels, biomagnification potential and comparisons with legacy contaminants. *Environ. Int.* 2020, *144*, 106037. 10.1016/j.envint.2020.106037
7. Du Y, Shi X, Liu C, Yu K, Zhou B (2009) Chronic effects of water-borne PFOS exposure on growth, survival and hepatotoxicity in zebrafish: a partial life-cycle test. Chemosphere 74(5):723–729. 10.1016/j.chemosphere.2008.09.075
8. Faxneld S, Berger U, Helander B, Danielsson S, Miller A, Miller A, Nyberg E, Persson J, Bignert A (2016) Temporal trends and geographical differences of perfluoroalkyl acids in Baltic Sea herring and white-tailed sea eagle eggs in Sweden. Environ Sci Technol 50(23):13070–13079. 10.1021/ACS.EST.6B03230
9. Fent K, Bätscher R (2000) Cytochrome P4501A induction potencies of polycyclic aromatic hydrocarbons in a fish hepatoma cell line: demonstration of additive interactions. Environ Toxicol Chem 19(8):2047–2058. https://doi.org/10.1002/etc.5620190813
10. Fenton SE, Ducatman A, Boobis A, DeWitt JC, Lau C, Ng C, Smith J, Roberts SM (2021) Per-and polyfluoroalkyl substance toxicity and human health review: Current state of knowledge and strategies for informing future research. Environ Toxicol Chem 40(3):606–630. 10.1002/etc.4890
11. Filipovic M, Berger U, McLachlan M (2013) Mass balance of perfluoroalkyl acids in the Baltic Sea. Environ Sci Technol 47(9):4088. https://doi.org/10.1021/es400174y
12. Gebbink WA, Bignert A, Berger U (2016) Perfluoroalkyl acids (PFAAs) and selected precursors in the Baltic Sea environment: Do precursors play a role in food web accumulation of PFAAs? Environ Sci

Technol 50(12):6354–6362. https://doi.org/10.1021/acs.est.6b01197

13. Haché GC, Harris TR, Pichette AM, Fung VY, Lamberti GA, Peaslee GF, De Silva A, Miranda D, Rand AA (2026) Effects of simulated maternal transfer of perfluorooctane sulfonate (PFOS) on oxidative stress levels in rainbow trout eggs. Available at SSRN 6827838
14. Hindar K, Hutchings JA, Diserud OH, Fiske P (2011) Stock, recruitment and exploitation. In Atlantic Salmon Ecology, pp. 299–332. Ed. by Ø. Aas, S. Einum, A. Klemetsen, and J. Skurdal. Wiley-Blackwell, Oxford, UK
15. ICES (2025) Working Group on Pathology and Diseases of Marine Organisms (WGPDMO). ICES Sci Rep 07:78. https://doi.org/10.17895/ices.pub.29413142
16. Jacobson P, Gårdmark A, Östergren J, Casini M, Huss M (2018) Size-dependent prey availability affects diet and performance of predatory fish at sea: a case study of Atlantic salmon. Ecosphere 9:1–13. https://doi.org/10.1002/ecs2.2081
17. Jacobson P, Gårdmark A, Huss M (2019) Population and size-specific distribution of Atlantic salmon Salmo salar in the Baltic Sea over five decades. Journal of Fish Biology. 2019;1–10. https://doi.org/10.1111/jfb.14213
18. Kanerva M, Tue NM, Kunisue T, Vuori K, Iwata H (2020) Effects on the liver transcriptome in Baltic salmon: contributions of contaminatin with organohalogen compounds and origin of salmon. Environ Sci Technol 54(23):15246–15256. 10.1021/acs.est.0c04763
19. Koponen R, Airaksinen A, Hallikainen PJ, Vuorinen J, Mannio H, Kiviranta (2015) Perfluoroalkyl acids in various edible Baltic, freshwater, and farmed fish in Finland. Chemosphere 129:186–191. 10.1016/j.chemosphere.2014.08.077
20. Kulmala S, Haapasaari P, Karjalainen T, Kuikka S, Pakarinen T, Parkkila K, Romakkaniemi A, Vuorinen PJ (2012) Ecosystem services provided by Baltic salmon–a regional perspective to the socio-economic benefits associated with a keystone migratory species. Synthesis context Econ Ecosyst Biodivers (TEEB) 2:266–276
21. Kursa MB, Rudnicki WR (2010) Feature selection with the Boruta package. J Stat Softw 36:1–13. 10.18637/jss.v036.i11
22. Liaw A, Wiener M (2002) Classification and regression by randomForest. R news 2(3):18–22
23. Mehdi Q, Sinkway TD, Blackman LE, Iorfida AL, Pittman AM, Sutterer C, Hinchliffe HL, Zautcke K, Morrow M, Shah N, Chambrier-Athias IA, Cohen KE, Fraser GJ, Adams DH, Bowden JA (2025) Occurrence and maternal transfer of per-and polyfluoroalkyl substances (PFAS) in pregnant sharks from Florida coastal waters. Environ Sci Technol 59(29):14981–14992. https://doi.org/10.1021/acs.est.5c03114
24. Reardon AJ, Moez EK, Dinu I, Goruk S, Field CJ, Kinniburgh DW, MacDonald A, Martin J, APrON Study (2019) Longitudinal analysis reveals early-pregnancy associations between perfluoroalkyl sulfonates and thyroid hormone status in a Canadian prospective birth cohort. Environ Int 129:389–399. 10.1016/j.envint.2019.04.023

25. Rozas-Serri M, Correa R, Walker-Vergara R, Coñuecar D, Barrientos S, Leiva I, Senn R, Peña C, A (2022) Reference intervals for blood biomarkers in farmed Atlantic salmon, coho salmon and rainbow trout in Chile: promoting a preventive approach in aquamedicine. Biology 11(7):1066. 10.3390/biology11071066
26. Schultes L, Sandblom O, Broeg K, Bignert A, Benskin JP (2020) Temporal trends (1981–2013) of per- and polyfluoroalkyl substances and total fluorine in Baltic cod (*Gadus morhua)*. Environ Toxicol Chem 39:300–309. 10.1002/etc.4615
27. Wang M, Chen J, Lin K, Chen Y, Hu W, Tanguay R, Huang C, Dong Q (2011) Chronic zebrafish PFOS exposure alters sex ratio and maternal related effects in F1 offspring. Environ Toxicol Chem 30(9):2073–2080. http://dx.doi.org/10.1002/etc.594
28. Weichert FG, Axén C, Förlin L, Inostroza PA, Kammann U, Welling A, Sturve J, Asker N (2021) A multi-biomarker study on Atlantic salmon (Salmo salar L.) affected by the emerging Red Skin Disease in the Baltic Sea. J Fish Dis 44(4):429–440. 10.1111/jfd.13288

Figures

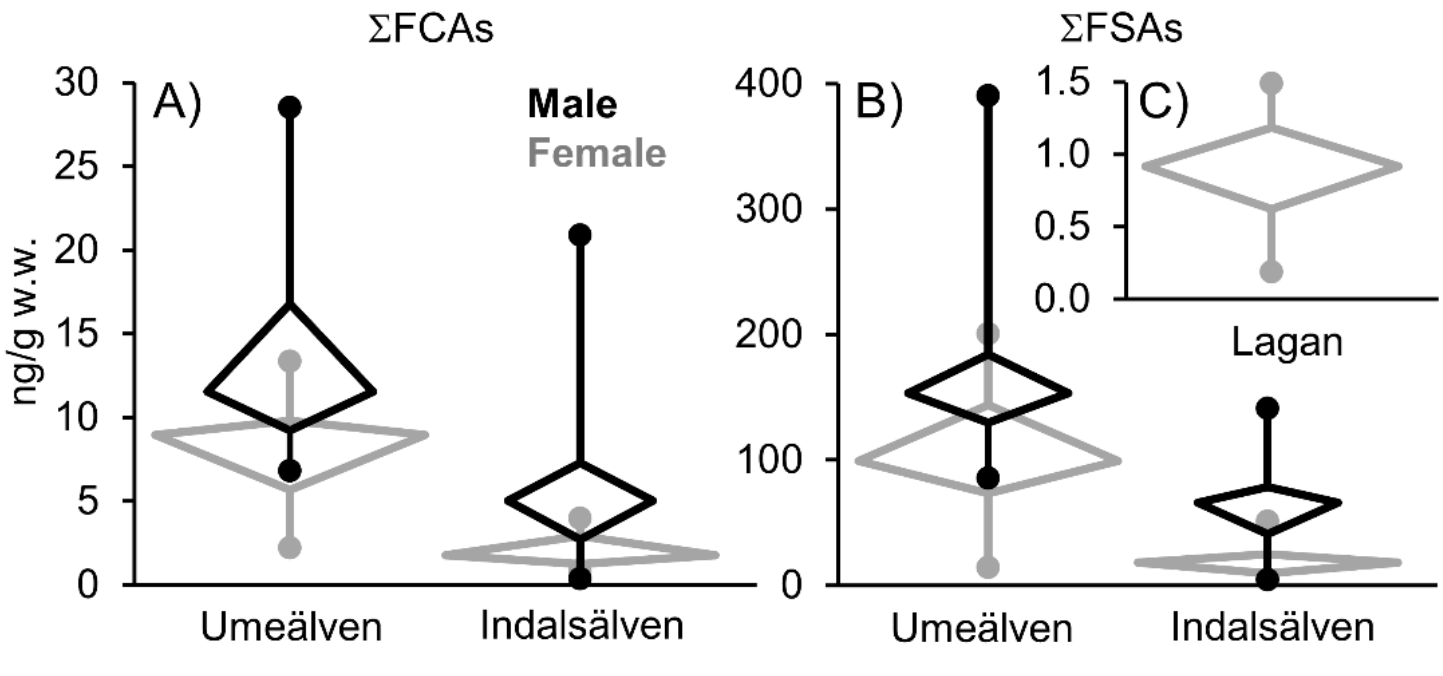


Figure 1

Box and whiskers plot showing the difference in serum levels between sexes (male in black and female in grey). The box is shaped like a diamond where the broadest point is the median and the top and bottom of the diamond are the 3rd and 1st quartile respectively, whiskers display high and low values, and the width of the diamond is proportional to the size of the sample set. Levels are given in ng/g w.w. as a sum of all FCAs and FSAs respectively. A) Difference in levels of SFCAs between sexes for both Ume/Vindelälven and

Indalsälven. B) Difference in levels of SFSAs between sexes for both Ume/Vindelälven and Indalsälven. C) Levels of SFSAs (PFOS only) in female specimens from river Lagan.