Malin Andersson Stavridis

Pollution in the Middle of Nowhere: Toxic Metals and PFAS in the Svalbard Reindeer

Doctoral thesis

NTNU
Norwegian University of
Science and Technology

UNIS
The University Centre in Svalbard

Malin Andersson Stavridis

Pollution in the Middle of Nowhere: Toxic Metals and PFAS in the Svalbard Reindeer

Thesis for the Degree of Philosophiae Doctor

Longyearbyen, March 2026

Norwegian University of Science and Technology
Faculty of Natural Sciences
Department of Biology

The University Centre in Svalbard
Department of Arctic Technology

NTNU
Norwegian University of
Science and Technology

UNIS
The University Centre in Svalbard

THESIS TITLE

Pollution in the Middle of Nowhere: Toxic Metals and PFAS in the Svalbard Reindeer

PhD CANDIDATE:
Malin Andersson Stavridis

SUPERVISORS:
Bjørn Munro Jenssen

Tomasz M Ciesielski

Øyvind Mikkelsen

AFFILIATIONS:
The University Centre in Svalbard (UNIS)

The Norwegian University of Science and Technology (NTNU)

ACKNOWLEDGEMENTS

First and foremost, I want to sincerely thank my main supervisor, Bjørn, for taking a chance on me, likely one of the most stubborn PhD applicants you have ever had. The idea of doing a PhD in ecotoxicology in the high Arctic got stuck with me years ago, and I still stand by what I told you during the interview for this position: I had been waiting forever for this opportunity to appear, and I would have kept on waiting for the next one if I hadn't gotten this one. This stubbornness and passion have kept me driven since. Thank you Bjørn for always being one WhatsApp message away, for guiding me, and letting me bounce ideas off you. I especially want to thank you for letting me take part in the planning and teaching of the AT-330 course, while encouraging change and new ideas. These have been some of the most educational and inspiring years of my life. Even though you were hundreds of kilometers away, you never felt too far.

This brings me to Tomek, my local supervisor, whom I recruited in my second year. I still remember the first time I was in contact with you, when you sent me back an abstract I had written for a conference, covered in red comments and edits. I thought, *why does this person hate me?* But I quickly learned that you are just **very** thorough and have an opinion on everything (which I have now learned not to always take too seriously). Over time, you not only became my supervisor, but also a friend. Even though you ended up chatting (read: disturbing) with me in my office more than helping me, haha. Still, I would not change a thing.

To my last supervisor, Øyvind, thank you for always providing fast and thorough feedback on any text, manuscript, or submission. I also want to especially thank you for your help and support at the Department of Chemistry at NTNU, and for making sure to arrange things for me to get my analyses done. I also really appreciate you involving me in the AT-331 course at UNIS, giving me more valuable teaching experience.

Besides my supervisors, a special thanks goes to Vebjørn Veiberg at NINA. I am so grateful that you let me be a part of the reindeer culling project; without it, I would not have any of the samples this thesis project is based on. Thank you for being welcoming, and your many (many!) comments on the manuscripts we co-author. I sincerely appreciate all the effort you have put into reading, commenting, and ultimately making the manuscripts better.

In contrast, I would like to <u>not</u> thank the analytical equipment I have been using these past years. Without exception, they made sure to always break just before my scheduled visit to NTNU to use them. A special non-thanks goes to the DMA-80 (or more commonly referred to as *Diva* among my students), which not once, but twice, exploded, and, in addition, ate tens, if not hundreds, of my samples without providing a reason why. Milestone claims that "analysis time is 5 minutes only", which is the biggest hoax of the century, as my samples always seem to average 12 minutes. All the blood, sweat, tears, and countless nights spent with this machine make me confident enough to now call myself a master of the DMA. On this topic, I do want to thank Kyyas at the Department of Chemistry for listening to all my complaints about Diva, and for trying to help the best he could.

To all the master students I supervised, I want to thank you for your invaluable contributions to this project and for making these years great. Thank you for the amazing moments in the field, in the lab, and while writing. A special thanks goes to my first students, Bjørg Marie and Valentina. I am very sorry about all the rookie supervisor mistakes I got to make with you. Andrea, you did such amazing and independent work on the Arctic fox samples. Görkem, I am so thankful for the PFAS results you contribute to the project. Tove and Valentin, thank you for making the fourth manuscript into what it is today. Without your amazing skills, we could not have managed to develop and optimize primers for so many genes. You are all amazing, and I wish you the best of luck in the future.

To my fellow PhD candidates at UNIS, we all managed to navigate the social experiment that is Svalbard together, and I cannot imagine doing this without you. Arthur, thank you for sharing this journey with me. This is not the end, only the start of a lifelong friendship. Cheshtaa, thank you for being the best ally possible through all the ups and downs, always ready for a quick rant or small panic cry. Thank you to all the nature guide students who adopted me during my first year here, with a special shoutout to Guri and Kristine, and to all my classmates here at UNIS, especially Mariane and Nicola. Again, thank you to Arthur for bringing his master's student, Kim, up here, who became a very close friend. Kim, thank you for all the laughs, cries, and for always being one phone call away.

Thank you to the Department of Arctic Technology at UNIS and especially Gijs. Even though I am a biologist at heart, the Technology Department was a great home these past four years. Even though you all thought I was odd for walking around in bloody clothes during a reindeer dissection session or being overly enthusiastic about reindeer poop, you made these four years feel safe. Thanks for all the lunches, coffee breaks, and unforgettable summer and Christmas parties.

To my family back home, thank you for supporting the "strange" decision to move to the Arctic, and for visiting even though you froze your assess off. To my friends in Sweden, thank you for forgiving me for leaving and being okay with only seeing me twice a year. And to those who came to visit, Lova, Caroline, Axel, Johanna, Elina, Anna Ida. I love you and thank you for wanting to come and see what my world looks like.

Finally, Jakub. You came into my life at what I call the worst timing (final year of PhD haha), but you have made my life better in every way possible since. Thank you for taking care of me, feeding me, and keeping our life up and running, allowing me to write this thesis in peace.

Coming to Svalbard changed my life, and I will forever be grateful for these four years. Although difficult at times, I would choose to do the PhD here again in a heartbeat. I will miss the view of Hiortfjellet on my walks to work. I will miss the deafening sound of silence surrounding Longyearbyen. I will miss the curiosity of the reindeer, the army of geese in summer, the beluga sightings from the UNIS staircase, and even the annoying calls of the common eider. A little piece of my heart is forever left here. This has been my greatest adventure so far.

In Sweden, we say: "Ingen nämnd, ingen glömd", which translates into "If you don't mention anyone, you don't forget anyone". I truly hope I have not forgotten to mention anyone, but if so, please know that I am very grateful for you, too. I am simply suffering from end-of-PhD brain. Thank you all.

SUMMARY

The Arctic is a sink for contaminants emitted at lower latitudes and transported northward via long-range atmospheric and oceanic pathways. As a result, Arctic wildlife is exposed to contaminants despite inhabiting one of the most remote regions on Earth. With rising temperatures due to climate change, there is a risk of remobilising legacy contaminants stored in the cryosphere, which may further increase exposure for both wildlife and humans in the Arctic. Previous studies have largely focused on assessing contaminants in the Arctic marine environment, while Arctic terrestrial ecosystems have received far less attention.

Studies addressing contaminant exposure in the Svalbard reindeer (*Rangifer tarandus platyrhynchus*) remain limited, despite its key role in the Arctic terrestrial environment as an herbivore, a food source for predators, and a species hunted by locals. To better understand contaminant dynamics in the Svalbard terrestrial environment, this thesis uses the Svalbard reindeer as a model organism and reports concentrations of mercury (Hg), cadmium (Cd), lead (Pb), and several per- and polyfluoroalkyl substances (PFAS) in a range of reindeer tissues and environmental samples. Potential biological effects of chronic, low-level contaminant exposure in Svalbard reindeer were evaluated using transcription levels of genes involved in key physiological processes as endpoints.

Contaminant concentrations in the Svalbard reindeer appear to fluctuate with season, with significantly higher concentrations in autumn compared to summer, which coincides with the annual maximum body mass of the reindeer. This suggests that ingestion of vegetation is the primary contaminant exposure route, with species such as polar willow (*Salix polaris*) likely being major contributors to this exposure. While Cd, Hg, and Pb concentrations in Svalbard reindeer were similar to, or lower than, concentrations reported in other *Rangifer* subspecies across the Arctic, concentrations of several PFAS (e.g., PFHxS, PFUnDA, and PFTriDA) were among the highest. PFAS concentrations in Svalbard reindeer have, moreover, increased significantly over the past decade, suggesting a change in exposure pathways.

The contaminant profile indicates that exposure in Svalbard reindeer is consistent with long-range transported contaminants, as concentrations appear to be independent of proximity to local settlements, and the profile includes compounds that are known to undergo long-range transport to the Arctic (e.g., Hg) or compounds with limited local emissions (e.g., long-chain perfluoroalkyl carboxylic acids, PFCAs). However, we cannot determine whether these contaminants reflect legacy inputs released from the thawing cryosphere or more recent influxes via atmospheric or oceanic pathways. Regardless, this thesis demonstrates that Svalbard reindeer are exposed to a range of contaminants despite living in one of the most remote terrestrial regions on Earth.

Although individual contaminant concentrations in the Svalbard reindeer do not exceed the current wildlife toxicity thresholds, we find that the Svalbard reindeer may be experiencing contaminant-driven biological effects. Exposure to contaminant mixtures in Svalbard reindeer, primarily driven by PFHxS, Hg, and Cd, was associated with altered transcription levels of genes involved in several key physiological processes, most notably in lipid

homeostasis. This indicates that while each compound the reindeer are exposed to may be considered safe on its own, mixtures can cause biological effects at far lower concentrations, supporting the notion that traditional toxicity assessments may underestimate mixture toxicity under real-world conditions. Furthermore, concentrations of both Cd and PFAS are high enough to warrant caution for human consumption, particularly of offal. However, as hunting of Svalbard reindeer is limited to one animal per hunter per year, the overall risk to human health remains low.

SAMMENDRAG

Arktis fungerer som et endepunkt for miljøgifter som slippes ut på lavere breddegrader og transporteres nordover via langtransporterte atmosfæriske og oseaniske prosesser. Som følge av dette eksponeres arktisk dyreliv for miljøgifter, til tross for at regionen er blant de mest avsidesliggende på jorden. Med økende temperaturer som følge av klimaendringer er det risiko for remobilisering av historiske forurensninger lagret i kryosfæren, noe som ytterligere kan øke eksponeringen for både dyreliv og mennesker i Arktis. Tidligere studier har i hovedsak fokusert på forurensninger i arktiske marine økosystemer, mens arktiske terrestriske økosystemer har fått langt mindre oppmerksomhet.

Studier som omhandler eksponering for miljøgifter hos svalbardrein (*Rangifer tarandus platyrhynchus*) er begrensede, til tross for artens sentrale rolle i det arktiske terrestriske økosystemet som planteeter, byttedyr for rovdyr og som en art som høstes av lokalbefolkningen. For å bedre forstå dynamikken til miljøgifter i det terrestriske miljøet på Svalbard er Svalbardrein brukt som modellorganisme i denne avhandlingen, og avhandlingen rapporterer konsentrasjoner av kvikksølv (Hg), kadmium (Cd), bly (Pb) og flere per- og polyfluorerte alkylstoffer (PFAS) i ulike vevstyper hos reinsdyr samt i miljøprøver. Potensielle biologiske effekter av kronisk eksponering for lave nivåer av miljøgifter hos svalbardrein ble vurdert ved bruk av transkripsjonsnivåer av gener involvert i sentrale fysiologiske prosesser som endepunkter.

Konsentrasjoner av miljøgifter hos svalbardrein ser ut til å variere sesongmessig, med signifikant høyere nivåer om høsten sammenlignet med sommeren, noe som sammenfaller med reinsdyrenes årlige maksimum i kroppsmasse. Dette indikerer at inntak av vegetasjon er den primære eksponeringsveien for miljøgifter, der arter som polarvier (*Salix polaris*) sannsynligvis bidrar vesentlig. Mens konsentrasjoner av Cd, Hg og Pb hos svalbardrein var på nivå med eller lavere enn det som er rapportert for andre *Rangifer*-underarter i Arktis, var konsentrasjonene av flere PFAS (f.eks. PFHxS, PFUnDA og PFTriDA) blant de høyeste. Videre har PFAS-konsentrasjonene hos svalbardrein økt betydelig det siste tiåret, noe som indikerer endrede eksponeringsveier.

Forurensningsprofilen indikerer at eksponeringen hos svalbardrein i hovedsak er forenlig med langtransporterte miljøgifter, ettersom konsentrasjonene ikke ser ut til å være påvirket av nærhet til lokale bosetninger, og profilen inkluderer forbindelser som er kjent for å gjennomgå langtransport til Arktis (f.eks. Hg) eller forbindelser med begrensede lokale utslipp (f.eks. langkjedede perfluorkarbonsyrer, PFCA). Det er imidlertid ikke mulig å fastslå om disse forurensningene reflekterer historiske bidrag frigjort fra en tinende kryosfære eller nyere tilførsler via atmosfæriske eller oseaniske transportveier. Uavhengig av dette viser avhandlingen at svalbardrein eksponeres for et bredt spekter av miljøgifter, til tross for at de lever i et av de mest avsidesliggende terrestriske områdene på jorden.

Selv om konsentrasjonene av enkeltstoffer hos svalbardrein ikke overskrider gjeldende toksisitetsgrenser for dyreliv, indikerer resultatene at reinsdyrene likevel kan oppleve forurensningsrelaterte biologiske effekter. Eksponering for blandinger av miljøgifter, primært

drevet av PFHxS, Hg og Cd, var assosiert med endrede transkripsjonsnivåer av gener involvert i flere sentrale fysiologiske prosesser, særlig lipidhomeostase. Dette indikerer at selv om hvert enkelt stoff kan anses som lite toksisk isolert sett, kan stoffblandinger gi biologiske effekter ved langt lavere konsentrasjoner, noe som støtter antakelsen om at tradisjonelle toksisitetsvurderinger kan undervurdere blandingstoksisitet under realistiske eksponeringsforhold. Videre er konsentrasjonene av både Cd og PFAS høye nok til å tilsi forsiktighet ved konsum for mennesker, særlig av innmat. Ettersom jakten på Svalbardrein er begrenset til ett dyr per jeger per år, vurderes den samlede helserisikoen for mennesker imidlertid som lav.

ABBREVIATIONS

Cd	Cadmium
CBR	Critical body residue
DDT	Dichlorodiphenyltrichloroethane
dw	Dry weight
FOSAs	Perfluoralkyl sulfonamides
FTOHs	Fluorotelomer-based compounds
Hg	Mercury
LRAT	Long-range atmospheric transport
LOD	Limits of detection
PAH	Polycyclic aromatic hydrocarbons
PCBs	Polychlorinated biphenyls
Pb	Lead
PBDEs	Polybrominated diphenyl ethers
PFAS	Per- and polyfluoroalkyl substances
PFCAs	Perfluoroalkyl carboxylic acids
PFSAs	Perfluorosulfonic acids
PCB	Polychlorinated biphenyl
POPs	Persistent organic pollutants
ww	Wet weight

TABLE OF CONTENTS

LIST OF PAPERS

This thesis is based on four papers that will be referred to in the text by roman numerals I-IV.

I. Andersson Stavridis, M., Røed, S.B., Hansen, B.B., Mikkelsen, Ø., Ciesielski, T.M., Jenssen, B.M., 2024. Tracing the footprints of Arctic pollution: Spatial variations in toxic and essential elements in Svalbard reindeer (*Rangifer tarandus platyrhynchus*) faeces. Sci. Total Environ. 906, 167562.

II. Andersson Stavridis, M., Pollestad, B.M., Veiberg, V., Mikkelsen, Ø., Ciesielski, T.M., Jenssen, B.M., 2025. Toxic metals and other elements in Svalbard reindeer: Establishing baselines and assessing non-invasive sampling for biomonitoring. Sci. Total Environ. 961, 178226.

III. Andersson Stavridis, M., Petersson, T., Kendir G. D., Sait, S., Mikkelsen, Ø., Veiberg, V., Ciesielski, T. M., Jenssen, B. M., 2025. Seasonal Shift in Exposure and Accumulation of PFAS and Heavy Metals in High Arctic Reindeer. *In review*.

IV. Andersson Stavridis, M., Petersson, T., Garreau, Mikkelsen, Ø., Veiberg, V., Reinardy, H. C., Jenssen, B. M., Ciesielski, T. M., 2025. Low-Level Chronic Exposure to PFAS and Toxic Metals Induces Transcriptional Changes in Svalbard Reindeer. *Manuscript ready for submission*.

DECLARATION OF CONTRIBUTIONS

Paper I was initiated by SBR, BMJ, and TMC with the help of BBH for sampling. Chemical analysis was performed by SBR under the supervision of TMC and BMJ. **MAS** performed all statistical analyses and manuscript writing under the supervision of ØM, TMC, and BMJ.

Paper II was initiated and planned by **MAS** in collaboration with ØM and BMJ. The sample collection was organised and performed by **MAS** and VV. The chemical analysis was organised and performed by **MAS** with the help of BMP. Statistical analyses and writing were done by **MAS** with support from all co-authors.

Paper III was initiated and planned by **MAS** with support from BMJ and TMC. The sample collection was organised and performed by **MAS**, TP, and VV. Metal analysis was performed by MAS and TP, while PFAS analysis was performed by GDK under the supervision of SS and TMC. Statistical analyses and writing were done by **MAS** with support from all co-authors.

Paper IV was initiated and planned by **MAS**. The sample collection was organised by and performed by VV. Primer design and optimisation, as well as gene expression analysis, were conducted by **MAS**, TP, and VG. Statistical analyses and writing were done by **MAS** with support from all co-authors.

ADDITIONAL PUBLICATIONS NOT INCLUDED IN THE THESIS

V. Andersson Stavridis, M., Pollestad, B. M., Piqué, V. A., Mikkelsen, Ø., Jenssen, B. M., Ciesielski, T. M., 2025. Faecal Biomonitoring of Mercury in Arctic Terrestrial Herbivores Reveals Spatial Trends Across Svalbard and Greenland. *Manuscript in preparation*.

INTRODUCTION

LEGACY AND EMERGING POLLUTANTS IN THE ENVIRONMENT

The anthropogenic disruption of the biogeochemical cycles of several metals has led to their increased presence in the biosphere since the onset of the Industrial Revolution (Amos et al., 2013). This especially applies to mercury (Hg), cadmium (Cd), and lead (Pb), which are associated with a wide range of adverse health effects, including neurotoxicity, nephrotoxicity, cytotoxicity, and immunotoxicity (Balali-Mood et al., 2021; Klaassen and Watkins, 2015). Activities such as non-ferrous metal mining and smelting, the burning of fossil fuels, and the historical use of leaded gasoline have led to a significant biospheric increase of these metals (Dudka and Adriano, 1997; Eisenreich et al., 1986; Pacyna and Pacyna, 2001). Once emitted into the environment, these metals can disperse globally via long-range oceanic (Søndergaard et al., 2025) or atmospheric transport (Gworek et al., 2020). As a result, their impacts are not limited to areas near emission sources but also extend to more remote regions. While the emissions of Hg, Cd, and Pb are regulated under several conventions and protocols (e.g., the Minamata Convention; United Nations Economic Commission for Europe, 1998; United Nations Environment Programme, 2024), legacy emissions of these metals are still circulating in the biosphere (Amos et al., 2013).

A similar pattern of global distribution can also be observed for persistent organic pollutants (POPs) (Wania and Mackay, 1993). Historically, POPs such as polychlorinated biphenyls (PCBs), dichlorodiphenyltrichloroethane (DDT), and polybrominated diphenyl ethers (PBDEs) were all designed for specific industrial and agricultural purposes, but their chemical properties also made them persistent to degradation, bioaccumulative, and toxic, leading to global environmental and human health issues (Jones, 2021). Similar to metals, regulatory frameworks, such as the Stockholm Convention, were established following the discovery of the toxic properties of these POPs (United Nations Environment Programme, 2001), leading to significantly reduced environmental emissions (Schuster et al., 2010). However, the introduction of new synthetic compounds into the environment continued, despite an increasing awareness of the potential environmental and human consequences. An example is the rise of per- and polyfluorinated substances (PFAS) in the biosphere.

PFAS are specifically designed to be resistant to heat, water, lipids, and oil, but as a consequence, they are also persistent to natural degradation. As a result of this stability, PFAS are often referred to as "forever chemicals" (Brunn et al., 2023). While some PFAS are listed under the Stockholm Convention (i.e., PFOS, PFOA, and PFHxS), the entire PFAS family currently includes more than 7 million individual compounds (Schymanski et al., 2023). These compounds can be divided into different PFAS classes, including perfluoroalkyl carboxylic acids (PFCAs), perfluoroalkane sulfonic acids (PFSAs), fluorotelomer-based compounds (FTOHs), and perfluoroalkyl sulfonamides (FOSAs) (Buck et al., 2011). While most PFAS are persistent to degradation, both FTOHs and FOSAs are labelled as precursor PFAS as they may undergo environmental transformation into more stable PFAS such as PFCAs and PFSAs (e.g., Ellis et al., 2004; Young and Mabury, 2010). These PFAS precursors are furthermore highly volatile and may travel far from point sources of emission via long-

range atmospheric transport (LRAT), which helps explain how PFAS are found in the most remote regions of Earth (Garnett et al., 2022; Hartz et al., 2023).

PFAS may enter the biosphere directly, such as through the use of aqueous film-forming foam (AFFF) used in firefighting (Moody and Field, 2000), or indirectly, through the production, use, and end-of-life of many everyday products that contain PFAS, such as non-stick cookware, water-repellent clothing, and food packaging (Herzke et al., 2012). Emissions of PFAS, mainly through the use of AFFS, have become a major global concern since PFAS, in addition to their persistent nature, are known as carcinogens, endocrine disruptors, immunosuppressants, and are associated with liver diseases in humans (Fenton et al., 2020). For example, higher rates of cancer have been reported in humans living in communities exposed to PFAS-contaminated groundwater (Li et al., 2022; Mastrantonio et al., 2018; Winquist et al., 2023).

POLLUTANTS IN THE ARCTIC

Although the Arctic often is perceived as a remote and untouched region, it has been characterised as a global sink for pollution (AMAP, 2005; Burkow and Kallenborn, 2000). Through long-range transport, especially semi-volatile pollutants emitted from all over the world may reach the Arctic (Muir et al., 2025; Søndergaard et al., 2025). Once they reach and deposit in the Arctic, the cold conditions of the region limit further mobilisation, resulting in their accumulation in the Arctic cryosphere (Beard et al., 2022; Wania and Mackay, 1993). For example, although far from any larger emission sources, PFAS have been detected in glaciers, snow, and meltwater throughout the Arctic, from Canada (Stock et al., 2007) to Svalbard (Ahrens et al., 2023; Hartz et al., 2023). Likewise, the storage of legacy Hg in Arctic permafrost is one of the largest sinks of Hg on Earth (Schuster et al., 2018).

In addition to remote sources, there are local sources contributing to Arctic pollution. In Svalbard, the use of AFFFs at the Longyearbyen firefighting training site or at the recently decommissioned (2022) Svea mine and airport has contaminated surrounding freshwater, soil, snow, and fauna with PFAS (Ahrens et al., 2023; Ali et al., 2021; Evenset et al., 2020). Moreover, the small-scale coal-fired power plants in the settlements of Svalbard are known sources of pollutants like polycyclic aromatic hydrocarbons and Hg (Drotikova et al., 2020; Steenhuisen and Van Den Heuvel-Greve, 2021).

With pollutants present in the Arctic environment, Arctic wildlife is also exposed, despite living in one of the most remote habitats on Earth. The pollutants of particular concern are those that bioaccumulate in organisms and biomagnify across trophic levels, resulting in apex predators having concentrations that are millions of times higher than those found in their surrounding environment (Lehnherr, 2014). The long marine food webs, with many trophic levels and interactions, result in a particular risk for high contaminant burdens in marine wildlife (Lehnherr, 2014; Scheuhammer et al., 2015). Therefore, monitoring of Arctic marine biota has been performed over the past decades to assess contaminant concentrations and temporal trends, with a predominant focus on wildlife of high concern, such as glaucous gulls (*Larus hyperboreus*; Melnes et al., 2017; Verreault et al., 2010), polar bears (*Ursus maritimus*; Dietz et al., 2022, 2004; Greaves et al., 2012), killer whales (*Orcinus*

orca; AMAP, 2018; Desforges et al., 2024, 2018), and seals (*Pusa* spp.; Houde et al., 2019; Rigét et al., 2020), which consistently show high contaminant burdens. In contrast, the Arctic terrestrial ecosystem remains relatively understudied, despite our knowledge of large contaminant reservoirs in the Arctic cryosphere.

With ongoing climate change and Arctic amplification, the Arctic region is warming more than four times faster than the global average (Rantanen et al., 2022). This rapid warming is causing the Arctic cryosphere to thaw, which may result in the stored pollutants remobilising into other media and becoming bioavailable again (Carrasco et al., 2024; Muir et al., 2025; Schuster et al., 2018). Once remobilised, pollutants can enter aquatic systems via freshwater transport, bind to organic matter, or be taken up by vegetation (MacInnis et al., 2022; Schaefer et al., 2020).

Numerous studies report the uptake of pollutants in Arctic vegetation, ranging from metals such as Hg, Cd, and Pb, to organic contaminants like PFAS. Vascular vegetation primarily absorbs pollutants from the surrounding soil via root uptake (e.g., Pourrut et al. 2011; Ismael et al. 2019), while non-vascular species such as bryophytes and lichens rely on atmospheric uptake (Bargagli, 2016; Zhou et al., 2021), resulting in distinct pollution profiles among different species of vegetation. The highest concentrations of Hg are commonly reported in bryophytes and lichen (Bargagli, 2016; Steinnes, 1995). As vascular uptake of Pb from surrounding soil is limited, the highest Pb concentrations are also reported in bryophytes and lichen (Krajcarová et al., 2016; Wojtuń et al., 2019, 2013). In contrast, the polar willow *Salix polaris* is a phytoaccumulator of Cd, with stems and leaves having concentrations severalfold higher than those of the surrounding soils (Wojtuń et al., 2019). Many vascular species of vegetation, including *S. polaris* and white dryas (*Dryas octopetala*), have also been found to accumulate PFAS from soil (Dai et al., 2025). As pollutants are present in Arctic vegetation, they may act as a vector of pollution, introducing both metals and PFAS into the Arctic terrestrial food chain.

THE SVALBARD REINDEER

Svalbard is an archipelago in the High Arctic (78°N). The islands are characterised by extensive glaciation and extreme seasonality, with months of polar night in winter and midnight sun in summer. Terrestrial wildlife is scarce, and is dominated by birds (i.e., rock ptarmigan, *Lagopus muta*, and snow bunting, *Plectrophenax nivalis*), Arctic fox (*Vulpes lagopus*), and the Svalbard reindeer (*Rangifer tarandus platyrhynchus*) (Descamps et al., 2017), which is the largest terrestrial mammal on the archipelago. Compared to many other species of reindeer and caribou, the Svalbard reindeer are relatively non-migratory and often remain in the same valley systems throughout their lifespan (Kinck, 2014; Tyler and Øritsland, 1989). Harsh climatic conditions, such as an extensive snow cover or rain-on-snow events, limit their access to feed in winter (Nov-Apr), resulting in pronounced seasonal variations in feeding behaviour, diet, and body mass (Albon et al., 2017; Kohler and Aanes, 2004). In the summer (Jun-Aug), when feed is abundant, the reindeer spend around 70 % of their time grazing (Tyler, 1987). They primarily feed on different graminoids (e.g., grasses), with minor contributions from different forbs and *S. polaris* (Bjørkvoll et al., 2009). As a result, the reindeer fatten during summer and early autumn and reach their peak body mass

by the end of October (Albon et al., 2017). With limited access to forage in winter, the diet of the Svalbard reindeer becomes more opportunistic. The winter diet is characterised by remnants of old graminoids, various bryophytes, and a relatively high fraction of *S. polaris* (Bjørkvoll et al., 2009; Hiltunen et al., 2022). In addition, during icy winter conditions, the Svalbard reindeer may feed closer to the shorelines, introducing marine biomass such as kelp into their diets (Hansen et al., 2019).

POLLUTANTS IN THE SVALBARD REINDEER

Studies on contaminants in Svalbard terrestrial mammals have so far predominantly focused on Arctic foxes, and long-term trends in both organic contaminants and Hg concentrations have been assessed in several studies (Aas et al., 2014; Hallanger et al., 2019; Wang-Andersen et al., 1993). In contrast, fewer studies have investigated other terrestrial mammals on the archipelago, such as the Svalbard reindeer.

The first study on pollutants in the Svalbard reindeer was performed in 1984-1986 by Borch-Iohnsen et al. (1996), who evaluated the influence of season and diet on metals in Svalbard reindeer. They reported hepatic and renal concentrations of Cd (0.73 µg/g and 4.30 µg/g, respectively) and Pb (1.30 µg/g and 0.8 µg/g, respectively). They report a strong seasonal influence on the accumulation of these metals, which was attributed to changes in physiological conditions and seasonal feeding patterns.

The following studies on pollutants in Svalbard reindeer were all performed using non-invasive samples, such as fur and faeces (Table 1). Both fur and faeces are practical matrices to use in biomonitoring, as the collection is relatively easy and there are minimal ethical concerns to consider (Jota Baptista et al., 2022). There are, however, limitations in using these kinds of samples. Yin et al. (2008) argue that while faeces may help compare pollution levels across species or different sampling locations, concentrations may be hard to interpret and use in toxicity assessments as the levels reflect a combination of both excreted and unabsorbed pollutants. Likewise, the use of hair in biomonitoring may also be challenging. Correlations between concentrations in hair and soft tissues have been established for specific contaminants and specific species, such as Hg concentrations in hair and blood in humans (Phelps et al., 1980), and Hg concentrations in hair and liver or kidney in polar bear (Born et al., 1991) and Arctic fox (Treu et al., 2018). Yet, due to interspecific differences in Hg metabolism and excretion via hair, such relationships are likely species-specific and need to be validated for a species before use (Dietz et al., 2009).

Pacyna et al. (2019) evaluated the use of Svalbard reindeer droppings for biomonitoring elements in the Arctic terrestrial environment, arguing that droppings provide useful information on the exposure and excretion of contaminants in the reindeer. They compared element concentrations in fresh summer faeces to those in older summer samples (<3 months) to evaluate the necessity of using fresh samples when assessing contaminant exposure. They concluded that for both Pb and Cd, older faecal samples are a suitable alternative when fresh samples cannot be obtained. Another study on Svalbard reindeer faeces assessed whether the differences in winter and summer diet would result in an annual variability in faecal heavy metal content (Węgrzyn et al., 2018). In general, the study

found a close relationship between the analysed heavy metals in faeces and in the feed of the reindeer, which varies between summer and winter. They also found higher levels of Cd in the summer faeces, which they attributed to a higher fraction of vascular vegetation that accumulates Cd from the surrounding soil. Similarly, Pacyna et al. (2018) used elemental concentrations in Svalbard reindeer fur to assess spatial differences in metal exposure between Svalbard reindeer living near Longyearbyen (78°N) or Hornsund (77°N). They did not find any significant differences in elemental composition or concentration between the two sampling locations. In a follow-up study, Pacyna-Kuchta et al. (2020) used Svalbard reindeer fur to screen exposure to trace elements and persistent organic pollutants. They reported low levels of Cd and Pb, while Hg levels were below the limits of detection (LOD). In addition, they detected dichlorodiphenyltrichloroethane (DDT), polycyclic aromatic hydrocarbons (PAHs), and one polychlorinated biphenyl (PCB) in the samples, demonstrating how fur can be used to assess elemental exposure.

Table 1. An overview of previous studies reporting contaminant concentrations in fur and faeces samples from the Svalbard reindeer (*Rangifer tarandus platyrhynchus*). All concentrations are presented as means and derive from the reported concentrations in the referenced studies. All concentrations are reported in µg/g dry weight (dw), unless annotated with an (a), indicating wet weight (ww).

	Tissue	n	Conc. in µg/g	Year	Reference
Hg	Fur	8	<LOD	2017	Pacyna-Kuchta et al., 2020
	Fur	9	0.20	2015-	Pacyna et al., 2018
	Faeces	23	0.10	2016	Pacyna et al., 2019
Cd	Fur	8	0.17	2017	Pacyna-Kuchta et al., 2020
	Fur	27	0.08	2015-	Pacyna et al., 2018
	Faeces	30	0.002 [a]	2015	Węgrzyn et al., 2018
	Faeces	54	1.12	2016	Pacyna et al., 2019
Pb	Fur	8	0.63	2017	Pacyna-Kuchta et al., 2020
	Fur	27	1.82	2015-	Pacyna et al., 2018
	Faeces	30	0.007 [a]	2015	Węgrzyn et al., 2018
	Faeces	54	4.28	2016	Pacyna et al., 2019

Overall, these studies provide a base for using non-invasive samples and demonstrate that the contaminant concentrations in Svalbard reindeer are relatively low. However, none of the studies on Svalbard reindeer conducted in the 21st century accounted for internal organ concentrations or assessed whether non-invasive samples may be representative of invasive concentrations. As a current assessment of renal or hepatic contaminant concentrations is missing, it is difficult to, for example, assess any temporal changes in exposure and accumulation of Cd and Pb since the study from the mid-1980s (Borch-Iohnsen et al., 1996). This may be of particular interest considering the future of the thawing Arctic and the risk of increased exposure to remobilised legacy contaminants in the Arctic terrestrial environment. As Svalbard reindeer are hunted by residents (Sysselmesteren,

2019), it is furthermore of value to monitor current contaminant concentrations in internal tissues to assess the potential risks associated with consumption.

EFFECTS OF CONTAMINANT EXPOSURE

Exposure to both heavy metals and PFAS is associated with adverse effects on vital physiological functions. Metals interact with various biological systems through different pathways of toxicity. Hg, Cd, and Pb are all known to cause oxidative stress, which in turn may result in damaged membranes, proteins, and DNA (Balali-Mood et al., 2021). Another main mechanism of action for both Hg and Cd involves their high affinity for thiol (-SH) groups of enzymes and proteins, resulting in irreversible binding that can alter or inactivate protein function (Joseph, 2009; Souza-Arroyo et al., 2022; Ynalvez et al., 2016). A main mechanism of action for Pb is ionic mimicry, since Pb ions are structurally similar to essential ions such as Ca^{2+}, Zn^{2+}, and Fe^{2+}, which allows Pb to replace these ions, and thus disrupts enzyme function and neurotransmitter activity (Jomova et al., 2025). Consequently, these mechanisms lead to a range of symptoms of toxicity, including neurological damage, histopathological changes in tissues, and immunosuppression (Genchi et al., 2020; Kim et al., 2016; Wani et al., 2015).

The most pronounced toxic effects of PFAS exposure are immunological (Schrenk et al., 2020). While the mechanisms of action are not yet fully understood, and most existing studies solely focus on PFOS and PFOA, it is evident that exposure to both PFSAs and PFCAs causes a range of immunological effects, including an increased susceptibility to infections and a reduced efficiency of vaccines (Ehrlich et al., 2023). PFAS have been found to inhibit both T-cell and antibody responses (e.g., Liang et al. 2022; Iulini et al. 2025), to directly induce the formation of ROS, causing oxidative stress (e.g., Wielsøe et al. 2015), and to modulate the transcription of several vital genes involved in, for example, the metabolism of fatty acids, adipogenesis, or immune system regulation (Ehrlich et al., 2023; Ishibashi et al., 2008; Lühmann et al., 2020; Routti et al., 2019).

With extensive knowledge on contaminant toxicity, toxicity thresholds derived from experimental exposure studies help distinguish between harmful concentrations and presumed safe concentrations, which are considered low enough to avoid any adverse biological effects. In wildlife, commonly used toxicity thresholds are hepatic concentrations of 4.2 µg/g (ww) for Hg (Dietz et al., 2022), renal concentrations of 100 µg/g (ww) for Cd (Larison et al., 2000), and a critical body residue (CBR) below 655 ng/g (ww) for PFAS (Dietz et al., 2018a). For Pb, renal concentrations above 15 µg/g (dw) have been associated with functional kidney damage and concentrations above 80 µg/g (dw) with severe weight loss and death in a range of mammals (Ma, 2011).

Generally, contaminant concentrations in Arctic terrestrial wildlife are below these established toxicity thresholds (Dietz et al., 2022). For example, hepatic ΣPFSA concentrations of around 100 ng/g (ww) were reported in Arctic foxes from Svalbard. Given that PFAS accumulate in the liver (Schrenk et al., 2020), these hepatic concentrations likely represent the highest in the body, suggesting that the total body burden remains below the CBR toxicity threshold. In contrast, hepatic Hg levels in Icelandic adult Arctic foxes range

between 0.28-46.2 µg/g (ww) in males and 0.96-35.5 µg/g (ww) in females (Treu et al., 2018), placing a relatively high proportion of the studied populations (~57 %) in a moderate or high-risk category for adverse effects from Hg exposure (Dietz et al., 2022).

By comparison, far lower concentrations of toxic metals and PFAS are reported in the different subspecies of reindeer and caribou across the Arctic (Gamberg et al., 2016, 2005; Roos et al., 2021). Some of the highest PFOS concentrations have been reported in reindeer and caribou from the Akia-Maniitsoq and Kangerlussuaq-Sisimiut regions, with mean concentrations of 14.3-15.6 ng/g (ww) (Roos et al., 2021), which are far below the critical body residues for PFAS. Similarly, some of the highest Hg concentrations are reported by (Gamberg et al., 2020) in Barren-ground caribou (*Rangifer tarandus groenlandicus*) from Baffin Island, with renal mean Hg concentration ranging between 1.3-1.7 µg/g (ww) (recalculated from dw using the renal moisture content (78%) reported in Gamberg et al. (2016)). Overall, Arctic reindeer and caribou have been placed in the no-risk category for Hg (Dietz et al., 2022), and their contaminant concentrations are considered unlikely to pose any immediate health risks for them.

To the best of my knowledge, no studies have attempted to assess the potential effects of exposure to contaminants in any of the *Rangifer* subspecies. Toxicity thresholds represent reference points rather than definitive cut-offs, as they typically derive from specific endpoints and may not capture sublethal effects occurring at lower concentrations (Cairns, 1992). For example, the threshold of 4.2 µg/g (ww) of Hg originates from exposure studies on mink (*Mustela vision*) using endpoints on tissue (e.g., histology) or behavioural (e.g., anorexia or ataxia) level (Wobeser et al., 1976; Wren et al., 1987). While it is known that exposure to high concentrations of heavy metals may result in severe acute symptoms, continuous chronic exposure to lower concentrations may lead to more subtle effects that are harder to detect (Balali-Mood et al., 2021). In addition, Arctic wildlife is exposed to a chemical cocktail (Sonne et al., 2021), where there can be cumulative effects from the combined exposure to several different pollutants (Balali-Mood et al., 2021). Hence, exposure to pollutant concentrations below the established thresholds may still cause biological effects that may go unnoticed.

GENE EXPRESSION AS AN ENDPOINT OF TOXICITY
While most studies on contaminant effects are conducted under controlled laboratory conditions, field-based studies are important as they both quantify the contaminant concentrations in natural populations and assess whether the concentrations are sufficiently high to cause biological effects in wildlife (Ankley et al., 2020). Wildlife biomonitoring often combines the quantification of contaminants with evaluations of biological endpoints of toxicity, frequently at a subcellular level, such as gene expression (e.g., Bjørneset et al., 2023; King et al., 2023; Lühmann et al., 2020; Lunardi et al., 2016).

Alterations in gene expression are one of the rapid biological responses to environmental stressors, such as increased exposure to contaminants (e.g., Robinson et al., 2012). Well-known examples include the rapid upregulation of genes encoding detoxification enzymes, such as CYP1A, following exposure to PAHs and PCBs (Bucheli and Fent, 1995). These

immediate transcriptional responses occur well before changes related to contaminant exposure are detected at higher levels of biological organisation, which may already indicate adverse effects (Joseph, 2022). As such, gene expression may serve as a more sensitive early warning signal of contaminant exposure, and transcriptional changes may indicate exposure before more severe or irreversible effects manifest.

Several genes have been proposed to serve as biomarkers of exposure for specific pollutants. Notable examples include genes that encode members of the peroxisome proliferator-activated receptor (PPAR) family, which regulate vital physiological functions, including fatty acid oxidation and lipid metabolism (Mandard et al., 2004). Studies across different species of mammals have consistently found genes from this family, especially *PPARA*, to be upregulated as a response to PFAS exposure, including in the Baikal seal (*Pusa sibirica*; Ishibashi et al., 2008), polar bear (Routti et al., 2019), and different species of cetaceans (Kurtz et al., 2019). Other examples include genes from the cytochrome P450 (CYP) family, which have shown a significant upregulation as a response to PFAS exposure in both cetaceans (Kurtz et al., 2019) and seabirds (King et al., 2023). These genetic responses to PFAS exposure have been evaluated across multiple taxa, revealing consistent patterns of gene expression regardless of species (Beccacece et al., 2023), further supporting the use of gene expression as a suitable predictive marker of PFAS exposure in wildlife.

Similarly, Hg exposure in wild seabirds has been associated with the upregulation of genes such as *CYP3a37* and selenoprotein (*SELENOP*) (King et al., 2023), while both Hg and Cd have shown positive relationships with the expression of genes involved in detoxification (e.g., metallothionein) and oxidative stress response (e.g., glutathione reductase), in in vitro studies on fish (Morcillo et al., 2016).

Many contaminants present in Arctic wildlife are capable of causing harmful biological effects. Consequently, even low concentrations, such as those reported in *Rangifer* subspecies across the Arctic, may influence lower levels of biological organisation, including gene expression. Even subtle alterations in transcriptional profiles may cascade into effects at higher levels of biological organisation. However, as species with relatively low contaminant burdens are often understudied, such subcellular effects may remain unnoticed.

THESIS AIMS

The primary aim of this thesis was to investigate the occurrence and effects of PFAS and toxic elements Hg, Cd, and Pb in the Arctic terrestrial environment using the Svalbard reindeer as the model organism. This was achieved by assessing the exposure to, and distribution and excretion of, contaminants in the reindeer. In addition, potential subcellular effects of contaminant exposure in the reindeer were assessed. The primary aims were divided into the following objectives:

I) To investigate the distribution and excretion of Hg, Cd, and Pb in the Svalbard reindeer by determining concentrations in soft tissues, blood, milk, faeces, and fur, and to evaluate the potential of non-invasive samples for monitoring internal metal burdens (**Paper II**).

II) To determine differences in elemental exposure between different populations of Svalbard reindeer and assess potential local anthropogenic contributions of Hg, Cd, Pb, and other elements by analysing the faecal elemental composition in two reindeer populations inhabiting areas of varying distance to human settlements (**Paper I**).

III) To determine whether contaminant concentrations (Hg, Cd, Pb, and PFAS) exceed the thresholds of toxicity in the reindeer, or the thresholds of human consumption (**Paper II-III**).

IV) To assess whether there is a seasonal variation in contaminant concentrations in Svalbard reindeer by comparing the contaminant profile (metals and PFAS) in reindeer culled in late summer (August) with individuals culled in autumn (October) (**Paper III**).

V) To investigate whether exposure to contaminants (Hg, Cd, Pb, and PFAS) induces molecular responses in Svalbard reindeer by assessing associations between mRNA transcription and contaminant concentrations (**Paper IV**).

METHODS

FIELDWORK PAPER I

Fresh faecal samples were collected from Svalbard reindeer in the spring (Mar-Apr) of 2014, 2015, and 2017 as part of the fieldwork in the AT-330 (Arctic Environmental Toxicology) course given at The University Centre in Svalbard (UNIS). The samples were collected from the Nordenskiöld coast (n=33) and Adventdalen (n=63), in central Spitsbergen (Fig. 1). The sex and approximate age (juvenile or adult) of the reindeer were estimated while observing them from a distance. Once defecation occurred, the subsequent collection of the faeces took place within a few minutes. All faecal samples were stored in polyethylene zip-lock bags in −20°C until further processing.

FIELDWORK PAPER II-IV

Over three years, a total of 68 female Svalbard reindeer were culled in an international and interdisciplinary research project (permission 16/01632–40 and 21/03815–4 from the Governor of Svalbard). All reindeer were culled in central Nordenskiöld land (around Reindalen and Colesdalen, 77°05'–78°20'N, 15°00'–15°60'E; Fig. 1) in August 2022 (n=12) and October 2021 (n=18), 2022 (n=18), and 2023 (n=20). All individuals were shot from a distance of approximately 80 meters, in the heart and lung region.

Figure 1. An overview of central Spitsbergen, Svalbard, marking out major sampling sites of the PhD project by name.

In August 2022, liver and muscle samples were collected within thirty minutes post-mortem in the field, while for all October cullings (2021-2023), the full necropsy took place within twelve hours at the University Centre in Svalbard (UNIS) (Fig. 2). The exception was blood, which was immediately collected post-mortem from the jugular vein into EDTA-lined vacutainer vials (VACUETTE, Greiner Bio-One, Austria). A complete list of collected samples is found in Table 2. All samples were collected using fresh nitrile gloves and single-use scalpels (Swann-Morton, Swann-Morton Limited, UK) to reduce cross-contamination between individuals. In addition, the outer layer of all tissue samples (a few mm) was removed to reduce potential external contamination. Half a kidney lobe was sampled, and the liver samples were primarily collected from the left liver lobe. Muscle samples were collected from the medial thigh muscle. The brain samples either consisted of an entire brain lobe, when the skull could be opened using a saw, or from the brainstem using a bovine brain sampling spoon. White guard hairs were collected using clean stainless-steel scissors from the rump region. Faecal pellets were collected from the large intestine. All samples were collected into polyethylene zip-lock bags. If the female reindeer was lactating, the udder was dissected, and milk was collected into a 20 mL centrifuge tube. All samples were kept at -80°C until further processing and subsequent contaminant analysis.
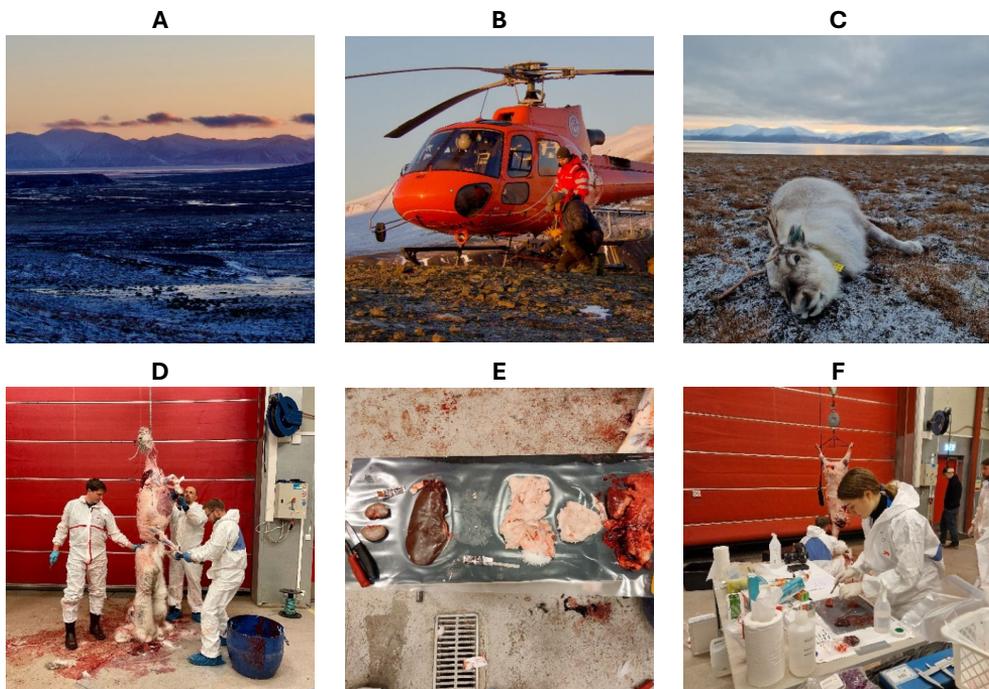
Figure 2. An overview of the annual culling procedure in October 2021-2023. A) Reindalen in mid-October, B) the helicopter used for transport out and back to the culling sites, C) an ID-tagged female Svalbard reindeer, D) carcass processing back at UNIS, E) kidney, liver, kidney fat, and heart from the Svalbard reindeer, and F) subsampling of liver samples. Photos: Malin Andersson Stavridis, UNIS/NTNU, and Stefaniya Kamenova, CEES.

In August 2022, samples were also collected for gene expression analysis, in addition to those collected for contaminant analysis. Small samples of liver and muscle (approximately 1 × 0.5 cm) were preserved in RNA later for subsequent extraction of mRNA. However, the RNA extracted from the muscle samples was of poor quality and was therefore excluded from downstream qPCR analyses.

Table 2. An overview of the samples collected during the respective culling.

| | October | | | August |
Tissue	2021	2022	2023	2022
Fur	× (a,b)	× (a)		
Faeces	× (a,b)	× (a)		
Brain	× (a)			
Blood		× (a)		
Liver	× (a,b)	× (a)	× (a,b,c)	× (a,b,c,d)
Kidney	× (a,b)	× (a)		
Muscle	× (a,b)	× (a)	× (a,b,c)	× (a,b,c)
Milk		× (a)		

Analysed for [a] mercury; [b] elemental analysis; [c] PFAS; [d] gene expression.

SAMPLE PREPARATION AND CHEMICAL ANALYSIS

All chemical analyses were conducted at the Department of Chemistry, Norwegian University of Science and Technology (NTNU), Norway. The full details on sample processing, analytical procedures, and quality assurance and control are found in the method description and supplementary materials of **Papers I-III**. The procedures are briefly described below.

For **Paper I**, faecal samples were analysed for 16 elements (Al, As, Ca, Cd, Cu, Fe, Hg, Mg, Na, Ni, P, Pb, S, Se, Si, and Zn), all of which were detected above the limit of detection (LOD). The analysis was conducted using a high-resolution inductively coupled plasma mass spectrometer (ICP-MS, Thermo Finnigan Element 2, Bremen, Germany) following the methods described in Dolan et al. (2017) and Halbach et al. (2017).

In **Paper II**, all faecal, fur, blood, brain, liver, kidney, muscle, and milk samples were analysed for Hg using a Direct Mercury Analyser (DMA-80, Milestone, Italy) following the US EPA method 7473. In addition, faeces, fur, liver, kidney, and muscle were analysed for eight elements (Al, Cd, Fe, P, Pb, S, Se, and Si) using an 8800 Triple Quadrupole ICP-MS system (Agilent, Santa Clara, USA) equipped with a prep FAST M5 autosampler (ESI, USA). All contaminants were detected above LOD in all samples.

In **Paper III**, liver and muscle samples were analysed for Hg, Cd, and Pb following the same protocol as in **Paper II** (Hg using DMA-80 and Cd and Pb using ICP-MS). In addition, samples were analysed for 41 per- and polyfluoroalkyl substances (PFAS), divided into perfluoroalkyl carboxylic acids (C5-C18 PFCAs), perfluoroalkyl sulfonic acids (C5-C11 PFSAs and PFECHS), fluorotelomer sulfonates (4:2, 6:2, 8:2 and 10:2 FTSs), sulfonamides and

precursor PFAS (FOSAA, MeFOSAA, EtFOSAA, PFOSA, MeFOSA, EtFOSA, MeFOSE, and EtFOSE) and emerging PFAS (GenX, NaDONA, 9Cl-PF3ONS, P37DMOA, SaMPAP and diSAMPAP) (See Supporting Information from **Paper III** for the full names of all analysed PFAS). Following the protocol described in Sait et al. (2023), PFAS analysis was performed using an Xevo TQ-XS Triple Quadrupole Mass Spectrometer coupled with an ACQUITY UPLC system (Waters, MA, USA), with manual mass peak integration using MassLynx software (Waters, MA, USA). Only PFAS detected in more than 25% of samples were reported, which included PFUnDA, PFTriDA, PFTDA, PFHxDA, PFHxS, PFOS, PFNS, PFDS, PFECHS, FOSAA, MeFOSAA, EtFOSAA, and NaDONA.

MOLECULAR ANALYSIS

Hepatic mRNA was extracted, reverse-transcribed into cDNA, and analysed for the expression of 21 genes using quantitative PCR (Applied Biosystems StepOne, Thermo Fisher Scientific, CA, USA). The genes were selected based on their stability as reference genes or their biological function, and their documented association with contaminant concentrations in previous studies on wildlife. Novel primers were designed based on highly conserved genetic regions in other mammals and optimised using PCR (Mastercycler X50s, Eppendorf, Hamburg, Germany), with assays run at different annealing temperatures. PCR products were initially controlled by using gel electrophoresis and later confirmed using Sanger sequencing. The results from the gene expression analysis are reported in **Paper IV**.

STATISTICAL ANALYSIS

All statistical analyses were performed using R version 4.2.2-4.3.0 (R Core Team, 2024). The full details on data handling are described in **Papers I-IV**. In brief, data were assessed for normality using the Shapiro-Wilk test, and equal variances were evaluated using Levene's test. Box-Cox transformations were applied in attempts to normalise variables. Still, non-parametric tests were primarily used to determine significance, as most data were non-normally distributed. Spearman's rank correlations were used to assess the relationship between variables in **Papers I-IV**. Significant differences in contaminant concentrations between groups are evaluated using Mann-Whitney or Kruskal-Wallis tests, followed by correction tests to limit false discovery rates, or a Student's t-test, depending on the normality of the data. Linear regression of raw or log-transformed data is used to visualise the relationship between Hg and selenium (Se) in faeces (**Paper I**) or between liver and faecal concentrations of Hg (**Paper II**).

Multivariate statistical analyses were applied to complete datasets, with missing values replaced by either the variable mean (for analytical errors) or a random value between zero and half the LOD (for concentrations below the LOD). Principal Component Analysis (PCA) was performed to investigate the possible relationship between variables and the spatial variation of the dataset. In **Paper I**, a PCA was used to evaluate the spatial differences in elemental concentrations between the two sampling sites. Orthogonal partial least squares (O-PLS) was performed in **Paper IV** to model the effects of contaminant concentrations and biometrics (predictor variables: age, body mass, Hg, Cd, Pb, and seven PFAS) on gene expression (response variables: gene expression of *JUN*, *FOS*, *CYP1A2*, *ECH1*, *FABP3*, *FADS1*, *FOXA2*, *GSR*, *HAMP*, *MET1A*, *NR0B2*, *POR*, *PPARG*, *SELENOP*, *SREBF1*, *THRA*, and

VLDLR). O-PLS is especially suitable for handling datasets with few observations and potential collinearity among explanatory variables (Sørmo et al., 2011).

SUMMARY OF MAIN FINDINGS

Paper I: Tracing the footprints of Arctic pollution: Spatial variations in toxic and essential elements in Svalbard reindeer (*Rangifer tarandus platyrhynchus*) faeces

With a limited number of contaminant studies in terrestrial wildlife on Svalbard, this study used Svalbard reindeer faeces as a non-invasive biomonitoring tool to investigate elemental exposure in reindeer, a key organism in the Svalbard terrestrial ecosystem.

The main objectives were to assess the elemental exposure in reindeer and to explore whether animals from remote areas had a different faecal elemental composition compared to those inhabiting areas close to human settlements. Faecal samples were collected from Adventdalen, close to the main settlement Longyearbyen, and the Nordenskiöld coast, 50 km away from Longyearbyen (Fig. 1).

The faecal elemental profile revealed that Svalbard reindeer are exposed to a range of elements, including the toxic metals Hg, Cd, and Pb. We also observed significant spatial differences in faecal elemental composition between the two studied subpopulations of reindeer. While reindeer faeces from Adventdalen was characterised by significantly higher concentrations of geogenic elements aluminium (Al), copper (Cu), iron (Fe), and nickel (Ni), reindeer faeces from Nordenskiöld coast were dominated by elements such as calcium (Ca), silicon (Si), and sulphur (S), indicating a marine influence on elemental exposure. In addition, faeces from the Nordenskiöld coast had significantly higher concentrations of both Cd and Pb.

Concentrations of Pb displayed site-specific patterns. In Adventdalen, Pb was strongly associated with other geogenic elements, suggesting a primarily geogenic origin of Pb. In contrast, the strong relationship between Pb and Hg in samples from the Nordenskiöld coast suggests a different, likely common source of origin at this site, such as local emissions from the coal-fired power plant in the Russian settlement of Barentsburg, located 15 km away.

Overall, this study highlights the spatial differences in elemental exposure in Svalbard reindeer and supports the use of faeces as a valuable non-invasive sample that can offer insight into spatial trends in contaminant exposure.

Paper II: Toxic metals and other elements in Svalbard reindeer: Establishing baselines and assessing non-invasive sampling for biomonitoring

The main objectives of this study were to determine the concentrations of Hg, Cd, and Pb in a range of Svalbard reindeer samples, including both invasive samples (e.g., kidney, liver or muscle) and non-invasive samples (e.g., fur and faeces), and to evaluate whether non-invasive samples could serve as reliable proxies for internal toxic metal concentrations, supporting less intrusive future biomonitoring of the Svalbard reindeer.

The highest concentrations of both Hg and Cd were found in the kidneys (0.68±0.31 µg/g (dw) and 16.3±11.2 µg/g (dw), respectively), followed by liver and faeces. In contrast, the highest concentrations of Pb were detected in faeces (2.62 ± 1.18 µg/g, dw), followed by liver and kidney. The molar ratio of Se and Hg was far above 1 in all samples, with average ratios of 18 in the kidney and 28 in the liver, indicating that there are sufficient levels of Se to counter the potential toxicity of Hg. A between-year difference in Hg concentrations was observed in faeces (0.06 µg/g versus 0.10 µg/g) and fur (9.22 ng/g versus 18.7 ng/g). A similar trend was, however, not observed for soft tissue Hg concentrations, which appeared stable across both sampling years (2021 and 2022).

There was a substantial covariation in metal concentrations among sample types, with strong positive correlations observed for Hg between faeces and liver, fur and muscle, and blood and liver, and for Cd between fur and kidney (Fig. 3). These findings suggest that several non-destructive and non-invasive sample types may be suitable for predicting internal concentrations of toxic metals in Svalbard reindeer. However, a strong covariation among geogenic elements (i.e., Pb, Al, Fe, and Si concentrations) in the fur samples suggests external contamination, which indicates that the cleaning procedure performed before the digestion and elemental analysis may have been insufficient. As external contamination may have confounded the results, it is difficult to assess the reliability of fur as a suitable non-invasive sample used in biomonitoring. A similar relationship among Pb, Al, and Fe concentrations in faeces suggests that the reindeer ingest soil particles during grazing, which increases their exposure to soil-derived contaminants, such as Pb.
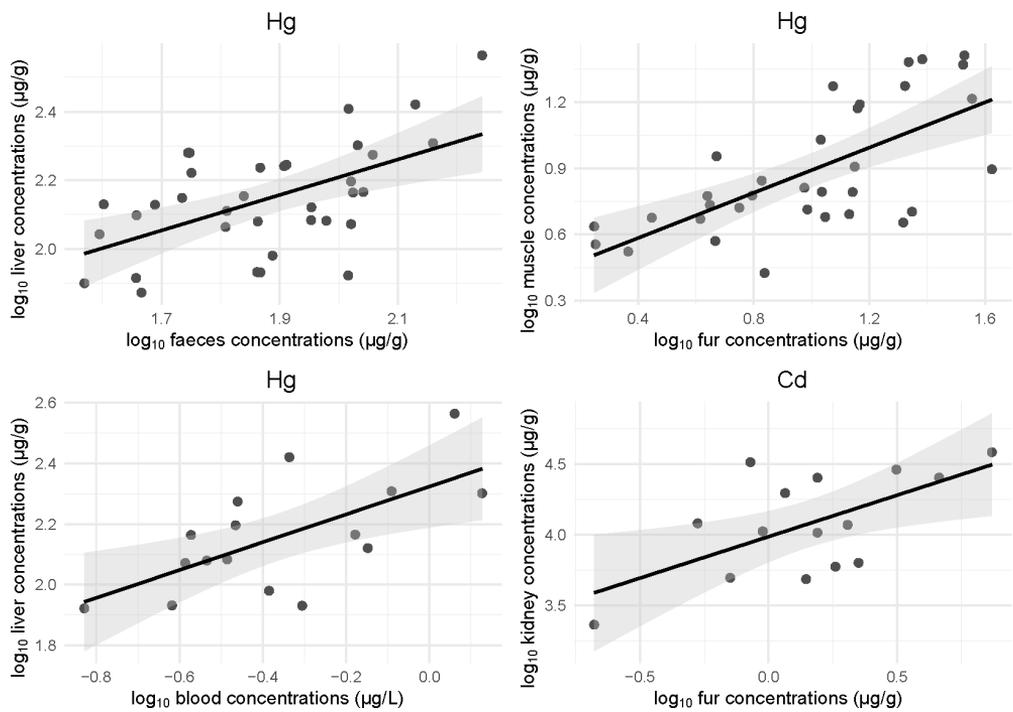
Figure 3. Correlation plots of significant (p<0.05) relationships between concentrations of Hg in faeces-liver, fur-muscle, and blood-liver, and Cd in fur-kidney of the Svalbard reindeer (based on data from **Paper II**).

Paper III: Seasonal Shift in Exposure and Accumulation of PFAS and Heavy Metals in High Arctic Reindeer

Malin Andersson Stavridis, Tove Petersson, Görkem Deniz Kendir, Shannen Sait, Øyvind Mikkelsen, Vebjørn Veiberg, Tomasz Maciej Ciesielski & Bjørn Munro Jenssen. In review.

The Svalbard reindeer undergoes annual cycles of fasting and fattening, driven by the seasonal availability of forage. By comparing the contaminant concentrations and profiles of Svalbard reindeer culled in late summer (August) and autumn (October), we investigated whether there are seasonal fluctuations in contaminant concentrations.

Levels of Hg, Cd, and several PFAS were higher in samples collected in October compared to August (Figs. 4-6). Hepatic concentrations of Hg increased from 11.1 ng/g in August to 21.5-42.6 ng/g in October, while Cd concentrations more than tripled (178 ng/g and 555 ng/g in August and October, respectively). Out of the 41 target PFAS, only thirteen were detected above the LOD in at least 25 % of the samples. These include PFUnDA, PFTriDA, PFTDA, PFHxDA, PFHxS, PFOS, PFNS, PFDS, PFECHS, FOSAA, MeFOSAA, EtFOSAA, and NaDONA. Following the same trend as Hg and Cd, hepatic ∑13PFAS concentrations were lower in August (18.8±13.0 ng/g) compared to in October (55.8±15.4 ng/g).

In addition to higher PFAS concentrations in the samples from October compared to August, there was a significant shift in the PFAS profile of the muscle samples. While August samples were characterised by high concentrations of PFOS and strong intercorrelations between PFOS and precursor PFAS, muscle samples from October were characterised by higher concentrations of PFCAs (PFUnDA and PFTriDA) and strong intercorrelations among them. Although very different, both profiles suggest that the reindeer are predominantly exposed to long-range transported PFAS.

Compared to previous studies on Svalbard reindeer, the result indicates that levels of PFAS have significantly increased during the past decade (since 2010). In contrast, concentrations of Cd and Pb have decreased over the past forty years (since the 1980s). Although the concentrations of PFAS, Hg, Cd, and Pb were below toxicity threshold levels, the hepatic concentrations of both ∑PFAS and Cd, especially in individuals culled in autumn, are high enough to warrant caution regarding human consumption.
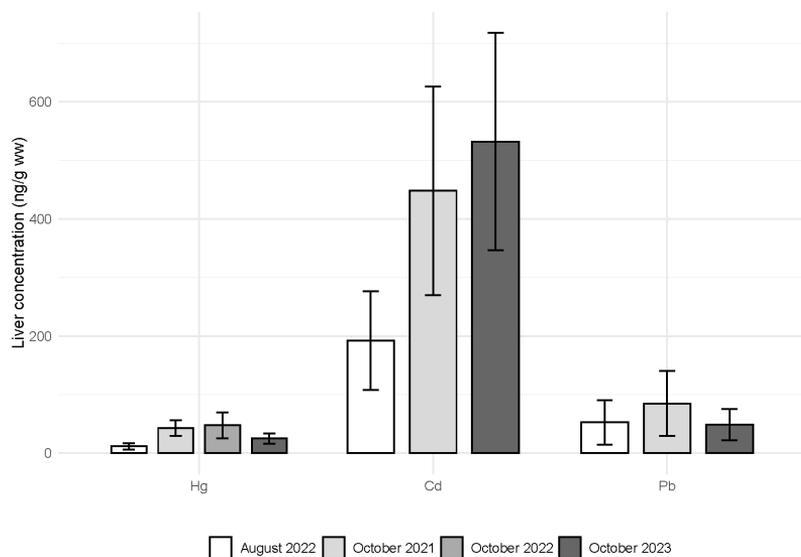
Figure 4. Bar plots showing average concentrations of Hg, Cd, and Pb in Svalbard reindeer liver across different sampling points (August 2022 and October 2021-2023). Error bars represent standard deviation. Data from October 2021 and 2022 are based on results from **Paper II**, while data from August 2022 and October 2023 were derived from **Paper III**.
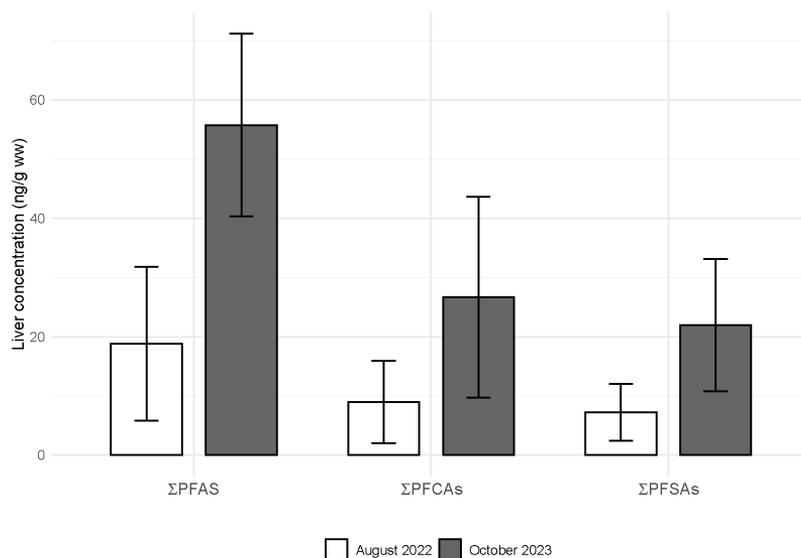
Figure 5. Bar plots showing the average ΣPFAS, ΣPFCAs, and ΣPFSAs in Svalbard reindeer liver between August 2022 and October 2023. Error bars represent standard deviation. Data is based on results from **Paper III**.
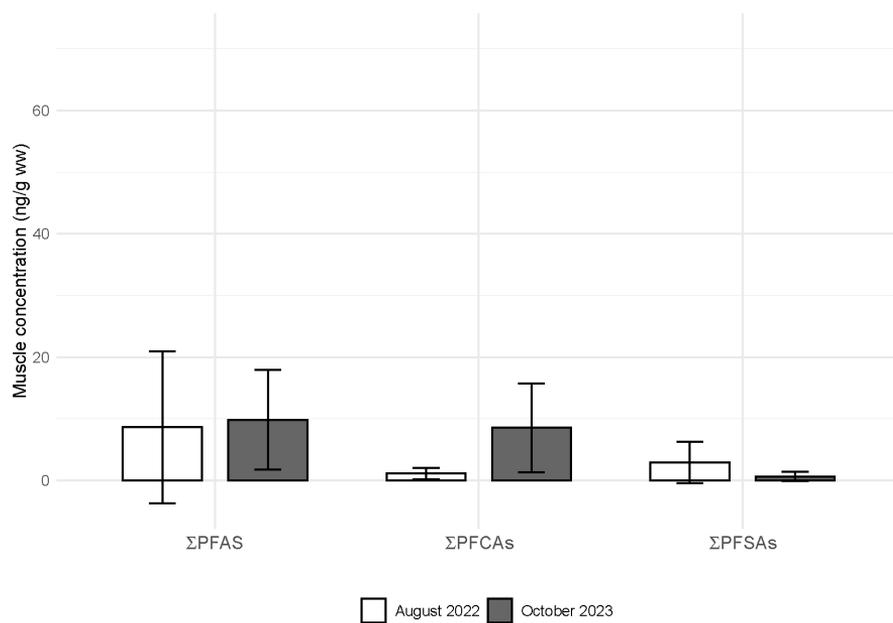
Figure 6. Bar plots showing the average ΣPFAS, ΣPFCAs, and ΣPFSAs in Svalbard reindeer muscle between August 2022 and October 2023. Error bars represent standard deviation. Data is based on results from **Paper III**.

Paper IV: Low-Level Chronic Exposure to PFAS and Toxic Metals Induces Transcriptional Changes in Svalbard Reindeer

Malin Andersson Stavridis, Tove Petersson, Valentin Garreau, Øyvind Mikkelsen, Vebjørn Veiberg, Helena Reinardy, Bjørn Munro Jenssen & Tomasz Maciej Ciesielski.

Svalbard reindeer are exposed to Hg, Cd, and various PFAS at concentrations far below the established toxicity thresholds. However, chronic exposure to low levels of contaminants may still cause biological effects. To explore this, we examined the mRNA expression of 17 genes previously linked to contaminant exposure in studies on other mammals. We compared gene expression with contaminant concentrations and biometric variables: body mass and age, to identify any potential associations among them.

Only a few direct relationships were observed between individual contaminants and specific target gene expression. Instead, multivariate statistical models revealed the expression of several target genes to be associated with both biometric variables and combinations of contaminants. More specifically, body mass and weight were consistently associated with an upregulation of target gene expression, while increasing concentrations of C6 perfluorosulfonic acid (PFHxS), Cd, and Hg primarily were associated with a decreased transcriptional activity (Fig. 7).

Among all target genes, significant multivariate models were mainly achieved for genes involved in lipid homeostasis (i.e., *FABP3*, *FOXA2*, *NR0B2*, *PPARG*, *SREBF1,* and *THRA*), which is concerning given that lipid homeostasis is critical for Arctic wildlife like Svalbard reindeer, which undergo annual fattening and fasting cycles and therefore rely on a functioning lipid metabolism. Multivariate models were also achieved for genes involved in other key physiological functions, such as xenobiotic metabolism (*CYP1A2*) and cell proliferation and apoptosis (*JUN*).

Overall, our findings suggest that chronic, low-level contaminant exposure is influencing molecular pathways in Svalbard reindeer, despite individual contaminant concentrations being below the current toxicity thresholds.
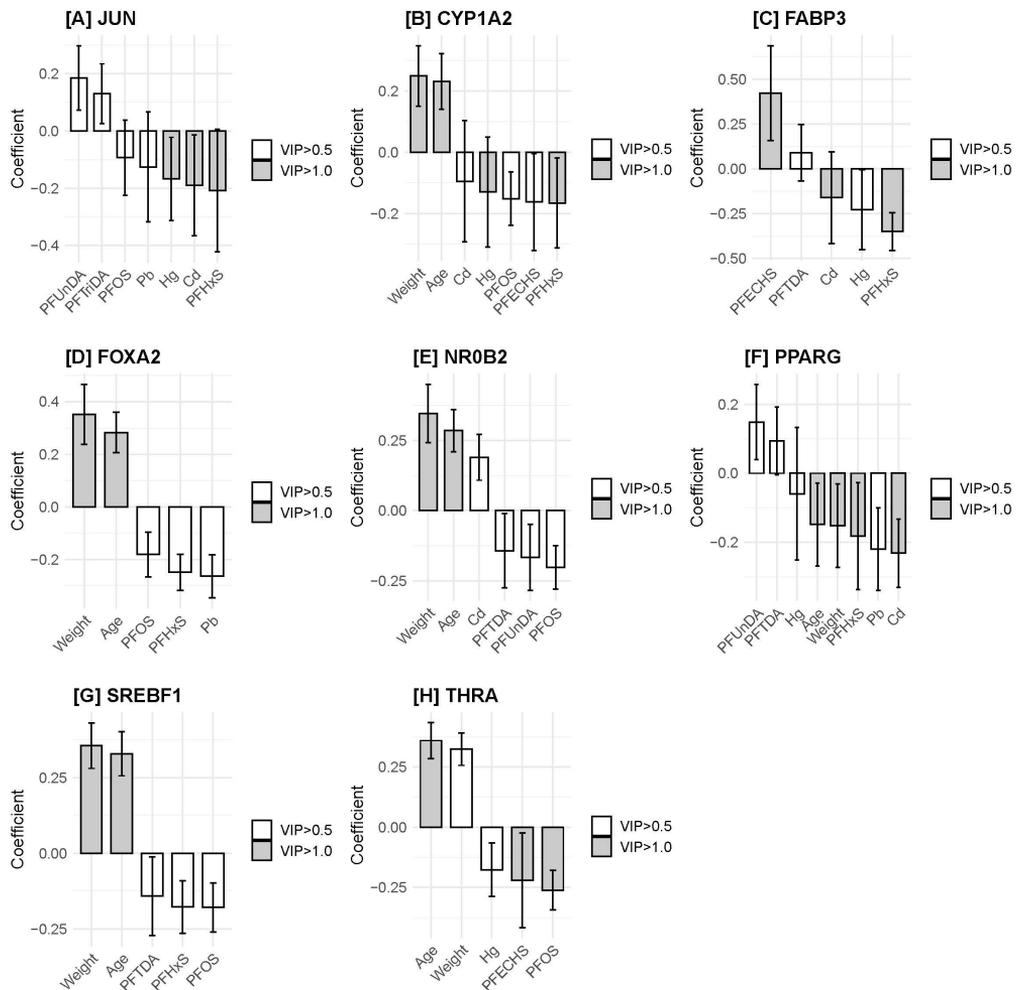
Figure 7. O-PLS regression coefficient for genes [A] *JUN*, [B] *CYP1A2*, [C] *FABP3*, [D] *FOXA2*, [E] *NR0B2*, [F] *PPARG*, [G] *SREBF1*, and [H] *THRA* with hepatic mRNA expression as a response to predictor variables (contaminant concentrations and biometrics). The bars represent the contribution (coefficient ± standard deviation) of each predictor variable to the model. Predictor variables with a greater contribution to the model have variable importance in projection (VIP) scores above 1 and are marked in grey (from **Paper IV**).

RESULTS AND DISCUSSION

CONTAMINANT CONCENTRATIONS IN THE SVALBARD REINDEER

Among the toxic metals studied, the highest concentrations of both Hg and Cd were found in the kidney, followed by the liver. Renal concentrations of Hg and Cd ranged between 0.03-0.29 µg/g (ww) and 0.45-7.68 µg/g (ww), respectively. In contrast, the highest concentration of Pb was found in faeces, while the highest internal concentration was obtained in liver (0.02-0.26 µg/g ww) (**Paper II**). Similarly, between muscle and liver, the highest concentrations of PFAS were observed in the liver, with the highest concentrations obtained for PFHxS >PFUnDA >PFTriDA >PFOS (**Paper III**).

Compared to other reindeer and caribou across the Arctic, concentrations of Hg and Pb in Svalbard reindeer were low, whereas Cd concentrations were similar to those previously reported in other subspecies (Table 3). A possible explanation for these spatial differences in metal concentrations among *Rangifer* subspecies may be diet, which is a known driver of contaminant exposure in various Arctic and sub-Arctic species, including marine mammals (e.g., McKinney et al., 2013; Remili et al., 2023), Arctic fox (Routti et al., 2017), ptarmigan (*Lagopus* spp.; Myklebust et al., 1993), and barren-ground caribou (*Rangifer tarandus groenlandicus*) (Gamberg et al., 2020).

For Arctic cervids, Gamberg et al. (2005) argue that different proportions of lichen and willows in the diet significantly drive the differences in tissue concentrations of both Hg and Cd. While lichen makes up a substantial part of the diet in many other *Rangifer* subspecies (Gamberg et al., 2020), studies on Svalbard reindeer rumen contents indicate a relatively low lichen intake (Bjørkvoll et al., 2009; Staaland, 1986). As lichen are efficient accumulators of atmospheric Hg (Bargagli, 2016), a smaller dietary fraction of lichen, and thus a reduced dietary exposure to Hg, likely contributes to their lower Hg concentrations compared to other subspecies. Instead, a consistent part of the Svalbard reindeer diet is *S. polaris* (Bjørkvoll et al., 2009), a known phytoaccumulator of Cd (Wojtuń et al., 2013). Notably, the proportion of *S. polaris* in the diet of other herbivores (i.e., ptarmigans) was strongly linked to internal tissue concentrations of Cd (Myklebust et al., 1993). Given that willows play a role in the diet of various *Rangifer* subspecies across the Arctic, similar Cd concentrations in different populations may be explained through willow intake.

As summarised in Table 3, reported Pb concentrations in reindeer and caribou varied widely, likely as a result of temporal and spatial differences across studies. The global use of leaded gasoline during the 1900s led to the enrichment of Pb in soils worldwide (Chen et al., 2025), reaching even the Arctic cryosphere (Boutron et al., 1995). While vegetation may have a limited uptake of Pb from the surrounding soil (Wojtuń et al., 2019, 2013), both reindeer and caribou are known to ingest soil during foraging (Gamberg et al., 2016; Reimers, 1983; **Paper I**; **Paper II**), increasing their dietary exposure to soil-derived Pb. As such, wildlife across the Arctic is exposed to Pb, regardless of location. However, proximity to point sources, such as coal power plants, may increase Pb concentrations in the local environment significantly (Chen et al., 2025; Steinnes, 1995), contributing to different exposures across the Arctic.

Table 3. A circumpolar overview of previous studies reporting toxic metal concentrations in kidney, liver, and muscle samples from the *Rangifer* family. All concentrations presented are derived means from the reported concentrations in the studies. Concentrations reported from **Paper II** are marked in bold. All concentrations are reported in µg/g wet weight.

	Tissue	n	Conc. in µg/g	Location	Year	Reference
Hg	**Kidney**	**36**	**0.14**	**Svalbard**	**2021-2022**	**Paper II**
	Kidney	315	0.51	Russia	2015-2020	Makarov et al., 2022
	Kidney	26	0.04	Canada	2010-2013	Larter et al., 2016
	Kidney	81	0.38 [a]	Greenland	2008-2009	Gamberg et al., 2016
	Kidney	63	0.36	Canada	1994-2006	Schuster et al., 2011
	Kidney	906	1.04 [a]	Canada	1991-2016	Gamberg et al., 2020
	Kidney	76	0.39 [a]	Greenland	1991-2016	Gamberg et al., 2020
	Liver	**36**	**0.04**	**Svalbard**	**2021-2022**	**Paper II**
	Liver	505	0.15	Russia	2015-2020	Makarov et al., 2022
	Liver	80	0.07 [a]	Greenland	2008-2009	Gamberg et al., 2016
	Liver	63	0.12	Canada	1994-2006	Schuster et al., 2011
	Muscle	**36**	**0.0027**	**Svalbard**	**2021-2021**	**Paper II**
	Muscle	10	0.001	Canada	2010-2013	Larter et al., 2016
	Muscle	40	0.01 [a]	Greenland	2008-2009	Gamberg et al., 2016
	Muscle	230	0.001	Sweden	1983-2003	Odsjö et al., 2005
Cd	**Kidney**	**18**	**3.31**	**Svalbard**	**2021-2021**	**Paper II**
	Kidney	315	3.01	Russia	2015-2020	Makarov et al., 2022
	Kidney	26	13.9	Canada	2010-2013	Larter et al., 2016
	Kidney	81	2.76 [a]	Greenland	2008-2009	Gamberg et al., 2016
	Kidney	906	5.88	Canada	1991-2016	Gamberg et al., 2020
	Kidney	76	2.23	Greenland	1991-2016	Gamberg et al., 2020
	Kidney	60	4.3	Svalbard	1984-1986	Borch-Iohnsen et al., 1996
	Liver	**18**	**0.45**	**Svalbard**	**2021-2021**	**Paper II**
	Liver	505	0.61	Russia	2015-2020	Makarov et al., 2022
	Liver	80	0.50 [a]	Greenland	2008-2009	Gamberg et al., 2016
	Liver	78	0.73	Svalbard	1984-1986	Borch-Iohnsen et al., 1996
	Muscle	**18**	**0.0014**	**Svalbard**	**2021-2021**	**Paper II**
	Muscle	22	0.003	Russia	2015-2020	Makarov et al., 2022
	Muscle	10	0.20	Canada	2010-2013	Larter et al., 2016
	Muscle	40	0.002 [a]	Greenland	2008-2009	Gamberg et al., 2016
	Kidney	**18**	**0.04**	**Svalbard**	**2021-2021**	**Paper II**
	Kidney	26	0.02	Canada	2010-2013	Larter et al., 2016
	Kidney	81	0.07 [a]	Greenland	2008-2009	Gamberg et al., 2016

	Tissue	n	Conc. in µg/g	Location	Year	Reference
	Kidney	64	2.61	Alaska	1994-1996	O'Hara et al., 2003
	Kidney	57	0.80	Svalbard	1984-1986	Borch-Iohnsen et al., 1996
	Liver	**18**	**0.08**	**Svalbard**	**2021-2021**	**Paper II**
	Liver	80	0.32 [a]	Greenland	2008-2009	Gamberg et al., 2016
Pb	Liver	64	1.09	Alaska	1994-1996	O'Hara et al., 2003
	Liver	78	1.30	Svalbard	1984-1986	Borch-Iohnsen et al., 1996
	Muscle	**18**	**0.0008**	**Svalbard**	**2021-2021**	**Paper II**
	Muscle	10	0.02	Canada	2010-2013	Larter et al., 2016
	Muscle	40	0.001 [a]	Greenland	2008-2009	Gamberg et al., 2016
	Muscle	64	0.47	Alaska	1994-1996	O'Hara et al., 2003

In contrast to both Hg and Pb, concentrations of PFAS are higher in Svalbard reindeer than in many of the previous studies reporting concentrations in various subspecies of Arctic *Rangifer* (Table 4). Similar PFAS concentrations to those presented in this thesis have been reported in reindeer and caribou from Greenland, where the proximity to direct point sources such as firefighting training sites was suggested as a primary source of exposure (Roos et al., 2021). This may partly explain the high concentrations of PFOS and PFHxS detected in Svalbard reindeer in **Paper III**.

On Svalbard, the Longyearbyen firefighting training site has been pointed out as a major source of PFAS into the local environment, significantly increasing the levels of, for example, PFHxS and PFOS, in the nearby marine and freshwater environment (Ali et al., 2021). Moreover, historical usage of AFFFs in sites such as the former mining town Svea (permanently ended operations in 2015) may now act as point sources of legacy contamination into the environment (Evenset et al., 2020). However, PFAS are not restricted to areas near these local sources but have been reported in both biotic and abiotic matrices all over the archipelago. While PFSAs (e.g., PFOS or PFHxS) are dominant in areas close to settlements (Ahrens et al., 2023), the PFAS profile in freshwater, sediments, and soil samples from further inland, or more remote coastal areas, is characterised by higher levels of PFCAs (e.g., PFUnDA or PFTriDA), which has been attributed to LRAT (Ahrens et al., 2023; Hartz et al., 2023). Traces of both PFAS precursors and PFCAs were found throughout a glacial ice core from Lomonosovfonna, central Spitsbergen, indicating continuous long-range transport of atmospheric precursors that degrade into PFCAs, which are subsequently stored in the glaciers (Hartz et al., 2023). Furthermore, glacial meltwater has been reported to have high concentrations of PFCAs, demonstrating that the legacy PFAS stored in glaciers remobilise with the annual thaw (Ahrens et al., 2023). With PFCAs present in meltwater, it becomes bioavailable to vegetation and wildlife, which may help explain the presence of PFCAs in the tissues of the Svalbard reindeer.

Table 4. A circumpolar overview of previous studies reporting PFAS concentrations in liver samples from the *Rangifer tarandus* family. Mean concentrations from **Paper III** are included in bold (Aug-Oct), and all other presented concentrations are derived means from the reported concentrations in Roos et al. (2021). All concentrations are reported in ng/g wet weight.

	n	Conc. in ng/g	Location	Year
PFHxS	**21**	**5.35-21.6**	**Svalbard**	**2022-2023**
	146	0.02	Canada	2002-2017
	30	0.43	Greenland	2008-2012
	7	0.43	Svalbard	2010
	60	0.07	Sweden	2002-2011
PFOS	**21**	**3.83-5.28**	**Svalbard**	**2022-2023**
	146	0.94	Canada	2002-2017
	30	10.4	Greenland	2008-2012
	7	0.60	Svalbard	2010
	60	5.86	Sweden	2002-2011
PFUnDA	**21**	**4.47-14.4**	**Svalbard**	**2022-2023**
	146	1.34	Canada	2002-2017
	30	5.10	Greenland	2008-2012
	7	0.17	Svalbard	2010
	60	1.41	Sweden	2022-2011
PFTriDA	**21**	**4.17-11.0**	**Svalbard**	**2022-2023**
	146	0.26	Canada	2002-2017
	30	0.50	Greenland	2008-2012
	7	0.22	Svalbard [a]	2010
	60	0.27	Sweden	2002-2011

LONG-RANGE AND LOCAL SOURCES OF PFAS

As above-mentioned, the PFAS fingerprint in the Svalbard reindeer was dominated by PFOS, PFHxS, PFUnDA, and PFTriDA, which indicates that the reindeer likely are exposed to a combination of locally emitted and long-range transported PFAS (**Paper III**).

Both PFOS and PFHxS are associated with the use of AFFFs around Svalbard settlements (Ali et al., 2021; Evenset et al., 2020). Thus, the elevated concentrations of PFOS and PFHxS in Svalbard reindeer may reflect exposure to locally emitted PFAS. However, PFOS concentrations in the reindeer tissues were strongly correlated with those of the PFAS precursors FOSAA and EtFOSAA (ρ=0.6-0.9, p<0.05). Given that both FOSAA and EtFOSAA are transported to the Arctic via LRAT (Kwok et al., 2013; Muir et al., 2025), and relationships between these precursors and PFOS have been reported in environmental samples from Svalbard (Hartz et al., 2023), the PFOS detected in Svalbard reindeer may partly reflect precursor degradation following long-range transport. Another plausible explanation for this

relationship and the high PFOS concentrations in the reindeer is exposure to precursor PFAS alone, as both FOSAA and EtFOSAA are known to metabolise into PFOS in biota (Martin et al., 2010), and these precursors have been suggested as an explanation behind high concentrations of PFOS in other Svalbard wildlife (Ali et al., 2021).

Similarly, the presence of PFCAs in Svalbard reindeer is also consistent with exposure to emissions from remote sources, as these PFAS are most frequently detected in areas at high latitudes (e.g., glaciers) or far away from local settlements on Svalbard (Ahrens et al., 2023; Ali et al., 2021; Hartz et al., 2023). Taken together, the observed PFAS profile in Svalbard reindeer tissues appears to mainly be driven by LRAT and the transformation of precursor compounds, rather than by local sources.

That the PFAS in Svalbard reindeer would originate from sources outside of the Arctic contrasts the findings of Roos et al. (2021), where similar ΣPFAS concentrations, but different PFAS profiles, in Greenlandic reindeer and caribou were attributed to proximity to local communities and anthropogenic activities. Our results further contrast the findings of Roos et al. (2021) regarding concentrations in Svalbard reindeer, with the present thesis not only reporting overall higher concentrations of PFAS, but also a different PFAS profile. Roos et al. (2021) report a PFAS fingerprint dominated by PFHxS, PFOS, and PFNA (0.43 ng/g, 0.60 ng/g, and 0.66 ng/g, respectively) in Svalbard reindeer liver sampled during the hunting season (mid-August to the end of September) in 2010. In **Paper III**, the PFAS profile of Svalbard reindeer is still partly dominated by PFHxS and PFOS, but instead of PFNA, we find other longer-chained PFCAs such as PFUnDA and PFTriDA. In addition, concentrations are far higher for both PFSAs and PFCAs. For example, hepatic PFOS concentrations were 3.83 ng/g in August 2022 and 5.28 ng/g in October 2023, compared to 0.60 ng/g in 2010, demonstrating a significant increase in exposure to PFAS over the past decade.

A possible explanation for the changes in hepatic contaminant concentrations in Svalbard reindeer since 2010 may be linked to the accelerated thawing of the Svalbard cryosphere and the subsequent remobilisation of legacy contaminants that were previously stored in ice, which would result in an altered contaminant exposure (Kwok et al., 2013; MacInnis et al., 2022; Xue et al., 2025). Since the 2010s, Svalbard has experienced a rapid melting of glaciers (Spolaor et al., 2024), with the total glacial melt of the 2024 summer being comparable to the melt of the entire Greenland ice sheet (Schuler et al., 2025), and glacial meltwater concentrations of PFCAs in the Longyear river nearly doubling from 2006 to 2021 (Ali et al., 2021). It is therefore not unlikely that exposure to legacy contaminants has changed over the last decade, which may explain both the shift in PFAS profile and increased PFAS concentrations in the Svalbard reindeer today.

LONG-RANGE OR LOCAL SOURCES OF TOXIC METALS
While long-range oceanic or atmospheric transport of contaminants from lower latitudes dominates the contribution of toxic metals such as Hg. Cd and Pb into the Arctic environment (Rudnicka-Kępa and Zaborska, 2021), there are also local sources of emissions in the Arctic (e.g., Beitveit, 2016; Rose et al., 2004; Zaborska et al., 2017). This was reflected

in the results of **Paper I**, which suggests that the Svalbard reindeer are exposed to a combination of metals deriving from both local and long-range sources.

Concentrations of Hg were similar in Svalbard reindeer faeces from Adventdalen (0.08 µg/g), Nordenskiöld coast (0.09 µg/g) (**Paper I**), Reindalen (0.08 µg/g; **Paper II**), and around Recherchefjorden (0.07-0.100 µg/g; Pacyna et al., 2019). Modelled atmospheric mercury deposition across Svalbard is relatively homogenous (Dastoor et al., 2022), which may explain the similar exposure of Hg across the different populations of Svalbard reindeer. In contrast to Hg, concentrations of both Cd and Pb significantly differed among reindeer in Adventdalen, Nordenskiöld coast, and Reindalen (**Papers I and II**). The faecal concentrations of Cd were the highest in samples from Reindalen (0.74 µg/g), followed by the Nordenskiöld coast (0.55 µg/g) and Adventdalen (0.37 µg/g). In contrast, the Pb concentrations were the highest in at Nordenskiöld coast (5.36 µg/g) followed by Adventdalen (3.81 µg/g) and Reindalen (2.62 µg/g).

In both Reindalen and Adventdalen, the relatively low concentrations of Pb were strongly positively related to elements of geogenic origin (e.g., Al and Fe). Both of these valleys share similar lithogenic characteristics and are predominantly composed of sandstone, whereas the Nordenskiöld coast has a distinctively different profile dominated by shale and siltstone (Norwegian Polar Institute, 2016). Given how cryoturbation contributes to mixing the mineral and organic layer of soil on Svalbard (Halbach et al., 2017), local differences in bedrock composition likely help explain the differences in faecal Pb concentrations across locations. This is especially relevant considering the high rates of soil ingestion by Svalbard reindeer during foraging (Reimers, 1983).

The significantly higher concentrations of Pb in faeces from the Nordenskiöld coast were strongly, positively correlated with Hg concentrations, suggesting either a common origin or similar environmental behaviour. With a higher annual precipitation at Nordenskiöld coast compared to Adventdalen (Norsk Klimaservicesenter, n.d.), a higher rate of wet deposition of metals is likely, which may contribute to the increased concentrations of Pb in the soil and subsequently in the diet of the reindeer. Furthermore, the proximity to Barentsburg, a Russian settlement located less than 20 km from the Nordenskiöld coast, may contribute to the elevated Pb concentrations observed, potentially due to emissions from its coal-fired power plant.

Faecal Cd concentrations were significantly higher in faeces from the Nordenskiöld coast relative to Adventdalen (**Paper I**), despite previous studies linking levels of Cd in Adventdalen with local, anthropogenic emissions (e.g., Beitveit, 2016). This may, similarly to Pb, be explained by higher levels of precipitation, thus wet deposition, or proximity to Barentsburg. However, the highest Cd concentrations were obtained in faeces from Reindalen, which is the most remote out of all three sampling locations (Adventdalen, Nordenskiöld coast, and Reindalen; Fig. 1), suggesting that there are other factors influencing Cd exposure. A possible explanation is differences in the dietary fraction of *S.polaris* across reindeer populations, as Svalbard reindeer prioritise forage quantity over quality (Van Der Wal et al., 2000). Consequently, the composition of the diet is shaped by local vegetation communities

within the different valley systems, which may lead to slight differences in *S. polaris* ingestion and thus Cd exposure.

While the results from **Paper I** demonstrate that present-day Svalbard reindeer are exposed to Cd and Pb, **Paper II** reports significantly lower soft tissue concentrations of both Cd and Pb in reindeer today (2020s), as compared to in the mid-1980s (Borch-Iohnsen et al., 1996). With better emission control techniques and the phase-out of leaded gasoline in the last century, the environmental emissions of both Cd and Pb have dramatically decreased (Pacyna et al., 2009). This is reflected in the atmospheric measurements of these metals, and on Svalbard, atmospheric concentrations of both Cd and Pb are decreasing by approximately $2.8\%y^{-1}$ and $4.6\%y^{-1}$, respectively (Platt et al., 2022). Although the melting of the Arctic cryosphere is known to remobilise legacy contaminants, including metals such as Cd and Pb (Xue et al., 2025), the additional exposure resulting from this does not appear sufficient to compensate for the overall decline in environmental concentrations driven by reduced emissions, and subsequently decreased atmospheric input to Svalbard.

Similarly to Cd and Pb, the atmospheric concentrations of Hg on Svalbard are decreasing, but at a considerably slower rate ($-0.57\%y^{-1}$) (MacSween et al., 2022). This is likely due to the non-uniform global reductions in Hg emissions, with decreases in Europe and the Americas offset by increases in Asia and other regions (Obrist et al., 2018; Pacyna et al., 2006). Recent studies on Arctic fox on Svalbard even report that Hg concentrations have been increasing over the past decades (Hallanger et al., 2019). If a similar pattern exists for the Svalbard reindeer remains unknown, as no previous study reports soft tissue concentrations of Hg in these reindeer. Monitoring Hg concentrations in Arctic terrestrial wildlife is especially valuable considering the ongoing thaw of the Arctic cryosphere, where stored Hg may be released into the atmosphere, nearby bodies of water, or taken up by vegetation (Schaefer et al., 2020), thus increasing its bioavailability and subsequently its exposure to biota. Given the unknown future biogeochemical fate of Hg in the Svalbard terrestrial environment, a continued monitoring of Hg in wildlife is warranted, where the data reported in this thesis provide an important temporal baseline to which future assessments in Svalbard reindeer can be compared.

USING FAECES AND FUR FOR METAL BIOMONITORING

In **Paper II**, strong positive correlations were found between faeces and liver ($R^2=0.33$, $p<0.001$), fur and muscle ($R^2=0.46$, $p<0.001$), and blood and liver ($R^2=0.43$, $p<0.01$) for Hg, and between fur and kidney ($R^2=0.38$, $p=0.015$), for Cd. Positive relationships between heavy metal concentrations in different tissues have been reported across a range of mammals. For example, Hg concentrations in hair are positively associated with those in soft tissues in both Arctic fox (Treu et al., 2018) and polar bear (Bechshoft et al., 2019), and with blood in humans (Phelps et al., 1980). Similar associations have been reported for Cd between hair and soft tissues in grey wolves (*Canis lupus;* Hernández-Moreno et al., 2013) and rabbits (*Oryctolagus cuniculus;* Gil-Jiménez et al., 2020), and for a range of metals and POPs between feathers and soft tissues in birds (Jaspers et al., 2019).

While hair or feathers are common non-invasive matrices for assessing contaminant concentrations in wildlife (e.g., Bechshoft et al., 2019; Jaspers et al., 2019), faeces has mainly been used to monitor exposure to contaminants (e.g., Pacyna et al., 2019; Węgrzyn et al., 2018), and have rarely been directly linked to soft tissue concentrations (Jota Baptista et al., 2022). As such, the significant relationship in Hg concentrations between faeces and liver in the Svalbard reindeer was unexpected.

Faecal contaminant concentrations mainly reflect the contaminants present in the diet of grazing animals (Roggeman et al., 2013). Consequently, the faecal Hg concentrations in Svalbard reindeer likely mirror their diet and would vary with seasonal dietary changes (Bjørkvoll et al., 2009; Staaland, 1986). This is supported by the findings of **Papers I, II,** and the unpublished manuscript (**Paper V**) by Andersson Stavridis et al. (2025), which together demonstrate significant seasonal fluctuations in faecal Hg concentrations, with higher concentrations found in faeces from late summer and autumn, relative to early summer and spring. Similarly, the hepatic concentrations of Hg in Svalbard reindeer also fluctuate with season, with **Paper III** demonstrating an increase from late summer to autumn, likely also mirroring an increased dietary exposure. The dietary influence on Hg concentrations in both faeces and liver helps explain the strong positive correlations that were found between these matrices (R2=0.33, p<0.001) in **Paper II**. Given this relationship, faecal Hg may serve as a useful proxy for hepatic Hg, and as such, a non-invasive method of assessing Hg in Svalbard reindeer soft tissues. Especially when considering the feasibility of faecal sampling relative to the sampling of internal tissues (Jota Baptista et al., 2022).

Concentrations of Hg and Cd in Svalbard reindeer fur had strong associations with concentrations in muscle and kidney, respectively (**Paper II**), indicating fur as another suitable non-invasive matrix for biomonitoring. However, the elemental composition of the fur revealed strong correlations between geogenic elements Pb, Al, Fe, and Si (Halbach et al., 2017), suggesting that the hair may not only reflect excreted levels of contaminants, but also external contamination. Both Hg and Cd are present in Svalbard soils (Gopikrishna et al., 2020; Krajcarová et al., 2016; Wojtuń et al., 2013), and the Svalbard reindeer allocate a substantial part of their daily time budget to resting and ruminating while lying down on the ground (Tyler, 1987), exposing their fur to soil and geogenic elements. Despite us following established procedures for cleaning the fur samples prior to digestion and elemental analysis (e.g., Dolan et al., 2017), the results from **Paper II** suggest that the cleaning methods were insufficient, which is a common case when studying contaminants in samples like fur or feathers (Gil-Jiménez et al., 2020; Løseth et al., 2019; Tajchman et al., 2023), which are in direct contact with external contaminant sources. As we cannot differentiate between internal and external contaminants in the Svalbard reindeer fur used in the study, any relationship between contaminant concentrations in fur and internal tissues may be confounded, and as such, no conclusions can be drawn on the suitability of using fur to monitor levels of Hg and Cd in soft tissues of Svalbard reindeer.

In addition, the seasonal changes in soft tissue contaminant concentrations reported in **Paper III** may introduce further issues with the usage of hair. Moulting in the Svalbard reindeer occurs annually in spring, with new winter fur being formed during late summer

(Cuyler and Øritsland, 2002). As fur is formed in close contact with the bloodstream, contaminant concentrations in fur will mirror those of the blood at the time of formation (Wang et al., 2014), which in turn depends on the current exposure in the reindeer. Therefore, the strong associations between fur and soft tissue contaminant concentrations in **Paper II** may not be consistent throughout the year, as internal tissue concentrations appear to fluctuate (**Paper III**), potentially affecting the relationship between tissues. Therefore, samples collected at different times of the year are required to determine whether relationships between contaminant concentrations in fur and internal tissues are constant across seasons.

BIOLOGICAL EFFECTS OF CHRONIC, LOW-LEVEL EXPOSURE TO CONTAMINANTS
Contaminant concentrations reported in the Svalbard reindeer consistently fall below the current toxicity thresholds for wildlife. Hepatic PFOS concentrations above 655 ng/g (ww) have been associated with reproductive, immune, and carcinogenic effects in polar bears (Dietz et al., 2018b). In comparison, the hepatic Σ_{13}PFAS reported in the most contaminated individual Svalbard reindeer (**Paper III**) is far lower (78.4 ng/g ww). Similarly, the hepatic concentrations of Hg (0.02-0.10 µg/g ww), and renal concentrations of Cd (0.45-7.68 µg/g ww) and Pb (0.02-0.11 µg/g ww) in Svalbard reindeer (**Paper II**), are also substantially lower than their respective toxicity thresholds for wildlife (Hg: 4.2 µg/g ww; Dietz et al., 2022, Cd: 100 µg/g ww; Larison et al., 2000, Pb: 15 µg/g dw; Ma, 2011). Despite this, the results in **Paper IV** suggest that the exposure to a combination of different PFAS and metals together causes biological changes on a molecular level in Svalbard reindeer.

In **Paper IV**, increasing concentrations of contaminants (primarily PFHxS, Hg, and Cd) were associated with the downregulation of genes related to lipid metabolism (*FABP3*, *PPARG*, *THRA*), xenobiotic metabolism (*CYP1A2*), and cell regulation (*JUN*), with genes related to lipid metabolism being most consistently affected. Similar to many other species of Arctic mammals, Svalbard reindeer heavily depend on normal lipid metabolic function, as they experience annual cycles of fattening and fasting driven by seasonal changes in forage availability (Albon et al., 2017; Reimers, 1984). Consequently, contaminant-modulated effects on adipogenesis or fatty acid mobilisation may disrupt vital processes in the reindeer.

Although the ultimate physiological outcome of these changes in gene expression cannot be determined from the data presented in this study, these findings can be seen as early-response indicators for potential adverse effects on higher levels of biological organization (Ishibashi et al., 2008; Piña et al., 2007; Trego et al., 2019). These results warrant further assessment of contaminant-driven effects on the lipid metabolic pathways in Svalbard reindeer, where the impact on endpoints such as total lipid content, triglyceride plasma levels, and enzymatic activity of receptors involved in lipid metabolism (Routti et al., 2019, 2016) would be relevant targets.

Previous studies have demonstrated that exposure to contaminant mixtures can cause effects at far lower concentrations than individual compounds, suggesting that traditional approaches to risk assessment may underestimate the effects of mixtures on physiological functions (e.g., Desforges et al., 2017). In line with these findings, the results from **Paper IV**

suggest that even contaminant concentrations below established toxicity thresholds may cause a biological response, supporting the need to establish new toxicity thresholds based on contaminant mixtures (Sonne et al., 2021).

HUMAN RELEVANCE OF THE FINDINGS

Traditional Arctic food sources, such as whale or polar bear, are known to contain high levels of contaminants due to their positions as apex predators (Dietz et al., 2022, 2013). Consequently, the dietary exposure in many Arctic communities has resulted in high contaminant burdens, even reaching concentrations associated with adverse biological effects (AMAP, 2021). For example, prenatal exposure to contaminants through maternal consumption of pilot whale (*Globicephala melaena*) has been linked to neurological and immunological effects in children from the Faroe Islands (Weihe and Grandjean, 2012).

In contrast to many other Arctic populations, residents of Svalbard have a relatively small fraction of traditional foods in their diet, and hunting in Svalbard is primarily recreational rather than for subsistence. Commonly hunted species of wildlife include seals, ptarmigans, and Svalbard reindeer (Sysselmesteren, 2019). The Svalbard reindeer hunting period is limited to five weeks, starting August 15th and ending September 20th.

In **Paper III**, we report significantly higher concentrations of Hg, Cd, and ΣPFAS in reindeer culled in October, compared to those culled in August. While contaminant concentrations are far lower in Svalbard reindeer compared to marine apex predators, the concentrations were high enough to consider the tolerable weekly intake (TWI), particularly for PFAS. The TWI for Σ_4PFAS (PFOA, PFNA, PFHxS, and PFOS) is set to 4.4 ng/kg body weight/week (Schrenk et al., 2020). The average Σ_4PFAS concentrations in Svalbard reindeer culled in August 2022 and October 2023 were 12.8 ng/g and 1.21 ng/g for liver and muscle, respectively. For a person weighing 70 kg, this corresponds to a maximum weekly consumption of 24 g of liver or 254 g of muscle, while for a child (30 kg: 10 g and 108 g, respectively), significantly lower amounts of both meat and offal can be consumed without exceeding the safe thresholds. However, given that each hunter is granted only one reindeer per hunting season (Sysselmesteren, 2024), the dietary exposure on an annual basis is limited, and the overall exposure to PFAS via the consumption of reindeer should remain low in Svalbard hunters and their families. Nonetheless, the significantly higher PFAS concentrations today (**Paper III**), compared to a decade ago (Roos et al., 2021), warrant further monitoring of PFAS in Svalbard reindeer with respect to human consumption to ensure that the concentrations are not increasing further.

Based on the findings of **Papers II** and **III**, a majority of the reported soft tissue concentrations of Hg and Pb fall below the maximum levels in food (500 ng/g and 200 ng/g for Hg and Pb, respectively) set by the European Commission (2023). However, a considerable number of reindeer individuals had hepatic Cd levels exceeding the maximum food level (500 ng/g) (**Papers II and III**), and when considering the recommended TWI of Cd (2.5 µg/kg body weight; EFSA, 2011), a 70 kg adult could consume approximately 300-400 g of liver weekly before exceeding the set thresholds. Nonetheless, similarly to PFAS, the

limitations in Svalbard reindeer consumption regulated by the hunting quotas reduce the overall dietary risk.

CONCLUDING REMARKS

Contaminants in Arctic terrestrial ecosystems have been far less studied compared to marine environments. This thesis helps close existing knowledge gaps by exploring the occurrence and effects of PFAS and toxic elements Hg, Cd, and Pb in the Svalbard terrestrial ecosystem using the Svalbard reindeer as our model organism. The main findings of this thesis highlight that even Svalbard reindeer, a terrestrial herbivore, are exposed to contaminants that likely have reached the Arctic through long-range transport.

CONTAMINANT EXPOSURE IN THE SVALBARD REINDEER

Contaminant exposure in Svalbard reindeer appears to be primarily driven by diet, through the consumption of different species of vegetation which accumulate contaminants from the surrounding soil and air. The highest concentration of toxic metals in Svalbard reindeer tissues generally followed the pattern Cd > Hg > Pb, with hepatic and renal tissues having greater concentrations than muscle. For PFAS, hepatic concentrations were dominated by perfluorosulfonic acids (PFHxS and PFOS) and perfluoroalkyl carboxylic acids (PFUnDA and PFTriDA). The soft tissue contaminant profiles suggest that long-range transported pollutants are the main source of exposure in the reindeer.

Concentrations of Cd and Pb in Svalbard reindeer appear to have declined over the past forty years. In contrast, the concentrations of PFAS have increased, along with a shift in PFAS profile towards longer-chain PFCA compounds, which may be linked to the remobilization of contaminants from the thawing Arctic cryosphere. For Hg, no previous data from Svalbard reindeer soft tissues are reported, making the findings of the present thesis an important baseline for future temporal comparisons.

SEASONAL FLUCTUATIONS IN CONTAMINANT CONCENTRATIONS

Soft tissue contaminant concentrations in Svalbard reindeer are significantly higher in October compared to August. This aligns with the annual peak in body mass of the reindeer, which occurs in October, suggesting that their contaminant burden reflects the cumulative dietary exposure from a continued ingestion of forage over the growing season.

Although the risk of exceeding human health thresholds through the consumption of Svalbard reindeer is low, mainly given the annual allocation of one animal per hunter, concentrations of both Cd and ΣPFAS are sufficiently high to warrant caution during consumption of both meat and offal. Furthermore, these findings suggest that hunting regulations could consider recommending earlier culling in the season to minimise contaminant exposure.

NON-INVASIVE CONTAMINANT MONITORING IN SVALBARD REINDEER

Concentrations of Hg and Cd in faeces and fur were significantly positively related to concentrations in soft tissues. However, due to external contamination of the fur, which is a commonly reported problem in existing literature, we consider fur to be unreliable as a non-invasive proxy for Cd. In contrast, faecal concentrations had a clear relationship with liver

Hg concentrations, suggesting that faecal sampling may be a more feasible, non-invasive method of monitoring internal Hg levels in Svalbard reindeer.

CONTAMINANT-DRIVEN EFFECTS IN SVALBARD REINDEER

Although all reported contaminant concentrations in Svalbard reindeer were below the current wildlife toxicity thresholds, our findings suggest that contaminant exposure, primarily driven by PFHxS, Cd, and Hg, influences the expression levels of genes involved in key physiological functions, such as lipid metabolism. This indicates that current toxicity thresholds may not fully reflect the potential for contaminant-driven biological effects in wildlife. Current toxicity thresholds are still largely based on single-contaminant studies, yet wildlife is exposed to complex chemical mixtures, which may lead to effects that are not predicted by the existing thresholds.

Like many Arctic species, Svalbard reindeer are facing the threat of multiple stressors, including rapid environmental changes driven by climate change. This PhD thesis demonstrates that in addition to these challenges, Svalbard reindeer are also subject to contaminant exposure and contaminant-driven biological effects, despite living in one of the most remote regions on Earth.

FUTURE PERSPECTIVES

While this thesis addresses a critical knowledge gap by providing new data on contaminants in the Arctic terrestrial ecosystem, it also creates opportunities for future research.

This thesis includes the first report of soft tissue Hg concentrations in Svalbard reindeer. Considering that permafrost is one of the largest sinks of Hg on Earth, the thawing cryosphere due to climate change warrants further monitoring of Hg in terrestrial wildlife to assess how concentrations may change in the future. As such, our findings will be a good temporal time point to compare any future measurements with.

PFAS concentrations in Svalbard reindeer appear to have significantly increased over the past decade, which may be the result of contaminant remobilisation from the thawing cryosphere. This requires further study, where PFAS in both vegetation and soil in proglacial areas could help determine the ultimate sources of PFAS in Svalbard reindeer. The increased PFAS concentrations also warrant additional research from a human health perspective. Collaboration with hunters during the entire hunting season (mid-August to September) could enable sampling of reindeer across large parts of Nordenskiöld land, providing valuable temporal and spatial data that can help determine whether the trends observed in this thesis also apply to reindeer from other areas than Reindalen.

Finally, our findings highlight the need for further research into contaminant-driven effects, especially on lipid metabolic pathways in Svalbard reindeer. Future studies should investigate whether the observed transcriptional changes in this thesis translate into physiological outcomes, focusing on endpoints such as total lipid content, plasma triglyceride levels, and the enzymatic activity of receptors involved in lipid metabolism. This will require a carefully planned sampling campaign that allows for rapid tissue collection and subsequent preservation suitable for such biochemical analyses, which may be challenging under Arctic working conditions.

REFERENCES

Aas, C.B., Fuglei, E., Herzke, D., Yoccoz, N.G., Routti, H., 2014. Effect of Body Condition on Tissue Distribution of Perfluoroalkyl Substances (PFASs) in Arctic Fox (*Vulpes lagopus*). Environ. Sci. Technol. 48, 11654–11661. https://doi.org/10.1021/es503147n

Ahrens, L., Rakovic, J., Ekdahl, S., Kallenborn, R., 2023. Environmental distribution of per- and polyfluoroalkyl substances (PFAS) on Svalbard: Local sources and long-range transport to the Arctic. Chemosphere 345, 140463. https://doi.org/10.1016/j.chemosphere.2023.140463

Albon, S.D., Irvine, R.J., Halvorsen, O., Langvatn, R., Loe, L.E., Ropstad, E., Veiberg, V., Van Der Wal, R., Bjørkvoll, E.M., Duff, E.I., Hansen, B.B., Lee, A.M., Tveraa, T., Stien, A., 2017. Contrasting effects of summer and winter warming on body mass explain population dynamics in a food-limited Arctic herbivore. Glob. Change Biol 23, 1374–1389. https://doi.org/10.1111/gcb.13435

Ali, A.M., Langberg, H.A., Hale, S.E., Kallenborn, R., Hartz, W.F., Mortensen, Å.-K., Ciesielski, T.M., McDonough, C.A., Jenssen, B.M., Breedveld, G.D., 2021. The fate of poly- and perfluoroalkyl substances in a marine food web influenced by land-based sources in the Norwegian Arctic. Environ. Sci.: Process Impacts 23, 588–604. https://doi.org/10.1039/D0EM00510J

AMAP, 2018. AMAP assessment 2018: Biological Effects of Contaminants on Arctic Wildlife and Fish. Oslo, Norway.

AMAP, 2005. AMAP Assessment 2002: Heavy metals in the Arctic. Arctic Monitoring and Assessment Programme (AMAP), Oslo.

AMAP, 2021, 2021. AMAP Assessment 2021: Human Health in the Arctic. Arctic Monitoring and Assessment Programme (AMAP), Tromsø, Norway.

Amos, H.M., Jacob, D.J., Streets, D.G., Sunderland, E.M., 2013. Legacy impacts of all-time anthropogenic emissions on the global mercury cycle: GLOBAL IMPACTS OF LEGACY MERCURY. Global Biogeochem. Cycles 27, 410–421. https://doi.org/10.1002/gbc.20040

Andersson Stavridis, M., Pollestad, B.M., Piqué, V.A., Mikkelsen, Ø., Jenssen, B.M., Ciesielski, T.M., 2025. Faecal Biomonitoring of Mercury in Arctic Terrestrial Herbivores Reveals Spatial Trends Across Svalbard and Greenland.

Ankley, G.T., Cureton, P., Hoke, R.A., Houde, M., Kumar, A., Kurias, J., Lanno, R., McCarthy, C., Newsted, J., Salice, C.J., Sample, B.E., Sepúlveda, M.S., Steevens, J., Valsecchi, S., 2020. Assessing the Ecological Risks of Per- and Polyfluoroalkyl Substances: Current State-of-the Science and a Proposed Path Forward. Environmental Toxicology and Chemistry 40, 564–605. https://doi.org/10.1002/etc.4869

Balali-Mood, M., Naseri, K., Tahergorabi, Z., Khazdair, M.R., Sadeghi, M., 2021. Toxic Mechanisms of Five Heavy Metals: Mercury, Lead, Chromium, Cadmium, and Arsenic. Front. Pharmacol. 12, 643972. https://doi.org/10.3389/fphar.2021.643972

Bargagli, R., 2016. Moss and lichen biomonitoring of atmospheric mercury: A review. Sci. Total Environ. 572, 216–231. https://doi.org/10.1016/j.scitotenv.2016.07.202

Beard, D.B., Clason, C.C., Rangecroft, S., Poniecka, E., Ward, K.J., Blake, W.H., 2022. Anthropogenic contaminants in glacial environments I: Inputs and accumulation. Progress in Physical Geography: Earth and Environment 46, 630–648. https://doi.org/10.1177/03091333221107376

Beccacece, L., Costa, F., Pascali, J.P., Giorgi, F.M., 2023. Cross-Species Transcriptomics Analysis Highlights Conserved Molecular Responses to Per- and Polyfluoroalkyl Substances. Toxics 11, 567. https://doi.org/10.3390/toxics11070567

Bechshoft, T., Dyck, M., St. Pierre, K.A., Derocher, A.E., St. Louis, V., 2019. The use of hair as a proxy for total and methylmercury burdens in polar bear muscle tissue. Sci. Total Environ. 686, 1120–1128. https://doi.org/10.1016/j.scitotenv.2019.06.087

Beitveit, G.M., 2016. Contamination in an Arctic Environment: Abiotic and Biotic Impacts of Local Pollution (Master thesis). Norwegian University of Science and Technology, Trondheim.

Bjørkvoll, E., Pedersen, B., Hytteborn, H., Jónsdóttir, I.S., Langvatn, R., 2009. Seasonal and Interannual Dietary Variation During Winter in Female Svalbard Reindeer (*Rangifer Tarandus Platyrhynchus*). AAAR 41, 88–96. https://doi.org/10.1657/1523-0430-41.1.88

Bjørneset, J., Blévin, P., Bjørnstad, P.M., Dalmo, R.A., Goksøyr, A., Harju, M., Limonta, G., Panti, C., Rikardsen, A.H., Sundaram, A.Y.M., Yadetie, F., Routti, H., 2023. Establishment of killer whale (Orcinus orca) primary fibroblast cell cultures and their transcriptomic responses to pollutant exposure. Environment International 174, 107915. https://doi.org/10.1016/j.envint.2023.107915

Borch-Iohnsen, B., Nilssen, K.J., Norheim, G., 1996. Influence of season and diet on liver and kidney content of essential elements and heavy metals in svalbard reindeer. Biol Trace Elem Res 51, 235–247. https://doi.org/10.1007/BF02784078

Born, E.W., Renzoni, A., Dietz, R., 1991. Total mercury in hair of polar bears (*Ursus maritimus*) from Greenland and Svalbard. Polar Res. 9, 113–120. https://doi.org/10.3402/polar.v9i2.6784

Boutron, C.F., Candelone, J.-P., Hong, S., 1995. Greenland snow and ice cores: unique archives of large-scale pollution of the troposphere of the Northern Hemisphere by lead and other heavy metals. Science of The Total Environment 160–161, 233–241. https://doi.org/10.1016/0048-9697(95)04359-9

Brunn, H., Arnold, G., Körner, W., Rippen, G., Steinhäuser, K.G., Valentin, I., 2023. PFAS: forever chemicals—persistent, bioaccumulative and mobile. Reviewing the status and the need for their phase out and remediation of contaminated sites. Environ Sci Eur 35. https://doi.org/10.1186/s12302-023-00721-8

Bucheli, T.D., Fent, K., 1995. Induction of cytochrome P450 as a biomarker for environmental contamination in aquatic ecosystems. Critical Reviews in Environmental Science and Technology 25, 201–268. https://doi.org/10.1080/10643389509388479

Buck, R.C., Franklin, J., Berger, U., Conder, J.M., Cousins, I.T., De Voogt, P., Jensen, A.A., Kannan, K., Mabury, S.A., Van Leeuwen, S.P., 2011. Perfluoroalkyl and polyfluoroalkyl substances in the environment: Terminology, classification, and origins. Integrated Environmental Assessment and Management 7, 513–541. https://doi.org/10.1002/ieam.258

Burkow, I.C., Kallenborn, R., 2000. Sources and transport of persistent pollutants to the Arctic. Toxicology Letters 112–113, 87–92. https://doi.org/10.1016/S0378-4274(99)00254-4

Cairns, J., 1992. The threshold problem in ecotoxicology. Ecotoxicology 1, 3–16. https://doi.org/10.1007/BF00702652

Carrasco, N., McGovern, M., Evenset, A., Søreide, J.E., Arts, M.T., Jonsson, S., Poste, A.E., 2024. Seasonal riverine inputs may affect diet and mercury bioaccumulation in Arctic coastal zooplankton. Science of The Total Environment 906, 167643. https://doi.org/10.1016/j.scitotenv.2023.167643

Chen, M., Gazze, L., DiTraglia, F.J., Das, R., Nriagu, J., Erel, Y., Boyle, E.A., Taylor, C.M., Weiss, D., 2025. Environmental lead risk in the 21st century. Commun Earth Environ 6, 776. https://doi.org/10.1038/s43247-025-02735-x

Cuyler, C., Øritsland, N.A., 2002. Do seasonal changes in Svalbard reindeer fur have relevance for heat transfer? Rangifer 22, 133. https://doi.org/10.7557/2.22.2.1532

Dai, S., Zhang, G., Dong, C., Yang, R., Pei, Z., Li, Y., Li, A., Zhang, Q., Jiang, G., 2025. Occurrence, bioaccumulation and trophodynamics of per- and polyfluoroalkyl substances (PFAS) in

terrestrial and marine ecosystems of Svalbard, Arctic. Water Res. 271, 122979. https://doi.org/10.1016/j.watres.2024.122979

Dastoor, A., Wilson, S.J., Travnikov, O., Ryjkov, A., Angot, H., Christensen, J.H., Steenhuisen, F., Muntean, M., 2022. Arctic atmospheric mercury: Sources and changes. Sci. Total Environ. 839, 156213. https://doi.org/10.1016/j.scitotenv.2022.156213

Descamps, S., Aars, J., Fuglei, E., Kovacs, K.M., Lydersen, C., Pavlova, O., Pedersen, Å.Ø., Ravolainen, V., Strøm, H., 2017. Climate change impacts on wildlife in a High Arctic archipelago – Svalbard, Norway. Global Change Biology 23, 490–502. https://doi.org/10.1111/gcb.13381

Desforges, J.-P., Ferguson, S.H., Remili, A., McKinney, M.A., Watt, C.A., Matthews, C.J.D., 2024. Assessment of persistent organic pollutants in killer whales (Orcinus orca) of the Canadian Arctic: Implications for subsistence consumption and conservation strategies. Environmental Research 244, 117992. https://doi.org/10.1016/j.envres.2023.117992

Desforges, J.-P., Hall, A., McConnell, B., Rosing-Asvid, A., Barber, J.L., Brownlow, A., De Guise, S., Eulaers, I., Jepson, P.D., Letcher, R.J., Levin, M., Ross, P.S., Samarra, F., Víkingson, G., Sonne, C., Dietz, R., 2018. Predicting global killer whale population collapse from PCB pollution. Science 361, 1373–1376. https://doi.org/10.1126/science.aat1953

Desforges, J.-P., Levin, M., Jasperse, L., De Guise, S., Eulaers, I., Letcher, R.J., Acquarone, M., Nordøy, E., Folkow, L.P., Hammer Jensen, T., Grøndahl, C., Bertelsen, M.F., St. Leger, J., Almunia, J., Sonne, C., Dietz, R., 2017. Effects of Polar Bear and Killer Whale Derived Contaminant Cocktails on Marine Mammal Immunity. Environ. Sci. Technol. 51, 11431–11439. https://doi.org/10.1021/acs.est.7b03532

Dietz, R., Desforges, J.-P., Gustavson, K., Rigét, F.F., Born, E.W., Letcher, R.J., Sonne, C., 2018a. Immunologic, reproductive, and carcinogenic risk assessment from POP exposure in East Greenland polar bears (Ursus maritimus) during 1983–2013. Environment International 118, 169–178. https://doi.org/10.1016/j.envint.2018.05.020

Dietz, R., Desforges, J.-P., Gustavson, K., Rigét, F.F., Born, E.W., Letcher, R.J., Sonne, C., 2018b. Immunologic, reproductive, and carcinogenic risk assessment from POP exposure in East Greenland polar bears (Ursus maritimus) during 1983–2013. Environment International 118, 169–178. https://doi.org/10.1016/j.envint.2018.05.020

Dietz, R., Letcher, R.J., Aars, J., Andersen, M., Boltunov, A., Born, E.W., Ciesielski, T.M., Das, K., Dastnai, S., Derocher, A.E., Desforges, J.-P., Eulaers, I., Ferguson, S., Hallanger, I.G., Heide-Jørgensen, M.P., Heimbürger-Boavida, L.-E., Hoekstra, P.F., Jenssen, B.M., Kohler, S.G., Larsen, M.M., Lindstrøm, U., Lippold, A., Morris, A., Nabe-Nielsen, J., Nielsen, N.H., Peacock, E., Pinzone, M., Rigét, F.F., Rosing-Asvid, A., Routti, H., Siebert, U., Stenson, G., Stern, G., Strand, J., Søndergaard, J., Treu, G., Víkingsson, G.A., Wang, F., Welker, J.M., Wiig, Ø., Wilson, S., Sonne, C., 2022. A risk assessment review of mercury exposure in Arctic marine and terrestrial mammals. Sci. Total Environ. 154445. https://doi.org/10.1016/j.scitotenv.2022.154445

Dietz, R., Outridge, P.M., Hobson, K.A., 2009. Anthropogenic contributions to mercury levels in present-day Arctic animals—A review. Sci. Total Environ. 407, 6120–6131. https://doi.org/10.1016/j.scitotenv.2009.08.036

Dietz, R., Riget, F., Sonne, C., Letcher, R., Born, E., Muir, D., 2004. Seasonal and temporal trends in polychlorinated biphenyls and organochlorine pesticides in East Greenland polar bears (Ursus maritimus), 1990–2001. Sci. Total Environ. 331, 107–124. https://doi.org/10.1016/j.scitotenv.2004.03.025

Dietz, R., Rune Dietz, Sonne, C., Basu, N., Braune, B.M., O'Hara, T.M., Letcher, R.J., Scheuhammer, T.M., Andersen, M., Andreasen, C., Andriashek, D., Asmund, G., Aubail, A., Baagøe, H.J.,

Born, E.W., Chan, H.M., Derocher, A.E., Grandjean, P., Knott, K.K., Kirkegaard, M., Krey, A., Lunn, N.J., Messier, F., Obbard, M., Olsen, M.T., Ostertag, S.K., Peacock, E., Renzoni, A., Rigét, F.F., Skaare, J.U., Stern, G.A., Stirling, I., Taylor, M.K., Wiig, Ø., Wilson, S., Aars, J., 2013. What are the toxicological effects of mercury in Arctic biota. Sci. Total Environ. 443, 775–790. https://doi.org/10.1016/j.scitotenv.2012.11.046

Dolan, K.J., Ciesielski, T.M., Lierhagen, S., Eulaers, I., Nygård, T., Johnsen, T.V., Gómez-Ramírez, P., García-Fernández, A.J., Bustnes, J.O., Ortiz-Santaliestra, M.E., Jaspers, V.L.B., 2017. Trace element concentrations in feathers and blood of Northern goshawk (Accipiter gentilis) nestlings from Norway and Spain. Ecotox. Environ. Saf. 144, 564–571. https://doi.org/10.1016/j.ecoenv.2017.06.062

Drotikova, T., Ali, A.M., Halse, A.K., Reinardy, H.C., Kallenborn, R., 2020. Polycyclic aromatic hydrocarbons (PAHs) and oxy- and nitro-PAHs in ambient air of the Arctic town Longyearbyen, Svalbard. Atmos. Chem. Phys. 20, 9997–10014. https://doi.org/10.5194/acp-20-9997-2020

Dudka, S., Adriano, D.C., 1997. Environmental Impacts of Metal Ore Mining and Processing: A Review. Journal of Environmental Quality 26, 590–602. https://doi.org/10.2134/jeq1997.00472425002600030003x

EFSA, 2011. Statement on tolerable weekly intake for cadmium. EFSA J 9. https://doi.org/10.2903/j.efsa.2011.1975

Ehrlich, V., Bil, W., Vandebriel, R., Granum, B., Luijten, M., Lindeman, B., Grandjean, P., Kaiser, A.-M., Hauzenberger, I., Hartmann, C., Gundacker, C., Uhl, M., 2023. Consideration of pathways for immunotoxicity of per- and polyfluoroalkyl substances (PFAS). Environ Health 22, 19. https://doi.org/10.1186/s12940-022-00958-5

Eisenreich, S.J., Metzer, N.A., Urban, N.R., Robbins, J.A., 1986. Response of atmospheric lead to decreased use of lead in gasoline. Environ. Sci. Technol. 20, 171–174. https://doi.org/10.1021/es00144a010

Ellis, D.A., Martin, J.W., De Silva, A.O., Mabury, S.A., Hurley, M.D., Sulbaek Andersen, M.P., Wallington, T.J., 2004. Degradation of Fluorotelomer Alcohols: A Likely Atmospheric Source of Perfluorinated Carboxylic Acids. Environ. Sci. Technol. 38, 3316–3321. https://doi.org/10.1021/es049860w

Evenset, A., Pedersen, K.B., Kvennås, M., Cappelen, P., 2020. Miljøtekniske undersøkelser i Svea 2019 (No. APN-60480-2). Akvaplan-niva.

Fenton, S.E., Ducatman, A., Boobis, A., DeWitt, J.C., Lau, C., Ng, C., Smith, J.S., Roberts, S.M., 2020. Per- and Polyfluoroalkyl Substance Toxicity and Human Health Review: Current State of Knowledge and Strategies for Informing Future Research. Environmental Toxicology and Chemistry 40, 606–630. https://doi.org/10.1002/etc.4890

Gamberg, M., Braune, B., Davey, E., Elkin, B., Hoekstra, P.F., Kennedy, D., Macdonald, C., Muir, D., Nirwal, A., Wayland, M., Zeeb, B., 2005. Spatial and temporal trends of contaminants in terrestrial biota from the Canadian Arctic. Sci. Total Environ. 351–352, 148–164. https://doi.org/10.1016/j.scitotenv.2004.10.032

Gamberg, M., Cuyler, C., Wang, X., 2016. Contaminants in two West Greenland caribou populations. Sci. Total Environ. 554–555, 329–336. https://doi.org/10.1016/j.scitotenv.2016.02.154

Gamberg, M., Pratte, I., Brammer, J., Cuyler, C., Elkin, B., Gurney, K., Kutz, S., Larter, N.C., Muir, D., Wang, X., Provencher, J.F., 2020. Renal trace elements in barren-ground caribou subpopulations: Temporal trends and differing effects of sex, age and season. Sci. Total Environ. 724, 138305. https://doi.org/10.1016/j.scitotenv.2020.138305

Garnett, J., Halsall, C., Winton, H., Joerss, H., Mulvaney, R., Ebinghaus, R., Frey, M., Jones, A., Leeson, A., Wynn, P., 2022. Increasing Accumulation of Perfluorocarboxylate Contaminants Revealed in an Antarctic Firn Core (1958–2017). Environ. Sci. Technol. 56, 11246–11255. https://doi.org/10.1021/acs.est.2c02592

Genchi, G., Sinicropi, M.S., Lauria, G., Carocci, A., Catalano, A., 2020. The Effects of Cadmium Toxicity. IJERPH 17, 3782. https://doi.org/10.3390/ijerph17113782

Gil-Jiménez, E., Mateo, R., De Lucas, M., Ferrer, M., 2020. Feathers and hair as tools for non-destructive pollution exposure assessment in a mining site of the Iberian Pyrite Belt. Environ. Pollut. 263, 114523. https://doi.org/10.1016/j.envpol.2020.114523

Gopikrishna, V.G., Sebastian, A., Akhilghosh, K.A., Mohan, M., 2020. Mercury and Other Trace Metals in the Surface Soils of Ny-Ålesund, Arctic. Water Air Soil Pollut 231, 377. https://doi.org/10.1007/s11270-020-04745-1

Greaves, A.K., Letcher, R.J., Sonne, C., Dietz, R., Born, E.W., 2012. Tissue-Specific Concentrations and Patterns of Perfluoroalkyl Carboxylates and Sulfonates in East Greenland Polar Bears. Environ. Sci. Technol. 46, 11575–11583. https://doi.org/10.1021/es303400f

Gworek, B., Dmuchowski, W., Baczewska-Dąbrowska, A.H., 2020. Mercury in the terrestrial environment: a review. Environ Sci Eur 32, 128. https://doi.org/10.1186/s12302-020-00401-x

Halbach, K., Mikkelsen, Ø., Berg, T., Steinnes, E., 2017. The presence of mercury and other trace metals in surface soils in the Norwegian Arctic. Chemosphere 188, 567–574. https://doi.org/10.1016/j.chemosphere.2017.09.012

Hallanger, I.G., Fuglei, E., Yoccoz, N.G., Pedersen, Å.Ø., König, M., Routti, H., 2019. Temporal trend of mercury in relation to feeding habits and food availability in arctic foxes (Vulpes lagopus) from Svalbard, Norway. Sci. Total Environ. 670, 1125–1132. https://doi.org/10.1016/j.scitotenv.2019.03.239

Hansen, B.B., Lorentzen, J.R., Welker, J.M., Varpe, Ø., Aanes, R., Beumer, L.T., Pedersen, Å.Ø., 2019. Reindeer turning maritime: Ice-locked tundra triggers changes in dietary niche utilization. Ecosphere 10, e02672. https://doi.org/10.1002/ecs2.2672

Hartz, W.F., Björnsdotter, M.K., Yeung, L.W.Y., Hodson, A., Thomas, E.R., Humby, J.D., Day, C., Jogsten, I.E., Kärrman, A., Kallenborn, R., 2023. Levels and distribution profiles of Per- and Polyfluoroalkyl Substances (PFAS) in a high Arctic Svalbard ice core. Sci. Total Environ. 871, 161830. https://doi.org/10.1016/j.scitotenv.2023.161830

Hernández-Moreno, D., De La Casa Resino, I., Fidalgo, L.E., Llaneza, L., Soler Rodríguez, F., Pérez-López, M., López-Beceiro, A., 2013. Noninvasive heavy metal pollution assessment by means of Iberian wolf (Canis lupus signatus) hair from Galicia (NW Spain): a comparison with invasive samples. Environ. Monit. Assess. 185, 10421–10430. https://doi.org/10.1007/s10661-013-3341-x

Herzke, D., Olsson, E., Posner, S., 2012. Perfluoroalkyl and polyfluoroalkyl substances (PFASs) in consumer products in Norway – A pilot study. Chemosphere 88, 980–987. https://doi.org/10.1016/j.chemosphere.2012.03.035

Hiltunen, T.A., Stien, A., Väisänen, M., Ropstad, E., Aspi, J.O., Welker, J.M., 2022. Svalbard reindeer winter diets: Long-term dietary shifts to graminoids in response to a changing climate. Global Change Biology 28, 7009–7022. https://doi.org/10.1111/gcb.16420

Houde, M., Wang, X., Colson, T.-L.L., Gagnon, P., Ferguson, S.H., Ikonomou, M.G., Dubetz, C., Addison, R.F., Muir, D.C.G., 2019. Trends of persistent organic pollutants in ringed seals (Phoca hispida) from the Canadian Arctic. Science of The Total Environment 665, 1135–1146. https://doi.org/10.1016/j.scitotenv.2019.02.138

Ishibashi, H., Iwata, H., Kim, E.-Y., Tao, L., Kannan, K., Tanabe, S., Batoev, V.B., Petrov, E.A., 2008. Contamination and Effects of Perfluorochemicals in Baikal Seal (*Pusa sibirica*). 2.

Molecular Characterization, Expression Level, and Transcriptional Activation of Peroxisome Proliferator-Activated Receptor α. Environ. Sci. Technol. 42, 2302–2308. https://doi.org/10.1021/es0720558

Ismael, M.A., Elyamine, A.M., Moussa, M.G., Cai, M., Zhao, X., Hu, C., 2019. Cadmium in plants: uptake, toxicity, and its interactions with selenium fertilizers. Metallomics 11, 255–277. https://doi.org/10.1039/C8MT00247A

Iulini, M., Bettinsoli, V., Maddalon, A., Galbiati, V., Janssen, A.W.F., Beekmann, K., Russo, G., Pappalardo, F., Fragki, S., Paini, A., Corsini, E., 2025. In vitro approaches to investigate the effect of chemicals on antibody production: the case study of PFASs. Arch Toxicol 99, 2075–2086. https://doi.org/10.1007/s00204-025-03993-6

Jaspers, V.L.B., Covaci, A., Herzke, D., Eulaers, I., Eens, M., 2019. Bird feathers as a biomonitor for environmental pollutants: Prospects and pitfalls. TrAC Trends in Analytical Chemistry 118, 223–226. https://doi.org/10.1016/j.trac.2019.05.019

Jomova, K., Alomar, S.Y., Nepovimova, E., Kuca, K., Valko, M., 2025. Heavy metals: toxicity and human health effects. Arch Toxicol 99, 153–209. https://doi.org/10.1007/s00204-024-03903-2

Jones, K.C., 2021. Persistent Organic Pollutants (POPs) and Related Chemicals in the Global Environment: Some Personal Reflections. Environ. Sci. Technol. 55, 9400–9412. https://doi.org/10.1021/acs.est.0c08093

Joseph, P., 2022. Transcriptomics: Applications in Toxicology and Medicine, in: Sahu, S.C. (Ed.), Genomic and Epigenomic Biomarkers of Toxicology and Disease. Wiley, pp. 133–161. https://doi.org/10.1002/9781119807704.ch7

Joseph, P., 2009. Mechanisms of cadmium carcinogenesis☆. Toxicology and Applied Pharmacology 238, 272–279. https://doi.org/10.1016/j.taap.2009.01.011

Jota Baptista, C., Seixas, F., Gonzalo-Orden, J.M., Oliveira, P.A., 2022. Biomonitoring metals and metalloids in wild mammals: invasive versus non-invasive sampling. Environ. Sci. Pollut. Res. 29, 18398–18407. https://doi.org/10.1007/s11356-022-18658-5

Kim, K.-H., Kabir, E., Jahan, S.A., 2016. A review on the distribution of Hg in the environment and its human health impacts. Journal of Hazardous Materials 306, 376–385. https://doi.org/10.1016/j.jhazmat.2015.11.031

Kinck, C., 2014. Reduced Forage Access Affects Home Range Size and Site Fidelity of Svalbard Reindeer (Rangifer tarandus platyrhynchus). NMBU.

King, M.D., Su, G., Crump, D., Farhat, A., Marlatt, V., Lee, S.L., Williams, T.D., Elliott, J.E., 2023. Contaminant biomonitoring augmented with a qPCR array indicates hepatic mRNA gene expression effects in wild-collected seabird embryos. Sci. Total Environ. 904, 166784. https://doi.org/10.1016/j.scitotenv.2023.166784

Klaassen, C.D., Watkins, J.B. (Eds.), 2015. Casarett & Doull's essentials of toxicology, Third edition. ed. McGraw-Hill, New York.

Kohler, J., Aanes, R., 2004. Effect of Winter Snow and Ground-Icing on a Svalbard Reindeer Population: Results of a Simple Snowpack Model. Arctic, Antarctic, and Alpine Research 36, 333–341. https://doi.org/10.1657/1523-0430(2004)036%255B0333:EOWSAG%255D2.0.CO;2

Krajcarová, L., Novotný, K., Chattová, B., Elster, J., 2016. Elemental analysis of soils and Salix polaris in the town of Pyramiden and its surroundings (Svalbard). Environ. Sci. Pollut. Res. 23, 10124–10137. https://doi.org/10.1007/s11356-016-6213-4

Kurtz, A.E., Reiner, J.L., West, K.L., Jensen, B.A., 2019. Perfluorinated Alkyl Acids in Hawaiian Cetaceans and Potential Biomarkers of Effect: Peroxisome Proliferator-Activated Receptor

Alpha and Cytochrome P450 4A. Environ. Sci. Technol. 53, 2830–2839. https://doi.org/10.1021/acs.est.8b05619

Kwok, K.Y., Yamazaki, E., Yamashita, N., Taniyasu, S., Murphy, M.B., Horii, Y., Petrick, G., Kallerborn, R., Kannan, K., Murano, K., Lam, P.K.S., 2013. Transport of Perfluoroalkyl substances (PFAS) from an arctic glacier to downstream locations: Implications for sources. Science of The Total Environment 447, 46–55. https://doi.org/10.1016/j.scitotenv.2012.10.091

Larison, J.R., Likens, G.E., Fitzpatrick, J.W., Crock, J.G., 2000. Cadmium toxicity among wildlife in the Colorado Rocky Mountains. Nature 406, 181–183. https://doi.org/10.1038/35018068

Larter, N.C., Macdonald, C.R., Elkin, B.T., Wang, X., Harms, N.J., Gamberg, M., Muir, D.C.G., 2016. Cadmium and other elements in tissues from four ungulate species from the Mackenzie Mountain region of the Northwest Territories, Canada. Ecotox. Environ. Saf. 132, 9–17. https://doi.org/10.1016/j.ecoenv.2016.05.018

Lehnherr, I., 2014. Methylmercury biogeochemistry: a review with special reference to Arctic aquatic ecosystems. Environ. Rev. 22, 229–243. https://doi.org/10.1139/er-2013-0059

Li, H., Hammarstrand, S., Midberg, B., Xu, Y., Li, Y., Olsson, D.S., Fletcher, T., Jakobsson, K., Andersson, E.M., 2022. Cancer incidence in a Swedish cohort with high exposure to perfluoroalkyl substances in drinking water. Environmental Research 204, 112217. https://doi.org/10.1016/j.envres.2021.112217

Liang, L., Pan, Y., Bin, L., Liu, Y., Huang, W., Li, R., Lai, K.P., 2022. Immunotoxicity mechanisms of perfluorinated compounds PFOA and PFOS. Chemosphere 291, 132892. https://doi.org/10.1016/j.chemosphere.2021.132892

Løseth, M.E., Briels, N., Flo, J., Malarvannan, G., Poma, G., Covaci, A., Herzke, D., Nygård, T., Bustnes, J.O., Jenssen, B.M., Jaspers, V.L.B., 2019. White-tailed eagle (Haliaeetus albicilla) feathers from Norway are suitable for monitoring of legacy, but not emerging contaminants. Science of The Total Environment 647, 525–533. https://doi.org/10.1016/j.scitotenv.2018.07.333

Lühmann, K., Lille-Langøy, R., Øygarden, L., Kovacs, K.M., Lydersen, C., Goksøyr, A., Routti, H., 2020. Environmental Pollutants Modulate Transcriptional Activity of Nuclear Receptors of Whales *In Vitro*. Environ. Sci. Technol. 54, 5629–5639. https://doi.org/10.1021/acs.est.9b06952

Lunardi, D., Abelli, L., Panti, C., Marsili, L., Fossi, M.C., Mancia, A., 2016. Transcriptomic analysis of bottlenose dolphin (Tursiops truncatus) skin biopsies to assess the effects of emerging contaminants. Marine Environmental Research 114, 74–79. https://doi.org/10.1016/j.marenvres.2016.01.002

Ma, W.-C., 2011. Lead in Mammals, in: Environmental Contaminants in Biota, Interpreting Tissue Concentrations. CRC Press, Taylor & Francis Group.

MacInnis, J., De Silva, A.O., Lehnherr, I., Muir, D.C.G., St. Pierre, K.A., St. Louis, V.L., Spencer, C., 2022. Investigation of perfluoroalkyl substances in proglacial rivers and permafrost seep in a high Arctic watershed. Environ. Sci.: Processes Impacts 24, 42–51. https://doi.org/10.1039/D1EM00349F

MacSween, K., Stupple, G., Aas, W., Kyllönen, K., Pfaffhuber, K.A., Skov, H., Steffen, A., Berg, T., Mastromonaco, M.N., 2022. Updated trends for atmospheric mercury in the Arctic: 1995–2018. Science of The Total Environment 837, 155802. https://doi.org/10.1016/j.scitotenv.2022.155802

Makarov, D.A., Ovcharenko, V.V., Nebera, E.A., Kozhushkevich, A.I., Shelepchikov, A.A., Turbabina, K.A., Kalantaenko, A.M., Bardyugov, N.S., Gergel, M.A., 2022. Geographical distribution of dioxins, cadmium, and mercury concentrations in reindeer liver, kidneys, and muscle in the

Russian Far North. Environ. Sci. Pollut. Res. 29, 12176–12187.
https://doi.org/10.1007/s11356-021-16310-2

Mandard, S., Muller, M., Kersten, S., 2004. Peroxisome proliferator-activated receptor a target genes. Cellular and Molecular Life Sciences (CMLS) 61, 393–416.
https://doi.org/10.1007/s00018-003-3216-3

Martin, J.W., Asher, B.J., Beesoon, S., Benskin, J.P., Ross, M.S., 2010. PFOS or PreFOS? Are perfluorooctane sulfonate precursors (PreFOS) important determinants of human and environmental perfluorooctane sulfonate (PFOS) exposure? J. Environ. Monit. 12, 1979.
https://doi.org/10.1039/c0em00295j

Mastrantonio, M., Bai, E., Uccelli, R., Cordiano, V., Screpanti, A., Crosignani, P., 2018. Drinking water contamination from perfluoroalkyl substances (PFAS): an ecological mortality study in the Veneto Region, Italy. European Journal of Public Health 28, 180–185.
https://doi.org/10.1093/eurpub/ckx066

McKinney, M.A., Iverson, S.J., Fisk, A.T., Sonne, C., Rigét, F.F., Letcher, R.J., Arts, M.T., Born, E.W., Rosing-Asvid, A., Dietz, R., 2013. Global change effects on the long-term feeding ecology and contaminant exposures of E ast G reenland polar bears. Global Change Biology 19, 2360–2372. https://doi.org/10.1111/gcb.12241

Melnes, M., Gabrielsen, G.W., Herzke, D., Sagerup, K., Jenssen, B.M., 2017. Dissimilar effects of organohalogenated compounds on thyroid hormones in glaucous gulls. Environmental Research 158, 350–357. https://doi.org/10.1016/j.envres.2017.06.007

Moody, C.A., Field, J.A., 2000. Perfluorinated Surfactants and the Environmental Implications of Their Use in Fire-Fighting Foams. Environ. Sci. Technol. 34, 3864–3870.
https://doi.org/10.1021/es991359u

Morcillo, P., Romero, D., Meseguer, J., Esteban, M.Á., Cuesta, A., 2016. Cytotoxicity and alterations at transcriptional level caused by metals on fish erythrocytes in vitro. Environ Sci Pollut Res 23, 12312–12322. https://doi.org/10.1007/s11356-016-6445-3

Muir, D., Gunnarsdóttir, M.J., Koziol, K., Von Hippel, F.A., Szumińska, D., Ademollo, N., Corsolini, S., De Silva, A., Gabrielsen, G., Kallenborn, R., Polkowska, Ż., Krümmel, E., Vorkamp, K., 2025. Local sources *versus* long-range transport of organic contaminants in the Arctic: future developments related to climate change. Environ. Sci.: Adv. 4, 355–408.
https://doi.org/10.1039/d4va00240g

Myklebust, I., Nybø, S., Kls, J.A., Pedersen, H.C., 1993. Cadmium accumulation in willow ptarmigan (Lagopus L. lagopus) and rock ptarmigan (L. mutus) in Central Norway. Sci. Total Environ. 134, 135–139. https://doi.org/10.1016/S0048-9697(05)80012-9

Norsk Klimaservicesenter, n.d. Observasjoner og værstatistikk.

Norwegian Polar Institute, 2016. Geological map of Svalbard (1:250000).
https://doi.org/10.21334/NPOLAR.2016.616F7504

Obrist, D., Kirk, J.L., Zhang, L., Sunderland, E.M., Jiskra, M., Selin, N.E., 2018. A review of global environmental mercury processes in response to human and natural perturbations: Changes of emissions, climate, and land use. Ambio 47, 116–140.
https://doi.org/10.1007/s13280-017-1004-9

Odsjö, T., Räikkönen, J., Bignert, A., 2005. Time trends of metals in liver and muscle of reindeer (Rangifer tarandus) from northern and central Lapland, Sweden, 1983-2003. Swedish monitoring programme in terrestrial biota. (Miljögifter i biota - fjäll No. 214 0330). Swedish Museum of Natural History, Stockholm.

O'Hara, T.M., George, J.C., Blake, J., Burek, K., Carroll, G., Dau, J., Bennett, L., McCoy, C.P., Gerard, P., Woshner, V., 2003. Investigation of Heavy Metals in a Large Mortality Event in Caribou of Northern Alaska. ARCTIC 56, 125–135. https://doi.org/10.14430/arctic608

Pacyna, A.D., Frankowski, M., Kozioł, K., Węgrzyn, M.H., Wietrzyk-Pełka, P., Lehmann-Konera, S., Polkowska, Ż., 2019. Evaluation of the use of reindeer droppings for monitoring essential and non-essential elements in the polar terrestrial environment. Sci. Total Environ. 658, 1209–1218. https://doi.org/10.1016/j.scitotenv.2018.12.232

Pacyna, A.D., Koziorowska, K., Chmiel, S., Mazerski, J., Polkowska, Ż., 2018. Svalbard reindeer as an indicator of ecosystem changes in the Arctic terrestrial ecosystem. Chemosphere 203, 209–218. https://doi.org/10.1016/j.chemosphere.2018.03.158

Pacyna, E.G., Pacyna, J.M., Steenhuisen, F., Wilson, S., 2006. Global anthropogenic mercury emission inventory for 2000. Atmospheric Environment 40, 4048–4063. https://doi.org/10.1016/j.atmosenv.2006.03.041

Pacyna, J.M., Pacyna, E.G., 2001. An assessment of global and regional emissions of trace metals to the atmosphere from anthropogenic sources worldwide. Environ. Rev. 9, 269–298. https://doi.org/10.1139/a01-012

Pacyna, J.M., Pacyna, E.G., Aas, W., 2009. Changes of emissions and atmospheric deposition of mercury, lead, and cadmium. Atmospheric Environment 43, 117–127. https://doi.org/10.1016/j.atmosenv.2008.09.066

Pacyna-Kuchta, A.D., Wietrzyk-Pełka, P., Węgrzyn, M.H., Frankowski, M., Polkowska, Ż., 2020. A screening of select toxic and essential elements and persistent organic pollutants in the fur of Svalbard reindeer. Chemosphere 245, 125458. https://doi.org/10.1016/j.chemosphere.2019.125458

Phelps, R.W., Clarkson, T.W., Kershaw, T.G., Wheatley, B., 1980. Interrelationships of blood and hair mercury concentrations in a North American population exposed to methylmercury. Arch. Environ. Health. 35, 161–168. https://doi.org/10.1080/00039896.1980.10667486

Piña, B., Casado, M., Quirós, L., 2007. Analysis of gene expression as a new tool in ecotoxicology and environmental monitoring. TrAC Trends in Analytical Chemistry 26, 1145–1154. https://doi.org/10.1016/j.trac.2007.09.009

Platt, S.M., Hov, Ø., Berg, T., Breivik, K., Eckhardt, S., Eleftheriadis, K., Evangeliou, N., Fiebig, M., Fisher, R., Hansen, G., Hansson, H.-C., Heintzenberg, J., Hermansen, O., Heslin-Rees, D., Holmén, K., Hudson, S., Kallenborn, R., Krejci, R., Krognes, T., Larssen, S., Lowry, D., Lund Myhre, C., Lunder, C., Nisbet, E., Nizzetto, P.B., Park, K.-T., Pedersen, C.A., Aspmo Pfaffhuber, K., Röckmann, T., Schmidbauer, N., Solberg, S., Stohl, A., Ström, J., Svendby, T., Tunved, P., Tørnkvist, K., Van Der Veen, C., Vratolis, S., Yoon, Y.J., Yttri, K.E., Zieger, P., Aas, W., Tørseth, K., 2022. Atmospheric composition in the European Arctic and 30 years of the Zeppelin Observatory, Ny-Ålesund. Atmos. Chem. Phys. 22, 3321–3369. https://doi.org/10.5194/acp-22-3321-2022

Pourrut, B., Shahid, M., Dumat, C., Winterton, P., Pinelli, E., 2011. Lead Uptake, Toxicity, and Detoxification in Plants, in: Whitacre, D.M. (Ed.), Reviews of Environmental Contamination and Toxicology Volume 213, Reviews of Environmental Contamination and Toxicology. Springer New York, New York, NY, pp. 113–136. https://doi.org/10.1007/978-1-4419-9860-6_4

R Core Team, 2024. R: A language and environment for statistical computing.

Rantanen, M., Karpechko, A.Yu., Lipponen, A., Nordling, K., Hyvärinen, O., Ruosteenoja, K., Vihma, T., Laaksonen, A., 2022. The Arctic has warmed nearly four times faster than the globe since 1979. Commun Earth Environ 3, 168. https://doi.org/10.1038/s43247-022-00498-3

Reimers, E., 1984. Body composition and population regulation of Svalbard reindeer. Ran 4, 16. https://doi.org/10.7557/2.4.2.499

Reimers, E., 1983. Mortality in Svalbard reindeer. Ecography 6, 141–149. https://doi.org/10.1111/j.1600-0587.1983.tb01075.x

Remili, A., Dietz, R., Sonne, C., Samarra, F.I.P., Letcher, R.J., Rikardsen, A.H., Ferguson, S.H., Watt, C.A., Matthews, C.J.D., Kiszka, J.J., Rosing-Asvid, A., McKinney, M.A., 2023. Varying Diet Composition Causes Striking Differences in Legacy and Emerging Contaminant Concentrations in Killer Whales across the North Atlantic. Environ. Sci. Technol. 57, 16109–16120. https://doi.org/10.1021/acs.est.3c05516

Rigét, F., Vorkamp, K., Eulaers, I., Dietz, R., 2020. Influence of climate and biological variables on temporal trends of persistent organic pollutants in Arctic char and ringed seals from Greenland. Environ. Sci.: Processes Impacts 22, 993–1005. https://doi.org/10.1039/C9EM00561G

Robinson, J.F., Pennings, J.L.A., Piersma, A.H., 2012. A Review of Toxicogenomic Approaches in Developmental Toxicology, in: Harris, C., Hansen, J.M. (Eds.), Developmental Toxicology, Methods in Molecular Biology. Humana Press, Totowa, NJ, pp. 347–371. https://doi.org/10.1007/978-1-61779-867-2_22

Roggeman, S., van den Brink, N., Van Praet, N., Blust, R., Bervoets, L., 2013. Metal exposure and accumulation patterns in free-range cows (Bos taurus) in a contaminated natural area: Influence of spatial and social behavior. Environ. Pollut. 172, 186–199. https://doi.org/10.1016/j.envpol.2012.09.006

Roos, A.M., Gamberg, M., Muir, D., Kärrman, A., Carlsson, P., Cuyler, C., Lind, Y., Bossi, R., Rigét, F., 2021. Perfluoroalkyl substances in circum-ArcticRangifer: caribou and reindeer. Environ. Sci. Pollut. Res. https://doi.org/10.1007/s11356-021-16729-7

Rose, N.L., Rose, C.L., Boyle, J.F., Appleby, P.G., 2004. Lake-Sediment Evidence for Local and Remote Sources of Atmospherically Deposited Pollutants on Svalbard. Journal of Paleolimnology 31, 499–513. https://doi.org/10.1023/B:JOPL.0000022548.97476.39

Routti, H., Aars, J., Fuglei, E., Hanssen, L., Lone, K., Polder, A., Pedersen, Å.Ø., Tartu, S., Welker, J.M., Yoccoz, N.G., 2017. Emission Changes Dwarf the Influence of Feeding Habits on Temporal Trends of Per- and Polyfluoroalkyl Substances in Two Arctic Top Predators. Environ. Sci. Technol. 51, 11996–12006. https://doi.org/10.1021/acs.est.7b03585

Routti, H., Berg, M.K., Lille-Langøy, R., Øygarden, L., Harju, M., Dietz, R., Sonne, C., Goksøyr, A., 2019. Environmental contaminants modulate the transcriptional activity of polar bear (Ursus maritimus) and human peroxisome proliferator-activated receptor alpha (PPARA). Sci Rep 9, 6918. https://doi.org/10.1038/s41598-019-43337-w

Routti, H., Lille-Langøy, R., Berg, M.K., Fink, T., Harju, M., Kristiansen, K., Rostkowski, P., Rusten, M., Sylte, I., Øygarden, L., Goksøyr, A., 2016. Environmental Chemicals Modulate Polar Bear (*Ursus maritimus*) Peroxisome Proliferator-Activated Receptor Gamma (PPARG) and Adipogenesis in Vitro. Environ. Sci. Technol. 50, 10708–10720. https://doi.org/10.1021/acs.est.6b03020

Rudnicka-Kępa, P., Zaborska, A., 2021. Sources, fate and distribution of inorganic contaminants in the Svalbard area, representative of a typical Arctic critical environment–a review. Environ. Monit. Assess. 193, 724. https://doi.org/10.1007/s10661-021-09305-6

Sait, S.T.L., Rinø, S.F., Gonzalez, S.V., Pastukhov, M.V., Poletaeva, V.I., Farkas, J., Jenssen, B.M., Ciesielski, T.M., Asimakopoulos, A.G., 2023. Occurrence and tissue distribution of 33 legacy and novel per- and polyfluoroalkyl substances (PFASs) in Baikal seals (Phoca sibirica). Sci. Total Environ. 889, 164096. https://doi.org/10.1016/j.scitotenv.2023.164096

Schaefer, K., Elshorbany, Y., Jafarov, E., Schuster, P.F., Striegl, R.G., Wickland, K.P., Sunderland, E.M., 2020. Potential impacts of mercury released from thawing permafrost. Nat. Commun. 11, 4650. https://doi.org/10.1038/s41467-020-18398-5

Scheuhammer, A., Braune, B., Chan, H.M., Frouin, H., Krey, A., Letcher, R., Loseto, L., Noël, M., Ostertag, S., Ross, P., Wayland, M., 2015. Recent progress on our understanding of the

biological effects of mercury in fish and wildlife in the Canadian Arctic. Sci. Total Environ. 509–510, 91–103. https://doi.org/10.1016/j.scitotenv.2014.05.142

Schrenk, D., Bignami, M., Bodin, L., Chipman, J.K., del Mazo, J., Grasl-Kraupp, B., Hogstrand, C., Hoogenboom, L. (Ron), Leblanc, J., Nebbia, C.S., Nielsen, E., Ntzani, E., Petersen, A., Sand, S., Vleminckx, C., Wallace, H., Barregård, L., Ceccatelli, S., Cravedi, J., Halldorsson, T.I., Haug, L.S., Johansson, N., Knutsen, H.K., Rose, M., Roudot, A., Van Loveren, H., Vollmer, G., Mackay, K., Riolo, F., Schwerdtle, T., 2020. Risk to human health related to the presence of perfluoroalkyl substances in food. EFS2 18. https://doi.org/10.2903/j.efsa.2020.6223

Schuler, T.V., Benestad, R.E., Isaksen, K., Kierulf, H.P., Kohler, J., Moholdt, G., Schmidt, L.S., 2025. Svalbard's 2024 record summer: An early view of Arctic glacier meltdown? Proc. Natl. Acad. Sci. U.S.A. 122, e2503806122. https://doi.org/10.1073/pnas.2503806122

Schuster, J.K., Gioia, R., Breivik, K., Steinnes, E., Scheringer, M., Jones, K.C., 2010. Trends in European Background Air Reflect Reductions in Primary Emissions of PCBs and PBDEs. Environ. Sci. Technol. 44, 6760–6766. https://doi.org/10.1021/es101009x

Schuster, P.F., Schaefer, K.M., Aiken, G.R., Antweiler, R.C., Dewild, J.F., Gryziec, J.D., Gusmeroli, A., Hugelius, G., Jafarov, E., Krabbenhoft, D.P., Liu, L., Herman-Mercer, N., Mu, C., Roth, D.A., Schaefer, T., Striegl, R.G., Wickland, K.P., Zhang, T., 2018. Permafrost Stores a Globally Significant Amount of Mercury. Geophys. Res. Lett. 45, 1463–1471. https://doi.org/10.1002/2017GL075571

Schuster, R.C., Gamberg, M., Dickson, C., Chan, H.M., 2011. Assessing risk of mercury exposure and nutritional benefits of consumption of caribou (Rangifer tarandus) in the Vuntut Gwitchin First Nation community of Old Crow, Yukon, Canada. Environ. Res. 111, 881–887. https://doi.org/10.1016/j.envres.2011.05.025

Schymanski, E.L., Zhang, J., Thiessen, P.A., Chirsir, P., Kondic, T., Bolton, E.E., 2023. Per- and Polyfluoroalkyl Substances (PFAS) in PubChem: 7 Million and Growing. Environ. Sci. Technol. 57, 16918–16928. https://doi.org/10.1021/acs.est.3c04855

Søndergaard, J., Elberling, B., Sonne, C., Larsen, M.M., Dietz, R., 2025. Stable isotopes unveil ocean transport of legacy mercury into Arctic food webs. Nat Commun 16, 5135. https://doi.org/10.1038/s41467-025-60356-6

Sonne, C., Dietz, R., Jenssen, B.M., Lam, S.S., Letcher, R.J., 2021. Emerging contaminants and biological effects in Arctic wildlife. Trends. Ecol. Evol. 36, 421–429. https://doi.org/10.1016/j.tree.2021.01.007

Sørmo, E.G., Ciesielski, T.M., Øverjordet, I.B., Lierhagen, S., Eggen, G.S., Berg, T., Jenssen, B.M., 2011. Selenium Moderates Mercury Toxicity in Free-Ranging Freshwater Fish. Environ. Sci. Technol. 45, 6561–6566. https://doi.org/10.1021/es200478b

Souza-Arroyo, V., Fabián, J.J., Bucio-Ortiz, L., Miranda-Labra, R.U., Gomez-Quiroz, L.E., Gutiérrez-Ruiz, M.C., 2022. The mechanism of the cadmium-induced toxicity and cellular response in the liver. Toxicology 480, 153339. https://doi.org/10.1016/j.tox.2022.153339

Spolaor, A., Scoto, F., Larose, C., Barbaro, E., Burgay, F., Bjorkman, M.P., Cappelletti, D., Dallo, F., De Blasi, F., Divine, D., Dreossi, G., Gabrieli, J., Isaksson, E., Kohler, J., Martma, T., Schmidt, L.S., Schuler, T.V., Stenni, B., Turetta, C., Luks, B., Casado, M., Gallet, J.-C., 2024. Climate change is rapidly deteriorating the climatic signal in Svalbard glaciers. The Cryosphere 18, 307–320. https://doi.org/10.5194/tc-18-307-2024

Staaland, H., 1986. Svalbardreinens ernæring, in: Svalbardreinen Og Dens Livsgrunnlag. Universitetsforlaget AS, pp. 72–91.

Steenhuisen, F., Van Den Heuvel-Greve, M., 2021. Exposure radius of a local coal mine in an Arctic coastal system; correlation between PAHs and mercury as a marker for a local mercury source. Environ Monit Assess 193, 499. https://doi.org/10.1007/s10661-021-09287-5

Steinnes, E., 1995. A critical evaluation of the use of naturally growing moss to monitor the deposition of atmospheric metals. Sci. Total Environ. 160–161, 243–249. https://doi.org/10.1016/0048-9697(95)04360-D

Stock, N.L., Furdui, V.I., Muir, D.C.G., Mabury, S.A., 2007. Perfluoroalkyl Contaminants in the Canadian Arctic: Evidence of Atmospheric Transport and Local Contamination. Environ. Sci. Technol. 41, 3529–3536. https://doi.org/10.1021/es062709x

Sysselmesteren, 2024. Søk om reinsjakt 2024 [WWW Document]. URL https://www.sysselmesteren.no/nyheter/2024/05/sok-om-reinsjakt-2024/ (accessed 10.14.25).

Sysselmesteren, 2019. Governor of Svalbard. Species that can be hunted [WWW Document]. URL https://www.sysselmesteren.no/en/hunting-trapping-and-fishing/jaktbare-arter/ (accessed 1.7.25).

Tajchman, K., Drabik, K., Ukalska-Jaruga, A., Janiszewski, P., Spustek, D., Wengerska, K., 2023. The screening method for use of wild pheasant feathers in the monitoring of environmental pollution with heavy metals. Sci. Rep. 13, 6540. https://doi.org/10.1038/s41598-023-33649-3

The European Commission, 2023. COMMISSION REGULATION (EU) 2023/915 of 25 April 2023 on maximum levels for certain contaminants in food and repealing Regulation (EC) No 1881/2006.

Trego, M.L., Hoh, E., Whitehead, A., Kellar, N.M., Lauf, M., Datuin, D.O., Lewison, R.L., 2019. Contaminant Exposure Linked to Cellular and Endocrine Biomarkers in Southern California Bottlenose Dolphins. Environ. Sci. Technol. 53, 3811–3822. https://doi.org/10.1021/acs.est.8b06487

Treu, G., Krone, O., Unnsteinsdóttir, E.R., Greenwood, A.D., Czirják, G.Á., 2018. Correlations between hair and tissue mercury concentrations in Icelandic arctic foxes (Vulpes lagopus). Sci. Total Environ. 619–620, 1589–1598. https://doi.org/10.1016/j.scitotenv.2017.10.143

Tyler, N.J.C., 1987. Natural limitation of the abundance of the high Arctic Svalbard reindeer. University Of Cambridge. https://doi.org/10.17863/CAM.20267

Tyler, N.J.C., Øritsland, N.A., 1989. Why don't Svalbard reindeer migrate? Ecography 12, 369–376. https://doi.org/10.1111/j.1600-0587.1989.tb00911.x

United Nations Economic Commission for Europe, 1998. The 1998 Aarhus Protocol on Heavy Metals.

United Nations Environment Programme, 2024. MINIMATA CONVENTION ON MERCURY.

United Nations Environment Programme, 2001. STOCKHOLM CONVENTION ON PERSISTENT ORGANIC POLLUTANTS (POPS).

Van Der Wal, R., Madan, N., Van Lieshout, S., Dormann, C., Langvatn, R., Albon, S.D., 2000. Trading forage quality for quantity? Plant phenology and patch choice by Svalbard reindeer. Oecologia 123, 108–115. https://doi.org/10.1007/s004420050995

Verreault, J., Gabrielsen, G.W., Bustnes, J.O., 2010. The Svalbard Glaucous Gull as Bioindicator Species in the European Arctic: Insight from 35 Years of Contaminants Research, in: Whitacre, D.M. (Ed.), Reviews of Environmental Contamination and Toxicology 205, Reviews of Environmental Contamination and Toxicology. Springer New York, New York, NY, pp. 77–116. https://doi.org/10.1007/978-1-4419-5623-1_2

Wang, W., Evans, R.D., Hickie, B.E., Rouvinen-Watt, K., Evans, H.E., 2014. Methylmercury accumulation and elimination in mink (Neovison vison) hair and blood: Results of a controlled feeding experiment using stable isotope tracers. Environ. Toxicol. Chem. 33, 2873–2880. https://doi.org/10.1002/etc.2762

Wang-Andersen, G., Utne Skaare, J., Prestrud, P., Steinnes, E., 1993. Levels and congener pattern of PCBs in arctic fox, Alopex lagopus, in Svalbard. Environmental Pollution 82, 269–275. https://doi.org/10.1016/0269-7491(93)90129-C

Wani, A.L., Ara, A., Usmani, J.A., 2015. Lead toxicity: a review. Interdisciplinary Toxicology 8, 55–64. https://doi.org/10.1515/intox-2015-0009

Wania, F., Mackay, D., 1993. Global Fractionation and Cold Condensation of Low Volatility Organochlorine Compounds in Polar Regions. Ambio 22, 10–18.

Węgrzyn, M.H., Wietrzyk, P., Lehmann-Konera, S., Chmiel, S., Cykowska-Marzencka, B., Polkowska, Ż., 2018. Annual variability of heavy metal content in Svalbard reindeer faeces as a result of dietary preferences. Environ Sci Pollut Res 25, 36693–36701. https://doi.org/10.1007/s11356-018-3479-8

Weihe, P., Grandjean, P., 2012. Cohort studies of Faroese children concerning potential adverse health effects after the mothers' exposure to marine contaminants during pregnancy. Acta Vet Scand 54, S7, 1751-0147-54-S1-S7. https://doi.org/10.1186/1751-0147-54-S1-S7

Wielsøe, M., Long, M., Ghisari, M., Bonefeld-Jørgensen, E.C., 2015. Perfluoroalkylated substances (PFAS) affect oxidative stress biomarkers in vitro. Chemosphere 129, 239–245. https://doi.org/10.1016/j.chemosphere.2014.10.014

Winquist, A., Hodge, J.M., Diver, W.R., Rodriguez, J.L., Troeschel, A.N., Daniel, J., Teras, L.R., 2023. Case–Cohort Study of the Association between PFAS and Selected Cancers among Participants in the American Cancer Society's Cancer Prevention Study II LifeLink Cohort. Environ Health Perspect 131. https://doi.org/10.1289/ehp13174

Wobeser, G., Nielsen, N.O., Schiefer, B., 1976. Mercury and Mink. II. Experimental methyl mercury intoxication. Can J Comp Med 40, 34–45.

Wojtuń, B., Polechońska, L., Pech, P., Mielcarska, K., Samecka-Cymerman, A., Szymański, W., Kolon, M., Kopeć, M., Stadnik, K., Kempers, A.J., 2019. Sanionia uncinata and Salix polaris as bioindicators of trace element pollution in the High Arctic: a case study at Longyearbyen, Spitsbergen, Norway. Polar Biol. 42, 1287–1297. https://doi.org/10.1007/s00300-019-02517-0

Wojtuń, B., Samecka-Cymerman, A., Kolon, K., Kempers, A.J., Skrzypek, G., 2013. Metals in some dominant vascular plants, mosses, lichens, algae, and the biological soil crust in various types of terrestrial tundra, SW Spitsbergen, Norway. Polar Biol. 36, 1799–1809. https://doi.org/10.1007/s00300-013-1399-0

Wren, C.D., Hunter, D.B., Leatherland, J.F., Stokes, P.M., 1987. The effects of poly chlorinated biphenyls and methylmercury, singly and in combination, on Mink. I: Uptake and toxic responses. Arch. Environ. Contam. Toxicol. 16, 441–447. https://doi.org/10.1007/BF01055265

Xue, Y., Tu, X., Nie, Y., Wu, L., Zhang, H., Wang, Z., Su, G., Liu, X., 2025. Secondary releases of legacy heavy metals to proglacial ecosystems from melting Arctic glaciers. Journal of Hazardous Materials 496, 139290. https://doi.org/10.1016/j.jhazmat.2025.139290

Yin, X., Xia, L., Sun, L., Luo, H., Wang, Y., 2008. Animal excrement: A potential biomonitor of heavy metal contamination in the marine environment. Sci. Total Environ. 399, 179–185. https://doi.org/10.1016/j.scitotenv.2008.03.005

Ynalvez, R., Gutierrez, J., Gonzalez-Cantu, H., 2016. Mini-review: toxicity of mercury as a consequence of enzyme alteration. Biometals 29, 781–788. https://doi.org/10.1007/s10534-016-9967-8

Young, C.J., Mabury, S.A., 2010. Atmospheric Perfluorinated Acid Precursors: Chemistry, Occurrence, and Impacts, in: De Voogt, P. (Ed.), Reviews of Environmental Contamination

and Toxicology Volume 208, Reviews of Environmental Contamination and Toxicology. Springer New York, New York, NY, pp. 1–109. https://doi.org/10.1007/978-1-4419-6880-7_1

Zaborska, A., Beszczyńska-Möller, A., Włodarska-Kowalczuk, M., 2017. History of heavy metal accumulation in the Svalbard area: Distribution, origin and transport pathways. Environmental Pollution 231, 437–450. https://doi.org/10.1016/j.envpol.2017.08.042

Zhou, J., Obrist, D., Dastoor, A., Jiskra, M., Ryjkov, A., 2021. Vegetation uptake of mercury and impacts on global cycling. Nat. Rev. Earth Environ. 2, 269–284. https://doi.org/10.1038/s43017-021-00146-y

Paper I

Check for updates

Tracing the footprints of Arctic pollution: Spatial variations in toxic and essential elements in Svalbard reindeer (*Rangifer tarandus platyrhynchus*) faeces

Malin Andersson Stavridis [a,b,*], Susanne Brix Røed [b], Brage Bremset Hansen [b,c], Øyvind Mikkelsen [a,d], Tomasz Maciej Ciesielski [a,b], Bjørn Munro Jenssen [a,b,*]

[a] *Department of Arctic Technology, University Centre in Svalbard (UNIS), P.O. Box 156, N-9171 Longyearbyen, Norway*
[b] *Department of Biology, Norwegian University of Science and Technology (NTNU), NO-7491 Trondheim, Norway*
[c] *Department of Terrestrial Ecology, Norwegian Institute for Nature Research (NINA), NO-7485 Trondheim, Norway*
[d] *Department of Chemistry, Norwegian University of Science and Technology (NTNU), NO-7491 Trondheim, Norway*

HIGHLIGHTS

- The elemental composition of Svalbard reindeer faeces was analysed.
- There were significant differences in several elements between populations.
- Local Arctic settlements may influence the bioavailability of toxic metals.
- Dietary differences between populations may also affect their toxic metal exposure.
- Faeces could be used to biomonitor trends in environmental pollution.

GRAPHICAL ABSTRACT
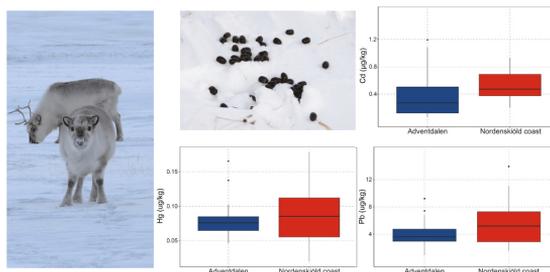
ARTICLE INFO

Editor: Frederic Coulon

Original content: Soil and Faecal Elemental Concentrations (Original data)

ABSTRACT

The Arctic is an accumulation zone of long-range transported pollution. In addition, local anthropogenic activities further contribute to regional pollution levels. The Svalbard reindeer (*Rangifer tarandus platyrhynchus*) is a suitable organism for studying and monitoring exposure to anthropogenic pollutants at the base of the terrestrial Arctic food web, and reindeer faeces have been promoted as non-invasive means of biomonitoring contaminant exposure. This study used HR-ICP-MS to analyse levels and composition of 16 elements in Svalbard reindeer faeces ($n = 96$) and soil ($n = 9$) from two locations on Svalbard, with the aim to assess whether local anthropogenic pollution influences element bioavailability. One of the sampling areas, the Nordenskiöld coast, is situated on the west coast of Spitsbergen, close to the Arctic Ocean and relatively far from local anthropogenic sources. The other sampling area, Adventdalen, is located further inland and close to Longyearbyen, the largest settlement of the archipelago. There was a significant difference in faecal elemental concentration and composition between the Adventdalen and Nordenskiöld coast reindeer populations. Elements of geogenic origin (e.g.,

Al, Cu and Fe) were found at higher levels in faeces from Adventdalen. In comparison, levels of Ca, Se and the toxic elements Cd and Pb were higher in faecal samples from the Nordenskiöld coast. The significantly higher levels of faecal Cd and Pb at Nordenskiöld coast may be due to marine input, dietary differences between the populations, or possible anthropogenic influence from the nearby settlement of Barentsburg. There was, however, a decoupling in elemental composition between faecal and soil samples, which may derive from a selective vegetational uptake of elements from the soil. The results suggest that reindeer are exposed to a range of elements and that faeces can be used to monitor the exposure to bioavailable environmental levels of both essential and toxic elements in terrestrial ecosystems.

1. Introduction

The anthropogenic disruption of the biogeochemical cycles of several heavy metals, such as mercury (Hg), cadmium (Cd) and lead (Pb), has led to their increase in the biosphere (Amos et al., 2013; Johansson et al., 2001). Besides several natural sources (e.g., volcanic eruptions or wildfires), there are many anthropogenic contributions to the presence of these metals in the environment. Emissions, for instance, occur during fossil fuel burning, waste incineration, chemical production and mining (Pacyna and Pacyna, 2001; Raj and Maiti, 2020). Heavy metal emissions occur globally, but the main contributions derive from highly industrialised regions. The chemical properties of these metals (e.g., high volatility or association with particles) allow them to travel long distances from their source of origin to pristine areas without much local pollution input, such as the Arctic (AMAP, 2005).

Once emitted to the atmosphere, the transport of these metals takes place either in gaseous (e.g., Hg) or fine particulate form (e.g., Cd and Pb) before being deposited through wet or dry deposition (Carpi, 1997; Thorne et al., 2018). Furthermore, Hg and Cd deposition is strongly associated with marine vectors such as sea spray aerosols and fog droplets (Garbarino et al., 2002). The residence time of gaseous Hg in the atmosphere is up to 18 months, which allows for long-range atmospheric transport to areas with few local emissions (Gworek et al., 2020). In contrast, the atmospheric transportation range of particulate matter strongly depends on variables such as the size of the particle, wind conditions and precipitation patterns. Elevated levels of metals such as Cd and Pb are mainly found close to their emission sources (AMAP, 2005). However, studies have found traceable levels of these metals several hundred km from where they were first emitted, demonstrating a gradient of pollution originating from highly polluting areas. This gradient has been observed in a range of matrices, such as moss (Berg and Steinnes, 1997), air (Strizhkina et al., 2022) and snow (Barrie et al., 1992), which indicates that low levels of these metals may travel from central Europe all the way to the Arctic (AMAP, 2005).

Svalbard is an archipelago in the high Arctic (74–81° N). The unique meteorological conditions, ice-bound environment and extreme changes in the light regime between seasons make the Arctic an accumulation zone of long-range transported (LRT) pollutants and thus a recipient of many toxic elements, including Hg, Cd and Pb (AMAP, 1997). In addition, local sources of emissions further contribute to the pollution of the Svalbard biosphere. These sources include burning fossil fuels in the local coal power plants in Longyearbyen and Barentsburg, vehicle exhaust and mining activities (Drotikova et al., 2020a, 2020b).

After being deposited, the pollutants can either undergo re-emission to the atmosphere, bind to soil organic matter or be taken up by vegetation via, for instance, leaf stomata or roots (Schaefer et al., 2020). The primary pathway of Hg into the terrestrial ecosystem is via vegetational (both vascular and non-vascular plants) atmospheric uptake, which accounts for approximately 90 % of all terrestrial Hg sequestration (Obrist et al., 2017; Zhou et al., 2021). In contrast, the major pathway for Cd and Pb accumulation in terrestrial environments is via root uptake from surrounding soils in vascular vegetation (Ismael et al., 2019; Pourrut et al., 2011). In general, non-vascular vegetation (e.g., bryophytes and lichens) efficiently accumulate atmospherically deposited heavy metals due to their overall high surface-to-mass ratio, long

lifespan, and slow growth rates (Bargagli, 2016; Gjengedal and Steinnes, 1990; Steinnes, 1995). This vegetational uptake of toxic elements subsequently acts as a pathway of exposure to herbivorous animals, such as the Svalbard reindeer (*Rangifer tarandus platyrhynchus*).

The Svalbard reindeer is the largest herbivore on Svalbard. It is found all over Spitsbergen and is mainly non-migratory (Le Moullec et al., 2019). It rarely leaves its limited home range unless motivated by restricted food accessibility in winter (Hansen et al., 2010). The primary diet of the reindeer consists of vascular plants (e.g., graminoids or polar willow) and bryophytes (Bjørkvoll et al., 2009), which may act as vectors of exposure to long-range and local pollution. It is therefore argued that the Svalbard reindeer is a suitable species for biomonitoring pollution levels in the Svalbard terrestrial ecosystem (Pacyna et al., 2018). As the Svalbard reindeer inhabit a relatively restricted area throughout their lifespan, their pollution levels further reflect any potential local contributions to the background environmental pollution load (Kinck, 2014).

Excretion via faeces is one of the main pathways for eliminating pollutants and excess nutrients from an organism. Faeces is, therefore, considered an indicator of environmental dietary exposure to elements. Faeces has also been promoted as a suitable matrix for biomonitoring since it can be collected using non-invasive methods (Pacyna-Kuchta et al., 2020).

There is currently a limited number of studies evaluating pollution levels in Arctic terrestrial ecosystems and organisms, especially few focusing on endemic species that do not migrate, such as Arctic herbivores (Dietz et al., 2022; Scheuhammer et al., 2015). In this study, we, therefore, investigated the composition of toxic (e.g., Hg, Cd and Pb) and essential elements in Svalbard reindeer faeces with the key aims to I) assess the Svalbard reindeer exposure to pollutants and II) evaluate whether reindeer inhabiting a presumed pristine area have a different faecal elemental composition as compared to reindeer living in areas more influenced by anthropogenic activities. Previous studies on Svalbard reindeer have not compared the faecal elemental composition between different reindeer populations to assess the potential anthropogenic influence on the bioavailability of elements, making this study design a novel approach to Arctic biomonitoring.

In addition to faeces, the elemental composition of soil was also analysed to evaluate to which extent spatial differences in soil composition are reflected in reindeer faeces.

2. Materials and methods

2.1. Study areas

The field sampling of faeces and soil was conducted at two separate areas on central Spitsbergen, the main island of the high Arctic archipelago of Svalbard. Adventdalen (78.2°N, 15.8°E) and Nordenskiöld coast (78.1°N, 13.6°E) were chosen as sampling sites due to the presumed difference in anthropogenic influence between them. Adventdalen is a valley close to Longyearbyen (Fig. 1), the largest settlement on the island (ca. 2500 inhabitants), where local sources of pollution include a coal power plant, an airport, vehicle exhaust, mining activities and releases from other local anthropogenic activities (Warner et al., 2019; Drotikova et al., 2020a, 2020b). In contrast, the Nordenskiöld

coast is likely more pristine as it is located near protected wildlife areas and further away from any direct point sources of pollution. Barentsburg, the second-largest settlement on Svalbard (ca. 500 inhabitants), is, however, situated approximately 15 km east of Nordenskiöld coast (Fig. 1), with pollution sources similar to Longyearbyen (Warner et al., 2019).

Although the sites are relatively close (~50 km), the weather conditions differ considerably between the two locations. The annual average precipitation in Adventdalen is around 190 mm, compared to 410 mm at the Nordenskiöld coast. Likewise, the annual average snow depth in Adventdalen is only half of that at Kapp Linné on the northern cape of the Nordenskiöld coast, 10.2 cm versus 20.1 cm (Førland et al., 2011; Norsk Klimaservicesenter, n.d.).

2.2. Field sampling

Faecal samples were collected in the spring (Feb-May) of 2014, 2015, and 2017. Reindeer were observed from 50 to 100 m away until faeces were excreted. Fresh faecal samples were subsequently collected within approximately 10 min. Nitrile gloves were worn during the collection to avoid sample contamination. Reindeer age (adult or calf), sex, and sampling coordinates were noted for each faecal sample. In total, 63 samples were collected from Adventdalen and 33 samples from the Nordenskiöld coast. The samples were kept in polyethylene zip-lock bags and stored frozen (−20 °C) until further analysis. Nine soil samples were collected from Adventdalen ($n = 5$) and Nordenskiöld coast ($n = 4$) in August 2021. The selected soil sampling sites had a developed organic surface layer and were entirely covered with vegetation. All samples were collected by cutting a square measuring $10 \times 10 \times 10$ cm using a stainless-steel knife. The samples were stored in paper bags until further sample preparations. For further details of the sampling, see Table A1-A2 and Fig. A1.

2.3. Element analysis

All faecal samples ($n = 96$) were freeze-dried using an Alpha 1–2 LDplus (Martin Christ Gefriertrocknungsanlagen GmbH, Osterode am Harz, Germany) for 24 h before being homogenised using a TissueLyser II with Teflon® chambers (Qiagen, Hilden, Germany).

The soil samples ($n = 9$) were left to airdry until all the water had evaporated (which was determined when weight changes were within ±5 % over a week). The edges of the dried samples were removed to avoid cross- or external contamination. The cores were homogenised using a cutting mill (RETSCH SM 100, 2-mm sieve, Retsch GmbH, Haan, Germany) and stored in polyethylene bags.

The dried, homogenised soil and faecal samples were digested in HNO_3. Approximately 0.5 g of faeces or 0.3 g of soil was transferred to an 18 ml polytetrafluoroethylene (PTFE) vial with 6.25 ml or 9 ml 50 % (v/v) HNO_3 acid (ultrapure grade, purified from HNO_3, AnalaR NOR-MAPUR®, VWR using a sub-boiling distillation system (Milestone, SubPur, Sorisole, BG, Italy)), respectively. The samples were digested using a high-pressure microwave system (Milestone Ultraclave, EMLS, Leutkirch, Germany) for 150 min and thereafter diluted with ultrapure water (Elga® Purelab Flex 4) up to 60 g (faecal samples) or 110 g (soil samples).

Additional blank (ultrapure water and HNO_3) and certified reference material samples (Virginia Tobacco leaves, INCT-PVTL-6, Institute of Nuclear Chemistry and Technology, Warszawa, Poland) were digested and analysed for quality assurance (for the results of the CRM analysis, see Table A3).

High-resolution inductively coupled plasma mass spectrometry (HR-ICP-MS, Thermo Finnigan model Element 2 instrument, Bremen, Germany) was used to identify and quantify the elemental composition of the samples. Sixteen elements (Al, As, Ca, Cd, Cu, Fe, Hg, Mg, Na, Ni, P, Pb, S, Se, Si, and Zn) were analysed, and all were detected above the limit of detection (LOD). The final concentration of each element was obtained by subtracting the average of the method blank concentration
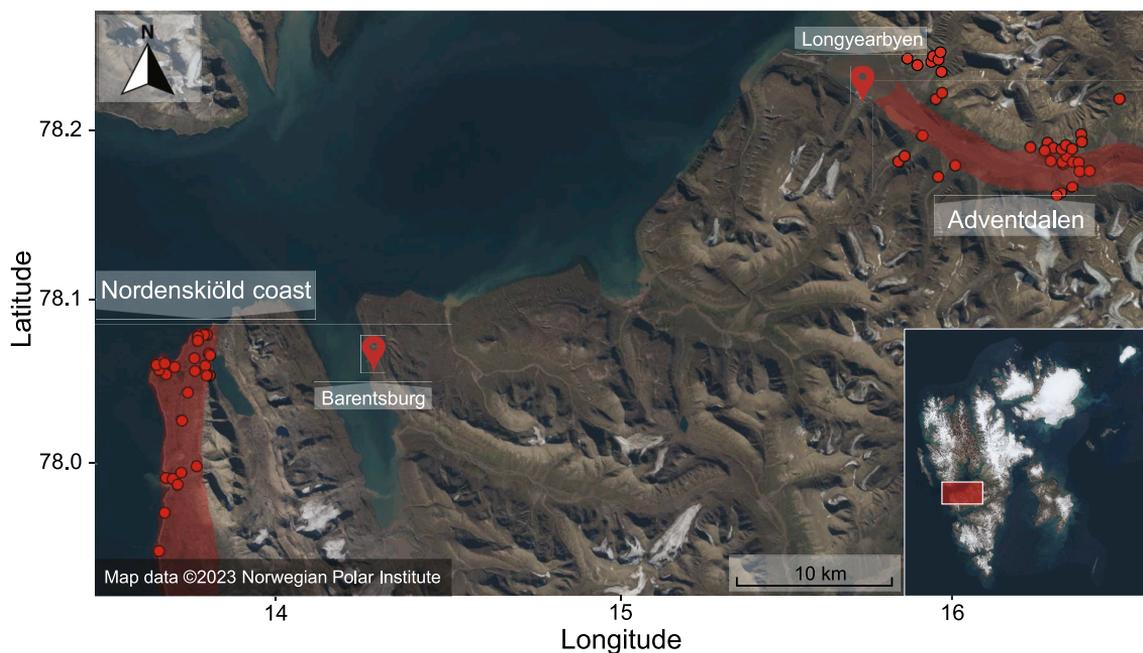

Fig. 1. Central Nordenskiöld land with Nordenskiöld coast and Adventdalen areas marked in dark red and the faecal sampling sites marked with red dots. The settlements of Barentsburg and Longyearbyen are indicated with red geotags.

of the respective element in all analysed samples.

The method detection limits for each element were either based on three times the standard deviation of the blanks or the instrument detection limits (IDL), depending on which method resulted in the highest value. The IDLs were estimated by analysing solutions with decreasing concentrations of the elements (see Table A4). The concentration that resulted in a relative standard deviation of approximately 25 % ($n = 3$ scans) was selected as IDL, with baseline corrections applied for these values.

The sixteen analysed elements include toxic metals (e.g., Cd, Pb, As and Hg) and essential elements (e.g., Se). Bedrock-related elements (i.e., Al, Ca, Cu, Fe, Mg, P, Si, Na, Ni, S and Zn) were primarily included as means to evaluate the origin of the toxic elements (Halbach et al., 2017; Klaassen et al., 2013).

2.4. Data treatment

All data were analysed for normal distribution using a Shapiro-Wilk test and for equal variances using Levene's test. None of the faecal elemental data was normally distributed, and a BoxCox transformation was applied to normalise the variables (using logarithmic or square root transformations). The transformed data did not fulfil the condition of equal variances, and non-parametric tests of analysis were therefore applied. A Mann-Whitney U test was employed to evaluate significant differences in faecal elemental concentrations between the two populations. Additionally, Spearman rank correlations were carried out to determine relationships among elements and between the elements and distance to the ocean at Nordenskiöld coast and Adventdalen, respectively. The results of the Spearman rank correlation were subsequently followed up using linear regression to plot the correlations. All soil elemental data (except for Pb) and parameters (TOC and pH) fulfilled the assumptions of parametric tests. A Student t-test was employed to evaluate significant differences in element concentration and soil parameters between the two locations (the Mann-Whitney U test was used for Pb).

Ocean distance was determined as the closest linear distance to the seashore from each sampling site. In Adventdalen, this distance was measured to the nearest point of Adventfjorden, while at Nordenskiöld coast, the distance was measured to the nearest open water of Isfjordbanken (see Fig. A1).

A principal component analysis (PCA) was conducted to explore the relationships among the elements and the spatial variation in the data

set. The variable ocean distance was excluded from the PCA due to considerable differences between the sampling sites, which contributed significantly to the PCs and thus masked the contributions of the elements. All numerical data were normalised by centring and scaling it (using the mean and standard deviation) before performing the PCA. All statistical analysis was done using R Statistical Software (V4.2.2, R Core Team, 2022).

3. Results

As presented in Table 1, the faecal elemental composition significantly differed between Adventdalen and the Nordenskiöld coast. Concentrations of Cu, Zn, Al, and Fe were significantly higher in faeces from Adventdalen than at Nordenskiöld coast ($p < 0.01$). In contrast, higher Cd, Se, Si, S, Ca, As and Pb concentrations were found in the Nordenskiöld coast faecal samples ($p < 0.01$ for all elements but $p < 0.05$ for As and Pb). The levels of Hg did not significantly differ between the two locations in the faeces. There were, however, significantly higher soil concentrations of Hg at the Nordenskiöld coast and higher levels of Cu, As and Zn in Adventdalen soil ($p < 0.05$). Total organic content and pH did not differ significantly between the soil samples from the two areas, where the samples from Nordenskiöld coast had an average TOC of 13.1 ± 6.9 % and an average pH of 5.3 ± 0.6, compared to the samples from Adventdalen which had an average TOC of 10.0 ± 5.1 % and an average pH of 5.0 ± 0.3.

The PCA for the faecal elemental composition resulted in three significant principal components (PCs), explaining 69.6 % of the total variation in the dataset. The observed spatial difference in reindeer faecal elemental composition (Table 1) is also indicated in the PCA score plot (Fig. 2A), which shows a slight separation between the two sampled reindeer populations along PC1. The clustering of elements in the loading plot (Fig. 2B) indicates that elements such as Al, Ni, Fe and Cu are strongly positively related. Many other elements are spread out along PC1 and PC2 without clustering. However, some elements are suggested to correlate positively, such as P and Ca or Cd and Si. The elements Hg, Se and Pb are somewhat grouped along PC2, which may suggest a positive relationship between these variables. PC3 (visualised in Fig. A2) likewise demonstrates similar loadings for Hg, Pb, Cd and Se, which supports a relationship between these elements. When comparing the score and loading plots, it is indicated that samples collected from Adventdalen had the highest levels of Al, Ni, Fe, and Cu. In contrast, faeces from the Nordenskiöld coast were characterised by higher

Table 1

Element concentrations in Svalbard reindeer (*R. tarandus platyrhynchus*) faeces and soil from Adventdalen and Nordenskiöld coast. Average elemental concentrations ± SD are provided in regular font. Concentrations given in italic font provide the median (min-max) values. All concentrations are given in µg/g (dry weight). Significantly different concentrations between the two sampling areas are indicated as (*) $p < 0.05$; (**) $p < 0.01$.

(n)	Faecal samples					Soil samples		
	Adventdalen (63) Mean ± SD	Median (min-max)	Sig.	Nordenskiöld coast (33) Mean ± SD	Median (min-max)	Adventdalen (5) Mean ± SD	Sig.	Nordenskiöld coast (4) Mean ± SD
Hg	0.0777 ± 0.0193	*0.0759 (0.0467-0.165)*		0.0869 ± 0.0429	*0.0853 (0.0200-0.178)*	0.112 ± 0.0426	(*)	0.178 ± 0.0291
Se	0.354 ± 0.104	*0.322 (0.184-0.655)*	(**)	0.560 ± 0.248	*0.516 (0.154-1.06)*	0.809 ± 0.385		0.725 ± 0.255
Cd	0.366 ± 0.288	*0.271 (0.0662-1.19)*	(**)	0.552 ± 0.297	*0.482 (0.203-1.72)*	0.374 ± 0.259		0.252 ± 0.123
As	3.22 ± 1.92	*3.05 (0.411-11.9)*	(*)	3.44 ± 3.97	*1.24 (0.322-13.4)*	11.3 ± 4.78	(*)	4.57 ± 1.11
Pb	3.81 ± 1.63	*3.62 (0.887-9.21)*	(*)	5.36 ± 2.91	*5.19 (1.51-13.9)*	12.5 ± 3.72		14.5 ± 1.91
Cu	8.41 ± 2.00	*8.22 (4.49-14.8)*	(**)	5.71 ± 2.01	*5.32 (2.50-13.3)*	16.2 ± 2.80	(*)	10.5 ± 3.8
Ni	10.6 ± 3.73	*10.5 (3.11-21.7)*	(**)	4.50 ± 2.59	*4.22 (1.85-17.5)*	24.3 ± 4.40		16.9 ± 3.91
Zn	78.7 ± 25.9	*75.8 (39.2-178)*	(**)	57.4 ± 16.6	*55.3 (28.9-87.1)*	80.0 ± 15.4	(*)	50.2 ± 14.1
Na	636 ± 360	*600 (110-2990)*		723 ± 644	*538 (187-3430)*	432 ± 78.8		1030 ± 682
Si	1390 ± 330	*1380 (780-2290)*	(**)	1960 ± 362	*2000 (1330-2530)*	1390 ± 707		1670 ± 458
P	1520 ± 459	*1360 (893-2930)*		1583 ± 482	*1550 (621-2980)*	802 ± 124		1050 ± 382
S	1780 ± 208	*1750 (1370-2270)*	(**)	3160 ± 2490	*2040 (679-10700)*	1270 ± 424		1290 ± 315
Mg	2600 ± 499	*2560 (1440-4110)*	(**)	4820 ± 3290	*3390 (1390-16500)*	4690 ± 563		5840 ± 981
Fe	7690 ± 3860	*6980 (1080-18000)*	(**)	3960 ± 1410	*3930 (1530-9120)*	25,700 ± 8890		20,900 ± 4080
Al	9950 ± 5260	*9950 (1340-28600)*	(**)	4220 ± 1480	*4260 (1550-9520)*	29,500 ± 7330		2570 ± 7030
Ca	16,000 ± 360	*15900 (8230-21500)*	(**)	30,700 ± 810	*30700 (11700-57000)*	6870 ± 3420		8560 ± 7130

Fig. 2. A. PCA score plot for Svalbard reindeer (*R. tarandus platyrhynchus*) faecal elemental composition from Adventdalen and Nordenskiöld coast. PC1 and PC2 explain 51.5% of the variance in the data set.

B. PCA loading plot for selected elements in Svalbard reindeer (*R. tarandus platyrhynchus*) faeces. PC1 and PC2 explain 51.5% of the variance in the data set. Elements with high loadings have long arrows, while elements with lower loading have shorter arrows. See Table A5 for the component matrix, presenting the loading of each element on the PCs.

Fig. 3. Spearman correlation heat maps demonstrating the relationship between faecal elemental concentrations and distance to the ocean (Ocean) in samples from Adventdalen (*n* = 63) and Nordenskiöld coast (*n* = 33). Values in each box represent the Spearman correlation coefficient (r), where |r| > 0.2 = p < 0.05, |r| > ±0.3 = p < 0.01 and |r| > ±0.4 = p < 0.001.

concentrations of Mg, S, Ca, Si, Se and Cd, P, and Na to a lesser extent.

The clustering in the PCA loading plot indicated positive relationships between many elements in the faecal samples. Site-specific correlations between variables are shown in Fig. 3 (Spearman correlation). Strong positive correlations between Hg and Se (p < 0.001) and between Cd and Zn (p < 0.001) were found in the faecal samples from both locations (see Fig. 4). The molar ratio between Se:Hg was, on average, 13.6. Elements Al, As, Cu, Fe and Mg were positively associated. The relationships between these elements were, however, stronger in the faecal samples from Adventdalen (p > 0.001) compared to Nordenskiöld

coast (significant correlation between Fe, Al and Ni, p > 0.001). Pb was strongly correlated to Hg in Nordenskiöld coast (p < 0.001) but had a stronger relationship to elements such as Al, Cu, Fe and Ni (p < 0.001) in Adventdalen. There were no strong correlations between the elements and ocean distance (Fig. 3). Overall, no indications of any relationship between faecal elemental concentrations and variables such as age, sex or year of sampling were identified (using Mann-Whitney *U* test).

4. Discussion

The faecal elemental concentrations of the toxic elements Hg, Pb and Cd are overall comparable to concentrations reported in a previous study, where Pacyna et al. (2019) detected similar ranges of Hg (0.069 ± 0.016 µg/g), Pb (5.23 ± 3.05 µg/g) and Cd (0.866 ± 0.411 µg/g) in fresh Svalbard reindeer faeces from Spitsbergen. There were, however, significant site-specific differences between the faecal elemental composition in samples from Adventdalen and Nordenskiöld coast. This suggests that, despite the two investigated sites being relatively close, there is a spatial influence on elemental exposure, uptake, or excretion of specific elements in the reindeer.
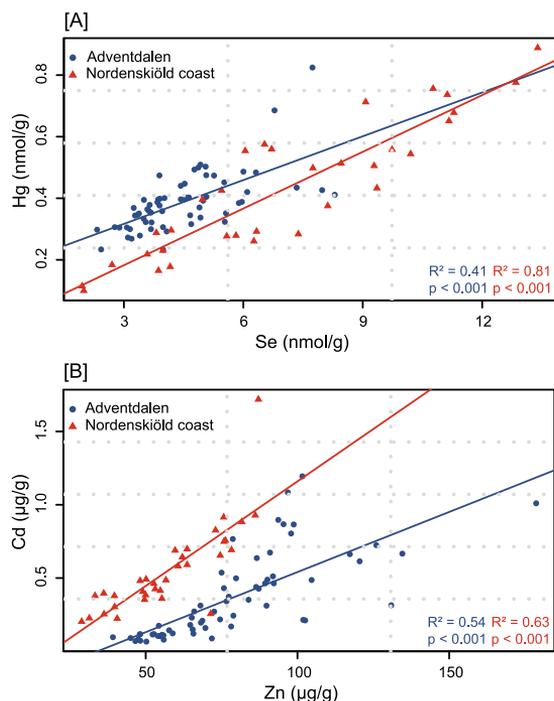
Fig. 4. Correlation plots between [A] Hg and Se and [B] between Cd and Zn in the faecal samples.

4.1. Dietary differences

Reindeer are exposed to elements through multiple pathways, but their exposure mainly derives from the vegetation on which they feed. The elemental composition of vegetation is affected by atmospheric deposition and soil composition, which further depends on the geochemical characteristics of the underlying bedrock (Ismael et al., 2019; Pourrut et al., 2011). For instance, Kłos et al. (2017) reported a species-dependent preference in metal absorption from the underlying soil when evaluating the metal accumulation capacity in different species of vascular plants on Svalbard. Mosses and lichen are known accumulators of atmospheric pollutants as they lack roots and depend on the ambient atmosphere for nutrients and water, making them susceptible to the absorption of pollutants from the air (Bargagli, 2016). Different species of plants will, therefore, differ in their elemental composition. When comparing the uptake of metals in lichen, bryophytes (i.e., mosses) and vascular vegetation, a study found the highest levels of Pb (2.1–2.4 µg/g dw) in lichen and mosses, Hg (0.033 µg/g dw) in mosses and Cd (0.52 µg/g dw) in vascular vegetation such as *Salix polaris* (Wojtuń et al., 2013).

The Svalbard reindeer diet consists of graminoids, forbs, bryophytes, and shrubs. Their diet undergoes seasonal shifts based on the availability of the different vegetation species and is in late winter dominated by *S. polaris* (Bjørkvoll et al., 2009). Similarly to other *Salix* spp., *S. polaris* is a phytoaccumulator (i.e., a plant with the capacity to absorb, accumulate and tolerate elevated levels of, for instance, heavy metals) (Pulford, 2003). A study conducted in Svalbard found levels of Cd up to sixty times higher in the stems of the plant than in the underlying soil (Wojtuń et al., 2019). *S. polaris* is widespread over the archipelago of Svalbard and has been identified in both Adventdalen and Nordenskiöld coast (Artsdatabanken, n.d.). The fraction of *S. polaris* in the diet of the reindeer depends on the presence of other species of vegetation, which may

influence their exposure to elements such as Cd. In Svalbard reindeer, it has been reported that season-dependent (i.e., winter versus summer) dietary shifts can result in variations in faecal concentrations of, for instance, Cd and Fe (Węgrzyn et al., 2018). As a result, differences in faecal elemental composition between the two populations may reflect potential dietary differences. There is, however, a lack of studies on the presence of different species of vegetation at the two sampling sites, as well as the dietary differences among these two reindeer populations, creating difficulties in proving this theory.

4.2. Element correlations in faeces

The excretion of elements via faeces is one elimination pathway of toxic metals in exposed organisms (Pacyna et al., 2019; Roggeman et al., 2013). Faecal concentrations of essential elements (i.e., nutrients that are vital for homeostasis) mainly depend on the current physiological requirements of the individual. Hence, to what extent the faecal concentration reflects the intake of essential elements will depend on the uptake rate, potential physiological needs, and elimination rates via faecal excretion.

Se is an essential element as it is a central part of the biological antioxidant defence system and, therefore, an active element in the protection against toxic metals. The protection mechanism includes reacting with metals such as Hg and forming an equimolar, inactive Se—Hg complex, often found in the liver (Ikemoto et al., 2004; Nève, 1991). High exposure to Hg and, thus, increased formation rates of these complexes can cause a deficiency of Se. However, Romero et al. (2016) and Sørmo et al. (2011) argue that a molar ratio of Se:Hg > 1 ensures a sufficient supply of Se to sustain its physiological function while, in addition, protecting against Hg. As the relationship between Se and Hg in this study far exceeded a molar ratio of 1 (minimum faecal Se:Hg = 7.8), Hg exposure does not seem to limit the Se availability for physiological functions or result in Hg toxicity. Although, considering that the analysed matrix is faeces, the elemental ratio in these samples provides us with little insight into the relationship between Hg and Se in internal organs such as the liver, where these Hg—Se complexes are stored, or whether there are sufficient internal concentrations of this essential element to sustain homeostasis.

As shown in Fig. 3, Cd concentrations strongly correlate to Zn in the faecal samples from both locations. To our knowledge, no previous studies have reported a correlation between these two elements in faeces. However, these elements are known to positively correlate in other matrices, such as in ore deposits or vegetation (Kabata-Pendias and Szteke, 2015). Multiple studies have evaluated the elemental composition of Svalbard vegetation. Some studies found Cd and Zn to positively correlate in species such as *S. polaris* or the moss *Dicranum angustum* (Ma et al., 2020; Wei et al., 2022; Wojtuń et al., 2019), while other studies did not (Krajcarová et al., 2016). Faecal levels of metals, including toxic metals, have been reported to correspond well with the dietary intake of these elements in grazers (*Bos taurus*) (Roggeman et al., 2013). This both suggests that increased exposure to metals does not necessarily result in higher intestinal uptake and that faecal metal levels depend on current exposure rather than representing previous assimilation that has been taken up, circulated, and subsequently eliminated via faeces. Consequently, the faecal elemental composition most likely depends on the elemental composition of the plants that comprise the diet. This may help explain the positive Cd—Zn relationship observed in the faeces, as these elements correlate in plant species consumed by reindeer (Ma et al., 2020; Wei et al., 2022; Wojtuń et al., 2019). Whether plant elemental composition also explains the significant correlation between faecal Hg and Se remains uncertain. To our knowledge, no studies have evaluated this correlation in Arctic vegetation or species included in the Svalbard reindeer diet. However, a relationship between Hg and Se has been observed in vegetation such as rice (*Oryza sativa*), suggesting a positive faecal Hg—Se correlation could derive from dietary exposure (Bai et al., 2019; Zhang et al., 2012).

4.3. Site-specific differences

Reindeer can consume both gravel and soil during grazing, further emphasising the influence of the elemental composition of the underlying bedrock on elemental exposure (Makarov et al., 2022; Orpin et al., 1985). The dominant bedrock material in Adventdalen is composed of different sandstone forms, which are dominated by Si, Al and Fe in descending order of concentrations (Middleton, 1960). In contrast, the Nordenskiöld coast has a more complex bedrock composition, which, in addition to sandstone, consists of chert and siliceous shale. These bedrocks contain high levels of Si, and limestone, which is composed of Ca (Cressman, 1967). However, despite the above-mentioned theoretical geochemical differences in parent bedrock material, only Zn, Cu, As and Hg concentrations differed significantly among all the analysed elements in the soil samples from the two sites. The differences in Zn, Cu and As in soil were reflected in the faeces, as these elements were significantly higher in faecal samples from Adventdalen, suggesting that these elements are of geogenic origin. There were, however, no differences in Hg faecal levels between the two locations.

Although the elemental distribution of the soil from the two locations may be similar, it is important to acknowledge that these elements might be incorporated into different types of minerals or be bound to different types of organic matter, which affects their bioavailability. Soil mobility of metals bound to organic matter is, for instance, higher than the mobility of metals bound to sulfide minerals, which have low solubility and are relatively stable under normal environmental conditions (John and Leventhal, 1995). Mobile elements are available to be taken up by vegetation and subsequently enter the food chain when the plants are grazed upon. Soil bioavailability may, therefore, partly explain the decoupling between the faecal and soil elemental composition at the two locations.

The organic content of Svalbard soils is, in general, far lower than in the soil from mainland Norway (Halbach et al., 2017). This results from slow rates of organic soil formation and cryoturbation, which allows for the mixing of the organic and mineral layers (Nygård et al., 2012). This can be observed in the low TOC content in the soil samples from both study sites, where the organic carbon percentage ranged between 3.3 % (in Adventdalen) and 21.6 % (at Nordenskiöld coast). Consequently, the sampled surface soils are highly characterised by the underlying mineral soils, which should reflect the differences in parent bedrock material between the two sampling areas better. The lack of significant difference in most analysed elements in the soil may be ascribed to poor statistical power, as few soil samples were included in the study ($n = 5$ and $n = 4$ for Adventdalen and Nordenskiöld coast, respectively).

In addition to the geochemical background, atmospheric deposition may influence the elemental composition in the sampling areas. The scavenging of airborne pollutants and aerosols through wet deposition has been found to contribute to levels of Cd and Hg in Arctic regions (Macdonald et al., 2005; Outridge et al., 2002; Pearson et al., 2019). Annual precipitation at Nordenskiöld coast is higher than in Adventdalen (Førland et al., 2011). The Nordenskiöld coast is furthermore closer to the sea and the Arctic Ocean when compared to Adventdalen. The on-land deposition of atmospheric Cd, to some extent, depends on the formation of particles with aerosols and, therefore, increases close to sources of open water where sea salt and water droplets act as scavengers (AMAP, 2005). This may explain the significantly higher levels of Cd in the Nordenskiöld coast faecal samples. However, no differences in Cd concentration were found in the soil samples from the two locations. On the contrary, Hg concentrations do not differ in faeces from the two study sites, while concentrations of Hg were significantly higher in soil samples from the Nordenskiöld coast than in Adventdalen. Atmospheric mercury depletion events (AMDEs) are enhanced in areas closer to the sea as the ocean is a source of halogens, which initiate depletion by oxidising atmospheric elemental mercury (Lindberg et al., 2002; Steffen et al., 2008). Increased deposition levels at Nordenskiöld coast may explain the higher levels of Hg in soil. The dominant Hg uptake pathway

in vegetation is, however, via the atmosphere, while root uptake from the soil is negligible (Zhou et al., 2021). Increased levels of Hg in the soil may, therefore, not translate into increased vegetation levels, which might explain the concentration decoupling between the soil and faecal Hg concentrations.

Another vector of pollution in Arctic terrestrial ecosystems is marine birds. Seabirds such as the little auk (*Alle alle*) feed off the marine ecosystem and breed in bird cliffs on the slopes of certain high land-based cliffs. Through processes such as the excretion of faeces or the dropping of food when flying, they transfer nutrients, elements and contaminants, such as toxic elements, from the marine to the terrestrial environment as they move between the ecosystems (Zwolicki et al., 2016). As a result, increased levels of soil nutrients such as Ca, Cu, Se, Zn and toxic elements such as Cd have been found in areas in which seabirds breed (Mallory et al., 2015; De La Peña-Lastra et al., 2022). This may help explain why levels of Ca, Se and Cd are significantly higher in the faecal samples from Nordenskiöld coast. There is moreover a strong correlation between Ca and P in Nordenskiöld coast, which is not observed in the samples from Adventdalen. P is a major component of bird guano (Duda et al., 2020) and would, therefore, correlate well with other guano-related elements such as Ca. There are, however, only marginal differences in P concentrations in the reindeer faeces from the two sampling sites. There is a bird reserve on the northernmost point of Nordenskiöld coast, but there are no seabird cliffs in the area. Therefore, it is not likely that the increased levels of Cd derive from bird guano.

Despite Cd levels being higher in Nordenskiöld coast faeces, the heat maps (Fig. 3) do not support a relationship between toxic metals and the distance to the ocean. It is, however, important to note that the reindeer's digestive retention time ranges from 21 to 69 h (Aagnes and Mathiesen, 1994). Hence, the location of excretion can differ from that of grazing, thus confounding the possible association between the distance from the ocean and the faecal elemental composition.

The heat maps, however, showed significant correlations between several elements in the faecal samples. At Nordenskiöld coast there was a significant positive relationship between Pb and Hg. In Adventdalen, the results instead showed significant positive relationships between Pb and several elements of mineral origin, such as Cu, Al, Fe, and As. The Pb levels were, moreover, higher in faeces from the Nordenskiöld coast, and we, therefore, suggest that the origin of Pb may differ at the two sampling sites. We propose that the Pb detected in the faecal samples from Adventdalen could be of mineral origin, while the positive relationship between Hg and Pb, together with increased levels of Pb at Nordenskiöld coast, may suggest anthropogenic input.

4.4. Local sources of pollution

Most Hg in the Arctic originates from anthropogenic activities and long-range atmospheric transport from sub-Arctic latitudes (Dastoor et al., 2022). However, as mentioned, a few local sources also contribute to the overall levels of several toxic elements in Svalbard (Drotikova et al., 2020a). As shown, the levels of the toxic elements Cd and Pb were higher in faeces from the Nordenskiöld coast than from Adventdalen. Additionally, Hg levels were higher in the soil at Nordenskiöld coast than in Adventdalen. This potentially contradicts our presumption that the Nordenskiöld coast is less affected by anthropogenic influences. There are no significant local point sources of pollutants on the Nordenskiöld coast. The area is, however, relatively close to the nearby settlement of Barentsburg (15 km), where there is an active coal power plant (Dekhtyareva et al., 2016). The annual general wind direction in Barentsburg is northeasterly, which may transport a considerable amount of the power plant exhaust towards the Nordenskiöld coast (Åkerman, 1980; The Norwegian Meteorological Institute, n.d.). Depending on the chemical composition of the coal used in these power plants, combustion can be a source of fossil fuel-related elements, such as heavy metals (Lewińska-Preis et al., 2009). Anthropogenic activity in Barentsburg could, therefore, to some extent, be the common origin of

both Hg and Pb, which may explain their relationship at this site. The same relationship between Hg and Pb was absent in faecal samples from Adventdalen despite its closeness to the coal power plant in Longyearbyen (3–18 km). The general annual wind direction in Adventdalen is, however, mostly southeasterly (Dekhtyareva et al., 2022), which is downwind from Longyearbyen and its associated point sources. The sampled reindeer in Adventdalen may, therefore, not be exposed to the potential anthropogenic contributions of toxic metals originating from the Longyearbyen powerplant.

The elemental composition of faeces is the final product of the exposure, intestinal uptake and excretion of surplus essential or toxic elements. Roggeman et al. (2013), for instance, reported that faecal concentration of elements corresponded well with dietary exposure. The levels of elements in faeces do, therefore, not necessarily reflect the levels in other tissues or organs of the organism. Although it has been argued that faecal samples can be used as a proxy for exposure to pollutants in terrestrial ecosystems (Pacyna et al., 2019), relationships between faecal concentrations of toxic metals and internal soft tissue and organ concentrations have been found in some Arctic species, but not yet been assessed in reindeer (Dietz et al., 2009). The biomonitoring of faeces should, therefore, instead be used as an indicator of exposure trends in Arctic terrestrial organisms rather than being used to derive the risk of exposure to, for instance, toxic metals. We furthermore suggest that faecal samples of reindeer can be applied to assess spatial and temporal differences and trends in patterns and levels of toxic elements (heavy metals) at the base of the food chain (i.e. in reindeer diet) in the Arctic.

5. Conclusions

In this study, we investigated the composition of toxic (e.g., Hg, Cd and Pb) and essential elements (e.g., Se) in Svalbard reindeer faeces with the key aim to I) assess the exposure of Svalbard reindeer to pollutants and II) evaluate whether reindeer inhabiting pristine areas have a different faecal elemental composition as compared to reindeer living in areas more influenced by anthropogenic activities. We found a significant difference in faecal elemental concentration and composition between Adventdalen and Nordenskiöld coast. Elements of geogenic origin (e.g., Al, Cu and Fe) were found at higher levels in faeces from Adventdalen. In comparison, levels of Ca, Se and the toxic elements Cd and Pb were higher in faecal samples from the Nordenskiöld coast. The significantly higher levels of Cd found in faeces from the Nordenskiöld coast may originate from the ocean or, considering the easterly wind patterns in late winter, from the coal combustion power plant in Barentsburg, located 15 km east of Nordenskiöld coast. The increased levels of Pb at Nordenskiöld coast faeces might also originate from the powerplant. These results indicate that reindeer living on the Nordenskiöld coast may be exposed to local anthropogenic pollution.

However, the same patterns in faecal elemental composition at the two study locations were not observed in the elemental composition of the soil samples from the same areas. This decoupling between matrices may be due to the reindeer diet, as plants have species-dependent uptake of elements from their surrounding environment. Potential dietary differences between the two sampled populations may also explain the differences in faecal elemental composition, but further studies on the elemental composition of the plants that comprise the reindeer diet are required to evaluate this.

We suggest that faecal samples of reindeer can be applied to assess spatial and temporal differences and trends in patterns and levels of toxic elements at the base of the terrestrial food chain in the Arctic.

Funding

CRediT authorship contribution statement

Malin Andersson Stavridis: Formal analysis, Writing – original draft, Visualization. **Susanne Brix Røed:** Conceptualization, Investigation. **Brage Bremset Hansen:** Investigation, Writing – review & editing. **Øyvind Mikkelsen:** Investigation, Writing – review & editing. **Tomasz Maciej Ciesielski:** Conceptualization, Writing – review & editing. **Bjørn Munro Jenssen:** Conceptualization, Supervision, Writing – review & editing.

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

Soil and Faecal Elemental Concentrations (Original data) (Mendeley Data).

Acknowledgements

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.scitotenv.2023.167562.

References

Aagnes, T.H., Mathiesen, S.D., 1994. Food and snow intake, body mass and rumen function in reindeer fed lichen and subsequently starved for 4 days. Rangifer 14, 33. https://doi.org/10.7557/2.14.1.1131.

Åkerman, J., 1980. Studies on Periglacial Geomorphology in West Spitsbergen, Meddelanden från Lunds Universitets Geografiska Institution. Avhandlingar. Inst., Univ. Lund.

AMAP, 1997. Arctic Pollution Issues: A State of the Arctic Environment Report. Arctic Monitoring and Assessment Programme (AMAP), Oslo, Norway xii+188 pp.

AMAP, 2005. AMAP Assessment 2002: Heavy Metals in the Arctic. Arctic Monitoring and Assessment Programme (AMAP), Oslo.

Amos, H.M., Jacob, D.J., Streets, D.G., Sunderland, E.M., 2013. Legacy impacts of all-time anthropogenic emissions on the global mercury cycle: global impacts of legacy mercury. Glob. Biogeochem. Cycles 27, 410–421. https://doi.org/10.1002/gbc.20040.

Artsdatabanken, n.d. *Salix polaris* on Svalbard [WWW Document]. URL https://artsdatabanken.no/ (accessed 10.11.22).

Bai, X., Li, Y., Liang, X., Li, H., Zhao, J., Li, Y.-F., Gao, Y., 2019. Botanic metallomics of mercury and selenium: current understanding of mercury-selenium antagonism in plant with the traditional and advanced technology. Bull. Environ. Contam. Toxicol. 102, 628–634. https://doi.org/10.1007/s00128-019-02628-8.

Bargagli, R., 2016. Moss and lichen biomonitoring of atmospheric mercury: a review. Sci. Total Environ. 572, 216–231. https://doi.org/10.1016/j.scitotenv.2016.07.202.

Barrie, L.A., Gregor, D., Hargrave, B., Lake, R., Muir, D., Shearer, R., Tracey, B., Bidleman, T., 1992. Arctic contaminants: sources, occurrence and pathways. Sci. Total Environ. 122, 1–74. https://doi.org/10.1016/0048-9697(92)90245-N.

Berg, T., Steinnes, E., 1997. Recent trends in atmospheric deposition of trace elements in Norway as evident from the 1995 moss survey. Sci. Total Environ. 208, 197–206. https://doi.org/10.1016/S0048-9697(97)00253-2.

Bjørkvoll, E., Pedersen, B., Hytteborn, H., Jónsdóttir, I.S., Langvatn, R., 2009. Seasonal and interannual dietary variation during winter in female Svalbard reindeer (*Rangifer tarandus platyrhynchus*). Arct. Antarct. Alp. Res. 41, 88–96. https://doi.org/10.1657/1523-0430-41.1.88.

Carpi, A., 1997. Mercury from combustion sources: a review of the chemical species emitted and their transport in the atmosphere. Water Air Soil Pollut. 98, 241–254. https://doi.org/10.1023/A:1026429911010.

Cressman, E.R., 1967. Nondetrital Siliceous Sediments, in: Data of Geochemistry, sixth ed.

Dastoor, A., Wilson, S.J., Travnikov, O., Ryjkov, A., Angot, H., Christensen, J.H., Steenhuisen, F., Muntean, M., 2022. Arctic atmospheric mercury: sources and changes. Sci. Total Environ. 839, 156213. https://doi.org/10.1016/j.scitotenv.2022.156213.

De La Peña-Lastra, S., Pérez-Alberti, A., Ferreira, T.O., Huerta-Díaz, M., Otero, X.L., 2022. Global deposition of potentially toxic metals via faecal material in seabird colonies. Sci. Rep. 12, 22392. https://doi.org/10.1038/s41598-022-26905-5.

Dekhtyareva, A., Edvardsen, K., Holmén, K., Hermanson, O., Hansson, H.-C., 2016. Influence of local and regional air pollution on atmospheric measurements in Ny-Ålesund. Int. J. Sustain. Dev. Plan. 11, 578–587. https://doi.org/10.2495/SDP-V11-N4-578-587.

Dekhtyareva, A., Hermanson, M., Nikulina, A., Hermansen, O., Svendby, T., Holmén, K., Graversen, R.G., 2022. Springtime nitrogen oxides and tropospheric ozone in Svalbard: results from the measurement station network. Atmos. Chem. Phys. 22, 11631–11656. https://doi.org/10.5194/acp-22-11631-2022.

Dietz, R., Outridge, P.M., Hobson, K.A., 2009. Anthropogenic contributions to mercury levels in present-day Arctic animals—a review. Sci. Total Environ. 407, 6120–6131. https://doi.org/10.1016/j.scitotenv.2009.08.036.

Dietz, R., Letcher, R.J., Aars, J., Andersen, M., Boltunov, A., Born, E.W., Ciesielski, T.M., Das, K., Dastnai, S., Derocher, A.E., Desforges, J.-P., Eulaers, I., Ferguson, S., Hallanger, I.G., Heide-Jørgensen, M.P., Heimbürger-Boavida, L.-E., Hoekstra, P.F., Jenssen, B.M., Kohler, S.G., Larsen, M.M., Lindstrøm, U., Lippold, A., Morris, A., Nabe-Nielsen, J., Nielsen, N.H., Peacock, E., Pinzone, M., Rigét, F.F., Rosing-Asvid, A., Routti, H., Siebert, U., Stenson, G., Stern, G., Strand, J., Søndergaard, J., Treu, G., Víkingsson, G.A., Wang, F., Welker, J.M., Wiig, Ø., Wilson, S., Sonne, C., 2022. A risk assessment review of mercury exposure in Arctic marine and terrestrial mammals. Sci. Total Environ. 154445. https://doi.org/10.1016/j.scitotenv.2022.154445.

Drotikova, T., Albinet, A., Halse, A.K., Reinardy, H.C., Ali, A.M., Kallenborn, R., 2020a. Svalbard local air contamination by PAHs and nitro- and oxy-PAHs. In: Norwegian Environmental Chemistry Symposium (NECS 2020).

Drotikova, T., Ali, A.M., Halse, A.K., Reinardy, H.C., Kallenborn, R., 2020b. Polycyclic aromatic hydrocarbons (PAHs) and oxy- and nitro-PAHs in ambient air of the Arctic town Longyearbyen, Svalbard. Atmos. Chem. Phys. 20, 9997–10014. https://doi.org/10.5194/acp-20-9997-2020.

Duda, M.P., Glew, J.R., Michelutti, N., Robertson, G.J., Montevecchi, W.A., Kissinger, J.A., Eickmeyer, D.C., Blais, J.M., Smol, J.P., 2020. Long-term changes in terrestrial vegetation linked to shifts in a colonial seabird population. Ecosystems 23, 1643–1656. https://doi.org/10.1007/s10021-020-00494-8.

Førland, E.J., Benestad, R., Hanssen-Bauer, I., Haugen, J.E., Skaugen, T.E., 2011. Temperature and precipitation development at Svalbard 1900–2100. Adv. Meteorol. 2011, 1–14. https://doi.org/10.1155/2011/893790.

Garbarino, J.R., Snyder-Conn, E., Leiker, T.J., Hoffman, G.L., 2002. Contaminants in Arctic snow collected over northwest Alaskan sea ice. Water Air Soil Pollut. 139, 183–214. https://doi.org/10.1023/A:1015808008298.

Gjengedal, E., Steinnes, E., 1990. Uptake of metal ions in moss from artificial precipitation. Environ. Monit. Assess. 14, 77–87. https://doi.org/10.1007/BF00394359.

Gworek, B., Dmuchowski, W., Baczewska-Dąbrowska, A.H., 2020. Mercury in the terrestrial environment: a review. Environ. Sci. Eur. 32, 128. https://doi.org/10.1186/s12302-020-00401-x.

Halbach, K., Mikkelsen, Ø., Berg, T., Steinnes, E., 2017. The presence of mercury and other trace metals in surface soils in the Norwegian Arctic. Chemosphere 188, 567–574. https://doi.org/10.1016/j.chemosphere.2017.09.012.

Hansen, B.B., Aanes, R., Sæther, B.-E., 2010. Partial seasonal migration in high-arctic Svalbard reindeer (Rangifer tarandus platyrhynchus). Can. J. Zool. 88, 1202–1209. https://doi.org/10.1139/Z10-086.

Ikemoto, T., Kunito, T., Anan, Y., Tanaka, H., Baba, N., Miyazaki, N., Tanabe, S., 2004. Association of heavy metals with metallothionein and other proteins in hepatic cytosol of marine mammals and seabirds. Environ. Toxicol. Chem. 23, 2008. https://doi.org/10.1897/03-456.

Ismael, M.A., Elyamine, A.M., Moussa, M.G., Cai, M., Zhao, X., Hu, C., 2019. Cadmium in plants: uptake, toxicity, and its interactions with selenium fertilisers. Metallomics 11, 255–277. https://doi.org/10.1039/C8MT00247A.

Johansson, K., Bergbäck, B., Tyler, G., 2001. Impact of atmospheric long range transport of lead, mercury and cadmium on the Swedish forest environment. Water Air Soil Pollut. Focus 1, 279–297. https://doi.org/10.1023/A:1017528826641.

John, D.A., Leventhal, J.S., 1995. Bioavailability of metals. In: Preliminary Compilation of Descriptive Geoenvironmental Mineral Deposit Models, pp. 10–18.

Kabata-Pendias, A., Szteke, B., 2015. Trace Elements in Abiotic and Biotic Environments. CRC Press, Taylor & Francis Group, Boca Raton.

Kinck, C., 2014. Reduced Forage Access Affects Home Range Size and Site Fidelity of Svalbard Reindeer (Rangifer tarandus platyrhynchus). NMBU.

Klaassen, C.D., Casarett, L.J., Doull, J. (Eds.), 2013. Casarett and Doull's Toxicology: The Basic Science of Poisons, eighth ed. McGraw-Hill Education, New York.

Kłos, A., Ziembik, Z., Rajfur, M., Dołhańczuk-Śródka, A., Bochenek, Z., Bjerke, J.W., Tømmervik, H., Zagajewski, B., Ziółkowski, D., Jerz, D., Zielińska, M., Krems, P., Godyń, P., 2017. The origin of heavy metals and radionuclides accumulated in the soil and biota samples collected in Svalbard, near Longyearbyen. Ecol. Chem. Eng. S 24, 223–238. https://doi.org/10.1515/eces-2017-0015.

Krajcarová, L., Novotný, K., Chattová, B., Elster, J., 2016. Elemental analysis of soils and Salix polaris in the town of Pyramiden and its surroundings (Svalbard). Environ. Sci. Pollut. Res. 23, 10124–10137. https://doi.org/10.1007/s11356-016-6213-4.

Le Moullec, M., Pedersen, Å.Ø., Stien, A., Rosvold, J., Hansen, B.B., 2019. A century of conservation: the ongoing recovery of Svalbard reindeer. J. Wildl. Manag. 83, 1676–1686. https://doi.org/10.1002/jwmg.21761.

Lewińska-Preis, L., Fabiańska, M.J., Ćmiel, S., Kita, A., 2009. Geochemical distribution of trace elements in Kaffiøyra and Longyearbyen coals, Spitsbergen, Norway. Int. J. Coal Geol. 80, 211–223. https://doi.org/10.1016/j.coal.2009.09.007.

Lindberg, S.E., Brooks, S., Lin, C.-J., Scott, K.J., Landis, M.S., Stevens, R.K., Goodsite, M., Richter, A., 2002. Dynamic oxidation of gaseous mercury in the Arctic troposphere at polar sunrise. Environ. Sci. Technol. 36, 1245–1256. https://doi.org/10.1021/es0111941.

Ma, H., Shi, G., Cheng, Y., 2020. Accumulation characteristics of metals and metalloids in plants collected from Ny-Ålesund, Arctic. Atmosphere 11, 1129. https://doi.org/10.3390/atmos11101129.

Macdonald, R.W., Harner, T., Fyfe, J., 2005. Recent climate change in the Arctic and its impact on contaminant pathways and interpretation of temporal trend data. Sci. Total Environ. 342, 5–86. https://doi.org/10.1016/j.scitotenv.2004.12.059.

Makarov, D.A., Ovcharenko, V.V., Nebera, E.A., Kozhushkevich, A.I., Shelepchikov, A.A., Turbabina, K.A., Kalantaenko, A.M., Bardyugov, N.S., Gergel, A.A., 2022. Geographical distribution of dioxins, cadmium, and mercury concentrations in reindeer liver, kidneys, and muscle in the Russian Far North. Environ. Sci. Pollut. Res. 29, 12176–12187. https://doi.org/10.1007/s11356-021-16310-2.

Mallory, M.L., Mahon, L., Tomlik, M.D., White, C., Milton, G.R., Spooner, I., 2015. Colonial marine birds influence Island soil chemistry through biotransport of trace elements. Water Air Soil Pollut. 226, 31. https://doi.org/10.1007/s11270-015-2314-9.

Middleton, G.V., 1960. Chemical composition of sandstones. Geol. Soc. Am. Bull. 71, 1011. https://doi.org/10.1130/0016-7606(1960)71[1011,CCOS]2.0.CO;2.

Nève, J., 1991. Physiological and nutritional importance of selenium. Experientia 47, 187–193. https://doi.org/10.1007/BF01945424.

Norsk Klimaservicesenter, n.d. Observasjoner og værstatistikk. URL https://seklima.met.no/observations/ (accessed 30.06.22).

Nygård, T., Steinnes, E., Røyset, O., 2012. Distribution of 32 elements in organic surface soils: contributions from atmospheric transport of pollutants and natural sources. Water Air Soil Pollut. 223, 699–713. https://doi.org/10.1007/s11270-011-0895-5.

Obrist, D., Agnan, Y., Jiskra, M., Olson, C.L., Colegrove, D.P., Hueber, J., Moore, C.W., Sonke, J.E., Helmig, D., 2017. Tundra uptake of atmospheric elemental mercury drives Arctic mercury pollution. Nature 547, 201–204. https://doi.org/10.1038/nature22997.

Orpin, C.G., Mathiesen, S.D., Greenwood, Y., Blix, A.S., 1985. Seasonal changes in the ruminal microflora of the high-arctic Svalbard reindeer (Rangifer tarandus platyrhynchus). Appl. Environ. Microbiol. 50, 144–151. https://doi.org/10.1128/aem.50.1.144-151.1985.

Outridge, P.M., Hermanson, M.H., Lockhart, W.L., 2002. Regional variations in atmospheric deposition and sources of anthropogenic lead in lake sediments across the Canadian Arctic. Geochim. Cosmochim. Acta 66, 3521–3531. https://doi.org/10.1016/S0016-7037(02)00955-9.

Pacyna, J.M., Pacyna, E.G., 2001. An assessment of global and regional emissions of trace metals to the atmosphere from anthropogenic sources worldwide. Environ. Rev. 9, 269–298. https://doi.org/10.1139/a01-012.

Pacyna, A.D., Koziorowska, K., Chmiel, S., Mazerski, J., Polkowska, Ż., 2018. Svalbard reindeer as an indicator of ecosystem changes in the Arctic terrestrial ecosystem. Chemosphere 203, 209–218. https://doi.org/10.1016/j.chemosphere.2018.03.158.

Pacyna, A.D., Frankowski, M., Kozioł, K., Węgrzyn, M.H., Wietrzyk-Pełka, P., Lehmann-Konera, S., Polkowska, Ż., 2019. Evaluation of the use of reindeer droppings for monitoring essential and non-essential elements in the polar terrestrial environment. Sci. Total Environ. 658, 1209–1218. https://doi.org/10.1016/j.scitotenv.2018.12.232.

Pacyna-Kuchta, A.D., Wietrzyk-Pełka, P., Węgrzyn, M.H., Frankowski, M., Polkowska, Ż., 2020. A screening of select toxic and essential elements and persistent organic pollutants in the fur of Svalbard reindeer. Chemosphere 245, 125458. https://doi.org/10.1016/j.chemosphere.2019.125458.

Pearson, C., Howard, D., Moore, C., Obrist, D., 2019. Mercury and trace metal wet deposition across five stations in Alaska: controlling factors, spatial patterns, and source regions. Atmos. Chem. Phys. 19, 6913–6929. https://doi.org/10.5194/acp-19-6913-2019.

Pourrut, B., Shahid, M., Dumat, C., Winterton, P., Pinelli, E., 2011. Lead uptake, toxicity, and detoxification in plants. In: Whitacre, D.M. (Ed.), Reviews of Environmental Contamination and Toxicology. Volume 213. Springer New York, New York, NY, pp. 113–136. https://doi.org/10.1007/978-1-4419-9860-6_4.

Pulford, I., 2003. Phytoremediation of heavy metal-contaminated land by trees—a review. Environ. Int. 29, 529–540. https://doi.org/10.1016/S0160-4120(02)00152-6.

R Core Team, 2022. R: A Language and Environment for Statistical Computing.

Raj, D., Maiti, S.K., 2020. Sources, bioaccumulation, health risks and remediation of potentially toxic metal(loid)s (As, Cd, Cr, Pb and Hg): an epitomised review. Environ. Monit. Assess. 192, 108. https://doi.org/10.1007/s10661-019-8060-5.

Roggeman, S., van den Brink, N., Van Praet, N., Blust, R., Bervoets, L., 2013. Metal exposure and accumulation patterns in free-range cows (Bos taurus) in a contaminated natural area: influence of spatial and social behavior. Environ. Pollut. 172, 186–199. https://doi.org/10.1016/j.envpol.2012.09.006.

Romero, M.B., Polizzi, P., Chiodi, L., Das, K., Gerpe, M., 2016. The role of metallothioneins, selenium and transfer to offspring in mercury detoxification in Franciscana dolphins (Pontoporia blainvillei). Mar. Pollut. Bull. 109, 650–654. https://doi.org/10.1016/j.marpolbul.2016.05.012.

Schaefer, K., Elshorbany, Y., Jafarov, E., Schuster, P.F., Striegl, R.G., Wickland, K.P., Sunderland, E.M., 2020. Potential impacts of mercury released from thawing permafrost. Nat. Commun. 11, 4650. https://doi.org/10.1038/s41467-020-18398-5.

Scheuhammer, A., Braune, B., Chan, H.M., Frouin, H., Krey, A., Letcher, R., Loseto, L., Noël, M., Ostertag, S., Ross, P., Wayland, M., 2015. Recent progress on our

understanding of the biological effects of mercury in fish and wildlife in the Canadian Arctic. Sci. Total Environ. 509–510, 91–103. https://doi.org/10.1016/j.scitotenv.2014.05.142.

Sørmo, E.G., Ciesielski, T.M., Øverjordet, I.B., Lierhagen, S., Eggen, G.S., Berg, T., Jenssen, B.M., 2011. Selenium moderates mercury toxicity in free-ranging freshwater fish. Environ. Sci. Technol. 45, 6561–6566. https://doi.org/10.1021/es200478b.

Steffen, A., Douglas, T., Amyot, M., Ariya, P., Aspmo, K., Berg, T., Bottenheim, J., Brooks, S., Cobbett, F., Dastoor, A., Dommergue, A., Ebinghaus, R., Ferrari, C., Gardfeldt, K., Goodsite, M.E., Lean, D., Poulain, A.J., Scherz, C., Skov, H., Sommar, J., Temme, C., 2008. A synthesis of atmospheric mercury depletion event chemistry in the atmosphere and snow. Atmos. Chem. Phys. 8, 1445–1482. https://doi.org/10.5194/acp-8-1445-2008.

Steinnes, E., 1995. A critical evaluation of the use of naturally growing moss to monitor the deposition of atmospheric metals. Sci. Total Environ. 160–161, 243–249. https://doi.org/10.1016/0048-9697(95)04360-D.

Strizhkina, I., Ilyin, I., Rozovskaya, O., Travnikov, O., 2022. Assessment of Heavy Metal and POP Pollution on Global, Regional and National Scales. Part I. Supplementary Materials for Heavy Metals. (No. 1/2022). EMEP.

The Norwegian Meteorological Institute, n.d. Wind Rose 1013: Isfjord Radio [WWW Document]. URL https://nwp.dmi.govcloud.dk/aviation/station.html?1013# (accessed 11.4.22).

Thorne, R.J., Pacyna, J.M., Sundseth, K., Pacyna, E.G., 2018. Fluxes of trace metals on a global scale. In: Encyclopedia of the Anthropocene. Elsevier, pp. 93–102. https://doi.org/10.1016/B978-0-12-809665-9.09918-3.

Warner, N.A., Sagerup, K., Kristoffersen, S., Herzke, D., Gabrielsen, G.W., Jenssen, B.M., 2019. Snow buntings (*Plectrophenax nivealis*) as bio-indicators for exposure differences to legacy and emerging persistent organic pollutants from the Arctic

terrestrial environment on Svalbard. Sci. Total Environ. 667, 638–647. https://doi.org/10.1016/j.scitotenv.2019.02.351.

Wegrzyn, M.H., Wietrzyk, P., Lehmann-Konera, S., Chmiel, S., Cykowska-Marzencka, B., Polkowska, Ż., 2018. Annual variability of heavy metal content in Svalbard reindeer faeces as a result of dietary preferences. Environ. Sci. Pollut. Res. 25, 36693–36701. https://doi.org/10.1007/s11356-018-3479-8.

Wei, Y., He, J., Xue, Y., Nie, Y., Liu, X., Wu, L., 2022. Spatial distribution of multi-elements in moss revealing heavy metal precipitation in London Island, Svalbard, Arctic. Environ. Pollut. 315, 120398. https://doi.org/10.1016/j.envpol.2022.120398.

Wojtuń, B., Samecka-Cymerman, A., Kolon, K., Kempers, A.J., Skrzypek, G., 2013. Metals in some dominant vascular plants, mosses, lichens, algae, and the biological soil crust in various types of terrestrial tundra, SW Spitsbergen, Norway. Polar Biol. 36, 1799–1809. https://doi.org/10.1007/s00300-013-1399-0.

Wojtuń, B., Polechońska, L., Pech, P., Mielcarska, K., Samecka-Cymerman, A., Szymański, W., Kolon, M., Kopeć, M., Stadnik, K., Kempers, A.J., 2019. *Sanionia uncinata* and *Salix polaris* as bioindicators of trace element pollution in the high Arctic: a case study at Longyearbyen, Spitsbergen, Norway. Polar Biol. 42, 1287–1297. https://doi.org/10.1007/s00300-019-02517-0.

Zhang, H., Feng, X., Zhu, J., Sapkota, A., Meng, B., Yao, H., Qin, H., Larssen, T., 2012. Selenium in soil inhibits mercury uptake and translocation in rice (*Oryza sativa* L.). Environ. Sci. Technol. 46, 10040–10046. https://doi.org/10.1021/es302245r.

Zhou, J., Obrist, D., Dastoor, A., Jiskra, M., Ryjkov, A., 2021. Vegetation uptake of mercury and impacts on global cycling. Nat. Rev. Earth Environ. 2, 269–284. https://doi.org/10.1038/s43017-021-00146-y.

Zwolicki, A., Zmudczyńska-Skarbek, K., Matuła, J., Wojtuń, B., Stempniewicz, L., 2016. Differential responses of Arctic vegetation to nutrient enrichment by plankton- and fish-eating colonial seabirds in Spitsbergen. Front. Plant Sci. 07 https://doi.org/10.3389/fpls.2016.01959.

SUPPORTING INFORMATION

Tracing the footprints of Arctic pollution: Spatial variations in toxic and essential elements in Svalbard reindeer (*Rangifer tarandus platyrhynchus*) faeces

Malin Andersson Stavridis[1,2*], Susanne Brix Røed[2], Brage Bremset Hansen[2,3], Øyvind Mikkelsen[1,4], Tomasz Maciej Ciesielski[1,2] & Bjørn Munro Jenssen[1,2*].

1. Department of Arctic Technology, University Centre in Svalbard (UNIS), P.O. Box 156 N-9171, Longyearbyen, Norway.

2. Department of Biology, Norwegian University of Science and Technology (NTNU), NO-7491, Trondheim, Norway.

3. Department of Terrestrial Ecology, Norwegian Institute for Nature Research (NINA), NO-7485 Trondheim, Norway.

4. Department of Chemistry, Norwegian University of Science and Technology (NTNU), NO-7491, Trondheim, Norway.

*Corresponding Author(s): Bjørn Munro Jenssen, bjorn.munro.jenssen@ntnu.no; Malin Andersson Stavridis, malins@unis.no.

1

Appendix A

Figures

Figure A1. Central Nordenskiöld land with the areas of Nordenskiöld coast and Adventdalen marked in dark red and the soil sampling sites marked with red dots. The settlements of Barentsburg and Longyearbyen are indicated with red geotags. Isfjordbanken is marked with an [I] and Adventfjorden marked with an [A].
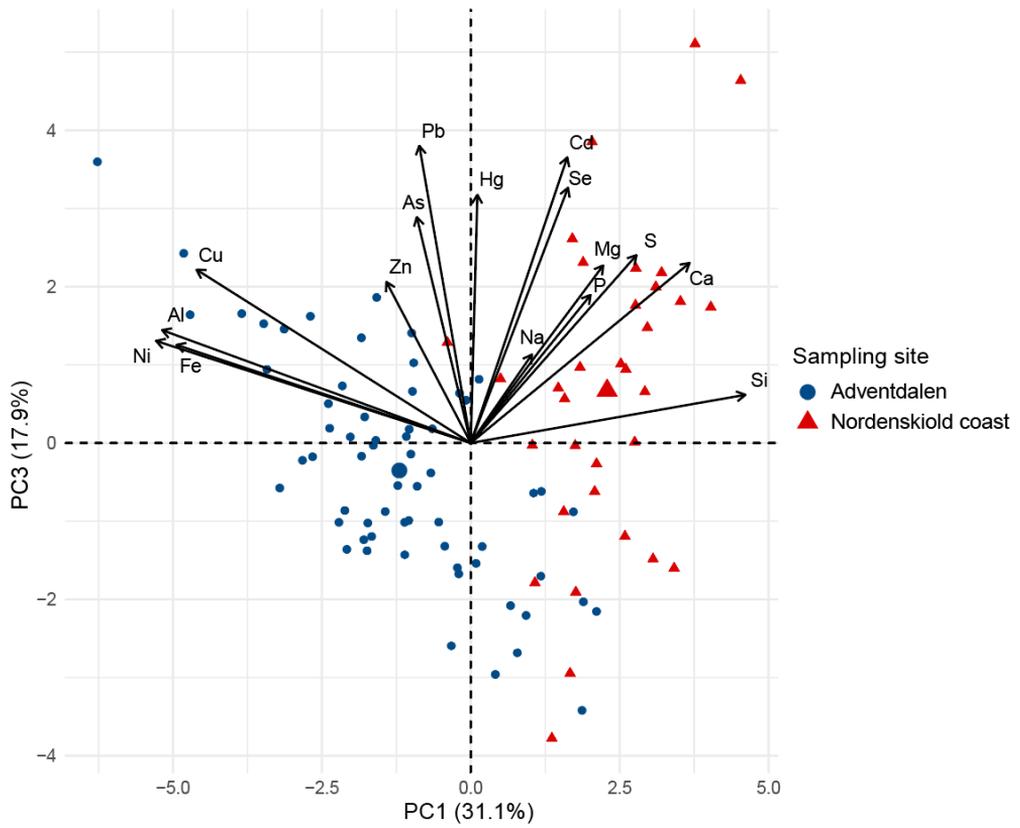
Figure A2. PCA biplot of the Svalbard reindeer (*Rangifer tarandus platyrhynchus*) faecal elemental composition. PC1 and PC3 explain 31.1% versus 17.9% of the variance in the data set.

Sampling information

Table A1. Additional soil sampling information, providing details of the sampling locations.

Sample ID	Location	Year	Latitude	Longitude	TOC	pH
1	Adventdalen	2021	78.16960	16.00585	7.3	5.0
2	Adventdalen	2021	78.16960	16.00585	13.1	4.8
3	Adventdalen	2021	78.16960	16.00585	3.3	4.8
4	Adventdalen	2021	78.16202	16.18628	9.6	5.4
5	Adventdalen	2021	78.16512	16.14622	16.5	5.2
6	Isfjord radio	2021	78.06002	13.64613	12.4	5.2
7	Isfjord radio	2021	78.06165	13.65083	4.8	5.2
8	Isfjord radio	2021	78.06307	13.65362	13.8	4.7
9	Isfjord radio	2021	78.05638	13.65778	21.6	6.2

Table A2. Additional faecal sampling information, providing details on annual average precipitation [mm] (Precipitation), the distance to the ocean [m] (Ocean), the sampling locations, year of sampling, sex and age of the sampled individuals and the sampling coordinates.

Sample ID	Precipitation	Ocean	Location	Year	Sex	Age	Latitude	Longitude
1	236.27	6340	Adventdalen	2014	F	A	78.181115	15.909956
2	209.43	22300	Adventdalen	2015	F	A	78.183544	16.19028
3	209.43	22300	Adventdalen	2015	F	C	78.183544	16.19028
4	209.43	21600	Adventdalen	2015	F	C	78.189679	16.218993

4

5	209.43	21600	Adventdalen	2015	F	A	78.189679	16.218993
6	209.43	18700	Adventdalen	2015	M	A	78.182299	16.230318
7	209.43	18700	Adventdalen	2015	M	A	78.182299	16.230318
8	209.43	18700	Adventdalen	2015	M	A	78.182299	16.230318
9	209.43	20900	Adventdalen	2015	M	A	78.197938	16.281426
10	209.43	20900	Adventdalen	2015	F	C	78.197938	16.281426
11	209.43	31700	Adventdalen	2015	M	A	78.21876	16.394247
12	285.19	2630	Adventdalen	2017	F	A	78.224342	15.876549
13	285.19	4510	Adventdalen	2017	M	A	78.220003	15.859204
14	285.19	2500	Adventdalen	2017	M	A	78.237457	15.877995
15	285.19	3970	Adventdalen	2017	F	A	78.24831	15.874836
16	285.19	3970	Adventdalen	2017	M	A	78.24831	15.874836
17	285.19	3970	Adventdalen	2017	M	A	78.24831	15.874836
18	285.19	3970	Adventdalen	2017	M	A	78.24831	15.874836
19	285.19	2600	Adventdalen	2017	M	A	78.246082	15.870714
20	285.19	2600	Adventdalen	2017	F	A	78.246082	15.870714
21	285.19	2330	Adventdalen	2017	M	A	78.246031	15.857957
22	285.19	2330	Adventdalen	2017	M	A	78.246031	15.857957

23	285.19	2420	Adventdalen	2017	F	A	78.243273	15.851163
24	285.19	2420	Adventdalen	2017	M	A	78.243273	15.851163
25	285.19	2420	Adventdalen	2017	M	A	78.243273	15.851163
26	285.19	2460	Adventdalen	2017	M	A	78.243086	15.823011
27	285.19	1040	Adventdalen	2017	F	A	78.241434	15.805313
28	285.19	1040	Adventdalen	2017	C	C	78.241434	15.805313
29	285.19	762	Adventdalen	2017	M	A	78.245009	15.776092
30	285.19	12800	Adventdalen	2017	F	A	78.161777	16.204893
31	285.19	13000	Adventdalen	2017	M	A	78.1619	16.205386
32	285.19	12700	Adventdalen	2017	M	A	78.163312	16.214875
33	285.19	13200	Adventdalen	2017	F	A	78.16707	16.253488
34	285.19	13700	Adventdalen	2017	M	A	78.176328	16.299539
35	285.19	13100	Adventdalen	2017	M	A	78.176329	16.275532
36	285.19	13700	Adventdalen	2017	C	C	78.176141	16.301399
37	285.19	12700	Adventdalen	2017	M	A	78.181067	16.266237
38	285.19	12700	Adventdalen	2017	M	A	78.181711	16.262368
39	285.19	12600	Adventdalen	2017	M	A	78.194161	16.286234
40	285.19	12600	Adventdalen	2017	F	A	78.194144	16.286188

41	285.19	12600	Adventdalen	2017	C	C	78.194144	16.286188
42	285.19	8500	Adventdalen	2017	M	A	78.148197	15.825676
43	285.19	6180	Adventdalen	2017	F	A	78.174543	15.865085
44	285.19	6180	Adventdalen	2017	F	A	78.174543	15.865085
45	285.19	6180	Adventdalen	2017	C	C	78.174734	15.864006
46	285.19	6320	Adventdalen	2017	M	A	78.162875	15.73043
47	285.19	6320	Adventdalen	2017	F	A	78.162875	15.73043
48	285.19	4220	Adventdalen	2017	F	A	78.182831	15.751287
49	285.19	4220	Adventdalen	2017	M	A	78.183154	15.751133
50	285.19	3850	Adventdalen	2017	F	A	78.186843	15.765673
51	285.19	3400	Adventdalen	2017	M	A	78.198641	15.815882
52	285.19	3420	Adventdalen	2017	C	C	78.191392	16.133662
53	285.19	10000	Adventdalen	2017	C	C	78.188483	16.174881
54	285.19	10600	Adventdalen	2017	F	A	78.19195	16.186211
55	285.19	10500	Adventdalen	2017	F	A	78.192425	16.184113
56	285.19	10500	Adventdalen	2017	F	A	78.19248	16.18368
57	285.19	10900	Adventdalen	2017	F	A	78.190485	16.197842
58	285.19	11500	Adventdalen	2017	M	A	78.180934	16.226853

59	285.19	12000	Adventdalen	2017	M	A	78.182143	16.229077
60	285.19	12000	Adventdalen	2017	F	A	78.185014	16.236854
61	285.19	11600	Adventdalen	2017	F	A	78.1914	16.235587
62	285.19	12000	Adventdalen	2017	F	A	78.189819	16.248863
63	285.19	12000	Adventdalen	2017	C	C	78.189827	16.247025
64	600.04	12	Nordenskiöld coast	2014	M	A	78.058703	13.591737
65	600.04	12	Nordenskiöld coast	2014	M	A	78.058703	13.591737
66	600.04	12	Nordenskiöld coast	2014	M	A	78.058703	13.591737
67	531.89	937	Nordenskiöld coast	2015	F	A	78.06287	13.702108
68	531.89	937	Nordenskiöld coast	2015	F	C	78.06287	13.702108
69	531.89	1380	Nordenskiöld coast	2015	M	A	78.055918	13.702853
70	531.89	1380	Nordenskiöld coast	2015	F	A	78.055918	13.702853
71	531.89	1980	Nordenskiöld coast	2015	F	A	78.042471	13.686559
72	531.89	477	Nordenskiöld coast	2015	M	A	77.986485	13.662672
73	531.89	34	Nordenskiöld coast	2015	M	A	77.94814	13.614251
74	724.29	356	Nordenskiöld coast	2017	M	A	78.05777	13.64301
75	724.29	328	Nordenskiöld coast	2017	F	A	78.07659	13.7176
76	724.29	284	Nordenskiöld coast	2017	M	A	78.07656	13.72288

77	724.29	236	Nordenskiöld coast	2017	C	C	78.07696	13.7261
78	724.29	565	Nordenskiöld coast	2017	M	A	78.07423	13.71163
79	724.29	310	Nordenskiöld coast	2017	F	A	78.07667	13.73904
80	724.29	1550	Nordenskiöld coast	2017	M	A	78.06633	13.74355
81	724.29	1650	Nordenskiöld coast	2017	F	A	78.06557	13.74297
82	724.29	1550	Nordenskiöld coast	2017	F	A	78.0657	13.74671
83	724.29	1800	Nordenskiöld coast	2017	M	A	78.05938	13.73339
84	724.29	2410	Nordenskiöld coast	2017	C	C	78.05339	13.74898
85	724.29	2500	Nordenskiöld coast	2017	C	C	78.05313	13.74548
86	724.29	362	Nordenskiöld coast	2017	N/A	N/A	78.05895	13.612767
87	724.29	431	Nordenskiöld coast	2017	M	A	78.058183	13.610917
88	724.29	759	Nordenskiöld coast	2017	F	A	78.053733	13.621933
89	724.29	2200	Nordenskiöld coast	2017	N/A	N/A	77.998717	13.717767
90	724.29	1050	Nordenskiöld coast	2017	M	A	78.026367	13.6678
91	724.29	0	Nordenskiöld coast	2017	M	A	77.991283	13.628917
92	724.29	974	Nordenskiöld coast	2017	F	A	77.994417	13.671783
93	724.29	192	Nordenskiöld coast	2017	M	A	77.99015	13.647167
94	724.29	190	Nordenskiöld coast	2017	M	A	77.99015	13.648333

| 95 | 724.29 | 112 | Nordenskiöld coast | 2017 | F | A | 77.970417 | 13.628967 |
| 96 | 724.29 | 330 | Nordenskiöld coast | 2017 | F | C | 78.0566 | 13.60215 |

Other data

Table A3. Certified reference material (CRM) analysis (n=4) on Virginia Tobacco leaves (INCT-PVTL-6, Institute of Nuclear Chemistry and Technology, Warszawa, Poland). We determined the recovery by comparing our measured concentrations to the certified values in the official CRM. N/A is given if the CRM is not certified for the element.

	(µg/kg)		
Element	**Certified CRM**	**Measured values**	**Recovery**
Hg	0.0232	0.0181	78
Se	N/A	0.031	N/A
Cd	2.23	2.328	104
As	0.138	0.116	84
Pb	0.972	0.657	68
Cu	5.12	4.57	89
Ni	1.49	1.22	82
Zn	43.6	40.3	92
Na	62.4	67.3	108
Si	N/A	901	N/A
P	3 780	3 310	88
S	2 420	2 577	106
Mg	2 410	2 330	97
Fe	258	256	99
Al	252	325	129
Ca	22 970	22 420	98

Table A4. Limits of detection (LOD) based on either three times the standard deviation or the instrument detection limit, depending on which gave the highest value. All values are provided in µg/g.

Element	LOD
Hg	0.000147
Pb	0.00406
Cd	0.00147
Se	0.00737
Ca	0.295
Zn	0.0182
Si	3.39
S	2.95
P	0.0590
Na	1.47
Cu	0.00442
Mg	0.0147
Al	0.155
Fe	0.168
As	0.00369
Ni	0.00221

Table A5. Contributions (loadings) to the first three principal components (PCs).

Element	PC1	PC2	PC3
Hg	0.01	17.60	10.86
Pb	0.46	7.45	15.55
Cd	1.62	1.04	14.36
Se	1.65	12.97	11.47
Ca	8.34	2.13	5.71

Zn	1.24	0.81	4.57
Si	13.14	1.25	0.41
S	13.10	0.28	5.29
P	16.60	0.80	2.24
Na	4.78	16.57	6.21
Cu	15.08	1.07	1.69
Mg	0.51	18.54	8.99
Al	0.65	1.19	1.38
Fe	3.06	17.83	5.56
As	2.50	0.23	3.86
Ni	17.26	0.23	1.84

Paper II

Toxic metals and other elements in Svalbard reindeer: Establishing baselines and assessing non-invasive sampling for biomonitoring

Malin Andersson Stavridis [a,b,*], Bjørg Marie Pollestad [a,b], Vebjørn Veiberg [c], Øyvind Mikkelsen [a,d], Tomasz Maciej Ciesielski [a,b], Bjørn Munro Jenssen [a,b,*]

[a] Department of Arctic Technology, University Centre in Svalbard (UNIS), PO Box 156, N-9171 Longyearbyen, Norway
[b] Department of Biology, Norwegian University of Science and Technology (NTNU), NO-7491 Trondheim, Norway
[c] Department of Terrestrial Ecology, Norwegian Institute for Nature Research (NINA), NO-7485 Trondheim, Norway
[d] Department of Chemistry, Norwegian University of Science and Technology (NTNU), NO-7491 Trondheim, Norway

HIGHLIGHTS

- We determined Hg, Cd and Pb in various Svalbard reindeer samples.
- Cd and Pb in some liver and kidney samples exceed safe consumption thresholds.
- Cd and Pb concentrations have decreased over the past forty years.
- The grazing behavior of the Svalbard reindeer affects their toxic metal exposure.
- Faeces is a moderate predictor of liver Hg concentrations.

GRAPHICAL ABSTRACT
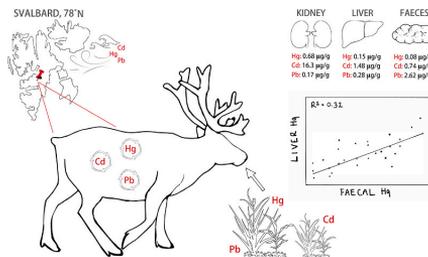
ARTICLE INFO

ABSTRACT

Pollutants emitted from all over the world may reach pristine areas, such as the Arctic. The Svalbard reindeer (Rangifer tarandus platyrhynchus) has been the subject of a few studies reporting toxic metal concentrations. However, these studies either date back a few decades or exclusively used non-invasive samples (e.g., faeces and fur), leaving us without an updated assessment of the concentrations in internal tissues and target organs.

This study is the first to present the concentrations of the toxic metals mercury (Hg), cadmium (Cd), and lead (Pb) in both invasive and non-invasive samples from female Svalbard reindeer. The highest concentrations of both Hg and Cd were found in the kidneys ($\bar{x}$=0.68 μg/g and 16.3 μg/g dw, respectively). The highest concentration of Pb was found in faeces ($\bar{x}$=2.62 μg/g dw), followed by liver ($\bar{x}$=0.28 μg/g dw). While both Cd and Pb concentrations in Svalbard reindeer were comparable to levels reported in other reindeer subspecies or circumpolar wildlife, Hg concentrations were lower than those reported in most other studies on reindeer. Conversely, Hg levels in Svalbard reindeer were still higher than levels reported in wildlife from central Europe, demonstrating the influence of long-range transport of Hg to the Arctic.

By comparing the toxic metal concentrations in different sample types, we confirm a relationship between Hg concentrations in invasive and non-invasive samples, with faeces being the most promising proxy for soft tissue Hg concentrations. Consequently, future Hg biomonitoring efforts may be carried out with relatively simple sampling procedures and without sacrificing Svalbard reindeer.

---

* Corresponding authors at: Department of Arctic Technology, University Centre in Svalbard (UNIS), PO Box 156, N-9171 Longyearbyen, Norway.
E-mail addresses: malins@unis.no (M. Andersson Stavridis), bjorn.munro.jenssen@ntnu.no (B.M. Jenssen).

1. Introduction

Anthropogenic activities have significantly increased the concentrations of toxic metals mercury (Hg), cadmium (Cd), and lead (Pb) in the biosphere since the onset of the industrial revolution (Dietz et al., 2009; Barbante et al., 2017). This trend is globally evident, including in remote Arctic regions like Svalbard (AMAP, 1998; Yang et al., 2020).

Hg, Cd, and Pb are known to cause adverse health effects and have been closely monitored in Arctic wildlife during the past decades (Klaassen and Watkins, 2015; AMAP, 2018). Monitoring efforts have primarily focused on marine ecosystems, while far fewer studies have been conducted in terrestrial environments. Consequently, there are limitations in the data on toxic metal concentrations in many Arctic terrestrial species, such as the Svalbard reindeer (*Rangifer tarandus platyrhynchus*) (Riget et al., 2004; Dietz et al., 2022). The Svalbard reindeer is among the least studied reindeer subspecies in the context of pollution. They are the largest terrestrial mammal on the Svalbard archipelago and play an important role in the local terrestrial ecosystem (Hansen et al., 2013), both as primary consumers and as prey for Arctic foxes (*Vulpes lagopus*; Prestrud, 1992; Eide et al., 2005) and polar bears (*Ursus maritimus*; Stempniewicz et al., 2021). In addition, Svalbard reindeer are hunted by local residents (MOSJ, 2021).

So far, most existing studies on contaminant concentrations in the Svalbard reindeer have used non-invasive samples (e.g., faeces and fur) without providing information on tissue distribution or toxic metal accumulation (Pacyna et al., 2018, 2019; Pacyna-Kuchta et al., 2020). This limits our understanding of concentrations in internal tissues, which are used for setting human consumption thresholds or toxicity evaluations (Andersson Stavridis et al., 2024).

Reindeer are considered ideal for biomonitoring pollutants in the Arctic terrestrial ecosystem (Pacyna et al., 2018). First, the circumpolar distribution of different reindeer subspecies enables a comparison between reindeer across different Arctic regions, which may help understand the global distribution of pollutants (Gamberg et al., 2020). Secondly, a relatively long lifespan of reindeer (Reimers, 1983) allows an accumulation of pollutants over long periods, which may provide a better overview of cumulative contaminant exposure. Thirdly, reindeer are key primary consumers in the Arctic terrestrial ecosystem and transfer pollutants from the environment into the Arctic food chain. Finally, the thawing of the Arctic permafrost may remobilise legacy contaminants, such as Hg, into the terrestrial ecosystem (Schaefer et al., 2020). It is, therefore, important to establish a baseline reference of metal concentrations in terrestrial organisms, such as reindeer, that can be used for future comparisons in monitoring programs.

Previous studies indicate that the Svalbard reindeer are exposed to a range of pollutants, including Hg, Cd, and Pb (e.g., Pacyna et al., 2018, 2019; Andersson Stavridis et al., 2024). To our knowledge, the only study reporting renal and hepatic metal concentrations in Svalbard reindeer dates back decades (Borch-Iohnsen et al., 1996), while modern studies focus solely on contaminant concentrations in non-invasive samples (e.g., faeces, fur), which are two key excretion pathways for metals (Wiener et al., 2003). While faecal and fur concentrations may indicate exposure to contaminants, they are not directly comparable to existing toxicity thresholds set for terrestrial mammals, which are based on soft tissue concentrations (Dietz et al., 2022). Jota Baptista et al. (2022) argue that there is a need to assess the information provided by invasive versus non-invasive samples and evaluate how contaminant levels in these different matrices are related to each other before using non-invasive samples in toxicity-related monitoring. Relationships between heavy metals in non-invasive and invasive matrices have previously been studied in various species, and correlations between, for instance, Hg in hair and soft tissues or blood were found in otters (*Lutra canadensis*; Evans et al., 1998; Mierle et al., 2009) and polar bears (Cardona-Marek et al., 2009), but not in seals (Ikemoto et al., 2004). Dietz et al. (2009) suggest that interspecific differences in metal excretion may explain why these relationships are found in certain species but

not others. Consequently, species-specific relationships between invasive and non-invasive matrices should be determined before using non-invasive matrices for biomonitoring.

The present study determined the concentrations of Hg, Cd, Pb, and other elements of interest in both invasive (kidney, liver, muscle, brain, and blood) and non-invasive (milk, fur, and faeces) samples from female Svalbard reindeer. The objectives of the study were to (1) explore potential relationships between concentrations of toxic metals and other selected elements in non-invasive and invasive samples, to determine if non-invasive samples can be effectively used to predict or reflect internal concentrations in reindeer, and (2) determine whether the concentrations of Hg, Cd, and Pb in the tissues exceed the thresholds set for adverse health effects in the reindeer, or human consumption. By evaluating metal contamination in Svalbard reindeer, this study contributes to the knowledge on contaminants in the Arctic terrestrial environment, which is especially important as permafrost thaw may accelerate the remobilisation of pollutants in a rapidly changing Arctic.

2. Methods

2.1. Sampling campaigns

As part of an interdisciplinary research collaboration (Research In Svalbard ID 2909), 36 Svalbard reindeer females were culled in October 2021 ($n = 18$) and 2022 ($n = 18$) in Nordenskiöld Land, Svalbard (77°50′–78°20′N, 15°00′–15°60′E; permission 16/01632–40 and 21/03815–4 from the Governor of Svalbard). All individuals were shot in the heart and lung region using regular hunting ammunition (calibers and bullet types: lead and copper-based 6.5 × 55 Federal Fusion and copper-based 30–06 Lapua Naturalis; bullet types had no significant effect on Pb tissue concentrations). The age of harvested animals ranged from one to 12 years. The age of yearlings and 2-year-olds was decided based on tooth eruption patterns, while older individuals were aged based on cementum annuli analyses (Veiberg et al., 2020).

Whole blood samples were collected post-mortem in the field. The blood was primarily collected from the jugular vein in a plastic container and subsequently transferred into EDTA-lined vacutainer tubes (VACUETTE®, Greiner Bio-One, Austria). The culled animals were transported to the University Centre in Svalbard by helicopter for further necropsy, which was conducted within 11 h of euthanasia. Soft biometrics (live and dress mass) were noted (±0.5 kg). Fur samples consisting of guard hairs were collected from the dorsal backside of the reindeer using stainless-steel scissors. Kidney, liver, and muscle samples from the medial thigh muscle were all collected using stainless-steel scalpels (Swann-Morton®, Swann-Morgon Limited, UK). The outer layers of the tissue and organ samples were removed using clean scalpel blades to reduce cross-contamination. The skull was opened using a saw, and brain samples were collected from the brainstem using a bovine brain sampling spoon or by collecting one complete brain lobe. Faecal samples were collected directly from the rectum. All samples were stored in polyethylene bags (VWR®). Milk samples were collected from lactating females by cutting through the udder tissue and collecting the milk directly into 15 mL plastic tubes (VWR®). All samples were stored frozen (−20 °C) until further preparation. For further details on the sampling, see Table S1.

Prior to analysis, fur samples were washed in cycles with ultrapure water (MilliQ, Millipore® MilliporeSigma, United States) and acetone (VWR BDH®, >99.5 %) to remove external contamination (Dolan et al., 2017). The fur was left to dry in a fume hood while covered with disposable wipes (KIMTECH®) for approximately 48 h.

The wet weight (ww) of all the samples was noted (Mettler Toledo®, ± 0.01 g). Brain, kidney, liver, muscle, and faecal samples were freeze-dried for 48-72 h at −50 °C and 0.04 mbar (FreeZone® Benchtop Freeze-drier, Labconco), and the dry weight (dw) was subsequently measured (the average moisture content in the kidneys were 78.9 %, liver 70.0 %, muscle 69.9 %, brain: 75.9 %, and faeces 78.0 %). The dried samples

were placed in zip-lock bags and homogenised using a plastic hammer until the entire sample was turned into a fine powder, ensuring a good homogenate based on visual inspection. Subsamples of blood and milk (approximately 1 mL) were homogenised using an ultrasonic processor (Cole Parmer GEX) for 2×10 s.

All collected samples were analysed for total mercury (THg, which includes all the different mercury species). In addition, kidney, liver, muscle fur, and faecal samples collected in 2021 ($n = 18$) were analysed for a range of additional trace elements. These included metal contaminants Cd and Pb, and selenium (Se), for its mitigating effects against Hg. We also included aluminium (Al), iron (Fe), sulphur (S), silica (Si) and phosphorous (P), as they may assist in indicating the origin of the Hg, Cd, and Pb (Halbach et al., 2017).

2.2. Mercury analysis

Approximately 40 mg of cleaned fur, freeze-dried and homogenised kidney, liver, muscle, brain, and faecal samples, and homogenised blood and milk samples were analysed for THg using thermal decomposition and atomic absorption spectrophotometry (Direct Mercury Analyser [DMA-80 evo] Milestone) at the Department of Chemistry, Norwegian University of Science and Technology (NTNU). All samples were analysed in duplicates. Samples that deviated >15 % between duplicates were analysed in triplicates. The cells of the DMA-80 were calibrated using a Hg standard (in 2 % HCl, LabKings B.V.®, Hilversum, Netherlands) with either a five- or seven-point calibration curve. A daily calibration check was performed by analysing a high and a low concentration standard in the working range of the analysis (following US EPA method 7473). The precision of the analysis was furthermore confirmed daily using certified reference materials (CRM), with recoveries ranging from 94 to 117 % (M-3 HerTis and INCT-OBTL-5 from the Institute of Nuclear Chemistry and Technology, Warsaw, Poland; and GBW 09101b Human hair, Shangai Institute of Nuclear Research, Academia Sinica, China; Table S2). Blanks were also analysed to determine LOD (three times the standard deviation of the blanks; Table S3). Hg was detected above the LOD in all samples except for three brain samples. All sample Hg concentrations were corrected for instrument drift and blank values. All Hg concentrations reported in the result section are given in dry weight (dw).

2.3. Element analysis

In addition to Hg, eight other elements are reported in this study: Cd, Pb, Se, Al, Fe, S, Si and P. The elemental analysis was conducted at the Department of Chemistry, Norwegian University of Science and Technology (NTNU). The cleaned fur and freeze-dried kidney, liver, muscle, and faeces homogenates were digested in ultrapure nitric acid (ultrapure grade, purified from HNO_3, AnalaR NORMAPUR®, VWR using a sub-boiling distillation system [Milestone, SubPur, Sorisole, BG, Italy]). Approximately 400 mg of sample was added to polytetrafluoroethylene (PTFE) vials with 5 mL of 50 % (v/v) HNO_3. The samples were digested using a high-pressure microwave system (Milestone Ultraclave, EMLS, Leutkirch, Germany) for 2.5 h. The digested samples were diluted with ultrapure water (Elga® Purelab Flex 4, High Wycombe, United Kingdom) to a final volume of 40 mL. Blanks (400 mg of ultrapure water and 5 mL of 50 % (v/v) HNO_3) and CRMs (1577b Bovine Liver, U.S. Department of Commerce National Institute of Standards and Technology, Gaithersburg, Md 20899 and GBW 09101b Human hair, Shangai Institute of Nuclear Research, Academia Sinica, China) were also digested for quality control. Reference material recovery rates ranged from 84 to 108 % for all the analysed elements herein, except for Pb (72 % recovery for 1577b Bovine Liver), Cd, and Al (75 % and 139 % recovery for GBW 09101b Human Hair, respectively; Table S4).

An 8800 Triple Quadrupole inductively coupled plasma mass spectrometry (ICP-MS) system (Agilent, Santa Clara, USA) equipped with a prepFAST M5 autosampler (ESI, USA) was used to identify and quantify

the elements. The limits of detection (LODs) were calculated for each element and were either based on three times the standard deviation of the blanks or the instrument detection limits (IDLs). The IDLs were determined by analysing solutions with decreasing concentrations of each element, where the elemental concentration that resulted in a relative standard deviation of 25 % in three scans was defined as the IDL (Table S5). All elements were detected above the LODs in all samples except for Pb in most muscle samples. All elemental concentrations reported in the result section are given in dry weight (dw).

2.4. Statistical analysis

All statistical analyses were conducted using R version 4.3.3 (R Core Team, 2024). The normality of the data was assessed using the Shapiro-Wilk test, and variance was analysed using Levene's test. For non-normally distributed variables, Box-Cox transformations were applied in an attempt to normalise them; however, not all variables could be transformed using the same transformation method. As a result, non-parametric tests were primarily used for the statistical analysis (Table S6).

A Kruskal-Wallis test, followed by a pairwise Wilcoxon rank sum test with a Bonferroni correction, was applied to the data to evaluate significant differences in elemental concentrations between the sample types. Spearman rank correlations were carried out to assess the relationship between Hg, Cd, and Pb concentrations in the different sample types and determine the relationship among the elements Hg, Se, Cd, Pb, Al, Fe, Si, P, and S in faeces and fur. The relationship between Hg concentrations in the liver and faeces was further explored using linear regression on log-transformed data. Linear regression was also used to determine a potential relationship between age and soft tissue Hg, Cd and Pb concentrations.

As kidney, liver, muscle, faeces, and fur samples from both 2021 and 2022 were analysed for Hg, interannual variation in Hg concentrations was assessed in these samples using a Student's *t*-test or Mann-Whitney *U* test for normal and non-normally distributed data, respectively (See Supplementary Information).

All data are presented as either mean concentrations or mean ± standard deviation (SD) throughout the article.

3. Results

The highest concentrations of Hg were found in the kidneys (0.68 ± 0.31 µg/g), followed by liver>faeces>fur>muscle>brain>milk>blood (Table 1). Similarly, the highest concentrations of Cd were also found in the kidneys (16.3 ± 11.2 µg/g), followed by liver>faeces>muscle>fur (Table 2). The highest concentrations of Pb were found in the faeces (2.62 ± 1.18 µg/g), followed by liver>kidney>fur>muscle.

The concentrations of Hg differed significantly between most sample types ($p < 0.05$), except between milk and brain, and muscle and fur. The concentrations of Cd and Pb also differed significantly between samples ($p < 0.05$), except for Cd concentrations in muscle and fur ($p = 0.18$).

The Se:Hg molar ratio was >1 for all sample types in all individuals, with the highest average ratio in muscle (Se:Hg = 283), followed by fur (Se:Hg = 147), liver (Se:Hg = 28.0), faeces (Se:Hg = 21.3) and kidney (Se:Hg = 18.0).

The correlation analyses of Pb, Cd, and Hg concentrations in the various sample types showed substantial variation in the degree of covariation. For Pb, the only significant correlation between sample types was found between liver and kidney (Fig. 1A). For Cd, strong positive intercorrelations were found among soft tissue (kidney, liver, muscle) concentrations ($p < 0.001$), and fur Cd concentrations correlated positively with liver and kidney concentrations ($p < 0.05$). Faecal Cd concentrations did not correlate with the concentrations in any of the other analysed sample types (Fig. 1B).

Among the invasive samples, Hg concentrations in blood, liver, and

Table 1

Dry weight concentrations of Hg in kidney, liver and faeces, muscle, brain and fur (µg/g), and wet weight concentrations of blood and milk (µg/L).

Tissue	n	Mean [Hg]	Median	SD	Min	Max
Kidney	36	0.68	0.64	0.31	0.15	1.50
Liver	35	0.15	0.14	0.06	0.07	0.37
Faeces	36	0.08	0.08	0.03	0.03	0.18
Muscle	36	9.50×10^{-3}	6.21×10^{-3}	6.95×10^{-3}	2.66×10^{-3}	25.9×10^{-3}
Brain	15	2.04×10^{-3}	1.68×10^{-3}	1.18×10^{-3}	1.08×10^{-3}	4.71×10^{-3}
Fur	33	13.5×10^{-3}	10.9×10^{-3}	10.7×10^{-3}	1.77×10^{-3}	42.0×10^{-3}
Blood	17	0.47×10^{-3}	0.33×10^{-3}	0.32×10^{-3}	0.14×10^{-3}	1.27×10^{-3}
Milk	7	1.55×10^{-3}	1.56×10^{-3}	0.75×10^{-3}	0.49×10^{-3}	2.49×10^{-3}

kidney showed significant, positive correlations (Fig. 1C). Hg levels in muscle tissue correlated significantly with liver, kidney, and blood levels, whereas Hg concentration in brain tissue showed no correlation with any of the other samples. The Hg concentration of the non-invasive faecal samples correlated significantly with blood, liver, and kidney concentrations (Fig. 1C). The strongest relationship was found between faecal and liver Hg concentration ($\beta = 0.52$, $R^2 = 0.33$, $p < 0.001$, Fig. 2).

The relationships between age and Hg, Cd, and Pb are visualised in Fig. 3, where kidney and liver Cd concentrations were strongly positively correlated to age ($R^2 = 0.83$ and $R^2 = 0.63$, respectively). In contrast, there were no clear indications of soft tissue accumulation of Hg or Pb with increasing age.

A between-year difference in Hg concentration was observed in both fur (9.22 ng/g versus 18.7 ng/g) and faecal (0.06 µg/g versus 0.10 µg/g) samples, with significantly higher concentrations obtained from samples

collected in 2022 ($p < 0.01$). No significant annual differences were found in muscle (7.56 ng/g versus 11.4 ng/g), liver (0.14 µg/g versus 0.15 µg/g) or kidney (0.69 µg/g versus 0.67 µg/g) samples. As only samples from 2021 were included in the elemental composition analysis (i.e., Cd, Pb, Se, Al, Fe, Si, S and P), the between-year variation could not be assessed.

The highest Al, Fe, and Si concentrations were found in reindeer faeces (5.56 mg/g, 3.78 mg/g, and 2.13 mg/g, respectively). In contrast, faeces generally contained the lowest concentrations of both S and P. Concentrations of S were highest in fur (25.1 mg/g), followed by the internal tissues muscle>kidney>liver, and faeces (Table 2).

The correlations between all the different element concentrations in the non-invasive samples (i.e., faeces and fur) from Svalbard reindeer are visualised in Fig. 4. The strongest relationships were among elements Pb, Al, Fe, and Se in faeces ($p < 0.001$) and between Pb, Al, Fe, and Si in fur ($p < 0.001$).

Table 2

Dry weight concentrations of Cd, Pb, Se, Al, Fe, Si, S and P (µg/g) in kidney, liver, muscle, faeces and fur from Svalbard reindeer ($n = 18$).

| Element	Tissue	Mean	Median	SD	Min	Max
Cd	Kidney	16.3	11.9	11.2	2.32	38.5
	Liver	1.48	1.55	0.58	0.42	2.63
	Muscle	4.43×10^{-3}	3.24×10^{-3}	3.25×10^{-3}	0.22×10^{-3}	10.5×10^{-3}
	Fur	1.94×10^{-3}	1.48×10^{-3}	1.82×10^{-3}	0.21×10^{-3}	7.39×10^{-3}
	Faeces	0.74	0.70	0.20	0.23	1.13
Pb	Kidney	0.17	0.13	0.13	0.07	0.54
	Liver	0.28	0.24	0.18	0.08	0.84
	Muscle	2.90×10^{-3}	2.44×10^{-3}	2.49×10^{-3}	0.41×10^{-3}	6.29×10^{-3}
	Fur	84.0×10^{-3}	50.3×10^{-3}	93.5×10^{-3}	9.05×10^{-3}	345×10^{-3}
	Faeces	2.62	2.59	1.18	0.76	5.05
Se	Kidney	3.95	4.02	0.29	3.08	4.27
	Liver	1.41	1.34	0.27	0.99	2.22
	Muscle	0.59	0.58	0.11	0.36	0.81
	Fur	0.28	0.27	0.08	0.14	0.43
	Faeces	0.45	0.46	0.12	0.28	0.72
Al	Kidney	1.41	1.11	0.77	0.63	3.52
	Liver	1.67	1.52	0.71	0.71	3.95
	Muscle	0.68	0.73	0.30	0.21	1.18
	Fur	125	64.4	190	2.48	807
	Faeces	5.56×10^3	5.06×10^3	2.74×10^3	1.50×10^3	12.6×10^3
Fe	Kidney	230	223	32.5	178	316
	Liver	1270	1280	353	661	2010
	Muscle	133	131	22.0	87.0	176
	Fur	85.4	48.7	121	5.51	521
	Faeces	3.78×10^3	3.47×10^3	1.72×10^3	1.31×10^3	8.38×10^3
Si	Kidney	26.5	27.5	4.76	16.6	31.9
	Liver	22.1	22.3	4.36	14.1	32.4
	Muscle	15.5	14.9	3.00	9.41	22.0
	Fur	238	125	360	6.38	1530
	Faeces	2.13×10^3	2.10×10^3	0.14×10^3	1.89×10^3	2.41×10^3
S	Kidney	9.21×10^3	9.26×10^3	0.38×10^3	8.35×10^3	9.83×10^3
	Liver	8.25×10^3	8.26×10^3	0.24×10^3	7.75×10^3	8.65×10^3
	Muscle	9.74×10^3	9.80×10^3	0.78×10^3	7.69×10^3	10.7×10^3
	Fur	25.1×10^3	25.5×10^3	4.68×10^3	10.7×10^3	30.6×10^3
	Faeces	2.98×10^3	2.87×10^3	0.44×10^3	2.49×10^3	4.33×10^3
P	Kidney	12.6×10^3	12.6×10^3	0.82×10^3	10.9×10^3	13.8×10^3
	Liver	11.2×10^3	11.35×10^3	0.43×10^3	10.1×10^3	11.8×10^3
	Muscle	7.31×10^3	7.42×10^3	0.61×10^3	5.43×10^3	8.00×10^3
	Fur	127	131	27.4	48.9	161
	Faeces	3.22×10^3	3.14×10^3	0.52×10^3	2.27×10^3	4.30×10^3
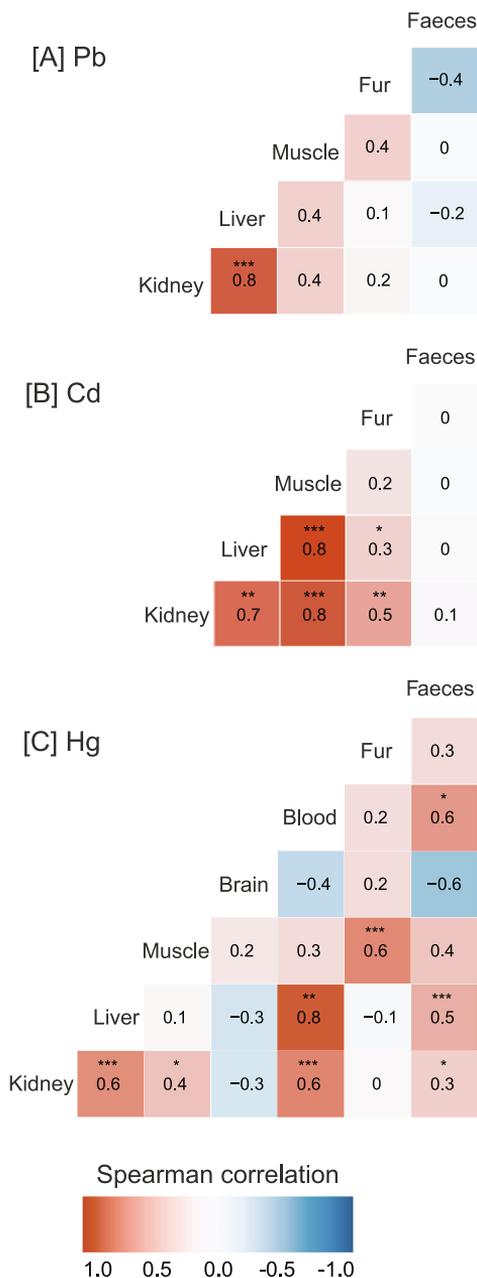
Fig. 1. Spearman correlation heat maps visualising the relationship between different tissue concentrations of [A] Pb ($n = 18$), [B] Cd ($n = 18$) and [C] Hg ($n = 36$). The Spearman correlation coefficient (ρ) is provided in the boxes. Significant relationships are marked as (*) if $p < 0.05$, as (**) if $p < 0.01$ and as (***) if $p < 0.001$.

4. Discussion

Among all the various sample types, the highest concentrations of both Hg and Cd were found in the kidneys (0.68 µg/g and 16.3 µg/g, respectively). In contrast, the highest concentration of Pb was found in
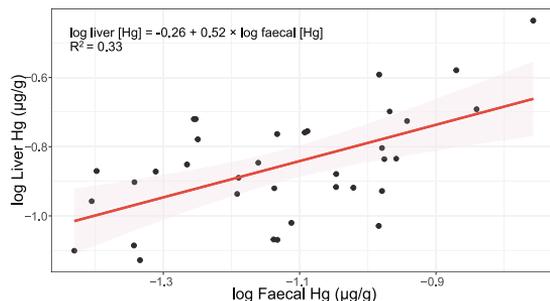

Fig. 2. Linear regression between log faecal and liver Hg concentrations ($n = 35$). Each dot represents an individual measurement, the red line represents the linear regression line, and the pink ribbon shows the 95 % confidence interval (CI). The equation predicts liver Hg concentrations with an accuracy of 107 ± 32 %.

faeces (2.62 µg/g), while the internal tissue concentrations were tenfold lower, with the highest concentrations found in the liver (0.28 µg/g). The different fates of Hg, Cd, and Pb in the Svalbard reindeer are most likely attributed to a combination of different levels of exposure, uptake, excretion pathways, and the chemical properties of the elements, which is further discussed in the following sections.

4.1. Hg in the Svalbard reindeer

Hg bioaccumulates and biomagnifies across trophic levels (Dietz et al., 2000). As a result, marine apex predators typically exhibit the highest concentrations of Hg among Arctic mammals (Scheuhammer et al., 2015). As primary consumers, it is, therefore, not surprising that the renal concentrations of Hg in the Svalbard reindeer (0.68 µg/g dw) are manifold lower than those of polar bears (hepatic:15.5 µg/g dw; Kannan et al., 2007) or Arctic foxes (hepatic: 2.66 µg/g dw, recalculated from ww; Hoekstra et al., 2003). The Svalbard reindeer are, in addition, among the least polluted subspecies of *Rangifer tarandus* when compared to studies from Canada and Greenland (Table 3 and S7).

Several studies have previously observed an east-to-west gradient of Hg in marine biota and seawater, with overall higher concentrations towards Greenland and the Canadian Arctic than in Svalbard (Rigét et al., 2011; Dietz et al., 2022). In line with previous circumpolar findings, we generally found the lowest Hg concentrations in reindeer from Svalbard and Scandinavia (Table 3), suggesting that geographical differences in Hg deposition and emission also may influence the exposure of terrestrial mammals.

Besides geography, differences in diet may also significantly affect Hg exposure, uptake, and accumulation as some sources of feed may be more proficient in the uptake of Hg than others. Lichen and bryophytes are, for instance, efficient accumulators of atmospheric Hg (Bargagli, 2016) and, therefore, tend to have higher Hg concentrations than vascular plant species (Wojtuń et al., 2013; Zhou et al., 2021). While lichen play an important role in the diet of some subspecies of reindeer (Gamberg et al., 2005), several studies on the botanical composition of Svalbard reindeer rumen content indicate that the proportion of lichen in their diet is relatively small (Staaland, 1986; Bjørkvoll et al., 2009; Węgrzyn et al., 2019). This low fraction of lichen is most likely due to past and present overgrazing of their preferred lichen species, which hinders regrowth and restricts their abundance on Svalbard (Staaland et al., 1993; Bjune, 2000). As a result, Svalbard reindeer may experience lower exposure to dietary Hg.

Although Hg concentrations in the Svalbard reindeer are lower than in many other reindeer subspecies, they are still comparable to, or even more contaminated than, cervids living closer to major Hg emission sources. For instance, studies on metal concentrations in populations of
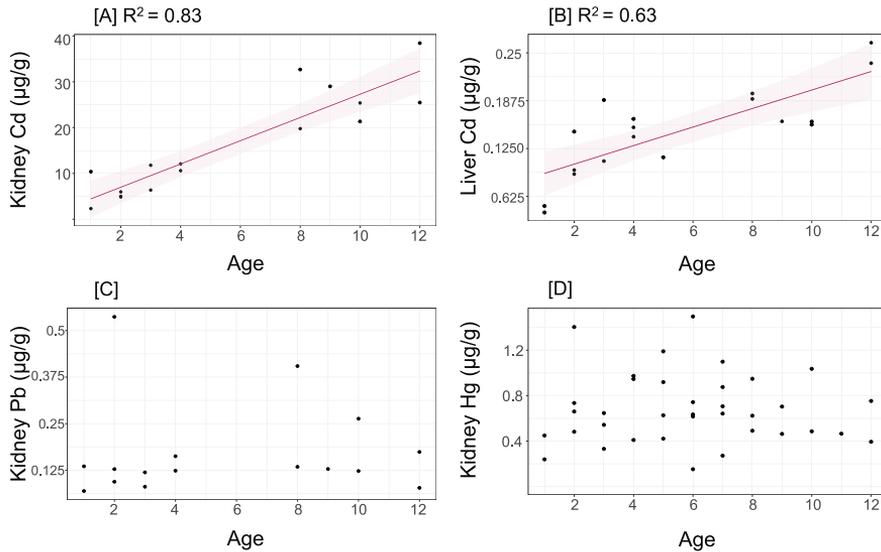
Fig. 3. Linear regression between kidney [A] and liver [B] Cd concentrations and age ($n = 16$ and $n = 18$, respectively). Plots [C] and [D] illustrate the kidney Pb ($n = 16$) and kidney Hg ($n = 35$) concentrations, respectively, as a function of age. Each dot represents an individual measurement. In [A] and [B], the red line represents the linear regression line, and the pink ribbon shows the 95 % confidence interval.
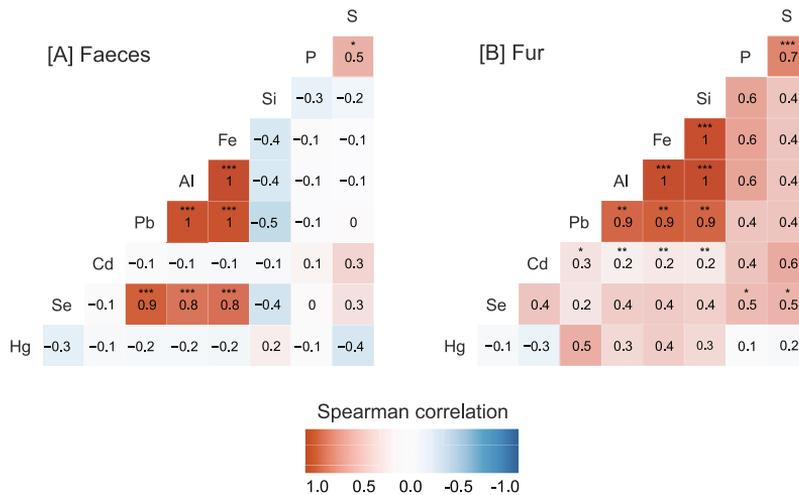
Fig. 4. Spearman correlation heat maps visualising the relationship between different element concentrations in [A] faeces and [B] fur from Svalbard reindeer females ($n = 18$). The Spearman correlation coefficient (ρ) is provided in the boxes. Significant relationships are marked as (*) if $p < 0.05$, as (**) if $p < 0.01$ and as (***) if $p < 0.001$.

red deer (*Cervus elaphus*), roe deer (*Capreolus capreolus*), or fallow deer (*Dama dama*) from central Europe report lower renal and hepatic Hg concentrations than the Svalbard reindeer, despite their closer proximity to large scale coal power plants or heavy industrial activities (Pokorny and Ribarič-Lasnik, 2002; Durkalec et al., 2015; Giżejewska et al., 2017; Vukšić et al., 2018). As herbivores living in one of the most remote areas of the Earth, the relatively high concentrations of Hg in the Svalbard reindeer emphasise the global distribution of pollutants, supporting the argument that the Earth is free from pristine reference sites (e.g., Muir et al., 1992).

The highest concentration of Hg was found in the kidneys of the

Svalbard reindeer, which is common for terrestrial mammals. This has been demonstrated under experimental conditions (Morcillo and Santamaria, 1996), as well as reported in many species of terrestrial wildlife, such as roe deer and wild boar (Srebočan et al., 2011) and grey wolves (*Canis lupus*; McGrew et al., 2014). In contrast, the highest concentrations of Hg in marine mammals are often found in the liver, as reported in several species of dolphins, whales, and seals (Dietz et al., 1990; Wagemann et al., 1998; Frodello et al., 2000; Capelli et al., 2008; AMAP, 2011). The higher renal accumulation of Hg in reindeer, in contrast to the higher hepatic accumulation of Hg in marine mammals, might be caused by exposure to different forms of Hg.

Table 3

A circumpolar overview of previous studies reporting Hg concentrations in kidney and liver samples from the *Rangifer tarandus* family. All concentrations are reported in μg/g and given in wet weight (ww). The studies are given in the order of concentrations, from low to high [Hg], with the current study marked in bold.

Tissue	n	[Hg] μg/g	Location	Year	Subspecies	Reference	Comment
Kidney	26	0.04	Canada	2010–2013	*R. caribou*	Larter et al., 2016	a, e
Kidney	**36**	**0.14**	**Svalbard**	**2021–2022**	***R. platyrhynchus***	**This study**	**b, d**
Kidney	63	0.36	Canada	1994–2006	*R. granti*	Schuster et al., 2011	b, d
Kidney	81	0.38	Greenland	2008–2009	*R. groenlandicus*	Gamberg et al., 2016	a, d
Kidney	315	0.51	Russia	2015–2020	N/A	Makarov et al., 2022	b, c
Kidney	1155	0.91	Greenland & Canada	1991–2016	*R. groenlandicus*	Gamberg et al., 2020	a, c
Kidney	310	0.99	Canada	1994–1995	N/A	Robillard et al., 2002	b, c
Kidney	60	1.90	Canada	1991–1992	N/A	Elkin and Bethke, 1995	b, c
Liver	280	0.04	Sweden	1983–2003	*R. tarandus*	Odsjö et al., 2005	b, e
Liver	**35**	**0.05**	**Svalbard**	**2021–2022**	***R. platyrhynchus***	**This study**	**b, d**
Liver	80	0.07	Greenland	2008–2009	*R. groenlandicus*	Gamberg et al., 2016	a, d
Liver	57	0.08	Norway	1978	N/A	Frøslie et al., 1984	b, c
Liver	101	0.08	Norway	1990–1991	N/A	Sivertsen et al., 1995	b, c
Liver	3	0.12	Canada	1994–2006	*R. granti*	Schuster et al., 2011	b, e
Liver	40	0.16	Russia	1997	*R. tarandus*	Bernhoft et al., 2002	b, c
Liver	126	0.23	Greenland	1995–1997	*R. groenlandicus*	Aastrup et al., 2000	b, c
Liver	505	0.34	Russia	2015–2020	N/A	Makarov et al., 2022	b, c
Liver	50	0.51	Canada	1991–1992	N/A	Elkin and Bethke, 1995	b, c
Liver	307	0.56	Canada	1994–1995	N/A	Robillard et al., 2002	b, c

The numbers in the comments provide additional information about the data, where a = original [Hg] was provided in dry weight (dw) and recalculated to wet weight (ww) according to the % water content provided by the article, b = original [Hg] provided as ww, c = both genders were sampled, d = only females were sampled, and e = only males were sampled.

The kidneys are a major target organ for inorganic Hg, as Hg ions have a high affinity towards sulfhydryl groups, which are abundant in the proteins of the kidney. In fact, up to 90 % of the inorganic Hg in adult mammals is accumulated in the kidneys (Scheuhammer, 1987). Organic mercury, specifically methylmercury (MeHg), is, on the other hand, more likely to be retained in the liver, where it can undergo demethylation and detoxification by forming mercuric selenide, HgSe (Wagemann et al., 1998). The molar Hg kidney:liver ratio can consequently assist in indicating which form of Hg an organism has been exposed to, where a ratio above 1 indicates higher exposure to inorganic Hg, while a ratio equal to or below 1 indicates higher concentrations of organic Hg (Scheuhammer, 1987). The Svalbard reindeer primarily feeds on vascular vegetation and bryophytes, with an occasional supplement of biomass from the marine food web, e.g., kelp and seaweed (Bjørkvoll et al., 2009; Hansen et al., 2019). Although certain species of macroalgae are capable of bioconcentrating MeHg from the sea, inorganic Hg dominates in terrestrial vegetation, and up to 90 % of all terrestrial sequestration of Hg derives from plant uptake of Hg^0 (i.e., gaseous elemental Hg which oxidates into Hg^{2+} after uptake; Obrist et al., 2017; Zhou et al., 2021). As the diet of the Svalbard reindeer is herbivorous and predominantly terrestrial, it is reasonable to assume that their Hg exposure is primarily to the inorganic form. This is supported by the results in this study where the kidney:liver ratio was, on average, 4.8, suggesting a higher exposure to inorganic Hg.

Hg exerts toxic effects even at low concentrations (Klaassen and Watkins, 2015). Consequently, toxicity thresholds for both wildlife and humans have been developed to assess environmental status and avoid adverse health effects in humans. While the Svalbard reindeer are more polluted with Hg than some central European cervids, their Hg concentrations are, similarly to most other reindeer subspecies, far below the current toxicity threshold of 4.2 μg/g (ww) set for terrestrial mammals (Dietz et al., 2022). This implies that the Svalbard reindeer are at low risk of suffering from Hg-related adverse effects. In addition, the Se: Hg molar ratios in the soft tissues (i.e., kidney, liver, muscle) are far higher than 1, meaning that the levels of Se are sufficiently high to both mitigate potential toxic Hg-derived effects while ensuring sufficient Se levels for other physiological functions (Romero et al., 2016).

Furthermore, the concentrations of Hg in Svalbard reindeer (0.07–0.29 μg/g ww in the kidney, see Table S8 for ww tissue concentrations) are lower than the advised threshold of 0.5 μg/g (ww) for human consumption set by The European Commission (2023). This implies that human consumption of both meat and offal (i.e., kidney or liver) from the Svalbard reindeer may be done without major concerns, especially when considering that inorganic Hg most likely is the dominant form of Hg in these reindeer, which is less toxic than MeHg, mostly found in, e.g., fish or marine mammals (EFSA, 2012).

4.2. Cd in the Svalbard reindeer

Similarly to Hg, the highest Cd reported in this study (renal concentrations of 16.3 μg/g dw or 3.31 μg/g ww) are lower than concentrations found in Arctic marine mammals. Dietz et al. (1996) report renal concentrations of Cd in polar bears ranging from 8.21 to 18.6 μg/g ww, in seals from 8.93 to 110 μg/g ww, and in toothed whales from 5.23 to 54.3 μg/g ww. Dietz et al. (2000) have previously demonstrated that herbivores, such as reindeer, can have up to 50 times lower Cd concentrations than marine mammals, such as seals.

The Cd concentrations reported in the present study are also lower than those in Borch-Iohnsen et al. (1996), who reported Cd levels ranging from 0.5 to 23 μg/g ww in renal samples from Svalbard reindeer collected in the mid-1980s. This overall indicates a decrease in Cd concentrations in the Svalbard reindeer over the past decades. Atmospheric Cd concentrations have generally decreased in Europe since the mid-1960s, mainly due to significant technical advancements in emission control techniques developed for key Cd sources such as smelters and powerplants (Pacyna et al., 2009). Likewise, atmospheric Cd concentrations in Svalbard have been reported to decrease by $2.8\%^{y-1}$ since the end of the last century (Platt et al., 2022). Consequently, we suggest that the decrease in Cd concentrations in Svalbard reindeer during the past forty years is due to reduced Cd emissions, resulting in less Cd being transported to the Arctic, hence a lower environmental exposure.

Likewise, the renal Cd concentrations in the Svalbard reindeer in the present study are seemingly similar to those of other *Rangifer tarandus* subspecies, with the exception of certain herds in Canada where renal concentrations as high as 882 μg/g (dw) have been reported (AMAP, 1998). The dietary composition of the reindeer may again explain some of the variations in Cd concentrations among different herds of reindeer. Specifically, high proportions of *Salix* spp. in the diet are strongly linked to increased Cd concentrations in other herbivores such as ptarmigans (*Lagopus L. lagopus and* L. *mutus*; Myklebust et al., 1993; Larison et al., 2000) and a similar relationship in reindeer can be expected.

The dietary composition of the Svalbard reindeer heavily depends on

the season, with consumption of high-biomass species dominating in summer and species growing in exposed, snow-free areas dominating in winter (Staaland, 1984). Some species are, however, omnipresent in their diet throughout the year, such as the dwarf shrub *Salix polaris* (Bjørkvoll et al., 2009). Willows are, in general, well-known for accumulating heavy metals (Luo et al., 2016). This also applies to *S. polaris*, where a study in Svalbard found 15–17 times higher Cd concentrations in the stems and leaves of the plant compared to the surrounding soil (Wojtuń et al., 2013). The concentrations of Cd in Svalbard *S. polaris* range between 0.2 and 8.5 µg/g (dw) (Wojtuń et al., 2013, 2019; Krajcarová et al., 2016), which is slightly lower than concentrations found in the reindeer in this study. Although dietary uptake of Cd is not very high (5–10 %), the reindeer consume a total of 600–700 kg (dw) of plant matter each year (Staaland, 1986; Klaassen and Watkins, 2015). Consequently, *S. polaris* may cause Cd exposure and subsequent accumulation in the kidneys of the Svalbard reindeer.

Unlike Hg, Cd is known to accumulate in the kidneys of both marine and terrestrial organisms (Muir et al., 1992; Riget et al., 2004). Absorbed Cd is initially stored in the liver bound to metallothioneins (MTs), rendering the Cd inert. These Cd-MT complexes may, however, be released from the liver into the circulatory system and reach the kidneys (Klaassen and Watkins, 2015). Through cellular uptake via specific receptors, both free Cd and MT-bound Cd bound can enter the cells of the kidneys. However, once internalised, Cd-MT may be degraded, liberating free Cd in the cell cytoplasm (Johri et al., 2010; Sabolić et al., 2010). The excretion of Cd from the kidney cells is limited, resulting in continuous renal Cd accumulation during chronic exposure (Nordberg, 1984).

Similarly to Hg, the ratios between the Cd concentrations in the liver and kidney can be used to determine the type of Cd exposure an organism has been subjected to. Chronic exposure to Cd will, as described above, result in renal accumulation. Conversely, an acute, high-dose exposure will result in higher levels of Cd in the liver, as the initial storage and binding to MTs occurs there (Scheuhammer, 1987). Consequently, a kidney:liver ratio below 1 would indicate an acute exposure event, while a ratio above 1 suggests chronic exposure. The present study found the average ratio between liver and kidney Cd concentrations to be 11 (liver:kidney ranged between 3.4 and 25), indicating that the Svalbard reindeer are chronically exposed to this toxic metal, which coincides well with their regular intake of *Salix polaris*.

Soft tissue Cd concentrations are known to significantly increase with age in wildlife (Muir et al., 1992; Dietz et al., 1996), which is related to the decadal half-life of Cd in organs such as the kidneys (Klaassen and Watkins, 2015). Thus, with chronic exposure, the Cd levels slowly accumulate and positively correlate with age. In line with these previous studies, we also found a strong correlation between age and soft tissue Cd concentrations (Fig. 3), further indicating chronic exposure to Cd in the Svalbard reindeer.

Cd concentrations exceeding 100 µg/g (ww) are known to cause toxic effects, including increased bone fragility and nephrotoxicity (Larison et al., 2000). The reindeer in the present study ranged from one to twelve years of age. However, not even the oldest sampled individuals (0.45–7.68 µg/g ww) exceeded this toxicity threshold. The oldest recorded female Svalbard reindeer was 17 years (Reimers, 1983), i.e., not much older than the reindeer analysed for in the present study. Consequently, it is unlikely that Svalbard reindeer would experience any adverse health effects related to Cd exposure.

However, with respect to human consumption of the Svalbard reindeer, there are additional thresholds to consider. The European Commission does, for instance, not recommend the consumption of offal with concentrations exceeding 0.5 µg/g ww (The European Commission, 2023). With nearly all kidney and half of the liver samples exceeding this threshold (Table 2 and Table S8), Svalbard reindeer offal should be consumed with caution.

4.3. Pb in the Svalbard reindeer

Pb does not typically biomagnify with increasing trophic levels (Dietz et al., 2000), and the highest Pb concentrations are generally found in soils and terrestrial vegetation compared to the marine environment (Stalwick et al., 2023). Exposure to Pb is thus dependent on the natural geology and the proximity to Pb point sources, which results in large inter- and intra-species variations in Pb concentrations (Muir et al., 1992; Dietz et al., 1996).

Among studies conducted on Arctic herbivores, a wide range of Pb concentrations have been reported, with hepatic concentrations of 0.11–2.88 µg/g (ww) in caribou from Northern Alaska, 0.15–0.52 µg/g (ww) in reindeer from northern Norway and 0.22–0.38 µg/g (ww) in reindeer from St. Lawrence Island in the northern Bering Sea (O'Hara et al., 2003; Welfinger-Smith et al., 2011; Ali Hassan et al., 2012). In comparison, the hepatic concentrations in Svalbard reindeer, ranging from 0.08 to 0.84 µg/g dw (0.02–0.26 µg/g ww), were slightly lower, most likely due to the greater distance to Pb emitting sources on the European or Asian continents (Stalwick et al., 2023).

Moreover, the Pb concentrations in Svalbard reindeer in the present study were lower than those reported by Borch-Iohnsen et al. (1996) in samples collected from the mid-1980s (1.3 µg/g ww). Atmospheric Pb levels in Svalbard have been reported to decrease by an average of 4.6 % annually, largely due to global reductions in Pb emissions following the phase-out of leaded gasoline by the late 20th century (Pacyna et al., 2009; Platt et al., 2022). Similar to Cd, the lower Pb concentrations in Svalbard reindeer today compared to forty years ago suggest reduced environmental exposure to Pb in recent decades. Although atmospheric concentrations of Pb are lower today, legacy emissions (e.g., leaded gasoline and smelting) have enriched the organic layer of Svalbard soils with Pb (Pourrut et al., 2011). Additionally, Pb is naturally present in the geosphere of Svalbard (Dallmann, 2015). Consequently, Svalbard reindeer are still exposed to Pb, though to a lesser extent than in the past.

Like Cd and Hg, grazing animals are primarily exposed to Pb through their diet, which mainly includes the ingestion of plant matter (Ma, 2011; Pareja-Carrera et al., 2014). Although accumulation of Pb from soil is limited in many plant species, Pb concentrations in Svalbard vegetation still range from 0.2 to 32 µg/g (dw) in mosses, 0.1–9.4 µg/g (dw) in vascular vegetation and 0.6–4.6 µg/g (dw) in lichen (Wojtuń et al., 2013, 2019; Krajcarová et al., 2016; Kłos et al., 2017). In addition to vegetation, it is well-known that the Svalbard reindeer often ingest soil particles during grazing, exposing them to soil-derived elements such as Pb (Reimers, 1983; Tyler, 1987), which influences their faecal elemental composition. A study on Svalbard found a strong, positive correlation between elements such as Fe, Al and Pb in soil samples (Halbach et al., 2016). Similarly, we observe a nearly identical positive correlation between Fe, Al, Pb (and Se) in the faecal samples from this study (Fig. 4), suggesting the presence of soil in the Svalbard reindeer faeces. Due to the limited bioavailability of Pb bound to the soil, such particles may pass through the gastrointestinal tract of the reindeer and be excreted with the faeces, with only a little Pb being taken up (Pourrut et al., 2011).

Unlike Hg and Cd, the highest concentration of Pb was found in the faeces, with far lower concentrations in the internal tissues (i.e., liver, kidney and muscle) or fur. The markedly difference between the internal organ and faecal concentration is most likely attributed to both low gastrointestinal absorption (3–10 %) in combination with low retention (around 5 %) of absorbed Pb (Klaassen and Watkins, 2015; ATSDR, 2020). The faecal Pb is, therefore, likely to derive from both unabsorbed Pb originating from plant matter, soil particles passing through the intestines undigested and previously absorbed Pb, which is excreted back into the faeces via the bile (Klaassen and Watkins, 2015).

After uptake from the intestines, Pb primarily distributes to the soft tissues before remobilising into fur and bone, including antlers, where bone stores up to 95 % of the total Pb body burden (Klaassen and Watkins, 2015). Consequently, while bone concentrations may indicate

general long-term exposure patterns, the concentrations in the liver or kidneys tend to mirror the levels in the blood, reflecting the recent exposure (Ma, 2011; ATSDR, 2020). The relatively low-level dietary exposure to bioavailable Pb, combined with the accumulation of Pb in bone, may help explain the low Pb concentrations in the internal tissues of the reindeer. Moreover, as Pb mainly accumulates in bone tissue, which was not analysed in the present study, and soft tissues represent recent exposure rather than an accumulation over time, the lack of a relationship between age and liver Pb concentrations observed in Fig. 3 was expected.

The highest Pb concentrations in soft tissues from this study were found in the liver and kidneys (0.28 µg/g dw and 0.17 µg/g dw, respectively), followed by muscle. This aligns well with previous literature, where the liver and kidneys typically contain the highest concentrations of Pb among soft tissues in mammals (Swaileh et al., 2009; Bischoff et al., 2016; Ma et al., 2020). Many argue that no Pb levels can be considered safe (Wani et al., 2015; Hydeskov et al., 2024). However, with relatively low concentrations in the reindeer soft tissues, we suggest that the Pb levels are most likely not harmful to the reindeer. In contrast, human consumption of Svalbard reindeer offal with regard to Pb levels should be done with some caution. The European Commission (2023) has set the Pb consumption threshold to 0.2 µg/g (ww) in offal, which was exceeded by the individuals with the highest hepatic concentrations in this study (0.02–0.26 µg/g ww; Table S8).

4.4. Using non-invasive samples as proxies for internal concentrations of contaminants

While non-invasive samples like faeces or hair are becoming increasingly popular in contaminant biomonitoring, few studies have attempted to correlate the concentrations of pollutants in faeces with those in soft tissues or target organs to validate the applicability of the non-invasive samples (Jota Baptista et al., 2022). In this study, we found faecal Hg concentrations to significantly correlate with liver Hg levels, fur Hg concentrations to significantly correlate with muscle Hg levels, and fur Cd concentrations to significantly correlate with kidney Cd levels, suggesting that faeces and fur have the potential to act as proxies to estimate internal tissue Hg and Cd in Svalbard reindeer, and potentially other *Rangifer* subspecies.

For non-invasive biomonitoring of Pb in Svalbard reindeer, neither faeces nor fur Pb concentrations correlated with soft tissue levels. In addition, the biomatrices that normally show high potential for non-destructive monitoring (i.e., blood) in other species (Skerfving and Bergdahl, 2007) were not assessed for Pb in our study. In central Europe, roe deer antlers have been used for Pb biomonitoring over the past decades (Pokorny, 2006). Since most absorbed Pb accumulates in bone tissue (Scheuhammer, 1987), reindeer antlers may be a viable option for future biomonitoring efforts of Pb.

4.4.1. Faeces

Faeces is a comparatively easy matrix to use in biomonitoring, as the collection imposes minimal disruption of the target organism and thus requires little ethical consideration. However, for the use of faeces in toxicity assessments, there may be some difficulties interpreting the relevance of the obtained data as faecal elemental concentrations are likely to both reflect the elements excreted from the body, as well as unabsorbed elements from ingestion (Klaassen and Watkins, 2015). Yin et al. (2008) conclude that animal excrement is suitable for monitoring heavy metal contamination when comparing species across different trophic levels or between geographical locations. However, measuring metal concentrations in other tissues is too necessary for a more precise toxicity assessment.

The observed relationship between Hg concentrations in the liver and faeces in the present study may be attributed to several factors. The faecal elemental composition of grazers is known to reflect their dietary intake of elements (Roggeman et al., 2013; Andersson Stavridis et al.,

2024). Granted a retention time of between 21 and 69 h for reindeer (Aagnes and Mathiesen, 1994), the faeces of the reindeer will only mirror the ingestion of vegetation and soil up to three days prior to defecation, meaning that the faeces represent a short-term-exposure window. In contrast, given the Hg biological half-life of around two months, the liver should represent a longer exposure time (Klaassen and Watkins, 2015). However, the confinement by geological features such as mountains and glaciers limit the Svalbard reindeer to a relatively small home range (5–25 km^2; Tyler, 1986; Kinck, 2014). As a result, they graze within a set area their entire lives, leading to relatively consistent exposure to the local environmental Hg. This is supported by the findings of Andersson Stavridis et al. (2024), who reported similar Svalbard reindeer faecal Hg concentrations (0.08 ± 0.02 µg/g dw) in samples collected during spring when compared with those reported in the present study (0.08 ± 0.03 µg/g dw). A consistent exposure to Hg allows the reindeer to potentially reach a steady state of accumulation and excretion, where the liver concentrations also reflect the ongoing exposure through dietary intake. With constant exposure over time, samples reflecting both acute and chronic exposure may mirror each other. The relationship between liver and faecal Hg concentrations may also be further enhanced by enterohepatic circulation, where Hg may cycle between the liver and intestines via the bile, influencing concentrations in both matrices simultaneously (Clarkson and Magos, 2006).

The hypothesis that the Svalbard reindeer reach a steady state of Hg accumulation and excretion is further supported by the lack of relationship between Hg tissue concentrations and age found in the present study, which indicates low Hg accumulation. These results are consistent with the findings of Robillard et al. (2002), who also found no relationship between reindeer age and soft tissue Hg concentrations in their study on caribou from northern Québec.

Similarly to Bechshoft et al. (2019), who developed conversion equations to predict the concentrations of Hg in muscle tissue from fur concentrations in polar bears, we used the relationship between liver and faecal Hg concentrations to produce a linear equation:

log liver [Hg] = −0.26 + 0.52 × log faecal [Hg], (Fig. 2), which may be used to estimate liver Hg concentrations based on the concentrations in faeces. This will allow an estimate of Hg concentrations in the liver without directly interacting with the reindeer, reducing animal stress and harm. Additionally, by relying on faecal samples, larger sample sizes across different populations may be collected without needing permits for lethal sampling, which is particularly beneficial in protected areas like Svalbard or other parts of the Arctic. Fresh faecal samples are favoured, as they have not been subjected to degradation or environmental contamination. Moreover, collecting fresh samples from observed individuals allows the gathering of more information (e.g., biometrics), which could be important for the study. Nonetheless, previous studies on contaminants in Svalbard reindeer found no differences between Hg, Cd, or Pb concentrations in fresh summer faeces and aged winter faeces, indicating that non-fresh faeces also are suitable for contaminant studies (Pacyna et al., 2019), further facilitating sampling for biomonitoring.

It is important to note that the relationship between faeces and liver Hg concentrations, while significant, is subject to certain variability, which may affect the precision of our estimates. However, when predictions based on faecal Hg concentrations were compared to the actual liver Hg concentrations, the model demonstrated an accuracy of 107 % ± 32 %, emphasising that the equation is suitable for indicative purposes, despite the relatively low R^2-value (0.33). Nonetheless, it is important to consider that this model is limited to the faecal concentrations used to create the linear regression, and applying the model to higher concentrations in faeces may result in less accurate predictions, with both possible overestimations and underestimations of internal concentrations. Despite these limitations, this model remains valuable for biomonitoring, as faecal samples can provide indications of internal tissue concentrations and help determine whether more extensive internal monitoring is needed. While it is worth noting that this study was conducted on female Svalbard reindeer and that there could be sex-

related differences in Hg exposure and accumulation, it is unclear if such differences would significantly impact the prediction of the equation presented above. For instance, Gamberg et al. (2020) reported higher renal Hg concentrations in female caribou compared to males, potentially due to increased energy requirements and feed intake during gestation. However, since Gamberg et al. (2020) did not analyse faecal Hg concentrations, it is difficult to conclude whether gender would affect the prediction of soft tissue Hg concentrations using faeces.

To our knowledge, no previous studies have attempted to correlate the concentrations of Hg, Cd or Pb in faeces to those in soft tissues. Based on our findings, we propose that such correlations should be conducted across other species to further evaluate the potential use of faeces in future biomonitoring efforts.

4.4.2. Hair

Hair and other keratinised tissues are among the most common non-invasive samples used for biomonitoring pollution (Jota Baptista et al., 2022). Hair is easily obtained without causing much stress to the animals, and it is easy to transport and store due to its stability and inert nature after formation (Duffy et al., 2005). As hair is formed under constant contact with blood, the concentration of toxic elements in the hair will reflect the blood concentrations during formation, which in turn are influenced by the recent exposure (Wang et al., 2014). There is a strong correlation between hair and blood Hg concentrations in humans (Phelps et al., 1980). Similarly, correlations between contaminant concentrations in hair and soft tissue concentrations have been reported in a range of biota, including for Hg in polar bears (Bechshoft et al., 2019; Born et al., 1991), Hg in Arctic foxes (Treu et al., 2018), Pb and Cd in wolves (Hernández-Moreno et al., 2013), Pb in hedgehogs (*Erinaceus europaeus*; D'Havé et al., 2006) and Pb and Cd in rabbits (*Oryctolagus cuniculus*; Gil-Jiménez et al., 2020). This promotes the use of hair as a predictor for soft tissue contaminant concentrations. However, since hair is in direct contact with the external environment, there are known issues with using hair as a sample matrix for contaminant studies, as it may reflect a combination of both internal offloading and external contamination. External contamination of fur and feathers is commonly reported (e.g., Scheuhammer, 1987; Gil-Jiménez et al., 2020), and feather concentrations of metals have, for instance, been found to correlate well with soil contamination levels (Tajchman et al., 2023).

The Svalbard reindeer undergoes annual moulting and starts forming new winter fur in late August each year (Cuyler and Øritsland, 2002). Consequently, contaminant levels in the fur from the present study are expected to reflect blood concentrations from late summer. Given the half-life of mercury in blood (up to 100 days; Klaassen and Watkins, 2015), blood concentrations measured during the culling in late October should closely reflect those in late August. Therefore, a positive correlation between fur and blood Hg concentrations was expected. Yet, the present study did not find strong relationships between fur and soft tissue concentrations of Hg, Cd, or Pb. This may be due to species-specific metabolism and excretion efficiencies, as suggested by Dietz et al. (2009), which could confound any direct relationship between the matrices. Additionally, as mentioned earlier, external contamination may further influence the connection between fur and blood contaminant levels.

The Svalbard reindeer spend a significant proportion of the day (29–45 %) lying down on the ground ruminating (Tyler, 1987), thus exposing their fur to soil-derived elements such as Al, Fe, and Pb (Halbach et al., 2016). Despite undergoing several cleaning cycles before the analysis, we report a strong intercorrelation between Al, Fe, Pb, and Si ($\rho \geq 0.9$) in the fur samples (Fig. 4), which may indicate external contamination from soil. Consequently, any relationship between elements in fur and other tissues would be affected, which may explain why there is a lack of strong relationships between these samples.

We did, however, find a moderate positive correlation between Hg in fur and muscle ($\rho = 0.6$), similar to Bechshoft et al. (2019), who reported a weak positive relationship ($R^2 = 0.18$–0.24) between Hg concentrations in the same matrices in polar bears, and Treu et al. (2018) who demonstrated positive correlations between hair and soft tissue (i.e., renal and hepatic) Hg concentrations in Arctic fox. We also found a moderate positive correlation between fur and kidney Cd concentrations ($\rho = 0.5$), a relationship previously reported in studies on bats (*Pipistrellus pipistrellus/pygmaeus*) and wood mice (*Apodemus sylvaticus L.*; Beernaert et al., 2007; Tête et al., 2014; Hernout et al., 2016).

As noted, the presence of lithogenic elements (Al, Fe, and Pb) in both fur and faeces suggests that Svalbard reindeer may be both externally exposed to or directly ingesting soil particles containing these elements. We found a strong correlation between Si and lithogenic elements in fur but not faeces. Since Si is a major component of soil, we would expect it to correlate with Al, Fe, and Pb in faeces as well. However, this discrepancy might result from limitations in the analytical method used in this study, specifically nitric acid digestion, which is ineffective at breaking down silicate minerals (Langenfeld and Bugbee, 2023). As a result, it may not fully extract Si from soil particles in the samples. This issue could be more pronounced in faecal samples than the fur samples, as faeces may contain compacted particles that reduce the effectiveness of digestion. In contrast, fur samples, which have a larger overall surface area for digestion, might allow for a more complete extraction of Si. This difference in digestion efficiency could lead to a biased Si recovery rate in faeces, affecting the observed relationship between Si and other lithogenic elements in the samples.

5. Conclusions

In summary, this study provides new insights into the concentration of Hg, Cd, and Pb in Svalbard reindeer. We found the highest concentrations of Hg and Cd in the kidneys, suggesting that the reindeer are chronically exposed to Cd and inorganic Hg. This exposure likely derives from their diet, where both *Salix polaris* (phytoaccumulator of Cd) and bryophytes (accumulators of atmospheric, inorganic Hg) play major roles.

While muscle tissues had low concentrations of Hg, Cd, and Pb, rendering Svalbard reindeer meat relatively safe for consumption, the high hepatic and renal Cd levels, combined with high hepatic Pb concentrations, suggest that Svalbard reindeer offal should be consumed with caution. Concentrations of both Cd and Pb have, however, decreased in Svalbard reindeer hepatic and renal tissues over the past forty years. This likely reflects the impact of technical advancements and stronger emission regulations, causing reduced atmospheric emissions and subsequent reduced bioavailability of these metal contaminants.

Hg concentrations in faeces and liver correlated significantly. Although there are limitations in using faeces as a predictor of soft tissue Hg concentrations, it may still serve as an indicative marker, supporting its use as a non-invasive matrix for Hg biomonitoring in Svalbard reindeer and potentially other *Rangifer* subspecies across the Arctic. Fur Cd and Hg concentrations were positively correlated with soft tissue concentrations. However, our results indicated that external contamination is a confounding factor, complicating the use of fur as a reliable non-invasive biomonitoring matrix for Hg and Cd.

Overall, our study contributes to the knowledge of contaminant exposure in Arctic terrestrial biota and highlights the potential of using non-invasive sampling methods for future biomonitoring efforts across the Arctic.

CRediT authorship contribution statement

Malin Andersson Stavridis: Writing – review & editing, Writing – original draft, Visualization, Formal analysis, Data curation, Conceptualization. **Bjørg Marie Pollestad:** Writing – review & editing, Methodology, Data curation. **Vebjørn Veiberg:** Writing – review & editing, Funding acquisition. **Øyvind Mikkelsen:** Writing – review & editing, Supervision. **Tomasz Maciej Ciesielski:** Writing – review & editing,

Supervision. **Bjørn Munro Jenssen:** Writing – review & editing, Supervision, Conceptualization.

Declaration of generative AI and AI-assisted technologies in the writing process

During the preparation of this work, the authors used ChatGPT (OpenAI., 2023, Mar 14 version) [Large language model] to improve the grammar and readability of the manuscript. After using this tool, the authors reviewed and edited the content as needed and take(s) full responsibility for the content of the publication.

Declaration of competing interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests: Vebjørn Veiberg reports financial support was provided by Research Council of Norway. Malin Andersson Stavridis reports financial support was provided by Research Council of Norway. If there are other authors, they declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.scitotenv.2024.178226.

Data availability

Toxic Metals and Other Elements in Svalbard Reindeer (Reference data) (Mendeley Data)

References

Aagnes, T.H., Mathiesen, S.D., 1994. Food and snow intake, body mass and rumen function in reindeer fed lichen and subsequently starved for 4 days. Rangifer 14, 33. https://doi.org/10.7557/2.14.1.1131.

Aastrup, P., Riget, F., Dietz, R., Asmund, G., 2000. Lead, zinc, cadmium, mercury, selenium and copper in Greenland caribou and reindeer (*Rangifer tarandus*). Sci. Total Environ. 245, 149–159. https://doi.org/10.1016/S0048-9697(99)00440-4.

Ali Hassan, A., Rylander, C., Brustad, M., Sandanger, T.M., 2012. Level of selected toxic elements in meat, liver, tallow and bone marrow of young semi-domesticated reindeer (*Rangifer tarandus tarandus L.*) from northern Norway. Int. J. Circumpolar Health 71, 18187. https://doi.org/10.3402/ijch.v71i0.18187.

AMAP, 1998. AMAP Assessment Report: Arctic Pollution Issues. The Programme, Oslo, Norway.

AMAP, 2011. AMAP assessment 2011. Arctic Monitoring and Assessment Programme, Oslo, Norway.

AMAP, 2018. AMAP Assessment 2018: Biological Effects of Contaminants on Arctic Wildlife and Fish. Oslo, Norway.

Andersson Stavridis, M., Røed, S.B., Hansen, B.B., Mikkelsen, Ø., Ciesielski, T.M., Jenssen, B.M., 2024. Tracing the footprints of Arctic pollution: spatial variations in toxic and essential elements in Svalbard reindeer (*Rangifer tarandus platyrhynchus*) faeces. Sci. Total Environ. 906, 167562. https://doi.org/10.1016/j.scitotenv.2023.167562.

ATSDR, 2020. Toxicological profile for lead. U.S. Department of Health and Human Services. Centers for Disease Control and Prevention (U.S.). Agency for Toxic Substances and Disease Registry. Agency for Toxic Substances and Disease Registry. Doi:10.15620/cdc:95222.

Barbante, C., Spolaor, A., Cairns, W.R., Boutron, C., 2017. Man's footprint on the Arctic environment as revealed by analysis of ice and snow. Earth Sci. Rev. 168, 218–231. https://doi.org/10.1016/j.earscirev.2017.02.010.

Bargagli, R., 2016. Moss and lichen biomonitoring of atmospheric mercury: A review. Sci. Total Environ. 572, 216–231. https://doi.org/10.1016/j.scitotenv.2016.07.202.

Bechshoft, T., Dyck, M., St. Pierre, K.A., Derocher, A.E., St. Louis, V., 2019. The use of hair as a proxy for total and methylmercury burdens in polar bear muscle tissue. Sci. Total Environ. 686, 1120–1128. https://doi.org/10.1016/j.scitotenv.2019.06.087.

Beernaert, J., Scheirs, J., Leirs, H., Blust, R., Verhagen, R., 2007. Non-destructive pollution exposure assessment by means of wood mice hair. Environ. Pollut. 145, 443–451. https://doi.org/10.1016/j.envpol.2006.04.025.

Bernhoft, A., Waaler, T., Mathiesen, S.D., Flåøyen, A., 2002. Trace elements in reindeer from Rybatsjij Ostrov, north western Russia. Rangifer 22, 67. https://doi.org/10.7557/2.22.1.690.

Bischoff, K., Hillebrandt, J., Erb, H.N., Thompson, B., Johns, S., 2016. Comparison of blood and tissue lead concentrations from cattle with known lead exposure. Food Addit. Contam. Part A 33, 1563–1569. https://doi.org/10.1080/19440049.2016.1230277.

Bjørkvoll, E., Pedersen, B., Hytteborn, H., Jónsdóttir, I.S., Langvatn, R., 2009. Seasonal and interannual dietary variation during winter in female Svalbard reindeer (*Rangifer Tarandus Platyrhynchus*). AAAR 41, 88–96. https://doi.org/10.1657/1523-0430-41.1.88.

Bjune, A.E., 2000. Pollen analysis of faeces as a method of demonstrating seasonal variations in the diet of Svalbard reindeer (*Rangifer tarandus platyrhynchus*). Polar Res. 19, 183–192. https://doi.org/10.3402/polar.v19i2.6544.

Borch-Iohnsen, B., Nilssen, K.J., Norheim, G., 1996. Influence of season and diet on liver and kidney content of essential elements and heavy metals in Svalbard reindeer. Biol. Trace Elem. Res. 51, 235–247. https://doi.org/10.1007/BF02784078.

Born, E.W., Renzoni, A., Dietz, R., 1991. Total mercury in hair of polar bears (*Ursus maritimus*) from Greenland and Svalbard. Polar Res. 9, 113–120. https://doi.org/10.3402/polar.v9i2.6784.

Capelli, R., Das, K., Pellegrini, R.D., Drava, G., Lepoint, G., Miglio, C., Minganti, V., Poggi, R., 2008. Distribution of trace elements in organs of six species of cetaceans from the Ligurian Sea (Mediterranean), and the relationship with stable carbon and nitrogen ratios. Sci. Total Environ. 390, 569–578. https://doi.org/10.1016/j.scitotenv.2007.10.036.

Cardona-Marek, T., Knott, K.K., Meyer, B.E., O'Hara, T.M., 2009. Mercury concentrations in southern Beaufort Sea polar bears: variation based on stable isotopes of carbon and nitrogen. Environ. Toxicol. Chem. 28, 1416–1424. https://doi.org/10.1897/08-557.1.

Clarkson, T.W., Magos, L., 2006. The toxicology of mercury and its chemical compounds. Crit. Rev. Toxicol. 36, 609–662. https://doi.org/10.1080/10408440600845619.

Cuyler, C., Øritsland, N.A., 2002. Do seasonal changes in Svalbard reindeer fur have relevance for heat transfer? Rangifer 22, 133. https://doi.org/10.7557/2.22.2.1532.

Dallmann, W.K., 2015. Geoscience Atlas of Svalbard. Norsk polarinstitutt, Tromsø.

D'Havé, H., Scheirs, J., Mubiana, V.K., Verhagen, R., Blust, R., De Coen, W., 2006. Non-destructive pollution exposure assessment in the European hedgehog (*Erinaceus europaeus*): II. Hair and spines as indicators of endogenous metal and as concentrations. Environ. Pollut. 142, 438–448. https://doi.org/10.1016/j.envpol.2005.10.021.

Dietz, R., Nielsen, C.O., Hansen, M.M., Hansen, C.T., 1990. Organic mercury in Greenland birds and mammals. Sci. Total Environ. 95, 41–51. https://doi.org/10.1016/0048-9697(90)90051-U.

Dietz, R., Riget, F., Johansen, P., 1996. Lead, cadmium, mercury and selenium in Greenland marine animals. Sci. Total Environ. 186, 67–93. https://doi.org/10.1016/0048-9697(96)05086-3.

Dietz, R., Riget, F., Cleemann, M., Aarkrog, A., Johansen, P., Hansen, J., 2000. Comparison of contaminants from different trophic levels and ecosystems. Sci. Total Environ. 245, 221–231. https://doi.org/10.1016/S0048-9697(99)00447-7.

Dietz, R., Outridge, P.M., Hobson, K.A., 2009. Anthropogenic contributions to mercury levels in present-day Arctic animals—A review. Sci. Total Environ. 407, 6120–6131. https://doi.org/10.1016/j.scitotenv.2009.08.036.

Dietz, R., Letcher, R.J., Aars, J., Andersen, M., Boltunov, A., Born, E.W., Ciesielski, T.M., Das, K., Dastnai, S., Derocher, A.E., Desforges, J.-P., Eulaers, I., Ferguson, S., Hallanger, I.G., Heide-Jørgensen, M.P., Heimbürger-Boavida, L.-E., Hoekstra, P.F., Jenssen, B.M., Kohler, S.G., Larsen, M.M., Lindstrøm, U., Lippold, A., Morris, A., Nabe-Nielsen, J., Nielsen, N.H., Peacock, E., Pinzone, M., Rigét, F.F., Rosing-Asvid, A., Routti, H., Siebert, U., Stenson, G., Stern, G., Strand, J., Søndergaard, J., Treu, G., Víkingsson, G.A., Wang, F., Welker, J.M., Wiig, Ø., Wilson, S., Sonne, C., 2022. A risk assessment review of mercury exposure in Arctic marine and terrestrial mammals. Sci. Total Environ. 154445 https://doi.org/10.1016/j.scitotenv.2022.154445.

Dolan, K.J., Ciesielski, T.M., Lierhagen, S., Eulaers, I., Nygård, T., Johnsen, T.V., Gómez-Ramírez, P., García-Fernández, A.J., Bustnes, J.O., Ortiz-Santaliestra, M.E., Jaspers, V.L.B., 2017. Trace element concentrations in feathers and blood of northern goshawk (*Accipiter gentilis*) nestlings from Norway and Spain. Ecotox. Environ. Saf. 144, 564–571. https://doi.org/10.1016/j.ecoenv.2017.06.062.

Duffy, L.K., Duffy, R.S., Finstad, G., Gerlach, C., 2005. A note on mercury levels in the hair of Alaskan reindeer. Sci. Total Environ. 339, 273–276. https://doi.org/10.1016/j.scitotenv.2004.12.003.

Durkalec, M., Szkoda, J., Kolacz, R., Opalinski, S., Nawrocka, A., Zmudzki, J., 2015. Bioaccumulation of lead, cadmium and mercury in roe deer and wild boars from areas with different levels of toxic metal pollution. IJER 9. https://doi.org/10.22059/ijer.2015.890.

EFSA, 2012. Scientific opinion on the risk for public health related to the presence of mercury and methylmercury in food. EFSA J. 10. https://doi.org/10.2903/j.efsa.2012.2985.

Eide, N.E., Eid, P.M., Prestrud, P., Swenson, J.E., 2005. Dietary responses of arctic foxes *Alopex lagopus* to changing prey availability across an Arctic landscape. Wildl. Biol. 11, 109–121. https://doi.org/10.2981/0909-6396(2005)11[109:DROAFA]2.0.CO;2.

Elkin, B.T., Bethke, R.W., 1995. Environmental contaminants in caribou in the Northwest Territories, Canada. Sci. Total Environ. 160–161, 307–321. https://doi.org/10.1016/0048-9697(95)04365-8.

Evans, R.D., Addison, E.M., Villeneuve, J.Y., MacDonald, K.S., Joachim, D.G., 1998. An examination of spatial variation in mercury concentrations in otter (*Lutra canadensis*) in south-central Ontario. Sci. Total Environ. 213, 239–245. https://doi.org/10.1016/S0048-9697(98)00096-5.

Frodello, J.P., Roméo, M., Viale, D., 2000. Distribution of mercury in the organs and tissues of five toothed-whale species of the Mediterranean. Environ. Pollut. 108, 447–452. https://doi.org/10.1016/S0269-7491(99)00221-3.

Frøslie, A., Norheim, G., Rambæk, J.P., Steinnes, E., 1984. Levels of trace elements in liver from Norwegian moose, reindeer and red deer in relation to atmospheric deposition. Acta Vet. Scand. 25, 333–345. https://doi.org/10.1186/BF03547248.

Gamberg, M., Braune, B., Davey, E., Elkin, B., Hoekstra, P.F., Kennedy, D., Macdonald, C., Muir, D., Nirwal, A., Wayland, M., Zeeb, B., 2005. Spatial and temporal trends of contaminants in terrestrial biota from the Canadian Arctic. Sci. Total Environ. 351–352, 148–164. https://doi.org/10.1016/j.scitotenv.2004.10.032.

Gamberg, M., Cuyler, C., Wang, X., 2016. Contaminants in two West Greenland caribou populations. Sci. Total Environ. 554–555, 329–336. https://doi.org/10.1016/j.scitotenv.2016.02.154.

Gamberg, M., Pratte, I., Brammer, J., Cuyler, C., Elkin, B., Gurney, K., Kutz, S., Larter, N.C., Muir, D., Wang, X., Provencher, J.F., 2020. Renal trace elements in barren-ground caribou subpopulations: temporal trends and differing effects of sex, age and season. Sci. Total Environ. 724, 138305. https://doi.org/10.1016/j.scitotenv.2020.138305.

Gil-Jiménez, E., Mateo, R., De Lucas, M., Ferrer, M., 2020. Feathers and hair as tools for non-destructive pollution exposure assessment in a mining site of the Iberian Pyrite Belt. Environ. Pollut. 263, 114523. https://doi.org/10.1016/j.envpol.2020.114523.

Giżejewska, A., Szkoda, J., Nawrocka, A., Żmudzki, J., Giżejewski, Z., 2017. Can red deer antlers be used as an indicator of environmental and edible tissues' trace element contamination? Environ. Sci. Pollut. Res. 24, 11630–11638. https://doi.org/10.1007/s11356-017-8798-7.

Halbach, K., Mikkelsen, Ø., Steinnes, E., 2016. Study of Mercury and Selected Trace Elements in Soil in the Norwegian Arctic, Svalbard (MSc thesis). The Norwegian University of Science and Technology, Trondheim.

Halbach, K., Mikkelsen, Ø., Berg, T., Steinnes, E., 2017. The presence of mercury and other trace metals in surface soils in the Norwegian Arctic. Chemosphere 188, 567–574. https://doi.org/10.1016/j.chemosphere.2017.09.012.

Hansen, B.B., Grøtan, V., Aanes, R., Sæther, B.-E., Stien, A., Fuglei, E., Ims, R.A., Yoccoz, N.G., Pedersen, Å.Ø., 2013. Climate events synchronize the dynamics of a resident vertebrate community in the High Arctic. Science 339, 313–315. https://doi.org/10.1126/science.1226766.

Hansen, B.B., Lorentzen, J.R., Welker, J.M., Varpe, Ø., Aanes, R., Beumer, L.T., Pedersen, Å.Ø., 2019. Reindeer turning maritime: ice-locked tundra triggers changes in dietary niche utilization. Ecosphere 10, e02672. https://doi.org/10.1002/ecs2.2672.

Hernández-Moreno, D., De La Casa Resino, I., Fidalgo, L.E., Llaneza, L., Soler Rodríguez, F., Pérez-López, M., López-Beceiro, A., 2013. Noninvasive heavy metal pollution assessment by means of Iberian wolf (*Canis lupus signatus*) hair from Galicia (NW Spain): a comparison with invasive samples. Environ. Monit. Assess. 185, 10421–10430. https://doi.org/10.1007/s10661-013-3341-x.

Hernout, B.V., McClean, C.J., Arnold, K.E., Walls, M., Baxter, M., Boxall, A.B.A., 2016. Fur: A non-invasive approach to monitor metal exposure in bats. Chemosphere 147, 376–381. https://doi.org/10.1016/j.chemosphere.2015.12.104.

Hoekstra, P., Braune, B., Elkin, B., Armstrong, F., Muir, D., 2003. Concentrations of selected essential and non-essential elements in arctic fox (*Alopex lagopus*) and wolverines (*Gulo gulo*) from the Canadian Arctic. Sci. Total Environ. 309, 81–92. https://doi.org/10.1016/S0048-9697(02)00684-8.

Hydeskov, H.B., Arnemo, J.M., Mills, C.L., Gentle, L.K., Uzal, A., 2024. A global systematic review of lead (Pb) exposure and its health effects in wild mammals. J. Wildl. Dis. 60. https://doi.org/10.7589/JWD-D-23-00055.

Ikemoto, T., Kunito, T., Watanabe, I., Yasunaga, G., Baba, N., Miyazaki, N., Petrov, E.A., Tanabe, S., 2004. Comparison of trace element accumulation in Baikal seals (*Pusa sibirica*), Caspian seals (*Pusa caspica*) and northern fur seals (*Callorhinus ursinus*). Environ. Pollut. 127, 83–97. https://doi.org/10.1016/S0269-7491(03)00251-3.

Johri, N., Jacquillet, G., Unwin, R., 2010. Heavy metal poisoning: the effects of cadmium on the kidney. Biometals 23, 783–792. https://doi.org/10.1007/s10534-010-9328-y.

Jota Baptista, C., Seixas, F., Gonzalo-Orden, J.M., Oliveira, P.A., 2022. Biomonitoring metals and metalloids in wild mammals: invasive versus non-invasive sampling. Environ. Sci. Pollut. Res. 29, 18398–18407. https://doi.org/10.1007/s11356-022-18658-5.

Kannan, K., Agusa, T., Evans, T.J., Tanabe, S., 2007. Trace element concentrations in livers of polar bears from two populations in northern and western Alaska. Arch. Environ. Contam. Toxicol. 53, 473–482. https://doi.org/10.1007/s00244-007-0018-x.

Kinck, C., 2014. Reduced Forage Access Affects Home Range Size and Site Fidelity of Svalbard Reindeer (*Rangifer tarandus platyrhynchus*). NMBU.

Klaassen, C.D., Watkins, J.B. (Eds.), 2015. Casarett & Doull's Essentials of Toxicology, Third, edition. ed. McGraw-Hill, New York.

Kłos, A., Ziembik, Z., Rajfur, M., Dołhańczuk-Śródka, A., Bochenek, Z., Bjerke, J.W., Tømmervik, H., Zagajewski, B., Ziółkowski, D., Jerz, D., Zielińska, M., Krems, P., Godyń, P., 2017. The origin of heavy metals and radionuclides accumulated in the soil and biota samples collected in Svalbard, near Longyearbyen. ECE S 24, 223–238. https://doi.org/10.1515/eces-2017-0015.

Krajcarová, L., Novotný, K., Chattová, B., Elster, J., 2016. Elemental analysis of soils and Salix polaris in the town of Pyramiden and its surroundings (Svalbard). Environ. Sci. Pollut. Res. 23, 10124–10137. https://doi.org/10.1007/s11356-016-6213-4.

Langenfeld, N.J., Bugbee, B., 2023. An improved digestion and analysis procedure for silicon in plant tissue. PloS One 18, e0289151. https://doi.org/10.1371/journal.pone.0289151.

Larison, J.R., Likens, G.E., Fitzpatrick, J.W., Crock, J.G., 2000. Cadmium toxicity among wildlife in the Colorado Rocky Mountains. Nature 406, 181–183. https://doi.org/10.1038/35018068.

Larter, N.C., Macdonald, C.R., Elkin, B.T., Wang, X., Harms, N.J., Gamberg, M., Muir, D.C.G., 2016. Cadmium and other elements in tissues from four ungulate species from the Mackenzie Mountain region of the Northwest Territories, Canada. Ecotox. Environ. Saf. 132, 9–17. https://doi.org/10.1016/j.ecoenv.2016.05.018.

Luo, Z.-B., He, J., Polle, A., Rennenberg, H., 2016. Heavy metal accumulation and signal transduction in herbaceous and woody plants: paving the way for enhancing phytoremediation efficiency. Biotechnol. Adv. 34, 1131–1148. https://doi.org/10.1016/j.biotechadv.2016.07.003.

Ma, H., Shi, G., Cheng, Y., 2020. Accumulation characteristics of metals and metalloids in plants collected from Ny-Ålesund. Arctic. Atmosphere 11, 1129. https://doi.org/10.3390/atmos11101129.

Ma, W.-C., 2011. Lead in mammals. In: Environmental Contaminants in Biota, Interpreting Tissue Concentrations. CRC Press, Taylor & Francis Group.

Makarov, D.A., Ovcharenko, V.V., Nebera, E.A., Kozhushkevich, A.I., Shelepchikov, A.A., Turbabina, K.A., Kalantaenko, A.M., Bardyugov, N.S., Gergel, M.A., 2022. Geographical distribution of dioxins, cadmium, and mercury concentrations in reindeer liver, kidneys, and muscle in the Russian far north. Environ. Sci. Pollut. Res. 29, 12176–12187. https://doi.org/10.1007/s11356-021-16310-2.

McGrew, A.K., Ballweber, L.R., Moses, S.K., Stricker, C.A., Beckmen, K.B., Salman, M.D., O'Hara, T.M., 2014. Mercury in gray wolves (*Canis lupus*) in Alaska: increased exposure through consumption of marine prey. Sci. Total Environ. 468–469, 609–613. https://doi.org/10.1016/j.scitotenv.2013.08.045.

Mierle, G., Addison, E.M., MacDonald, K.S., Joachim, D.G., 2009. Mercury levels in tissues of otters from Ontario, Canada: variation with age, sex, and location. Environ. Toxicol. Chem. 19, 3044–3051. https://doi.org/10.1002/etc.5620191226.

Morcillo, M.A., Santamaria, J., 1996. Mercury distribution and renal metallothionein induction after subchronic oral exposure in rats. Biometals 9, 213–220. https://doi.org/10.1007/BF00817918.

MOSJ, 2021. Uttak av svalbardrein. https://mosj.no/indikator/pavirkning/jakt-og-fangst/uttak-av-svalbardrein/. (Accessed 26 September 2024).

Muir, D.C.G., Wageman, R., Hargrave, B.T., Thomas, D.J., Peakall, D.B., Norstrom, R.J., 1992. Arctic marine ecosystem contamination. Sci. Total Environ. 122, 75–134. https://doi.org/10.1016/0048-9697(92)90246-O.

Myklebust, I., Nybø, S., Kls, J.A., Pedersen, H.C., 1993. Cadmium accumulation in willow ptarmigan (*Lagopus l. lagopus*) and rock ptarmigan (*L. mutus*) in Central Norway. Sci. Total Environ. 134, 135–139. https://doi.org/10.1016/S0048-9697(05)80012-9.

Nordberg, M., 1984. General aspects of cadmium: transport, uptake and metabolism by the kidney. Environ. Health Perspect. 54, 13–20. https://doi.org/10.1289/ehp.845413.

Obrist, D., Agnan, Y., Jiskra, M., Olson, C.L., Colegrove, D.P., Hueber, J., Moore, C.W., Sonke, J.E., Helmig, D., 2017. Tundra uptake of atmospheric elemental mercury drives Arctic mercury pollution. Nature 547, 201–204. https://doi.org/10.1038/nature22997.

Odsjö, T., Räikkönen, J., Bignert, A., 2005. Time trends of metals in liver and muscle of reindeer (*Rangifer tarandus*) from northern and central Lapland, Sweden, 1983-2003. Swedish monitoring programme in terrestrial biota. (Miljögifter i biota -. In: fjäll no, vol. 214. Swedish Museum of Natural History, Stockholm.

O'Hara, T.M., George, J.C., Blake, J., Burek, K., Carroll, G., Dau, J., Bennett, L., McCoy, C.P., Gerard, P., Woshner, V., 2003. Investigation of heavy metals in a large mortality event in caribou of northern Alaska. ARCTIC 56, 125–135. https://doi.org/10.14430/arctic608.

Pacyna, A.D., Koziorowska, K., Chmiel, S., Mazerski, J., Polkowska, Ż., 2018. Svalbard reindeer as an indicator of ecosystem changes in the Arctic terrestrial ecosystem. Chemosphere 203, 209–218. https://doi.org/10.1016/j.chemosphere.2018.03.158.

Pacyna, A.D., Frankowski, M., Kozioł, K., Węgrzyn, M.H., Wietrzyk-Pełka, P., Lehmann-Konera, S., Polkowska, Ż., 2019. Evaluation of the use of reindeer droppings for monitoring essential and non-essential elements in the polar terrestrial environment. Sci. Total Environ. 658, 1209–1218. https://doi.org/10.1016/j.scitotenv.2018.12.232.

Pacyna, J.M., Pacyna, E.G., Aas, W., 2009. Changes of emissions and atmospheric deposition of mercury, lead, and cadmium. Atmos. Environ. 43, 117–127. https://doi.org/10.1016/j.atmosenv.2008.09.066.

Pacyna-Kuchta, A.D., Wietrzyk-Pełka, P., Węgrzyn, M.H., Frankowski, M., Polkowska, Ż., 2020. A screening of select toxic and essential elements and persistent organic pollutants in the fur of Svalbard reindeer. Chemosphere 245, 125458. https://doi.org/10.1016/j.chemosphere.2019.125458.

Pareja-Carrera, J., Mateo, R., Rodríguez-Estival, J., 2014. Lead (Pb) in sheep exposed to mining pollution: implications for animal and human health. Ecotox. Environ. Saf. 108, 210–216. https://doi.org/10.1016/j.ecoenv.2014.07.014.

Phelps, R.W., Clarkson, T.W., Kershaw, T.G., Wheatley, B., 1980. Interrelationships of blood and hair mercury concentrations in a north American population exposed to methylmercury. Arch. Environ. Health 35, 161–168. https://doi.org/10.1080/00039896.1980.10667486.

Platt, S.M., Hov, Ø., Berg, T., Breivik, K., Eckhardt, S., Eleftheriadis, K., Evangeliou, N., Fiebig, M., Fisher, H., Hansen, G., Hansson, H.-C., Heintzenberg, J., Hermansen, O., Heslin-Rees, D., Holmén, K., Hudson, O., Kallenborn, R., Krejci, R., Krognes, T., Larssen, S., Lowry, D., Lund Myhre, C., Lunder, C., Nisbet, E., Nizzetto, P.B., Park, K.-T., Pedersen, C.A., Aspmo Pfaffhuber, K., Röckmann, T., Schmidbauer, N., Solberg, S., Stohl, A., Ström, J., Svendby, T., Tunved, P., Tørnkvist, K., Van Der Veen, C., Vratolis, S., Yoon, Y.J., Yttri, K.E., Zieger, P., Aas, W., Tørseth, K., 2022. Atmospheric composition in the European Arctic and 30 years of the Zeppelin observatory. Ny-Ålesund. Atmos. Chem. Phys. 22, 3321–3369. https://doi.org/10.5194/acp-22-3321-2022.

Pokorny, B., 2006. Roe deer (*Capreolus capreolus L.*) antlers as an accumulative and reactive bioindicator of lead pollution near the largest Slovene thermal power plant. Arch. Vet. 76, S131–S142.

Pokorny, B., Ribarič-Lasnik, C., 2002. Seasonal variability of mercury and heavy metals in roe deer (*Capreolus capreolus*) kidney. Environ. Pollut. 117, 35–46. https://doi.org/10.1016/S0269-7491(01)00161-0.

Pourrut, B., Shahid, M., Dumat, C., Winterton, P., Pinelli, E., 2011. Lead uptake, toxicity, and detoxification in plants. In: Whitacre, D.M. (Ed.), Reviews of Environmental Contamination and Toxicology Volume 213, Reviews of Environmental Contamination and Toxicology. Springer, New York, New York, NY, pp. 113–136. https://doi.org/10.1007/978-1-4419-9860-6_4.

Prestrud, P., 1992. Food habits and observations of the hunting behaviour of arctic foxes, *Alopex lagopus*. In: Svalbard. Can. Field-Nat., 106, pp. 225–236.

R Core Team, 2023. R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing, Vienna, Austria. https://www.R-project.org/.

Reimers, E., 1983. Mortality in Svalbard reindeer. Ecography 6, 141–149. https://doi.org/10.1111/j.1600-0587.1983.tb01075.x.

Riget, F., Dietz, R., Vorkamp, K., Johansen, P., Muir, D., 2004. Levels and spatial and temporal trends of contaminants in Greenland biota: an updated review. Sci. Total Environ. 331, 29–52. https://doi.org/10.1016/j.scitotenv.2004.03.022.

Rigét, F., Braune, B., Bignert, A., Wilson, S., Aars, J., Born, E., Dam, M., Dietz, R., Evans, M., Evans, T., Gamberg, M., Gantner, N., Green, N., Gunnlaugsdóttir, H., Kannan, K., Letcher, R., Muir, D., Roach, P., Sonne, C., Stern, G., Wiig, Ø., 2011. Temporal trends of Hg in Arctic biota, an update. Sci. Total Environ. 409, 3520–3526. https://doi.org/10.1016/j.scitotenv.2011.05.002.

Robillard, S., Beauchamp, G., Paillard, G., Bélanger, D., 2002. Levels of cadmium, lead, mercury and 137caesium in caribou (*Rangifer tarandus*) tissues from northern Québec. Arctic 55 (1), 1–9.

Roggeman, S., van den Brink, N., Van Praet, N., Blust, R., Bervoets, L., 2013. Metal exposure and accumulation patterns in free-range cows (*Bos taurus*) in a contaminated natural area: influence of spatial and social behavior. Environ. Pollut. 172, 186–199. https://doi.org/10.1016/j.envpol.2012.09.006.

Romero, M.B., Polizzi, P., Chiodi, L., Das, K., Gerpe, M., 2016. The role of metallothioneins, selenium and transfer to offspring in mercury detoxification in Franciscana dolphins (*Pontoporia blainvillei*). Mar. Pollut. Bull. 109, 650–654. https://doi.org/10.1016/j.marpolbul.2016.05.012.

Sabolić, I., Breljak, D., Škarica, M., Herak-Kramberger, C.M., 2010. Role of metallothionein in cadmium traffic and toxicity in kidneys and other mammalian organs. Biometals 23, 897–926. https://doi.org/10.1007/s10534-010-9351-z.

Schaefer, K., Elshorbany, Y., Jafarov, E., Schuster, P.F., Striegl, R.G., Wickland, K.P., Sunderland, E.M., 2020. Potential impacts of mercury released from thawing permafrost. Nat. Commun. 11, 4650. https://doi.org/10.1038/s41467-020-18398-5.

Scheuhammer, A., Braune, B., Chan, H.M., Frouin, H., Krey, A., Letcher, R., Loseto, L., Noël, M., Ostertag, S., Ross, P., Wayland, M., 2015. Recent progress on our understanding of the biological effects of mercury in fish and wildlife in the Canadian Arctic. Sci. Total Environ. 509–510, 91–103. https://doi.org/10.1016/j.scitotenv.2014.05.142.

Scheuhammer, A.M., 1987. The chronic toxicity of aluminium, cadmium, mercury, and lead in birds: A review. Environ. Pollut. 46, 263–295. https://doi.org/10.1016/0269-7491(87)90173-4.

Schuster, R.C., Gamberg, M., Dickson, C., Chan, H.M., 2011. Assessing risk of mercury exposure and nutritional benefits of consumption of caribou (*Rangifer tarandus*) in the Vuntut Gwitchin first nation community of old crow, Yukon, Canada. Environ. Res. 111, 881–887. https://doi.org/10.1016/j.envres.2011.05.025.

Sivertsen, T., Daae, H.L., Godal, A., Sand, G., 1995. Ruminant uptake of nickel and other elements from industrial air pollution in the Norwegian-Russian border area. Environ. Pollut. 90, 75–81. https://doi.org/10.1016/0269-7491(94)00091-Q.

Skerfving, S., Bergdahl, I.A., 2007. Lead. In: Handbook on the toxicology of metals. Elsevier, pp. 599–643. https://doi.org/10.1016/B978-012369413-3/50086-0.

Srebočan, E., Prevendar Crnić, A., Ekert-Kabalin, A.M., Lazarus, M., Jurasović, J., Tomljanović, K., Andreić, D., Strunjak Perović, I., Čož-Rakovac, R., 2011. Cadmium, lead, and mercury concentrations in tissues of roe deer (*Capreolus capreolus L.*) and wild boar (*Sus scrofa L.*) from lowland Croatia. Czech J. Food Sci. 29, 624–633. https://doi.org/10.17221/249/2010-CJFS.

Staaland, H., 1984. On the quality of Svalbard reindeer pasture in the summer and autumn. Rangifer 4, 16. https://doi.org/10.7557/2.4.1.487.

Staaland, H., 1986. Svalbardreinens ernæring. In: Svalbardreinen Og Dens Livsgrunnlag. Universitetsforlaget AS, pp. 72–91.

Staaland, H., Scheie, J.O., Grøndahl, F.A., Persen, E., Leifseth, A.B., Holand, Ø., 1993. The introduction of reindeer to Brøggerhalvøya, Svalbard: grazing preference and effect on vegetation. Rangifer 13, 15. https://doi.org/10.7557/2.13.1.1067.

Stalwick, J.A., Ratelle, M., Gurney, K.E.B., Drysdale, M., Lazarescu, C., Comte, J., Laird, B., Skinner, K., 2023. Sources of exposure to lead in Arctic and subarctic regions: a scoping review. Int. J. Circumpolar Health 82, 2208810. https://doi.org/10.1080/22423982.2023.2208810.

Stempniewicz, L., Kulaszewicz, I., Aars, J., 2021. Yes, they can: polar bears *Ursus maritimus* successfully hunt Svalbard reindeer *Rangifer tarandus platyrhynchus*. Polar Biol. 44, 2199–2206. https://doi.org/10.1007/s00300-021-02954-w.

Swaileh, K.M., Abdulkhaliq, A., Hussein, R.M., Matani, M., 2009. Distribution of toxic metals in organs of local cattle, sheep, goat and poultry from the West Bank, Palestinian authority. Bull. Environ. Contam. Toxicol. 83, 265–268. https://doi.org/10.1007/s00128-009-9704-x.

Tajchman, K., Drabik, K., Ukalska-Jaruga, A., Janiszewski, P., Spustek, D., Wengerska, K., 2023. The screening method for use of wild pheasant feathers in the monitoring of environmental pollution with heavy metals. Sci. Rep. 13, 6540. https://doi.org/10.1038/s41598-023-33649-3.

Tête, N., Afonso, E., Crini, N., Drouhot, S., Prudent, A.-S., Scheifler, R., 2014. Hair as a noninvasive tool for risk assessment: do the concentrations of cadmium and lead in the hair of wood mice (*Apodemus sylvaticus*) reflect internal concentrations? Ecotoxicol. Environ. Saf. 108, 233–241. https://doi.org/10.1016/j.ecoenv.2014.07.010.

The European Commission, 2023. COMMISSION REGULATION (EU) 2023/915 of 25 April 2023 on Maximum Levels for Certain Contaminants in Food and Repealing Regulation (EC) no 1881/2006.

Treu, G., Krone, O., Unnsteinsdóttir, E.R., Greenwood, A.D., Czirják, G.Á., 2018. Correlations between hair and tissue mercury concentrations in Icelandic arctic foxes (*Vulpes lagopus*). Sci. Total Environ. 619–620, 1589–1598. https://doi.org/10.1016/j.scitotenv.2017.10.143.

Tyler, N.J.C., 1986. Reinen i Adventdalen. In: Svalbardreinen Og Dens Livsgrunnlag. Universitetsforlaget AS, pp. 143–159.

Tyler, N.J.C., 1987. Natural limitation of the abundance of the high Arctic Svalbard reindeer. University Of Cambridge. https://doi.org/10.17863/CAM.20267.

Veiberg, V., Nilsen, E.B., Rolandsen, C.M., Heim, M., Andersen, R., Holmstrøm, F., Meisingset, E.L., Solberg, E.J., 2020. The accuracy and precision of age determination by dental cementum annuli in four northern cervids. Eur. J. Wildl. Res. 66, 91. https://doi.org/10.1007/s10344-020-01431-9.

Vukšić, N., Šperanda, M., Lončarić, Z., Didara, M., Ludek, E., Budor, I., 2018. The effect of dietary selenium addition on the concentrations of heavy metals in the tissues of fallow deer (*Dama dama L.*) in Croatia. Environ. Sci. Pollut. Res. 25, 11023–11033. https://doi.org/10.1007/s11356-018-1406-7.

Wagemann, R., Trebacz, E., Boila, G., Lockhart, W., 1998. Methylmercury and total mercury in tissues of arctic marine mammals. Sci. Total Environ. 218, 19–31. https://doi.org/10.1016/S0048-9697(98)00192-2.

Wang, W., Evans, R.D., Hickie, B.E., Rouvinen-Watt, K., Evans, H.E., 2014. Methylmercury accumulation and elimination in mink (*Neovison vison*) hair and blood: results of a controlled feeding experiment using stable isotope tracers. Environ. Toxicol. Chem. 33, 2873–2880. https://doi.org/10.1002/etc.2762.

Wani, A.L., Ara, A., Usmani, J.A., 2015. Lead toxicity: a review. Interdiscip. Toxicol. 8, 55–64. https://doi.org/10.1515/intox-2015-0009.

Węgrzyn, M.H., Wietrzyk-Pełka, P., Galanty, A., Cykowska-Marzencka, B., Sundset, M.A., 2019. Incomplete degradation of lichen usnic acid and atranorin in Svalbard reindeer (*Rangifer tarandus platyrhynchus*). Polar Res. 38. https://doi.org/10.33265/polar.v38.3375.

Welfinger-Smith, G., Minholz, J.L., Byrne, S., Waghiyi, V., Gologergen, J., Kava, J., Apatiki, M., Ungott, E., Miller, P.K., Arnason, J.G., Carpenter, D.O., 2011. Organochlorine and metal contaminants in traditional foods from St. Lawrence Island, Alaska. J. Toxicol. Environ. Health 74, 1195–1214. https://doi.org/10.1080/15287394.2011.590099.

Wiener, J.G., Krabbenhoft, D.P., Heinz, G.H., Scheuhammer, A., Burton Jr., G.A., Cairns Jr., J., 2003. Ecotoxicology of mercury. In: Hoffman, D.J., Rattner, B.A. (Eds.), Handbook of Ecotoxicology. Lewis Publishers, Boca Raton, pp. 409–465.

Wojtuń, B., Samecka-Cymerman, A., Kolon, K., Kempers, A.J., Skrzypek, G., 2013. Metals in some dominant vascular plants, mosses, lichens, algae, and the biological soil crust in various types of terrestrial tundra, SW Spitsbergen, Norway. Polar Biol. 36, 1799–1809. https://doi.org/10.1007/s00300-013-1399-0.

Wojtuń, B., Polechońska, L., Pech, P., Mielcarska, K., Samecka-Cymerman, A., Szymański, W., Kolon, M., Kopeć, M., Stadnik, K., Kempers, A.J., 2019. Sanionia uncinata and Salix polaris as bioindicators of trace element pollution in the high Arctic: a case study at Longyearbyen, Spitsbergen, Norway. Polar Biol. 42, 1287–1297. https://doi.org/10.1007/s00300-019-02517-0.

Yang, Z., Yuan, L., Xie, Z., Wang, J., Li, Z., Tu, L., Sun, L., 2020. Historical records and contamination assessment of potential toxic elements (PTEs) over the past 100 years in Ny-Ålesund, Svalbard. Environ. Pollut. 266, 115205. https://doi.org/10.1016/j.envpol.2020.115205.

Yin, X., Xia, L., Sun, L., Luo, H., Wang, Y., 2008. Animal excrement: A potential biomonitor of heavy metal contamination in the marine environment. Sci. Total Environ. 399, 179–185. https://doi.org/10.1016/j.scitotenv.2008.03.005.

Zhou, J., Obrist, D., Dastoor, A., Jiskra, M., Ryjkov, A., 2021. Vegetation uptake of mercury and impacts on global cycling. Nat. Rev. Earth Environ. 2, 269–284. https://doi.org/10.1038/s43017-021-00146-y.

SUPPORTING INFORMATION

Toxic Metals and Other Elements in Svalbard Reindeer:

Establishing Baselines and Assessing Non-Invasive

Sampling for Biomonitoring

Malin Andersson Stavridis[1,2], Bjørg Marie Pollestad[1,2], Vebjørn Veiberg[3], Øyvind Mikkelsen[1,4], Tomasz Maciej Ciesielski[1,2] & Bjørn Munro Jenssen[1,2].

1. Department of Arctic Technology, University Centre in Svalbard (UNIS), PO Box 156 N-9171, Longyearbyen, Norway.

2. Department of Biology, Norwegian University of Science and Technology (NTNU), NO-7491, Trondheim, Norway.

3. Department of Terrestrial Ecology, Norwegian Institute for Nature Research (NINA), NO-7485 Trondheim, Norway.

4. Department of Chemistry, Norwegian University of Science and Technology (NTNU), NO-7491, Trondheim, Norway.

Corresponding Author(s): Malin Andersson Stavridis: malins@unis.no & Bjørn Munro Jenssen: bjorn.munro.jenssen@ntnu.no.

Supplementary

Sampling information

All samples were collected during two culling campaigns in October 2021 and 2022. The prerequisites for the cullings differed, resulting in slightly different sample sizes for each type. The total number of samples collected at each culling is summarised in Table S1 below.

Table S1. An overview of the samples collected during the culling campaigns in 2021 and 2022.

Sample type	n (2021)	n (2022)
Kidney	18	18
Liver	18	17
Muscle	18	18
Brain	15	-
Blood	-	17
Fur	18	15
Faeces	18	18
Milk	-	7

Quality assurance and control

Table S2. Certified reference material recoveries (CRMs) of Hg using a DMA-80 evo. The CRMs used are MODAS-3 Herring Tissue (227 ± 21 µg/kg), GBW 09101b Human hair (1.06 ± 0.28) and Oriental Basma Tobacco Leaves INCT-OBLT-5 (20.9 ± 1.35 µg/kg).

CRM	Tissue	Year	Minimum recovery %	Maximum recovery %
MODAS-3 Herring Tissue	Faeces	2021	1.01	1.04
MODAS-3 Herring Tissue	Muscle	2021	1.04	1.06
MODAS-3 Herring Tissue	Brain	2021	1.02	1.10
MODAS-3 Herring Tissue	Liver	2021	1.09	1.14
MODAS-3 Herring Tissue	Kidney	2021	1.05	1.07
MODAS-3 Herring Tissue	Faeces	2022	1.06	1.15
MODAS-3 Herring Tissue	Muscle	2022	1.03	1.10
MODAS-3 Herring Tissue	Liver	2022	1.04	1.07
MODAS-3 Herring Tissue	Kidney	2022	1.05	1.10
INCT-OBTL-5	Blood	2022	0.94	0.97
INCT-OBTL-5	Fur	2022	0.95	1.00
GBW 09101b Human hair	Fur	2021	1.06	1.17

Table S3. Limits of detection (LOD) for Hg. The LODs are based on three times the standard deviation of the signal height of the blanks that were analysed using a DMA-80. At least three blanks were run during each analysis batch, and an LOD was calculated based on these for each separate analysis.

Tissue	Year	Average blank height	SD	LOD
Faeces	2021	0.001	0.0003	0.0009
Muscle	2021	0.0018	0.00002	0.00005
Brain	2021	0.0016	0.0009	0.0026
Liver	2021	0.0051	0.0067	0.02
Kidney	2021	0.0077	0.0062	0.0186
Fur	2021	0.0022	0.0019	0.0057
Faeces	2022	0.0047	0.0039	0.0116
Muscle	2022	0.0026	0.0011	0.0033
Liver	2022	0.0078	0.0055	0.0165
Kidney	2022	0.0121	0.0118	0.0354
Blood	2022	0.0019	0.0008	0.0024

Fur	2022	0.0017		0.0011	0.0032
Milk	2022	0.002		0.002	0.006

Table S4. Certified reference material recoveries (CRMs) of Cd, Pb, Se, Al, Fe, Si, P and S using HR-ICP-MS. The CRMs used are 1577b Bovine Liver and GBW 09101b Human hair. All concentrations are given in µg/g. We determined the recovery by comparing our measured concentrations to the certified values of the official CRM. N/A is given if the CRM is not certified for the element.

	1577b Bovine Liver			GBW 09101b Human hair		
Element	Official CRM	Our conc.	Recovery %	Official CRM	Our conc.	Recovery %
Cd	0.5 ± 0.03	0.48	97	0.072 ± 0.01	0.054	75
Pb	0.129 ± 0.004	0.09	72	3.83 ± 0.18	3.21	84
Se	0.73 ± 0.06	0.66	91	0.59 ± 0.04	0.63	108
Al	N/A	N/A	N/A	23.2 ± 2.0	32.3	139
Fe	184 ± 15	179	97	160 ± 16	151	94
Si	N/A	N/A	N/A	N/A	N/A	N/A
P	1.10 ± 0.03	1.09	99	174 ± 43	161	93
S	0.785 ± 0.006	0.78	100	N/A	N/A	N/A

Table S5. Limits of detection (LODs) are based on either three times the standard deviation of the blanks or the instrument detection limit, depending on which gave the highest value, based on the HR-ICP-MS results. All values are provided in µg/g.

Element	LOD
Cd	0.0007
Pb	0.0076
Se	0.0059
Al	0.0506
Fe	0.0523
Si	3.1982
P	0.8978
S	1.6521

Shapiro Wilk and Box-Cox transformations of the data

All data were assessed for normal distribution before the statistical analysis. A Box-Cox transformation was applied to determine the most suitable transformation to achieve normal distribution for variables that were not normally distributed. The results from these tests are summarised below.

Table S6. A description of the distribution of Hg, Cd, and Pb concentrations in the analysed samples, indicating whether the data were normally distributed or required transformation.

	Tissue	Normal distribution	Required log10 transformation	Required other transformation
Hg	Kidney	×		
	Liver		×	
	Muscle			$\text{sqrt}(x)^{-1}$
	Brain			$\text{sqrt}(x)$
	Faeces		×	
	Milk	×		
	Blood		×	
	Fur		×	
Cd	Kidney	×		
	Liver	×		
	Muscle			$\text{sqrt}(x)$
	Faeces	×		
	Fur		×	
Pb	Kidney			x^{-1}
	Liver		×	
	Muscle		×	
	Faeces	×		
	Fur	×		

As illustrated above, different sample types required different transformations to become normally distributed. Consequently, non-parametric statistical tests were primarily used to compare different samples, allowing us to use the raw data without ensuring normal distribution first.

Most Hg concentration data from 2021 and 2022 were not normally distributed. Only kidney and faeces Hg data had a normal distribution with equal variances between the years.

However, liver and muscle data could be transformed using $\log^{10}$ and x^{-1}, respectively, to achieve normal distribution. Therefore, a t-test was used to determine significant interannual differences in these four sample types. However, Fur Hg concentratios could not be successfully transformed, and a Mann-Whitney U test was used when assessing significant differences in fur Hg concentrations between years.

Derivation of the equation predicting the liver concentrations

As the liver and faeces Hg concentration data was not normally distributed, we applied a $\log^{10}$ transformation before running a linear regression between Hg concentrations in liver (y) and faeces (x) ($R^2 = 0.33$). The following linear equation was obtained from the regression: y = -0.23 + 0.56 × x, where y = log liver [Hg] and x = log faeces [Hg]. As this equation was produced to predict liver concentrations, the equation was solved for y as follows:

(1)　　　$y = -0.26 + 0.52 \times x$

(2)　　　$\Rightarrow \log liver\,[Hg] = -0.26 + 0.52\ \times \log faecal\,[Hg]$

(3)　　　$\Rightarrow liver\,[Hg] = 10^{-0.26} \times faecal\,[Hg]^{0.52}$

Similar equations were calculated for the lower (log liver [Hg] = 0.26 × log faecal [Hg] – 0.57) and upper (log liver [Hg] = 0.79 × log faecal [Hg] + 0.04) limits of the confidence interval (CI) to calculate the fit of the model. The predicted values from the regression model ranged from -1.02 to -0.66, with 95% CIs spanning from -1.11 to -0.55. This indicates that, while the model predictions were generally precise, the width of the confidence intervals varied slightly across the dataset, reflecting differences in the underlying data distribution and leverage of individual observations. When using the equation above to predict liver Hg concentrations from faecal Hg concentrations, the results are on average 107 ± 32% of the observed liver Hg concentrations (min: 61%, max: 181%).

Raw data from the literature review of Rangifer tarandus studies

Table S7. The raw data from the literature review used for Table 4. The data were taken directly from the articles or found in the study appendices. The **Source** refers to the article from which the data was obtained. **Tissue** refers to the analysed tissue, **(n)** describes the sample size and **[Hg]**, **Unit**, and **ww/dw** describes the concentration of Hg, unit and whether the authors provide the concentrations in wet weight (ww) or dry weight (dw). **Location** and **year** provide information on when and where the sampling took place, while Gender explains whether males (M) or females (F) were sampled. The column **[Hg] µg/g ww** is our recalculations into ww when the original data was provided in dw. These calculations were made based on the water content presented in the article, or in a previous article from the same author. For those articles that lacked water content data, the water content in Gamberg et al. (2016) was used.

Source	Tissue	(n)	[Hg]	Unit	ww/dw	Location	Year	Gender	[Hg] µg/g ww
(Elkin and Bethke 1995)	Kidney	20	0.52	µg/g	ww	Bathurst, Canada	1991-1992	M/F	0.52
(Elkin and Bethke 1995)	Kidney	10	2.93	µg/g	ww	Arviat, Canada	1991-1992	M/F	2.93
(Elkin and Bethke 1995)	Kidney	10	2.22	µg/g	ww	SH Island, Canada	1991-1992	M/F	2.22
(Elkin and Bethke 1995)	Kidney	10	1.25	µg/g	ww	Cape Dorset, Canada	1991-1992	M/F	1.25
(Elkin and Bethke 1995)	Kidney	10	2.56	µg/g	ww	Lake Harbour, Canada	1991-1992	M/F	2.56
(Elkin and Bethke 1995)	Liver	20	0.16	µg/g	ww	Bathurst, Canada	1991-1992	M/F	0.16
(Elkin and Bethke 1995)	Liver	10	0.92	µg/g	ww	Arviat, Canada	1991-1992	M/F	0.92
(Elkin and Bethke 1995)	Liver	10	0.38	µg/g	ww	Cape Dorset, Canada	1991-1992	M/F	0.38
(Elkin and Bethke 1995)	Liver	10	0.58	µg/g	ww	Lake Harbour, Canada	1991-1992	M/F	0.58
(Gamberg et al. 2020)	Kidney	173	1.65	µg/g	dw	Porcupine, Alaska	2005-2016	M/F	0.36
(Gamberg et al. 2020)	Kidney	16	7.8	µg/g	dw	Baffin Island, Greenland	1991-2016	M/F	1.72
(Gamberg et al. 2020)	Kidney	35	2.1	µg/g	dw	Bathurst, Canada	1991-2016	M/F	0.46
(Gamberg et al. 2020)	Kidney	16	7.1	µg/g	dw	Beverly, Canada	1991-2016	M/F	1.56
(Gamberg et al. 2020)	Kidney	22	4.2	µg/g	dw	Bluenose East, Canada	1991-2016	M/F	0.92
(Gamberg et al. 2020)	Kidney	11	6.8	µg/g	dw	Cape Bathurst, Canada	1991-2016	M/F	1.50
(Gamberg et al. 2020)	Kidney	260	1.4	µg/g	dw	Porcupine, Canada	1991-2016	M/F	0.31
(Gamberg et al. 2020)	Kidney	61	3.6	µg/g	dw	Qamanirjuaq, Canada	1991-2016	M/F	0.79
(Gamberg et al. 2020)	Kidney	27	5.9	µg/g	dw	Baffin Islan, Greenland	1991-2016	M/F	1.30
(Gamberg et al. 2020)	Kidney	73	4	µg/g	dw	Bathurst, Canada	1991-2016	M/F	0.88
(Gamberg et al. 2020)	Kidney	51	6.3	µg/g	dw	Beverly, Canada	1991-2016	M/F	1.39
(Gamberg et al. 2020)	Kidney	16	4.1	µg/g	dw	Bluenose East, Canada	1991-2016	M/F	0.90
(Gamberg et al. 2020)	Kidney	249	1.9	µg/g	dw	Porcupine, Canada	1991-2016	M/F	0.42
(Gamberg et al. 2020)	Kidney	69	5.8	µg/g	dw	Qamanirjuaq, Canada	1991-2016	M/F	1.28
(Gamberg et al. 2020)	Kidney	41	2.1	µg/g	dw	Akia-Maniitsoq, Greenland	1991-2016	M/F	0.46
(Gamberg et al. 2020)	Kidney	35	1.4	µg/g	dw	Kangerlussuaq-Sisimiut, Greenland	1991-2016	M/F	0.31
(Larter et al. 2016)	Kidney	26	0.21	mg/kg	dw	NW Territories, Canada	2010-2013	M	0.04
(Larter et al. 2016)	Muscle	10	0.005	mg/kg	dw	NW Territories, Canada	2010-2013	M	0.001
(Robillard et al. 2002)	Muscle	161	0.0285	µg/g	ww	Leaf River Region, Canada	1994-1995	M/F	0.03
(Robillard et al. 2002)	Kidney	264	1.38	µg/g	ww	Leaf River Region, Canada	1994-1995	M/F	1.38
(Robillard et al. 2002)	Liver	260	0.7	µg/g	ww	Leaf River Region, Canada	1994-1995	M/F	0.7
(Robillard et al. 2002)	Muscle	47	0.02	µg/g	ww	George River Region, Canada	1994-1995	M/F	0.02
(Robillard et al. 2002)	Kidney	46	0.59	µg/g	ww	George River Region, Canada	1994-1995	M/F	0.59

(Robillard et al. 2002)	Liver	47	0.415	µg/g	ww	George River Region, Canada	1994-1995	M/F	0.42
(Schuster et al. 2011)	Muscle	75	0.003	mg/kg	ww	Porcupine, Canada	1994-2006	M	0.003
(Schuster et al. 2011)	Kidney	63	0.36	mg/kg	ww	Porcupine, Canada	1994-2006	M	0.36
(Schuster et al. 2011)	Liver	3	0.12	mg/kg	ww	Porcupine, Canada	1994-2006	M	0.12
(Rintala et al. 1995)	Muscle	325	0.0055	mg/kg	ww	Lapland, Finland	1990-1992	N/A	0.006
(Aastrup et al. 2000)	Muscle	8	0.043	µg/g	ww	Isortoq, Greenland	1995-1997	M/F	0.04
(Aastrup et al. 2000)	Muscle	23	0.04	µg/g	ww	Itinnera, Greenland	1995-1997	M/F	0.04
(Aastrup et al. 2000)	Muscle	47	0.007	µg/g	ww	Kangerlussuaq, Greenland	1995-1997	M/F	0.007
(Aastrup et al. 2000)	Muscle	49	0.02	µg/g	ww	Akia, Greenland	1995-1997	M/F	0.02
(Aastrup et al. 2000)	Liver	7	0.618	µg/g	ww	Isortoq, Greenland	1995-1997	M/F	0.62
(Aastrup et al. 2000)	Liver	23	0.058	µg/g	ww	Itinnera, Greenland	1995-1997	M/F	0.06
(Aastrup et al. 2000)	Liver	47	0.051	µg/g	ww	Kangerlussuaq, Greenland	1995-1997	M/F	0.05
(Aastrup et al. 2000)	Liver	49	0.194	µg/g	ww	Akia, Greenland	1995-1997	M/F	0.19
(Gamberg et al. 2016)	Muscle	20	0.05	µg/g	dw	Akia-Maniitsoq, Greenland	2008-2009	F	0.01
(Gamberg et al. 2016)	Liver	40	0.46	µg/g	dw	Akia-Maniitsoq, Greenland	2008-2009	F	0.10
(Gamberg et al. 2016)	Kidney	41	2.22	µg/g	dw	Akia-Maniitsoq, Greenland	2008-2009	F	0.47
(Gamberg et al. 2016)	Muscle	20	0.02	µg/g	dw	Kangerlussuaq-Sisimiut, Greenland	2008-2009	F	0.00
(Gamberg et al. 2016)	Liver	40	0.24	µg/g	dw	Kangerlussuaq-Sisimiut, Greenland	2008-2009	F	0.05
(Gamberg et al. 2016)	Kidney	40	1.41	µg/g	dw	Kangerlussuaq-Sisimiut, Greenland	2008-2009	F	0.30
(Frøslie et al. 1984)	Liver	2	67	ppb	ww	Setedalsheiene, Norway	1978	N/A	0.07
(Frøslie et al. 1984)	Liver	5	192	ppb	ww	Dovre, Norway	1978	N/A	0.19
(Frøslie et al. 1984)	Liver	7	46	ppb	ww	Lierne, Norway	1978	N/A	0.05
(Frøslie et al. 1984)	Liver	8	16	ppb	ww	Hattfjelldal, Norway	1978	N/A	0.02
(Frøslie et al. 1984)	Liver	10	59	ppb	ww	Sortland, Norway	1978	N/A	0.06
(Frøslie et al. 1984)	Liver	12	137	ppb	ww	Karasjok, Norway	1978	N/A	0.14
(Frøslie et al. 1984)	Liver	13	45	ppb	ww	Karasjok, Norway	1978	N/A	0.05
(Makarov et al. 2022)	Liver	154	0.21	mg/kg	ww	Kola, Russia	2015-2020	M/F	0.21
(Makarov et al. 2022)	Liver	149	0.21	mg/kg	ww	Nenets, Russia	2015-2020	M/F	0.21
(Makarov et al. 2022)	Liver	42	0.13	mg/kg	ww	Komi, Russia	2015-2020	M/F	0.13
(Makarov et al. 2022)	Liver	119	0.17	mg/kg	ww	Yamalo-Nenets, Russia	2015-2020	M/F	0.17
(Makarov et al. 2022)	Liver	19	0.06	mg/kg	ww	Taymir, Russia	2015-2020	M/F	0.06
(Makarov et al. 2022)	Liver	22	0.11	mg/kg	ww	Kamchatka, Russia	2015-2020	M/F	0.11
(Makarov et al. 2022)	Kidney	90	0.62	mg/kg	ww	Kola, Russia	2015-2020	M/F	0.62
(Makarov et al. 2022)	Kidney	53	0.67	mg/kg	ww	Nenets, Russia	2015-2020	M/F	0.67
(Makarov et al. 2022)	Kidney	42	0.43	mg/kg	ww	Komi, Russia	2015-2020	M/F	0.43
(Makarov et al. 2022)	Kidney	100	0.52	mg/kg	ww	Yamalo-Nenets, Russia	2015-2020	M/F	0.52
(Makarov et al. 2022)	Kidney	10	0.05	mg/kg	ww	Taymir, Russia	2015-2020	M/F	0.05
(Makarov et al. 2022)	Kidney	20	0.75	mg/kg	ww	Kamchatka, Russia	2015-2020	M/F	0.75
(Makarov et al. 2022)	Muscle	10	0.05	mg/kg	ww	Taymir, Russia	2015-2020	M/F	0.05
(Bernhoft et al. 2002)	Liver	40	0.16	µg/g	ww	Rybatsjij Ostrov, Russia	1997	M/F	0.16
(Sivertsen et al. 1995)	Liver	31	0.07	µg/g	ww	Jarfjord, Norway	1990-1991	M/F	0.07
Sivertsen et al. 1995)	Liver	30	0.06	µg/g	ww	Pasvik, Norway	1990-1991	M/F	0.06
Sivertsen et al. 1995)	Liver	40	0.12	µg/g	ww	Finnmark, Norway	1990-1991	M/F	0.12
(Odsjö et al. 2005)	Liver	200	0.041	µg/g	ww	Lapland, Sweden	1983-2003	M	0.04
(Odsjö et al. 2005)	Muscle	150	0.001	µg/g	ww	Lapland, Sweden	1983-2003	M	0.001

| (Odsjö et al. 2005) | Liver | 80 | 0.032 | μg/g | ww | Lapland, Sweden | 1983-2003 | M | 0.03 |
| (Odsjö et al. 2005) | Muscle | 80 | 0.001 | μg/g | ww | Lapland, Sweden | 1983-2003 | M | 0.001 |

Table S8. Wet weight concentrations (in µg/g) of Hg (n=36), Cd and Pb (n=18) in kidney, liver and muscle samples from Svalbard reindeer.

Element	Tissue	Mean	Median	SD	Min	Max
Hg	Kidney	0.14	0.13	0.07	0.03	0.29
	Liver	0.04	0.04	0.02	0.02	0.10
	Muscle	2.7×10^{-3}	1.8×10^{-3}	1.9×10^{-3}	0.9×10^{-3}	7.2×10^{-3}
	Brain	0.4×10^{-3}	0.4×10^{-3}	0.3×10^{-3}	<LOD	1.1×10^{-3}
Cd	Kidney	3.31	2.39	2.36	0.45	7.68
	Liver	0.45	0.44	0.18	0.12	0.78
	Muscle	1.36×10^{-3}	0.89×10^{-3}	1.06×10^{-3}	0.07×10^{-3}	3.33×10^{-3}
Pb	Kidney	0.04	0.03	0.03	0.02	0.11
	Liver	0.08	0.07	0.06	0.02	0.26
	Muscle	0.88×10^{-3}	0.81×10^{-3}	0.85×10^{-3}	<LOD	1.77×10^{-3}

Paper III

1 # Seasonal Shift in Exposure and Accumulation of

2 # PFAS and Heavy Metals in High Arctic Reindeer

3 *Malin Andersson Stavridis[1,2]*, Tove Petersson[1,3], Görkem Deniz Kendir[2,4], Shannen Sait[4], Øyvind*

4 *Mikkelsen[1,4], Vebjørn Veiberg[5], Tomasz Maciej Ciesielski[1] & Bjørn Munro Jenssen[1,2]*.*

5 1. Department of Arctic Technology, University Centre in Svalbard (UNIS), PO Box 156 N-9171, Longyearbyen, Norway.

6 2. Department of Biology, Norwegian University of Science and Technology (NTNU), NO-7491, Trondheim, Norway.

7 3. Department of Ecoscience, Marine Mammal Research, Aarhus University, DK-4000, Roskilde, Denmark.

8 4. Department of Chemistry, Norwegian University of Science and Technology (NTNU), NO-7491, Trondheim, Norway.

9 5. Land and Biodiversity, Norwegian Institute for Nature Research (NINA), NO-7485 Trondheim, Norway.

10 **KEYWORDS**: Contaminants; Metals; PFAS; Svalbard Reindeer; Arctic.

11 ## ABSTRACT

12 Seasonal fluctuations in contaminant concentrations are an important, yet often overlooked,

13 aspect of Arctic wildlife monitoring, particularly in species like the Svalbard reindeer

14 (*Rangifer tarandus platyrhynchus*), which exhibit annual fattening and fasting cycles. While

15 seasonal variation in metal concentrations has been observed in this species, little is known

16 about how other contaminant groups, such as per- and polyfluoroalkyl substances (PFAS),

17 vary with season. In the present study, we report concentrations of total mercury (THg),

18 cadmium (Cd), lead (Pb), and 13 PFAS in the liver and muscle of Svalbard reindeer culled in

19 August 2022 and October 2023. Seasonal differences were observed, with higher

20 concentrations of THg, Cd, and several PFAS in October, likely reflecting an extended

21 foraging time following the winter fasting. The PFAS profiles, dominated by PFOS and long-

22 chain PFCAs, primarily suggest exposure to long-range transported contaminants rather than

23 local sources, highlighting that even remote terrestrial wildlife is affected by emissions from

24 industrialised regions. While Cd and Pb concentrations have decreased compared to levels

25 reported in the 1980s, PFAS concentrations have increased over the past decade. These

26 findings underscore the need for continued monitoring of Arctic terrestrial wildlife,

27 particularly in the context of environmental change and remobilisation of legacy

28 contaminants.

29 SYNOPSIS

30 Minimal research exists on seasonal contaminant levels in Arctic wildlife. This study reveals

31 elevated autumn concentrations in Svalbard reindeer, linked to extended foraging after winter

32 fasting and long-range pollutant exposure.

33 INTRODUCTION

34 Arctic wildlife is increasingly exposed to a complex mixture of environmental contaminants,

35 often referred to as a chemical cocktail [1,2]. This cocktail includes toxic metals such as

36 mercury (Hg), cadmium (Cd), and lead (Pb), and per- and polyfluoroalkyl substances (PFAS),

37 which are all known to cause adverse health effects, such as neurotoxicity, nephrotoxicity,

38 and endocrine disruption [3,4].

39 Most contaminants found in the Arctic originate from industrial regions at lower latitudes and

40 are transported northward through long-range atmospheric and oceanic transport [5–7]. Over

41 time, many of these pollutants have accumulated in the Arctic cryosphere. As the region

42 warms, thawing glaciers, snow, and permafrost may remobilise legacy contaminants,

43 increasing the risk of exposure to Arctic biota [8,9]. While marine mammals have been the

44 primary focus of Arctic contaminant research, far less is known about terrestrial species,

45 which may face increasing exposure as pollutants re-enter the environment and become

46 available to biota again, for instance, via vegetational uptake [10], and subsequent ingestion by

47 herbivores [11].

48   The Svalbard reindeer (*Rangifer tarandus platyrhynchus*) is the largest terrestrial herbivore in

49   Svalbard and plays a key ecological role in the local Arctic terrestrial food web [12]. However,

50   contaminant data for this species remain limited. Borch-Iohnsen et al. [13] reported strong

51   seasonal variation in hepatic and renal concentrations of toxic metals in Svalbard reindeer

52   culled at the end of the 1980s. Notably, Cd levels were lower in reindeer culled at the end of

53   summer (July/August) compared to autumn (October), while Pb concentrations showed the

54   opposite trend, being higher in late summer and lower in autumn. These shifts were linked to

55   seasonal changes in diet, which reflect variations in exposure to metals throughout the year. In

56   a more recent study, Andersson Stavridis et al. [14] reported lower Cd and Pb levels in

57   Svalbard reindeer now, compared to four decades ago [13], likely due to international emission

58   regulations. In terms of organic contaminants, the study by Roos et al. [15] remains the only

59   published study on PFAS in Svalbard reindeer. They found that Svalbard reindeer had the

60   lowest PFAS concentrations among the circumarctic *Rangifer* subspecies. However, given the

61   small sample size (n=7) and single time point (2010), it remains unclear whether PFAS

62   concentrations may have changed over the past decade, or whether they, like metals, also

63   follow seasonal patterns.

64   In this study, we characterise the contaminant profile of Svalbard reindeer liver and muscle

65   tissue collected in August 2022 and October 2023. We quantified concentrations of toxic

66   metals (i.e., Hg, Cd, Pb) and 13 PFAS compounds, including perfluorocarboxylic acids

67   (PFCAs), perfluorosulfonic acids (PFSAs), precursor PFAS, and emerging PFAS. By

68   sampling individuals from the same geographic area across two seasons, we provide new

69   insight into the temporal dynamics of contaminant exposure in Arctic herbivores living in a

70   rapidly changing Arctic environment, where prolonged vegetation growth due to climate

71   warming and pollutant remobilisation may alter their exposure.

MATERIALS AND METHODS

SAMPLES

The tissue samples used in this study were collected from female Svalbard reindeer, culled for a transdisciplinary and international research project [14] under the permission of the Governor of Svalbard (reference number 16/01632-38).

The culling campaigns took place in early August 2022 and late October 2023 around Reindalen, Svalbard (77°90′–78°10′N, 15°30′–16°00′E). The reindeer were shot through the heart and lung region with regular hunting ammunition and expanding bullets (using bullet types 6.5x55 Federal Fusion and 30-06 Lapua Naturalis). All culled reindeer appeared to be in good health, with no visible signs of disease or injury. The age of the reindeer ranged from one to 11 years old, with age being determined using tooth eruption patterns and cementum annuli analysis [16]. Reproductive maturity was assessed based on the presence of a calf at the time of culling and visible milk production. None of the reindeer culled in August were accompanied by calves, whereas six females culled in October had calves by their side.

In 2022, liver (n=12) and medial thigh muscle (n=12) were sampled within thirty minutes post-mortem. In 2023, a complete necropsy of the reindeer was performed within 6-12 hours post-mortem, during which both liver (n = 18) and medial thigh muscle (n = 18) samples were collected. To prevent cross-contamination, all samples were collected using clean scalpel blades (Swann-Morton, Swann-Morton Limited, UK) while wearing nitrile gloves. Given the use of lead-containing bullets, samples were exclusively collected from individuals where the entry wound was confined to the heart and lung region, ensuring distance from the tissues of interest. Furthermore, the outer layers (a few mm) of all tissue samples were removed to reduce external contamination of the samples. The samples were stored in polyethylene zip-lock bags (VWR, PA, USA) at -80°C until further processing.

96   All samples were freeze-dried (using a FreeZone Benchtop Freeze-Dryer, Labconco, MO,

97   USA) for 72h at -50ºC and 0.04 mbar. The samples were then placed in clean zip-lock bags

98   before being homogenised using a rubber mallet until a fine powder was obtained [14]. The

99   weight of the samples was noted (Mettler Toledo, $\pm$ 0.01 g) before and after freeze-drying, to

100 determine the moisture content of the samples (an average moisture content of 70.1% in liver

101 and 72.0% in muscle). All subsequent chemical analyses used the freeze-dried sample

102 homogenates. The analyses were conducted at the Department of Chemistry, Norwegian

103 University of Science and Technology (NTNU).

104 A subset of the collected samples was selected for contaminant analysis, prioritising muscle

105 samples as Svalbard reindeer are recreationally hunted and consumed by locals [17]. In

106 addition, liver samples representing a range of ages from both culling years were included in

107 the contaminant analysis. Figure 1 illustrates the individual PFAS profiles, indicating which

108 samples were analysed (coloured bars) and which were omitted from the PFAS analysis

109 (white bars). For further details on the culling and sampling sizes, see Table S1.

110 MERCURY ANALYSIS

111 Liver (n=21) and muscle (n=30) samples were analysed for total Hg (THg) using a Direct

112 Mercury Analyser (DMA-80 evo, Milestone, Italy). The analysis followed the US EPA

113 method 7473 with Hg calibration standards, certified reference materials (CRMs), and

114 blanks to ensure analytical accuracy (see Materials and Methods in Supporting Information).

115 ELEMENTAL ANALYSIS

116 For analysis of Cd, Pb, and Se, liver (n=21) and muscle (n=20) samples were digested in

117 ultrapure nitric acid (purified from HNO_3, AnalaR NORMAPUR, VWR) in a sub-boiling

118 distillation system (Milestone, SubPur, Sorisole, BG, Italy) using a high-pressure microwave

119 system (Milestone Ultraclave, EMLS, Leutkirch, Germany) for 150 min. CRMs and blanks

120 were digested following the same protocol as the tissue samples and included in the analysis

121   to ensure analytical accuracy. The elemental concentrations were quantified in the samples,

122   CRMs, and blanks using an 8800 Triple Quadrupole inductively coupled plasma mass

123   spectrometry (ICP-MS) system (Agilent Technologies, USA) equipped with a prepFAST M5

124   autosampler (ESI, USA), following the protocol described in Andersson Stavridis et al. [14]. In

125   the present study, we report concentrations of toxic elements cadmium (Cd) and lead (Pb), as

126   well as selenium (Se), due to its protective properties against toxic metals [18,19]. See Materials

127   and Methods in Supporting Information for further details on the analysis, quality assurance,

128   and quality control.

129   PFAS ANALYSIS

130   Liver (n=21) and muscle (n=30) samples were analysed for 41 different PFAS compounds

131   grouped into perfluoroalkyl carboxylic acids (PFCAs; PFBA, PFPeA, PFHxA, PFHpA,

132   PFOA, PFNA, PFDA, PFUnDA, PFDoDA, PFTriDA, PFTDA, PFHxDA, and PFOcDA),

133   perfluoroalkyl sulfonic acids (PFSAs; PFBS, PFPeS, PFHxS, PFHpS, PFOS, PFNS, PFDS,

134   PFDoDS, and PFECHS), fluorotelomer sulfonates (FTSs; 4:2 FTS, 6:2 FTS, 8:2 FTS, and

135   10:2 FTS), sulfonamides and precursor PFAS (FOSAA, MeFOSAA, EtFOSAA, PFOSA,

136   MeFOSA, EtFOSA, MeFOSE, and EtFOSE), and emerging PFAS (GenX, NaDONA, 9Cl-

137   PF3ONS, P37DMOA, SaMPAP and diSAMPAP). The analytical method targeted linear

138   PFAS isomers only. The full names of the PFAS compounds are listed in Table S2. PFAS

139   concentrations were determined using an Xevo TQ-XS Triple Quadrupole Mass Spectrometer

140   coupled with an ACQUITY UPLC system (Waters, Milford, MA, USA) following the

141   methods described in Sait et al. [20]. The peak integration was performed manually using

142   MassLynx software (Waters, Milford, MA, USA). A detailed extraction protocol is described

143   under Materials and Methods in Supporting Information.

144   DATA ANALYSIS

145   All contaminant concentrations are presented as average concentrations in ng/g, unless

146   otherwise stated. Only compounds detected above the limit of detection (LOD) in at least 25%

147   of the samples are reported and included in the statistical analysis. Accordingly,

148   concentrations of THg, Cd, Pb, and 13 PFAS are presented for liver, and THg, Pb, and 10

149   PFAS for muscle. The detection rates for all compounds are provided in Table S3. For these

150   compounds, average and median concentrations were calculated using values above the LOD,

151   while non-detects were excluded from the calculations.

152   All statistical analyses were conducted using RStudio version 4.3.0 [21]. The data were

153   analysed for normality using the Shapiro-Wilk test and for equal variances using Levene's

154   test. Given the non-normal distribution of the raw data, non-parametric Spearman rank

155   correlations were used to assess the relationship between PFAS and biometrics. Annual

156   variations in contaminant concentrations were analysed using Mann-Whitney U tests with

157   Benjamin-Hochberg corrections applied to limit false discovery rates. Mann-Whitney U tests

158   were also used to assess potential maternal transfer by comparing contaminant concentrations

159   in females with calves to those without among individuals culled in October, but no

160   significant differences were observed.

161   Statistical analyses were conducted on dry weight (dw) concentrations (Table S3) to avoid

162   any bias introduced by tissue water content. As such, the significant differences shown in

163   Tables 1-2 and Fig. 2 refer to dw, whereas all concentrations in the article are reported on a

164   wet weight (ww) basis, unless otherwise stated.

RESULTS AND DISCUSSION

SEASONAL VARIATION IN SVALBARD REINDEER CONTAMINANT LEVELS

In general, we found notable variations in both contaminant levels and profiles between sampling seasons (Table 1-2), and Svalbard reindeer culled in autumn (October) had significantly higher hepatic concentrations of Se, toxic metals (i.e., Hg and Cd), and PFAS, compared to the individuals sampled in late summer (August).

In the present study, average hepatic Hg concentrations for August 2022 and October 2023 were 12 ng/g (ww) and 25 ng/g (ww), respectively. Although previously reported average Hg concentrations for October 2021 (43 ng/g ww) and 2022 (47 ng/g ww) in Svalbard reindeer [14] were significantly higher than those measured in October 2023, all October data (2021-2023) show consistently higher Hg levels than those measured in August 2022 (p=0.002). A similar pattern between seasons was observed for Cd with significantly higher concentrations in October 2021 (448 ng/g ww) [14] and October 2023 (532 ng/g ww), compared to the 192 ng/g (ww) measured in August 2022 (p=0.006). Hepatic concentrations of PFAS were also significantly higher in October relative to August, with higher levels of ΣPFAS, ΣPFCAs, and ΣPFSAs (p=0.006, p=0.008, and p=0.015, respectively). Specifically, concentrations of both PFHxS (p=0.008), PFUnDA (p=0.009), PFTriDA (p=0.008), and PFTDA (p=0.009) were severalfold higher in October relative to August.

Similarly to liver, average levels of ΣPFCAs in muscle (Table 2) were significantly higher in October (8.54 ng/g ww) relative to August (1.12 ng/g ww; p=0.002). Conversely, muscle PFOS concentrations displayed an opposite behaviour, peaking in August, resulting in a significantly higher ΣPFSAs in 2022 (p=0.023).

Seasonal effects on tissue accumulation of metal concentrations in Svalbard reindeer were previously documented by Borch-Iohnsen et al. [13], who suggest that dietary changes

189 influenced by seasonal availability have a significant effect on metal exposure. Seasonal

190 fluctuations in contaminant concentrations are also documented in other Arctic mammals that

191 undergo annual shifts in diet and cycles of fattening and fasting, such as polar bears (*Ursus*

192 *maritimus*) [22,23].

193 **Table 1**. Contaminant concentrations (ww) in liver samples from August 2022 and October 2023. Significant

194 differences are marked as (*) when p<0.05, and (**) when p<0.01.

Liver concentrations (ng/g ww)									
	August 2022				October 2023				
Contaminant	Mean ± SD	Median	Min	Max	Mean ± SD	Median	Min	Max	Sig.
THg	11.6±5.27	11.1	5.71	25.6	24.8±8.59	21.5	13.1	37.0	(**)
Se[a]	394±57.0	382	314	496	477±56.6	474	393	575	(*)
Cd	192±84.2	178	113	423	532±186	555	329	927	(**)
Pb	52.5±37.8	43.2	14.2	127	48.7±26.5	42.1	13.8	92.7	
Σ_{13}PFAS[b]	18.8±13.0	17.1	5.95	54.1	55.8±15.4	53.8	34.8	78.4	(**)
ΣPFCAs	8.93±6.96	7.64	1.93	27.4	26.7±17.0	22.0	11.2	58.3	(**)
ΣPFSAs	7.24±4.81	6.15	2.56	15.1	21.9±11.2	22.9	1.26	39.3	(**)
PFUnDA	4.47±3.13	4.24	1.34	12.4	14.4±11.3	10.1	5.28	36.1	(**)
PFTriDA	4.17±3.17	3.45	1.59	12.8	11.0±5.45	10.3	5.29	22.3	(**)
PFTDA	0.40±0.22	0.40	0.09	0.80	0.87±0.29	0.84	0.42	1.33	(**)
PFHxDA	0.91±0.97	0.62	0.10	2.28	2.32±2.90	2.32	0.27	4.37	
PFHxS	5.35±4.10	3.28	1.23	10.6	21.6±8.30	17.7	12.5	36.7	(**)
PFOS	3.83±1.32	3.84	2.16	6.60	5.28±3.22	4.05	2.44	12.1	
PFNS	0.19±0.21	0.09	0.04	0.42	0.23±0.16	0.31	0.04	0.39	
PFDS	0.36±0.42	0.20	0.10	1.20	0.30±0.41	0.13	0.05	0.91	
PFECHS	0.19±0.10	0.18	0.11	0.39	0.22±0.06	0.23	0.13	0.30	
FOSAA	2.07±5.44	0.19	0.05	16.5	0.78±0.94	0.39	0.01	2.56	
MeFOSAA	0.08±0.10	0.03	0.01	0.20	0.67±0.26	0.52	0.51	0.97	
EtFOSAA	0.93±1.99	0.21	0.01	5.44	0.68±0.88	0.45	0.04	2.63	
NaDONA	0.44±0.46	0.30	0.08	1.49	6.41±7.63	4.94	0.20	23.6	(*)

[a] Se:Hg molar ratio was on average 99.9 in August and 54.4 in October.

[b] Σ_{13}PFAS includes PFUnDA, PFTriDA, PFTDAA, PFHxDA, PFHxS, PFOS, PFNS, PFDS, PFECHS, FOSAA, MeFOSAA, EtFOSAA, NaDONA.

195

196   The Svalbard reindeer undergoes significant seasonal fluctuations in weight, with a peak in

197   body mass (~70 kg) in October, followed by a decline to approximately 50 kg by April, due to

198   limited winter forage [24]. This is consistent with the individuals used in the present study,

199   where body mass was slightly lower in August compared to peak mass in October (65 ± 9 kg

200   and 72 ± 9 kg, respectively; Table S1). Given that ingestion of vegetation is the primary

201   exposure route for contaminants in Svalbard reindeer, both the duration of foraging and

202   seasonal shifts in diet are likely to influence their contaminant burden. Reindeer culled in

203   October have had two additional months of foraging compared to those culled in August. We

204   suggest that this extended feeding period may increase cumulative exposure to contaminants

205   present in their diet, or that shifts in diet due to plant phenology and senescence may result in

206   a larger dietary proportion of plant species or parts that contain higher levels of THg, Cd, and

207   ΣPFCAs.

208   In addition to two additional months of foraging in October, there is a seasonal shift in the

209   types of vegetation consumed from late summer to autumn. While reindeer generally

210   prioritise forage quantity over quality [25], changes in plant availability and nutritional content

211   throughout the growing season likely influence dietary composition. Eikeland [26] reported

212   that certain plant species were frequently grazed in both late summer and autumn, including

213   willow (*Salix polaris*) and grass (*Poa arctica*), while others, such as the flower *Eriophorum*

214   *scheuchzeri*, were primarily consumed in summer, and the herb *Saxifraga oppositifolia*

215   mainly in autumn. These patterns suggest a shift in grazing preference that may reflect

216   biological changes in nutritional needs or the availability of different types of vegetation as

217   the season progresses. Such dietary shifts are relevant for contaminant exposure, as the

218   concentration of contaminants may vary widely among different species of plants. For

219   example, average ΣPFAS concentrations are higher in vascular vegetation such as *Dryas*

220   *octopetala* ($\bar{x} = 8.39$ ng/g dw) and *S. polaris* ($\bar{x} = 2.37$ ng/g dw) than in bryophytes ($\bar{x} = 1.03$

221 ng/g dw) [10]. Likewise, *S. polaris* is known to accumulate Cd, resulting in it having

222 concentrations several times higher than those reported in other vascular plant species [27]. In

223 contrast, Hg accumulation is more efficient in bryophytes, compared to vascular vegetation

224 [28,29]. Contaminant burdens in other Arctic cervids have been linked to dietary shifts, with

225 proportions of bryophytes and willows such as *Salix* spp. being important drivers of Hg and

226 Cd exposure [30]. Consequently, changes in the proportion of different species of vegetation in

227 the diet of the Svalbard reindeer may influence their overall contaminant exposure. In contrast

228 to many other reindeer and caribou across the Arctic, the Svalbard reindeer are non-migratory,

229 with a home range limited to a few square kilometres [31,32]. As such, this overall increase in

230 contaminant concentrations between August to October is more likely related to a dietary

231 shift, rather than seasonal spatial changes in grazing grounds.

232 **Table 2**. Contaminant concentrations (ww) in muscle samples from August 2022 and October 2023. Significant

233 differences are marked as (*) when $p<0.05$, and (**) when $p<0.01$.

	Muscle concentrations (ng/g ww)								
	August 2022				October 2023				
Contaminant	Mean ± SD	Median	Min	Max	Mean ± SD	Median	Min	Max	Sig.
THg	2.43±1.49	2.37	0.73	6.37	1.74±0.76	1.41	0.92	3.19	
Se[a]	192±90.0	156	141	424	180±21.3	183	152	205	
Pb	0.23±0.15	0.30	0.03	0.36	0.28±0.24	0.25	0.03	0.73	
Σ_{10}PFAS[b]	8.63±12.3	3.45	0.73	33.9	12.7±8.08	6.97	3.09	29.5	
ΣPFCAs	1.12±0.92	1.06	0.06	2.56	8.54±7.19	5.70	1.86	28.5	(**)
ΣPFSAs	2.92±3.37	1.70	0.35	10.9	0.62±0.73	0.44	0.07	3.04	(*)
PFUnDA		Below LOD			4.48±4.63	2.68	0.30	17.9	(**)
PFTriDA	0.98±0.55	0.93	0.27	1.77	4.37±2.67	3.41	1.68	10.6	(**)
PFTDA	0.49±0.57	0.35	0.06	1.84	0.46±0.31	0.35	0.13	1.27	
PFPeS	0.14±0.07	0.14	0.08	0.19	0.10±0.06	0.07	0.04	0.19	
PFOS	2.47±2.49	1.27	0.35	6.72	0.57±0.79	0.32	0.07	3.04	(**)
PFDS	1.20±0.98	1.55	0.09	1.94	0.14±0.10	0.10	0.05	0.37	
FOSAA	4.33±7.10	0.36	0.03	15.7	0.35±0.53	0.14	0.01	2.20	
MeFOSAA	2.26±1.88	2.34	0.13	4.21	0.14±0.09	0.14	0.04	0.23	

EtFOSAA	1.21±1.84	0.23	0.02	4.16	0.25±0.44	0.07	0.01	1.75
NaDONA	0.39±0.69	0.08	0.03	1.77	0.18±0.45	0.04	0.01	1.67

[a] Se:Hg molar ratio was on average 228 in August and 321 in October.

[b] Σ_{10}PFAS includes PFUnDA, PFTriDA, PFTDAA, PFPeS, PFOS, PFDS, FOSAA, MeFOSAA, EtFOSAA, and NaDONA.

TISSUE DISTRIBUTION OF CONTAMINANTS

234

235   In addition to differences between seasons, contaminant concentrations also varied

236   significantly between tissues. Overall, liver exhibited the highest concentrations of toxic

237   metals (i.e., Hg, Cd, and Pb), ΣPFAS, as well as the essential element Se (Tables 1-2).

238   Notably, Cd concentrations in all muscle samples were below LOD. Despite the large

239   variation in total hepatic PFCA and PFSA concentrations, the PFAS profile in the liver

240   remained relatively homogenous across both sampling occasions, with PFOS and the long-

241   chained PFCAs PFUnDA and PFTriDA dominating the composition (Fig. 1A, 1C). In

242   contrast, the PFAS profile in muscle was more heterogeneous, with a clear shift between

243   August and October. While PFOS dominated in August, PFUnDA and PFTriDA were most

244   abundant in October (Fig. 1B, 1D).

245   The differences in contaminant concentrations and profiles between liver and muscle likely

246   reflect several physiological and chemical factors. The overall higher contaminant

247   concentrations in the liver are expected, given that it functions as a major detoxification

248   organ. This may also explain the higher Se concentrations in the liver compared to muscle,

249   given its role in mitigating metal toxicity through the formation of non-toxic complexes [33–35],

250   especially in the liver. While Se concentrations in muscle remained stable between late

251   summer and autumn (Table 2), hepatic Se was significantly higher in October than in August

252   (Table 1), possibly due to an increased dietary intake or an enhanced uptake in response to the

253   higher contaminant burdens. Although hepatic concentrations of both Se and Hg increased in

254   October compared to August, the Se:Hg molar ratio was lower in October (54.4) compared to

255   August (99.9), indicating a greater seasonal hepatic accumulation of Hg relative to Se.

256 However, this does not indicate a risk of Se limitations, as a 1:1 molar ratio generally is

257 considered sufficient to mitigate Hg toxicity [19], which is far surpassed in both liver and

258 muscle regardless of season.

259 Higher PFAS concentrations have been reported in the liver compared to muscle in a range of

260 wildlife, including seabirds [36], freshwater fish [37], and polar bears [38]. Different mechanisms

261 may influence the distribution of PFAS across tissues, such as variations in chain length and

262 functional groups. Chemical properties, such as lipophilicity, play an important role in tissue

263 distribution, as long-chain PFCAs, with higher $logK_ow$ values, tend to accumulate in lipid-

264 rich tissues, such as the liver [39]. In addition to chain length, the functional group of a PFAS

265 influences its distribution in the body by affecting its binding affinity to specific proteins,

266 resulting in tissue-specific accumulation [38,40]. Additionally, the PFAS half-lives may also be

267 tissue-specific. For example, Drew et al. [41] found that PFOS has a significantly longer half-

268 life in the liver (116 days) compared to muscle (77 days) in cattle, suggesting that muscle

269 tissue may reflect more recent exposure, while liver reflects longer-term accumulation.

270 Muscle tissue may also be more prone to PFAS remobilisation during periods of energy

271 demand. Trondrud et al. [42] found that 13-22 % of the total energy reserve of Svalbard

272 reindeer is stored in protein. It is, therefore, possible that muscle may be utilised during the

273 annual fasting in spring, resulting in PFAS stored in muscle being remobilised and potentially

274 excreted. Taken together, these mechanisms may help explain the temporal stability observed

275 in the liver PFAS profiles, compared to the more dynamic shifts reported in muscle.
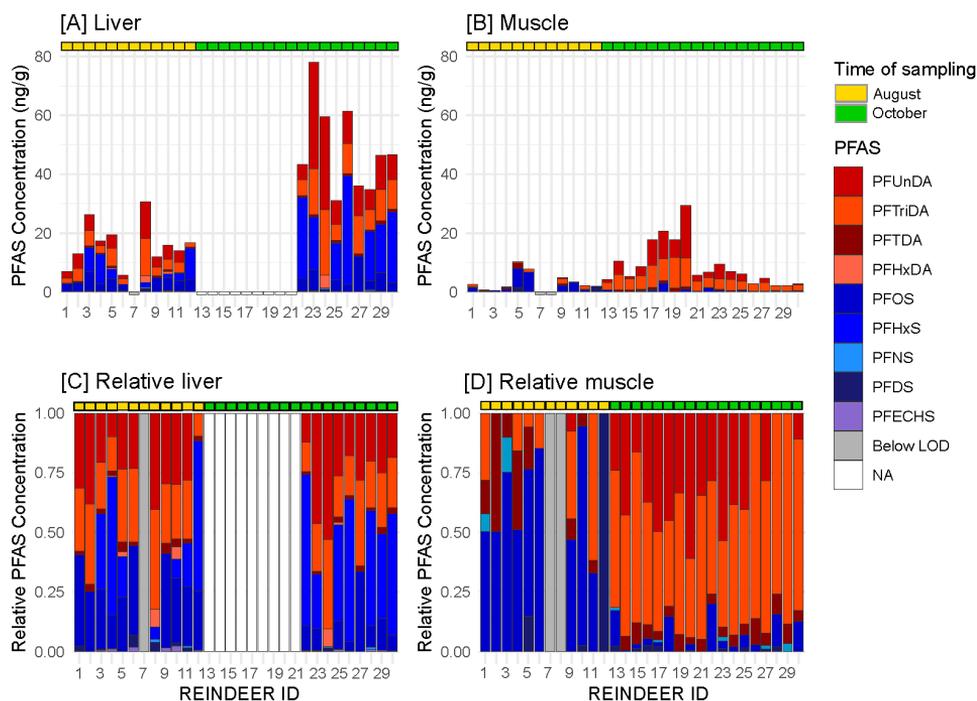
Figure 1. PFCA and PFSA concentrations (ww) in Svalbard reindeer liver [A] and muscle [B], and their relative abundance in liver [C] and muscle [D]. Each bar represents the PFAS fingerprint of a single individual. PFCAs are shown in red and orange, and PFSAs in blue and purple. Light grey bars indicate individuals with PFAS concentrations below the limit of detection (LOD), while white bars (NA) represent hepatic samples that were not analysed for PFAS. Individuals culled in August are marked in yellow, and those from October are marked in green.

LONG-RANGE VERSUS LOCAL SOURCES OF PFAS

The seasonal shift in the PFAS profile of muscle suggests changes in contaminant exposure between the two sampling seasons (Fig. 2). Muscle samples from August were mainly characterised by a strong positive relationship between PFOS and its precursors, FOSAA (ρ=0.93, p=0.003) and EtFOSAA (ρ=0.83, p=0.007), whereas samples from October showed stronger intercorrelations among PFDS and long-chain PFCAs (PFUnDA, PFTriDA, and PFTDA; ρ=0.83-0.96, p<0.001). Although the precursors FOSAA and EtFOSAA remained strongly correlated in October (ρ=0.81, p<0.001), their relationships with PFOS were

14

insignificant. Instead, FOSAA was significantly correlated with PFUnDA, PFTriDA, and
PFTDA (ρ=0.63-0.66, p≤0.005). PFOS concentrations were also lower in October (Table 2).

Figure 2. Spearman rank correlation analysis illustrating the relationship between contaminants (dw) in Svalbard reindeer muscle tissue from [A] August 2022 and [B] October 2023. Significant correlations (p < 0.05) are visualised with colour and ρ-value.

Although the muscle PFAS profiles differed between August and October, both likely reflect exposure dominated by long-range atmospheric transport. However, the specific sources or processes contributing to this long-range input remain uncertain. Volatile precursors such as FOSAA, EtFOSAA, and MeFOSAA reach remote regions like the Arctic via atmospheric transport, and then undergo degradation into both PFSAs (e.g., PFOS) and PFCAs [9,43]. Hartz et al. [8], for example, detected both PFOS, PFAS precursors, and long-chain PFCAs throughout a glacial ice core from Svalbard, demonstrating their continuous deposition following long-range transport. Once deposited, these PFAS may be remobilised into the local environment with meltwater runoff, providing a pathway of exposure to wildlife [8,44], such as Svalbard reindeer.

15

307   In addition to PFOS and its precursors co-occurring in the Svalbard environment, making

308   them available for direct exposure to wildlife, the PFOS in Svalbard reindeer may partly

309   reflect exposure to the precursor compounds alone, as both FOSAA and EtFOSAA are well-

310   documented to metabolise into PFOS in mammals [45], and exposure to these compounds has

311   been suggested to contribute to the accumulation of PFOS in other Svalbard wildlife [46]. This

312   explanation is supported by the strong intercorrelation between PFOS, FOSAA, and

313   EtFOSAA in August 2022, when concentrations of both PFOS and its precursors were

314   highest. Meanwhile, the absence of a significant relationship in October 2023, when

315   concentrations of both PFOS and its precursors were lower, further suggests that precursor

316   exposure and subsequent biotransformation may have played a role in the PFAS composition

317   of the Svalbard reindeer in August.

318   It should be noted that the significantly higher muscle concentrations of PFOS observed in

319   August 2022 compared to October 2023 (Table 2) may, to some extent, also be attributed to

320   local emissions. Local sources of PFAS, such as firefighting foam at Longyearbyen Airport

321   [46] or historical use at the former mining site of Svea [47], could contribute to PFOS exposure

322   through local transport to Reindalen. Specifically, the large-scale soil remediation project in

323   Svea between 2020 and 2022 [48] may have remobilised PFAS-contaminated particles, which

324   may have spread with the wind to nearby locations, temporarily increasing exposure to PFAS.

325   However, while local sources may play a role in exposure, the relationship in muscle between

326   PFOS and its precursors in August 2022 and the strong intercorrelation between long-chain

327   PFCAs and FOSAA in October 2023 (Fig. 2), suggests that long-range transport, rather than

328   local emissions, is the dominant influence on the PFAS profiles in the Svalbard reindeer of

329   the present study. While both profiles (i.e., in August and October) suggest that the PFAS

330   origin is dominated by long-range transport, the shift between the sampling times remains

331   unexplained but likely reflects a combination of changes in atmospheric transport patterns,

332   degradation processes, and seasonal melting of the cryosphere, all of which can influence the

333   availability and composition of PFAS in Arctic ecosystems.

334   THE SVALBARD REINDEER COMPARED WITH OTHER *RANGIFER* SUBSPECIES

335   In line with recent findings [14], the concentrations of both Hg and Pb in the present study are

336   among the lowest reported for other *Rangifer* subspecies across the Arctic ([43–47]; Figures S1-

337   S2), while Cd concentrations appear similar across circumpolar Rangifer ([43,45,46]; Fig. S3).

338   In comparison, few studies have quantified PFAS concentrations in Arctic terrestrial wildlife

339   [54]. Among those studying reindeer, considerable regional and population differences in both

340   PFAS levels and homologue patterns have been observed. For example, while PFNA and

341   PFDA concentrations were below the limit of detection in both liver and muscle in the present

342   study, these compounds were among the most abundant in both reindeer from Greenland

343   (5.77-17.2 ng/g ww) [15] and caribou from Canada (1.43-2.85 ng/g ww) [55]. Roos et al. [15]

344   documented significant differences in hepatic PFAS concentrations across reindeer

345   populations, with PFUnDA levels ranging from 2.66–4.04 ng/g (ww) in Bathurst (Canada),

346   7.63–22.4 ng/g (ww) in Akia-Maniitsoq (Greenland), and 0.02–0.37 ng/g (ww) in Svalbard.

347   Notably, there were large variations in hepatic PFAS profiles within the same regions, too,

348   with reindeer from Akia-Maniitsoq having PFUnDA concentrations (7.63-22.4 ng/g ww)

349   nearly fivefold those of the reindeer in the neighbouring region of Kangerlussuaq-Sisimiut

350   (1.38–4.01 ng/g ww)[15]. These differences were attributed to factors such as proximity to

351   military installations and airports, local differences in vegetation and climate, and exposure to

352   sea spray aerosols. Hepatic PFUnDA concentrations in the present study (1.34-12.4 ng/g ww)

353   are comparable to those in the caribou from Canada and Greenland, but far higher than what

354   was previously reported in Svalbard reindeer [15].

355   Roos et al. [15] is the only published study reporting PFAS concentrations in Svalbard

356   reindeer. Although the sample size in their study was limited (n=7), the reported

357   concentrations remain interesting to discuss as their samples were collected during the

358   hunting season of 2010, providing a temporal data point to compare our results with. In

359   general, their findings revealed relatively low hepatic PFAS concentrations, and a homologue

360   pattern dominated by PFNA and PFOS. In contrast, our study reports higher overall hepatic

361   concentrations of both PFSAs and PFCAs, along with a shift in the PFAS signature, with

362   long-chain PFCAs such as PFUnDA and PFTriDA dominating instead (Figures S4-S7). These

363   differences may be attributed to spatial variations in sampling in Svalbard or temporal

364   changes in PFAS exposure over the past decade.

365   Global emissions of PFOS have decreased following the industrial phase-out [56] and

366   implementation of restrictions on its use and production in 2009 [57]. Yet, PFOS continues to

367   be detected at high concentrations in the tissues of many wildlife species. Recent findings

368   suggest that concentrations in marine predators such as polar bears and ringed seals (*Pusa*

369   *hispida*) initially decreased following the restriction, but have been increasing again since

370   2014 [58], possibly due to enhanced environmental remobilisation of legacy PFAS or dietary

371   shifts. The fact that we observe a similar trend of increasing concentrations of both PFOS and

372   long-chain PFCAs in our study suggests that terrestrial herbivores, such as Svalbard reindeer,

373   may also be affected by these broader environmental changes, affecting their exposure to

374   PFAS.

375   HUMAN CONSUMPTION OF SVALBARD REINDEER

376   Based on the combined intake of PFOA, PFOS, PFNA, and PFHxS (Σ_4PFAS), which are the

377   four PFAS most commonly found in human blood, a tolerable weekly intake (TWI) of 4.4

378   ng/kg body weight was established by the European Food Safety Authorities (EFSA) in 2020

379   [59]. Regarding human consumption of Svalbard reindeer, intake should be approached with

380   caution. According to the present study, Svalbard reindeer in October have an average

381   Σ_4PFAS of 26.9 ng/g (ww) in liver and 0.57 ng/g (ww) in muscle, respectively. For a 70 kg

382   adult, the TWI corresponds to 308 ng Σ_4PFAS per week, which would be exceeded by

383   consuming 11.5 g of liver or 540 g of meat. It is worth noting that the PFAS profile of

384   Svalbard reindeer meat and offal not only includes PFOS and PFHxS, but is dominated by

385   long-chain PFCAs, which are not included in the EFSA Σ_4PFAS calculations, making the

386   comparison between our findings and the TWI less certain. If considering all the PFAS in the

387   ΣPFAS of muscle and liver in the present study, far lower amounts could be consumed

388   without exceeding the set threshold. In addition, although hepatic and muscle concentrations

389   of Hg and Pb remain below toxicity thresholds, hepatic Cd levels in most individuals culled in

390   October exceed the limit of 500 ng/g (ww) set by the European Commission [60], which is in

391   line with previous findings by Andersson Stavridis et al. (2025) [14]. Nevertheless, since

392   Svalbard reindeer hunting occurs on a small scale, with each hunter being allocated a quota of

393   one animal per year, the overall contaminant exposure risk is likely low but should still be

394   considered.

395   High concentrations of PFAS in wild game meat are not limited to Arctic wildlife, and studies

396   from different regions around the world report concentrations of contaminants in game that

397   exceed intake thresholds. For instance, elevated levels of PFAS have been documented in

398   white-tailed deer (*Odocoileus virginianus*) in the midwestern USA [61], and in wild boar (*Sus*

399   *scrofa*) from the Czech Republic [62], Poland [63], and Germany [64]. These findings have led to

400   governmental actions, such as hunting restrictions, and highlight the need for further studies

401   on wildlife living close to local PFAS emission sources or in environments such as the Arctic,

402   where legacy pollutants can remobilise in the future.

403   IMPLICATIONS

404   The current study reveals significant seasonal differences in contaminant concentrations, with

405   higher concentrations in Svalbard reindeer culled in October compared to those culled in

406   August. With the current annual hunting period starting in mid-August and ending at the end

407   of September [65], our findings suggest that hunters should aim to harvest their licensed

408   animals as early as possible in the allowed period to minimise their dietary exposure to

409   contaminants. The seasonal differences observed in this study also highlight the importance of

410   carefully planning future monitoring programs to ensure that all samples are collected at the

411   same time of year, as consistency is necessary for the results to be comparable across years.

412   While the concentrations of toxic metals appear to be relatively stable over the past three

413   years, there has been a significant increase in PFAS in Svalbard reindeer over the past decade.

414   The shift in the PFAS composition of the tissues, from a profile dominated by PFOS to one

415   where long-chain PFCAs are equally abundant, may be due to the remobilisation of legacy

416   contaminants from the melting cryosphere as the Arctic warms. This increase in PFAS

417   highlights the need for further monitoring to assess the impact of a rapidly changing Arctic on

418   terrestrial wildlife.

AUTHOR INFORMATION

CORRESPONDING AUTHORS (*)

Malin Andersson Stavridis - *Department of Arctic Technology, University Centre in Svalbard (UNIS), NO-9171, Longyearbyen, Norway, and the Department of Biology, Norwegian University of Science and Technology (NTNU), NO-7491, Trondheim, Norway*. Orcid: 0000-0002-9958-6093.

Email: malins@unis.no

Bjørn Munro Jenssen - *Department of Arctic Technology, University Centre in Svalbard (UNIS), NO-9171, Longyearbyen, Norway, and the Department of Biology, Norwegian University of Science and Technology (NTNU), NO-7491, Trondheim, Norway*. Orcid: 0000-0002-7042-2191.

Email: bjorn.munro.jenssen@ntnu.no

AUTHOR CONTRIBUTIONS

M.A.S. and B.M.J. designed the study. M.A.S., T.P., and V.V. collected the samples. M.A.S. and T.P. analysed the samples for elements, while G.D.K., S.S., and T.M.C. analysed the samples for PFAS. M.A.S. analysed the data and wrote the manuscript. All authors discussed the results and commented on the manuscript throughout all stages of writing. All authors have approved the final version of the manuscript.

FUNDING SOURCES

SUPPORTING INFORMATION

Supporting Information Table S1 provides supplementary culling and chemical analysis information. Table S2 provides the full name and recovery rate of all analysed PFAS. Table S3 provides all contaminant concentrations in dry weight (dw). Materials and methods provide detailed descriptions of the analytical methods used to determine Hg, elements Cd,

444   Pb, and Se, and PFAS. The supporting figures (Figs S1-S7) compare the concentrations of

445   Hg, Cd, Pb, PFUnDA, PFTriDA, PFHxS, and PFOS with the concentrations reported in other

446   reindeer and caribou across the Arctic.

DATA AVAILABILITY

448   All data used during this study are available in the Mendeley Data repository: [DOI not

449   available yet].

ACKNOWLEDGEMENTS

REFERENCES

459   (1)   Routti, H.; Atwood, T. C.; Bechshoft, T.; Boltunov, A.; Ciesielski, T. M.; Desforges,
460   J.-P.; Dietz, R.; Gabrielsen, G. W.; Jenssen, B. M.; Letcher, R. J.; McKinney, M. A.; Morris,
461   A. D.; Rigét, F. F.; Sonne, C.; Styrishave, B.; Tartu, S. State of Knowledge on Current
462   Exposure, Fate and Potential Health Effects of Contaminants in Polar Bears from the
463   Circumpolar Arctic. Sci. Total Environ. 2019, 664, 1063–1083.
464   https://doi.org/10.1016/j.scitotenv.2019.02.030.

465   (2)   Sonne, C.; Dietz, R.; Jenssen, B. M.; Lam, S. S.; Letcher, R. J. Emerging
466   Contaminants and Biological Effects in Arctic Wildlife. Trends. Ecol. Evol. 2021, 36 (5),
467   421–429. https://doi.org/10.1016/j.tree.2021.01.007.

468   (3)   Dietz, R.; Sonne, C.; Basu, N.; Braune, B.; O'Hara, T.; Letcher, R. J.; Scheuhammer,
469   T.; Andersen, M.; Andreasen, C.; Andriashek, D.; Asmund, G.; Aubail, A.; Baagøe, H.; Born,
470   E. W.; Chan, H. M.; Derocher, A. E.; Grandjean, P.; Knott, K.; Kirkegaard, M.; Krey, A.;
471   Lunn, N.; Messier, F.; Obbard, M.; Olsen, M. T.; Ostertag, S.; Peacock, E.; Renzoni, A.;

472 Rigét, F. F.; Skaare, J. U.; Stern, G.; Stirling, I.; Taylor, M.; Wiig, Ø.; Wilson, S.; Aars, J.
473 What Are the Toxicological Effects of Mercury in Arctic Biota? Sci. Total Environ. 2013,
474 443, 775–790. https://doi.org/10.1016/j.scitotenv.2012.11.046.

475 (4)　Sunderland, E. M.; Hu, X. C.; Dassuncao, C.; Tokranov, A. K.; Wagner, C. C.; Allen,
476 J. G. A Review of the Pathways of Human Exposure to Poly- and Perfluoroalkyl Substances
477 (PFASs) and Present Understanding of Health Effects. J Expo Sci Environ Epidemiol 2019,
478 29 (2), 131–147. https://doi.org/10.1038/s41370-018-0094-1.

479 (5)　Stock, N. L.; Furdui, V. I.; Muir, D. C. G.; Mabury, S. A. Perfluoroalkyl
480 Contaminants in the Canadian Arctic: Evidence of Atmospheric Transport and Local
481 Contamination. Environ. Sci. Technol. 2007, 41 (10), 3529–3536.
482 https://doi.org/10.1021/es062709x.

483 (6)　Dastoor, A.; Wilson, S. J.; Travnikov, O.; Ryjkov, A.; Angot, H.; Christensen, J. H.;
484 Steenhuisen, F.; Muntean, M. Arctic Atmospheric Mercury: Sources and Changes. Sci. Total
485 Environ. 2022, 839, 156213. https://doi.org/10.1016/j.scitotenv.2022.156213.

486 (7)　Søndergaard, J.; Elberling, B.; Sonne, C.; Larsen, M. M.; Dietz, R. Stable Isotopes
487 Unveil Ocean Transport of Legacy Mercury into Arctic Food Webs. Nat Commun 2025, 16
488 (1), 5135. https://doi.org/10.1038/s41467-025-60356-6.

489 (8)　Hartz, W. F.; Björnsdotter, M. K.; Yeung, L. W. Y.; Hodson, A.; Thomas, E. R.;
490 Humby, J. D.; Day, C.; Jogsten, I. E.; Kärrman, A.; Kallenborn, R. Levels and Distribution
491 Profiles of Per- and Polyfluoroalkyl Substances (PFAS) in a High Arctic Svalbard Ice Core.
492 Sci. Total Environ. 2023, 871, 161830. https://doi.org/10.1016/j.scitotenv.2023.161830.

493 (9)　Muir, D.; Gunnarsdóttir, M. J.; Koziol, K.; Von Hippel, F. A.; Szumińska, D.;
494 Ademollo, N.; Corsolini, S.; De Silva, A.; Gabrielsen, G.; Kallenborn, R.; Polkowska, Ż.;
495 Krümmel, E.; Vorkamp, K. Local Sources versus Long-Range Transport of Organic
496 Contaminants in the Arctic: Future Developments Related to Climate Change. Environ. Sci.:
497 Adv. 2025, 4 (3), 355–408. https://doi.org/10.1039/d4va00240g.

498 (10)　Dai, S.; Zhang, G.; Dong, C.; Yang, R.; Pei, Z.; Li, Y.; Li, A.; Zhang, Q.; Jiang, G.
499 Occurrence, Bioaccumulation and Trophodynamics of per- and Polyfluoroalkyl Substances
500 (PFAS) in Terrestrial and Marine Ecosystems of Svalbard, Arctic. Water Res. 2025, 271,
501 122979. https://doi.org/10.1016/j.watres.2024.122979.

502 (11)　Węgrzyn, M. H.; Wietrzyk, P.; Lehmann-Konera, S.; Chmiel, S.; Cykowska-
503 Marzencka, B.; Polkowska, Ż. Annual Variability of Heavy Metal Content in Svalbard
504 Reindeer Faeces as a Result of Dietary Preferences. Environ Sci Pollut Res 2018, 25 (36),
505 36693–36701. https://doi.org/10.1007/s11356-018-3479-8.

506 (12)　Ims, R. A.; Alsos, I. G.; Fuglei, E.; Pedersen, Å. Ø.; Yoccoz, N. G. An Assessment of
507 MOSJ: The State of the Terrestrial Environment in Svalbard; Rapportserie / Norsk
508 Polarinstitutt; Norsk Polarinst: Tromsø, 2014.

509   (13)   Borch-Iohnsen, B.; Nilssen, K. J.; Norheim, G. Influence of Season and Diet on Liver
510   and Kidney Content of Essential Elements and Heavy Metals in Svalbard Reindeer. Biol
511   Trace Elem Res 1996, 51 (3), 235–247. https://doi.org/10.1007/BF02784078.

512   (14)   Andersson Stavridis, M.; Pollestad, B. M.; Veiberg, V.; Mikkelsen, Ø.; Ciesielski, T.
513   M.; Jenssen, B. M. Toxic Metals and Other Elements in Svalbard Reindeer: Establishing
514   Baselines and Assessing Non-Invasive Sampling for Biomonitoring. Sci. Total Environ. 2025,
515   961, 178226. https://doi.org/10.1016/j.scitotenv.2024.178226.

516   (15)   Roos, A. M.; Gamberg, M.; Muir, D.; Kärrman, A.; Carlsson, P.; Cuyler, C.; Lind, Y.;
517   Bossi, R.; Rigét, F. Perfluoroalkyl Substances in Circum-ArcticRangifer: Caribou and
518   Reindeer. Environ. Sci. Pollut. Res. 2021. https://doi.org/10.1007/s11356-021-16729-7.

519   (16)   Veiberg, V.; Nilsen, E. B.; Rolandsen, C. M.; Heim, M.; Andersen, R.; Holmstrøm, F.;
520   Meisingset, E. L.; Solberg, E. J. The Accuracy and Precision of Age Determination by Dental
521   Cementum Annuli in Four Northern Cervids. Eur J Wildl Res 2020, 66 (6), 91.
522   https://doi.org/10.1007/s10344-020-01431-9.

523   (17)   MOSJ. Uttak av svalbardrein. https://mosj.no/indikator/pavirkning/jakt-og-
524   fangst/uttak-av-svalbardrein/ (accessed 2024-09-26).

525   (18)   Khan, M. A. K.; Wang, F. Mercury-Selenium Compounds and Their Toxicological
526   Significance: Toward a Molecular Understanding of the Mercury-Selenium Antagonism.
527   Environmental Toxicology and Chemistry 2009, 28 (8), 1567–1577.
528   https://doi.org/10.1897/08-375.1.

529   (19)   Sørmo, E. G.; Ciesielski, T. M.; Øverjordet, I. B.; Lierhagen, S.; Eggen, G. S.; Berg,
530   T.; Jenssen, B. M. Selenium Moderates Mercury Toxicity in Free-Ranging Freshwater Fish.
531   Environ. Sci. Technol. 2011, 45 (15), 6561–6566. https://doi.org/10.1021/es200478b.

532   (20)   Sait, S. T. L.; Rinø, S. F.; Gonzalez, S. V.; Pastukhov, M. V.; Poletaeva, V. I.; Farkas,
533   J.; Jenssen, B. M.; Ciesielski, T. M.; Asimakopoulos, A. G. Occurrence and Tissue
534   Distribution of 33 Legacy and Novel Per- and Polyfluoroalkyl Substances (PFASs) in Baikal
535   Seals (Phoca Sibirica). Sci. Total Environ. 2023, 889, 164096.
536   https://doi.org/10.1016/j.scitotenv.2023.164096.

537   (21)   R Core Team. R: A Language and Environment for Statistical Computing, 2024.
538   https://www.R-project.org/.

539   (22)   Polischuk, S. C.; Norstrom, R. J.; Ramsay, M. A. Body Burdens and Tissue
540   Concentrations of Organochlorines in Polar Bears (Ursus Maritimus) Vary during Seasonal
541   Fasts. Environmental Pollution 2002, 118 (1), 29–39. https://doi.org/10.1016/S0269-
542   7491(01)00278-0.

543   (23)   Dietz, R.; Riget, F.; Sonne, C.; Letcher, R.; Born, E.; Muir, D. Seasonal and Temporal
544   Trends in Polychlorinated Biphenyls and Organochlorine Pesticides in East Greenland Polar
545   Bears (Ursus Maritimus), 1990–2001. Sci. Total Environ. 2004, 331 (1–3), 107–124.
546   https://doi.org/10.1016/j.scitotenv.2004.03.025.

547 (24)   Albon, S. D.; Irvine, R. J.; Halvorsen, O.; Langvatn, R.; Loe, L. E.; Ropstad, E.;
548 Veiberg, V.; Van Der Wal, R.; Bjørkvoll, E. M.; Duff, E. I.; Hansen, B. B.; Lee, A. M.;
549 Tveraa, T.; Stien, A. Contrasting Effects of Summer and Winter Warming on Body Mass
550 Explain Population Dynamics in a Food-limited Arctic Herbivore. Glob. Change Biol 2017,
551 23 (4), 1374–1389. https://doi.org/10.1111/gcb.13435.

552 (25)   Van Der Wal, R.; Madan, N.; Van Lieshout, S.; Dormann, C.; Langvatn, R.; Albon, S.
553 D. Trading Forage Quality for Quantity? Plant Phenology and Patch Choice by Svalbard
554 Reindeer. Oecologia 2000, 123 (1), 108–115. https://doi.org/10.1007/s004420050995.

555 (26)   Eikeland, O. Nutritious and Delicious? A Non-Invasive Study of Food Plant Choices
556 by Svalbard Reindeer in Summer and Late Autumn, The Arctic University of Norway, 2023.
557 https://hdl.handle.net/10037/32151 (accessed 2025-06-26).

558 (27)   Wojtuń, B.; Polechońska, L.; Pech, P.; Mielcarska, K.; Samecka-Cymerman, A.;
559 Szymański, W.; Kolon, M.; Kopeć, M.; Stadnik, K.; Kempers, A. J. Sanionia Uncinata and
560 Salix Polaris as Bioindicators of Trace Element Pollution in the High Arctic: A Case Study at
561 Longyearbyen, Spitsbergen, Norway. Polar Biol. 2019, 42 (7), 1287–1297.
562 https://doi.org/10.1007/s00300-019-02517-0.

563 (28)   Wojtuń, B.; Samecka-Cymerman, A.; Kolon, K.; Kempers, A. J.; Skrzypek, G. Metals
564 in Some Dominant Vascular Plants, Mosses, Lichens, Algae, and the Biological Soil Crust in
565 Various Types of Terrestrial Tundra, SW Spitsbergen, Norway. Polar Biol. 2013, 36 (12),
566 1799–1809. https://doi.org/10.1007/s00300-013-1399-0.

567 (29)   Bargagli, R. Moss and Lichen Biomonitoring of Atmospheric Mercury: A Review.
568 Sci. Total Environ. 2016, 572, 216–231. https://doi.org/10.1016/j.scitotenv.2016.07.202.

569 (30)   Gamberg, M.; Braune, B.; Davey, E.; Elkin, B.; Hoekstra, P. F.; Kennedy, D.;
570 Macdonald, C.; Muir, D.; Nirwal, A.; Wayland, M.; Zeeb, B. Spatial and Temporal Trends of
571 Contaminants in Terrestrial Biota from the Canadian Arctic. Sci. Total Environ. 2005, 351–
572 352, 148–164. https://doi.org/10.1016/j.scitotenv.2004.10.032.

573 (31)   Kinck, C. Reduced Forage Access Affects Home Range Size and Site Fidelity of
574 Svalbard Reindeer (Rangifer Tarandus Platyrhynchus)., NMBU, 2014.

575 (32)   Tyler, N. J. C.; Øritsland, N. A. Why Don't Svalbard Reindeer Migrate? Ecography
576 1989, 12 (4), 369–376. https://doi.org/10.1111/j.1600-0587.1989.tb00911.x.

577 (33)   Zwolak, I. The Role of Selenium in Arsenic and Cadmium Toxicity: An Updated
578 Review of Scientific Literature. Biol Trace Elem Res 2020, 193 (1), 44–63.
579 https://doi.org/10.1007/s12011-019-01691-w.

580 (34)   Ikemoto, T.; Kunito, T.; Anan, Y.; Tanaka, H.; Baba, N.; Miyazaki, N.; Tanabe, S.
581 ASSOCIATION OF HEAVY METALS WITH METALLOTHIONEIN AND OTHER
582 PROTEINS IN HEPATIC CYTOSOL OF MARINE MAMMALS AND SEABIRDS.
583 Environ Toxicol Chem 2004, 23 (8), 2008. https://doi.org/10.1897/03-456.

584   (35)   Romero, M. B.; Polizzi, P.; Chiodi, L.; Das, K.; Gerpe, M. The Role of
585   Metallothioneins, Selenium and Transfer to Offspring in Mercury Detoxification in
586   Franciscana Dolphins (Pontoporia Blainvillei). Mar. Pollut. Bull. 2016, 109 (1), 650–654.
587   https://doi.org/10.1016/j.marpolbul.2016.05.012.

588   (36)   Robuck, A. R.; McCord, J. P.; Strynar, M. J.; Cantwell, M. G.; Wiley, D. N.;
589   Lohmann, R. Tissue-Specific Distribution of Legacy and Novel Per- and Polyfluoroalkyl
590   Substances in Juvenile Seabirds. Environ. Sci. Technol. Lett. 2021, 8 (6), 457–462.
591   https://doi.org/10.1021/acs.estlett.1c00222.

592   (37)   Hoa, N. T. Q.; Lieu, T. T.; Anh, H. Q.; Huong, N. T. A.; Nghia, N. T.; Chuc, N. T.;
593   Quang, P. D.; Vi, P. T.; Tuyen, L. H. Perfluoroalkyl Substances (PFAS) in Freshwater Fish
594   from Urban Lakes in Hanoi, Vietnam: Concentrations, Tissue Distribution, and Implication
595   for Risk Assessment. Environ Sci Pollut Res 2022, 29 (34), 52057–52069.
596   https://doi.org/10.1007/s11356-022-19532-0.

597   (38)   Greaves, A. K.; Letcher, R. J.; Sonne, C.; Dietz, R.; Born, E. W. Tissue-Specific
598   Concentrations and Patterns of Perfluoroalkyl Carboxylates and Sulfonates in East Greenland
599   Polar Bears. Environ. Sci. Technol. 2012, 46 (21), 11575–11583.
600   https://doi.org/10.1021/es303400f.

601   (39)   Jing, P.; Rodgers, P. J.; Amemiya, S. High Lipophilicity of Perfluoroalkyl Carboxylate
602   and Sulfonate: Implications for Their Membrane Permeability. J. Am. Chem. Soc. 2009, 131
603   (6), 2290–2296. https://doi.org/10.1021/ja807961s.

604   (40)   Shi, Y.; Vestergren, R.; Nost, T. H.; Zhou, Z.; Cai, Y. Probing the Differential Tissue
605   Distribution and Bioaccumulation Behavior of Per- and Polyfluoroalkyl Substances of
606   Varying Chain-Lengths, Isomeric Structures and Functional Groups in Crucian Carp. Environ.
607   Sci. Technol. 2018, 52 (8), 4592–4600. https://doi.org/10.1021/acs.est.7b06128.

608   (41)   Drew, R.; Hagen, T. G.; Champness, D.; Sellier, A. Half-Lives of Several
609   Polyfluoroalkyl Substances (PFAS) in Cattle Serum and Tissues. Food Addit. Contam. Part
610   A. 2022, 39 (2), 320–340. https://doi.org/10.1080/19440049.2021.1991004.

611   (42)   Trondrud, L. M.; Pigeon, G.; Król, E.; Albon, S.; Evans, A. L.; Arnold, W.; Hambly,
612   C.; Irvine, R. J.; Ropstad, E.; Stien, A.; Veiberg, V.; Speakman, J. R.; Loe, L. E. Fat Storage
613   Influences Fasting Endurance More than Body Size in an Ungulate. Functional Ecology 2021,
614   35 (7), 1470–1480. https://doi.org/10.1111/1365-2435.13816.

615   (43)   Ahrens, L.; Rakovic, J.; Ekdahl, S.; Kallenborn, R. Environmental Distribution of Per-
616   and Polyfluoroalkyl Substances (PFAS) on Svalbard: Local Sources and Long-Range
617   Transport to the Arctic. Chemosphere 2023, 345, 140463.
618   https://doi.org/10.1016/j.chemosphere.2023.140463.

619   (44)   MacInnis, J.; De Silva, A. O.; Lehnherr, I.; Muir, D. C. G.; St. Pierre, K. A.; St. Louis,
620   V. L.; Spencer, C. Investigation of Perfluoroalkyl Substances in Proglacial Rivers and
621   Permafrost Seep in a High Arctic Watershed. Environ. Sci.: Processes Impacts 2022, 24 (1),
622   42–51. https://doi.org/10.1039/D1EM00349F.

623 (45) Martin, J. W.; Asher, B. J.; Beesoon, S.; Benskin, J. P.; Ross, M. S. PFOS or PreFOS?
624 Are Perfluorooctane Sulfonate Precursors (PreFOS) Important Determinants of Human and
625 Environmental Perfluorooctane Sulfonate (PFOS) Exposure? J. Environ. Monit. 2010, 12
626 (11), 1979. https://doi.org/10.1039/c0em00295j.

627 (46) Ali, A. M.; Langberg, H. A.; Hale, S. E.; Kallenborn, R.; Hartz, W. F.; Mortensen, Å.-
628 K.; Ciesielski, T. M.; McDonough, C. A.; Jenssen, B. M.; Breedveld, G. D. The Fate of Poly-
629 and Perfluoroalkyl Substances in a Marine Food Web Influenced by Land-Based Sources in
630 the Norwegian Arctic. Environ. Sci.: Process Impacts 2021, 23 (4), 588–604.
631 https://doi.org/10.1039/D0EM00510J.

632 (47) Evenset, A.; Pedersen, K. B.; Kvennås, M.; Cappelen, P. Miljøtekniske Undersøkelser
633 i Svea 2019; APN-60480-2; Akvaplan-niva, 2020.

634 (48) Perpetuum. PFAS from airports: How Perpetuum deposited 50.000 tonnes of AFFF
635 PFAS soil. https://perpetuum.no/en/about-pfas/disposal-of-afff-foam-from-airport/.

636 (49) Gamberg, M.; Cuyler, C.; Wang, X. Contaminants in Two West Greenland Caribou
637 Populations. Sci. Total Environ. 2016, 554–555, 329–336.
638 https://doi.org/10.1016/j.scitotenv.2016.02.154.

639 (50) Schuster, R. C.; Gamberg, M.; Dickson, C.; Chan, H. M. Assessing Risk of Mercury
640 Exposure and Nutritional Benefits of Consumption of Caribou (Rangifer Tarandus) in the
641 Vuntut Gwitchin First Nation Community of Old Crow, Yukon, Canada. Environ. Res. 2011,
642 111 (6), 881–887. https://doi.org/10.1016/j.envres.2011.05.025.

643 (51) Makarov, D. A.; Ovcharenko, V. V.; Nebera, E. A.; Kozhushkevich, A. I.;
644 Shelepchikov, A. A.; Turbabina, K. A.; Kalantaenko, A. M.; Bardyugov, N. S.; Gergel, M. A.
645 Geographical Distribution of Dioxins, Cadmium, and Mercury Concentrations in Reindeer
646 Liver, Kidneys, and Muscle in the Russian Far North. Environ. Sci. Pollut. Res. 2022, 29 (8),
647 12176–12187. https://doi.org/10.1007/s11356-021-16310-2.

648 (52) O'Hara, T. M.; George, J. C.; Blake, J.; Burek, K.; Carroll, G.; Dau, J.; Bennett, L.;
649 McCoy, C. P.; Gerard, P.; Woshner, V. Investigation of Heavy Metals in a Large Mortality
650 Event in Caribou of Northern Alaska. ARCTIC 2003, 56 (2), 125–135.
651 https://doi.org/10.14430/arctic608.

652 (53) Ali Hassan, A.; Rylander, C.; Brustad, M.; Sandanger, TorkjelM. Level of Selected
653 Toxic Elements in Meat, Liver, Tallow and Bone Marrow of Young Semi-Domesticated
654 Reindeer (Rangifer Tarandus Tarandus L.) from Northern Norway. International Journal of
655 Circumpolar Health 2012, 71 (1), 18187. https://doi.org/10.3402/ijch.v71i0.18187.

656 (54) Muir, D.; Bossi, R.; Carlsson, P.; Evans, M.; De Silva, A.; Halsall, C.; Rauert, C.;
657 Herzke, D.; Hung, H.; Letcher, R.; Rigét, F.; Roos, A. Levels and Trends of Poly- and
658 Perfluoroalkyl Substances in the Arctic Environment – An Update. Emerg. Contam. 2019, 5,
659 240–271. https://doi.org/10.1016/j.emcon.2019.06.002.

660 (55) Müller, C. E.; De Silva, A. O.; Small, J.; Williamson, M.; Wang, X.; Morris, A.; Katz,
661 S.; Gamberg, M.; Muir, D. C. G. Biomagnification of Perfluorinated Compounds in a Remote

Terrestrial Food Chain: Lichen–Caribou–Wolf. Environ. Sci. Technol. 2011, 45 (20), 8665–8673. https://doi.org/10.1021/es201353v.

(56)   Paul, A. G.; Jones, K. C.; Sweetman, A. J. A First Global Production, Emission, And Environmental Inventory For Perfluorooctane Sulfonate. Environ. Sci. Technol. 2009, 43 (2), 386–392. https://doi.org/10.1021/es802216n.

(57)   Stockholm Convention on Persistent Organic Pollutants. Listing of PFOS, its salts and PFOSF in Annex B (Decision SC-4/17). https://chm.pops.int/.

(58)   Lohmann, R.; Abass, K.; Bonefeld-Jørgensen, E. C.; Bossi, R.; Dietz, R.; Ferguson, S.; Fernie, K. J.; Grandjean, P.; Herzke, D.; Houde, M.; Lemire, M.; Letcher, R. J.; Muir, D.; De Silva, A. O.; Ostertag, S. K.; Rand, A. A.; Søndergaard, J.; Sonne, C.; Sunderland, E. M.; Vorkamp, K.; Wilson, S.; Weihe, P. Cross-Cutting Studies of per- and Polyfluorinated Alkyl Substances (PFAS) in Arctic Wildlife and Humans. Sci. Total Environ. 2024, 954, 176274. https://doi.org/10.1016/j.scitotenv.2024.176274.

(59)   Schrenk, D.; Bignami, M.; Bodin, L.; Chipman, J. K.; del Mazo, J.; Grasl-Kraupp, B.; Hogstrand, C.; Hoogenboom, L. (Ron); Leblanc, J.; Nebbia, C. S.; Nielsen, E.; Ntzani, E.; Petersen, A.; Sand, S.; Vleminckx, C.; Wallace, H.; Barregård, L.; Ceccatelli, S.; Cravedi, J.; Halldorsson, T. I.; Haug, L. S.; Johansson, N.; Knutsen, H. K.; Rose, M.; Roudot, A.; Van Loveren, H.; Vollmer, G.; Mackay, K.; Riolo, F.; Schwerdtle, T. Risk to Human Health Related to the Presence of Perfluoroalkyl Substances in Food. EFS2 2020, 18 (9). https://doi.org/10.2903/j.efsa.2020.6223.

(60)   The European Commission. COMMISSION REGULATION (EU) 2023/915 of 25 April 2023 on Maximum Levels for Certain Contaminants in Food and Repealing Regulation (EC) No 1881/2006, 2023. https://eur-lex.europa.eu/legal-content/EN/TXT/PDF/?uri=CELEX:32023R0915 (accessed 2024-01-03).

(61)   Michigan Department of Health and Human Services. PFAS Levels in Michigan Deer and Eat Safe Wild Game Guidelines; Division of Environmental Health, Michigan Fish Consumption Advisory Program, 2018. https://www.michigan.gov/-/media/Project/Websites/PFAS-Response/Reports/2018-01-11-Eat-Safe-Wild-Game-Deer-Guidelines.pdf (accessed 2025-07-24).

(62)   Schröder, T.; Müller, V.; Preihs, M.; Borovička, J.; Gonzalez De Vega, R.; Kindness, A.; Feldmann, J. Fluorine Mass Balance Analysis in Wild Boar Organs from the Bohemian Forest National Park. Science of The Total Environment 2024, 922, 171187. https://doi.org/10.1016/j.scitotenv.2024.171187.

(63)   Mikolajczyk, S.; Warenik-Bany, M.; Pajurek, M.; Marchand, P. Perfluoroalkyl Substances in the Meat of Polish Farm Animals and Game – Occurrence, Profiles and Dietary Intake. Science of The Total Environment 2024, 945, 174071. https://doi.org/10.1016/j.scitotenv.2024.174071.

(64)   Felder, C.; Trompeter, L.; Skutlarek, D.; Färber, H.; Mutters, N. T.; Heinemann, C. Exposure of a Single Wild Boar Population in North Rhine-Westphalia (Germany) to

701 Perfluoroalkyl Acids. Environ Sci Pollut Res 2022, 30 (6), 15575–15584.
702 https://doi.org/10.1007/s11356-022-23086-6.

703 (65)   Sysselmesteren. Governor of Svalbard. Species that can be hunted.
704 https://www.sysselmesteren.no/en/hunting-trapping-and-fishing/jaktbare-arter/ (accessed
705 2025-01-07).

706 FOR TABLE OF CONTENTS ONLY
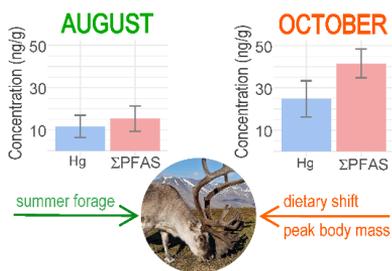
707

SUPPORTING INFORMATION

Title: Seasonal Shift in Exposure and Accumulation of PFAS and Heavy Metals in High Arctic Reindeer

Authors: Malin Andersson Stavridis[1,2]*, Tove Petersson[1,3], Görkem Deniz Kendir[2,4], Shannen Sait[4], Øyvind Mikkelsen[1,4], Vebjørn Veiberg[5], Tomasz Maciej Ciesielski[1] & Bjørn Munro Jenssen[1,2]*.

Affiliations:
1. Department of Arctic Technology, University Centre in Svalbard (UNIS), PO Box 156 N-9171, Longyearbyen, Norway.
2. Department of Biology, Norwegian University of Science and Technology (NTNU), NO-7491, Trondheim, Norway.

3. Department of Ecoscience, Marine Mammal Research, Aarhus University, DK-4000, Roskilde, Denmark.
4. Department of Chemistry, Norwegian University of Science and Technology (NTNU), NO-7491, Trondheim, Norway.
5. Land and Biodiversity, Norwegian Institute for Nature Research (NINA), NO-7485 Trondheim, Norway.

*corresponding author(s): Malin Andersson Stavridis: malins@unis.no & Bjørn Munro Jenssen: bjorn.munro.jenssen@ntnu.no.

SUPPORTING INFORMATION CONTAINS

18 pages: S1-S18

Tables S1-S3

Materials and Methods supplementary (with further details on chemical analysis)

Figures S1-S7

TABLE S1. Supplementary culling and analysis information.

| | | | | | Samples analysed for contaminants | | | | | |
| | | | | | Hg | | Elements | | PFAS | |
ID	Culling time	Age	Weight	Location	Liver	Muscle	Liver	Muscle	Liver	Muscle
1	Aug-2022	5	78	Reindalen	×	×	×	×	×	×
2	Aug-2022	4	65	Reindalen	×	×	×	×	×	×
3	Aug-2022	2	62	Semmeldalen	×	×	×	×	×	×
4	Aug-2022	10	73	Semmeldalen	×	×	×	×	×	×
5	Aug-2022	2	56.5	Reindalen	×	×	×	×	×	×
6	Aug-2022	9	70.5	Reindalen	×	×	×	×	×	×
7	Aug-2022	8	70	Reindalen	×	×	×	×	×	×
8	Aug-2022	9	67	Semmeldalen	×	×	×	×	×	×
9	Aug-2022	8	71	Kalvdalen	×	×	×	×	×	×
10	Aug-2022	2	58	Kalvdalen	×	×	×	×	×	×
11	Aug-2022	1	44	Kalvdalen	×	×	×	×	×	×
12	Aug-2022	3	60	Kalvdalen	×	×	×	×	×	×
13	Oct-2023	8	71	Colesdalen		×		×	×	×
14	Oct-2023	11	66	Colesdalen		×		×		×
15	Oct-2023	7	71	Colesdalen		×		×		×
16	Oct-2023	7	80	Colesdalen		×			×	×
17	Oct-2023	7	76	Colesdalen		×		×		×
18	Oct-2023	9	68.5	Colesdalen		×		×		×
19	Oct-2023	5	78	Colesdalen		×				×
20	Oct-2023	6	69	Reindalen		×		×		×
21	Oct-2023	7	73	Reindalen		×		×	×	×
22	Oct-2023	10	67	Reindalen	×	×	×	×	×	×
23	Oct-2023	10	75	Colesdalen	×	×	×		×	×
24	Oct-2023	7	73.5	Colesdalen	×	×	×			×
25	Oct-2023	8	73.5	Colesdalen	×	×	×			×
26	Oct-2023	4	67	Reindalen	×	×	×		×	×
27	Oct-2023	7	74.5	Reindalen	×	×	×			×
28	Oct-2023	6	72	Colesdalen	×	×	×		×	×
29	Oct-2023	4	70.5	Colesdalen	×	×	×		×	×
30	Oct-2023	6	78	Reindalen	×	×	×		×	×
				n=	21	30	21	20	21	30

TABLE S2. Full name, abbreviation, and recovery rate of all PFAS analysed in the study. Grouped by functional group and chemical structure.

	Full name	Abbreviation	Recovery (%)
PFCAs	Perfluorobutanoic acid	PFBA	100
	Perfluoropentanoic acid	PFPeA	103
	Perfluorohexanoic acid	PFHxA	100
	Perfluoroheptanoic acid	PFHpA	101
	7H-dodecafluoroheptanoic acid	PFHeA	63
	Perfluorooctanoic acid	PFOA	100
	Perfluorononanoic acid	PFNA	107
	Perfluorodecanoic acid	PFDA	105
	Perfluoroundecanoic acid	PFUnDA	55
	Perfluorododecanoic acid	PFDoDA	55
	Perfluorotridecanoic acid	PFTriDA	45
	Perfluorotetradecanoic acid	PFTDA	58
	Perfluoro-n-hexadecanoic acid	PFHxDA	91
	Perfluorooctadecanoic acid	PFOcDA	135
PFSAs	Perfluorobutanoic acid sulfonate	PFBS	100
	Perfluoropentane sulfonic acid	PFPeS	61
	Perfluorohexane sulfonic acid	PFHxS	64
	Perfluoro-1-heptanesulfonate	PFHpS	63
	Perfluorooctano sulfonic acid	PFOS	61
	Perfluorononane sulfonic acid	PFNS	63
	Perfluorodecane sulfonic acid	PFDS	61
	Perfluorododecane sulfonic acid	PFDoDS	63
	Perfluoroethylcyclohexane sulfonic acid	PFECHS	63
FTSs	1H,2H-perfluorohexan sulfonate 4:2	4:2 FTS	62
	1H,2H-perfluorooctane sulfonate 6:2	6:2 FTS	66
	1H,2H-perfluorodecan sulfonate 8:2	8:2 FTS	69
	1H,2H-perfluorododecan sulfonate 10:2	10:2 FTS	62
Sulfonamides and precursor PFAS	Perfluoro-1-octanesulfonamidoacetic acid	FOSAA	81
	2-(N-methylperfluoro-1-octansulfonamido)acetic acid	MeFOSAA	97
	N-ethylperfluoro-1-octanesulfonamide acetic acid	EtFOSAA	94
	Perfluorooctane sulfonamide	PFOSA	80
	N-methylperfluoro-1-octanesulfonamide	MeFOSA	69
	Sulfluramid	EtFOSA	74
	N-(2-hydroxyethyl)-N-methylperfluorooctane sulfonamide	MeFOSE	93
	N-ethyl-N-(2-hydroxyethyl)-N-methylperfluorooctane sulfonamide	EtFOSE	16
Emerging PFAS	2,3,3,3-tetrafluoro-2-(1,1,2,2,3,3,3-heptafluoropropoxy)propanoate	GenX	69
	Sodium dodecafluoro-3H-4,8-dioxanonanoate	NaDONA	69
	9-chlorohexadecafluoro-3-oxanonane-1-sulfonate	9Cl-PF3ONS	67
	2-(N-ethylperfluorooctane-1-sulfonamido)ethyl phosphate	SaMPAP	66
	Bis[2-(N-ethylperfluorooctane-1-sulfonamido)ethyl] phosphate	diSAMPAP	0
	Perfluoro-3,7-dimethyloctanoic acid	P37DMOA	66

TABLE S3. Dry weight (dw) concentrations of contaminants in Svalbard reindeer liver and muscle in August 2022 and October 2023. For each contaminant, columns refer to the detection rate, LOD, mean ± SD, median, minimum, and maximum contaminant concentrations.

Tissue	Contaminant	%>LOD	LOD	Concentration (ng/g dw)							
				August 2022				October 2023			
				Mean±SD	Median	Min	Max	Mean±SD	Median	Min	Max
Liver	THg	100	0.0003	40.4±19.0	37.2	20.4	92.1	83.5±28.5	67.9	49.7	130.2
	Se	100	0.003	1370±209	1340	1030	1700	1610±144	1589	1383	1901
	Cd	100	0.14	671±308	622	407	1520	1811±664	1850	1040	3130
	Pb	100	0.003	180±123	151	52.2	414	163±88.3	142	52.5	326
	PFUnDA	91	0.006	15.6±11.2	14.7	4.59	44.4	46.7±40.1	34.6	16.7	122
	PFTriDA	95	0.006	14.5±11.3	12.3	5.73	43.6	37.9±21.2	33.3	16.8	84.9
	PFTDA	86	0.006	1.37±0.73	1.34	0.31	2.69	2.90±0.94	2.83	1.40	4.29
	PFHxDA	29	0.006	3.17±3.49	2.09	0.31	8.18	8.78±11.1	8.78	0.94	16.6
	PFHxS	67	0.006	18.2±14.0	11.0	4.17	38.0	70.9±25.3	59.7	44.0	117
	PFOS	86	0.02	13.2±4.31	13.7	7.40	21.5	17.8±11.5	13.5	7.75	42.8
	PFNS	38	0.006	0.66±0.74	0.34	0.14	1.51	0.80±0.57	1.00	0.13	1.31
	PFDS	48	0.006	1.27±1.50	0.71	0.31	4.30	1.11±1.58	0.41	0.16	3.47
	PFECHS	57	0.006	0.65±0.35	0.57	0.39	1.32	0.47±0.21	0.77	0.44	0.96
	FOSAA	81	0.006	7.42±19.5	0.70	0.17	59.3	2.70±3.32	1.28	0.04	9.01
	MeFOSAA	29	0.006	0.28±0.37	0.11	0.02	0.71	2.38±1.15	1.82	1.62	3.70
	EtFOSAA	67	0.006	3.31±7.14	0.74	0.05	19.5	2.36±3.11	1.47	0.15	9.26
	NaDONA	76	0.006	1.53±1.66	1.02	0.29	5.34	22.3±27.1	16.32	0.70	83.0
Muscle	THg	100	0.0003	8.53±4.96	8.09	2.74	21.5	6.35±3.04	5.19	3.30	12.9
	Se	95	0.003	678±320	582	490	1500	654±108	670	523	831
	Pb	47	0.003	0.83±0.51	0.98	0.10	1.33	1.00±0.88	0.87	0.09	2.77
	PFUnDA	53	0.006	Below the LOD				16.2±16.2	10.1	1.05	61.1

				Concentration (ng/g dw)							
				August 2022				October 2023			
Tissue	Contaminant	%>LOD	LOD	Mean±SD	Median	Min	Max	Mean±SD	Median	Min	Max
	PFTriDA	80	0.006	3.41±1.93	3.19	1.08	6.08	15.8±9.76	12.0	6.01	38.8
	PFTDA	83	0.006	1.68±1.84	1.22	0.22	5.95	1.68±1.13	1.17	0.46	4.68
	PFPeS	27	0.02	0.49±0.24	0.49	0.32	0.67	0.35±0.24	0.25	0.13	0.73
	PFOS	77	0.02	8.47±8.19	4.42	1.23	22.8	2.06±2.93	1.14	0.26	11.5
	PFDS	40	0.006	3.97±3.24	5.03	0.33	6.55	0.50±0.39	0.38	0.18	1.38
	FOSAA	83	0.006	14.3±23.4	1.29	0.12	50.7	1.27±1.95	0.52	0.03	8.09
	MeFOSAA	27	0.006	7.48±6.14	7.93	0.47	13.6	0.50±0.34	0.53	0.13	0.80
	EtFOSAA	77	0.006	4.02±6.11	0.79	0.07	14.1	0.93±1.63	0.27	0.03	6.46
	NaDONA	63	0.006	1.33±2.31	0.28	0.13	5.97	0.65±1.66	0.17	0.05	6.16

MATERIALS AND METHODS SUPPLEMENTARY

THg determination

Before analysis, Hg calibration standards were made by preparing solutions containing 2%
hydrochloric acid, ultrapure water (Elga® Purelab Flex 4), and Hg from stock (LabKings
B.V.®; 0.01-100 ng) following the instructions provided by Milestone® for ultratrace analysis
of Hg. The calibration standards were used to create calibration curves in both cell 0
(R^2=0.999) and cell 1 (R^2=0.998. Both certified reference materials (CRMs) and calibration
standards were analysed to ensure the precision of the analysis, with MODAS–3 Herring
Tissue (M–3 HerTis, 221±21 µg/kg, recovery ranging between 107-108%) from MODAS,
Gdańsk (2015) and ORIENTAL BASMA TOBACCO LEAVES (INC-OBTL-5, 23.2±1.6
µg/kg, recovery ranging between 80-87%) from the Institute of Nuclear Chemistry and
Technology, Warszawa (2010) used as CRMs in cell 0 and cell 1, respectively.

Blanks were run at the beginning and end of each analysis day, with blank heights not
exceeding 0.003. All samples were run in singletons with periodic triplicates (about every 10
samples). Approximately 40 mg of sample material was analysed, with less mass analysed for
samples with higher fat content (22-40 mg). Background absorbance was accounted for by
averaging the blank heights from each day and subtracting this from the height of the sample
before calculating the final concentration.

Element quantification

Approximately 400 mg of muscle, 300 mg of liver, 300 mg of CRM (1577b Bovine Liver, US
Department of Commerce National Institute of Standards and Technology, Gaithersburg, Md
20899), 300 g of ultrapure water were added to polytetrafluoroethylene (PTFE) vials
containing 5 ml of 50% (v/v) HNO_3 (purified from HNO3, AnalaR NORMAPUR®, VWR) in
a sub-boiling distillation system (Milestone, SubPur, Sorisole, BG, Italy). Once digested using
a high-pressure microwave system (Milestone Ultraclave, EMLS, Leutkirch, Germany), the
samples were diluted to a final weight of approximately 54g. Concentrations of elements Cd,

Pb, and Se were quantified in the liver and muscle, CRM (Bovine Liver Standard Reference Material 1577b), and blank digests using an 8800 Triple Quadrupole inductively coupled plasma mass spectrometry (ICP-MS) system (Agilent, USA) equipped with a prepFAST M5 autosampler (ESI, USA). The final sample and CRM concentrations were corrected for total dilution volume and blank value concentrations.

The Bovine Liver 1577b certified values were as follows for Cd: 0.5±0.03 µg/g, Pb: 0.129±0.004, and Se: 0.73±0.06. Reference material recovery rates ranged from 79.9-84.3% for Pb, 84.7-106% for Cd, and 90.2-97.3% for Se.

The limits of detection (LOD) were determined as either three times the standard deviation of the blanks or the instrument detection limits based on the highest obtained value. The LOD for Cd was 0.47 µg/kg, for Pb 0.02 µg/kg, and for Se 0.01 µg/kg. All elements were detected above the LOD in the liver samples. In contrast, Cd was not detected above LOD in a majority of the muscle samples and therefore not reported.

PFAS quantification

Quantification of target analytes was achieved using internal standards and matrix-matched calibration curves, which were prepared by spiking target analytes and internal standards into the matrix prior to extraction, as described in Sait et al., (2023). Calibration was performed using a 13-point calibration curve (0.00, 0.01, 0.02, 0.05, 0.10, 0.20, 0.50, 1.00, 2.00, 5.00, 10.0, 20.0, and 50.0 ng/ml) with R^2 values ranging from 0.935-0.999. The method LOD was determined using the lowest detected concentration in the calibration curve range, corrected for the dilution factor, with LODs ranging between 0.013-0.152 ng/g ww. Solvent blanks were run every ten samples to monitor contamination and were subtracted from sample measurements to correct final concentrations.

The efficiency of the extraction was determined through absolute recovery rates using the method blanks, matrix-matched, and spiked samples. Out of the 41 targeted PFAS, EtFOSE

and diSAMPAP had recoveries below 45% and were excluded from the analysis. Recovery rates for the remaining PFAS ranged between 45-135%, with RSDs <10%. Several PFAS were not detected above the LOD in at least 25 % the samples and are consequently not reported in the current study. These include PFBA, PFPeA, PFHxA PFHpA, PFOA, PFNA, PFDA, PFDoDA, PFOcDA, PFBS, PFHpS, PFDoDS, 4:2 FTS, 6:2 FTS, 8:2 FTS, 10:2 FTS, PFOSA, MeFOSA, EtFOSA, MeFOSE, EtFOSE, GenX, 9Cl-PF3ONS, P37DMOA, SaMPAP, and diSAMPAP in both liver and muscle samples, PFHxDA, PFHxS, PFNS, PFECHS in muscle samples, and PFPeS in liver samples.

Analytical standards (purity ≥98%) were supplied by Wellington Laboratories Inc. (Ontario, Canada). Internal standards (purity ≥99%) of isotopically labelled perfluorooctanoic acid (PFOA-13C8), perfluorooctanesulfonate sodium salt (PFOS-13C8), and 1H,2H-perfluorooctane sulfonate (6:2) (6:2 FTS-13C2) were supplied by Cambridge Isotope Laboratories, Inc. (Tewksbury, MA, US). Stock solutions of target analyte (200 ppb) and internal standards (1 ppm) were prepared in methanol (MeOH) and stored at -20ºC.

Protocol for PFAS extraction
Approximately 100 mg of homogenised and freeze-dried liver and muscle samples were transferred into individual 15 mL polypropylene (PP) test tubes. To these tubes, 3 mL of ethyl acetate, 300 μL of 1.0 M ammonium acetate, and 10 μL of internal standard mix were added. The tubes were vortexed for 10 seconds until fully mixed, then ultrasonicated for 45 minutes. Following centrifugation (4000×g, 10 min) for protein precipitation, the top 3 mL ethyl acetate layer was transferred to a fresh PP tube. An additional 3 mL of ethyl acetate was added to the original sample tube, followed by vortexing, ultrasonication, and centrifugation. The top 3 mL ethyl acetate layer was again transferred to the fresh PP tube. This step was repeated once more, resulting in a total of 9 mL of ethyl acetate extract. Following the last transfer, 1 mL of ultrapure water was added to the extracts.

The new mixtures were centrifuged once more (4000×g, 10 min), and the supernatant of each tube was transferred to fresh PP tubes. The supernatant solutions were evaporated under a gentle stream of nitrogen gas (~2.5 PSI) to near dryness. The dried residue was reconstituted in 500 μL of a 1:1 MeOH-water solution, vortexed for 10s, and transferred to liquid chromatography (LC) autosampler vials. Vials were stored at -20°C until further analysis.

Analysis of PFAS was conducted at the Norwegian University of Science and Technology (NTNU) using ultra-performance liquid chromatography (UPLC) on a Waters Acquity I-Class system (Waters, Milford, USA), coupled to a Xevo TQ-S triple quadrupole mass spectrometer operated in multiple reaction monitoring (MRM) mode. Ionization was performed using a ZSpray electrospray ion source in negative mode (ESI–). Chromatographic separation was achieved using a Kinetex C18 column (30 × 2.1 mm, 1.3 μm, 100 Å; Phenomenex, Denmark) equipped with a matching C18 guard column (10 × 2.1 mm). The column temperature was held at 30 °C.

Mobile phases consisted of (A) water with 2 mM ammonium acetate and (B) methanol. A gradient elution was applied as follows: 20% B for 0.1 min, ramped to 50% at 0.1 min, 70% at 0.6 min, 80% at 0.7 min, 85% at 1.3 min, and 100% at 1.7 min. This was held for 1 min before returning to 20% B over 0.1 min, followed by re-equilibration for 0.4 min. The total run time was 6 minutes, with a flow rate of 0.25 mL/min and an injection volume of 4 μL.

Instrument parameters were optimized as follows: capillary voltage at 2.0 kV, source temperature 150 °C, desolvation temperature 450 °C, cone gas flow 150 L/h, desolvation gas flow 650 L/h, and nebulizer pressure at 6 bar.

Peak integration was carried out manually using MassLynx software. Final concentrations were corrected for sample mass and dilution factor.
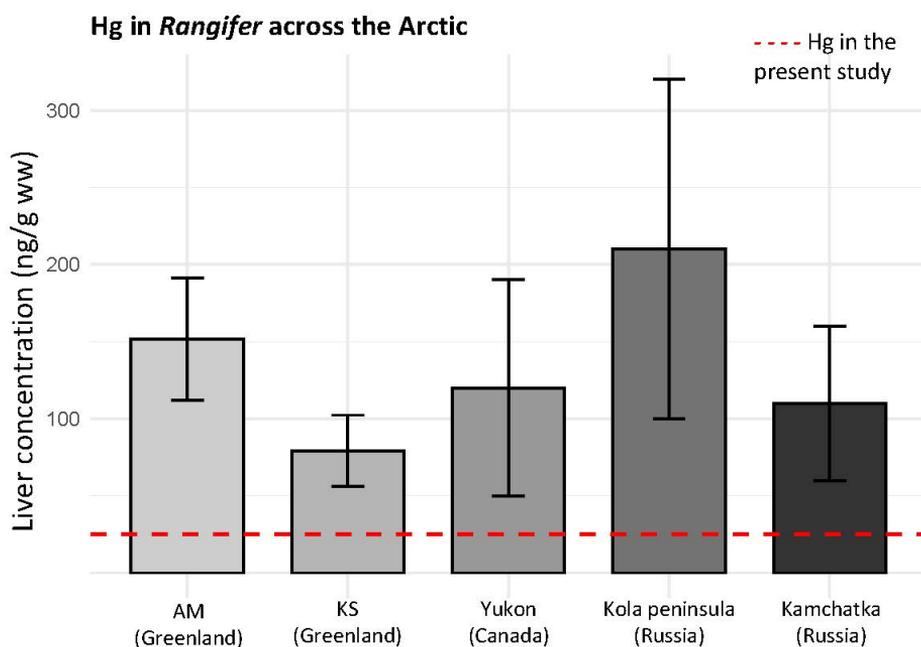
Figure S1. Hepatic Hg concentrations (ww) from various Rangifer subspecies across the Arctic. The dashed red line represents the average Hg concentration reported in individuals culled in October in the present study (25 ng/g ww). Concentrations from Akia-Maniitsoq (AM) and Kangerlussuaq-Sisimiut (KS) are reported in Gamberg et al. (2016), concentrations from Yukon derive from Schuster et al., (2011), and the data from Kola Peninsula and Kamchatka from Makarov et al. (2022).
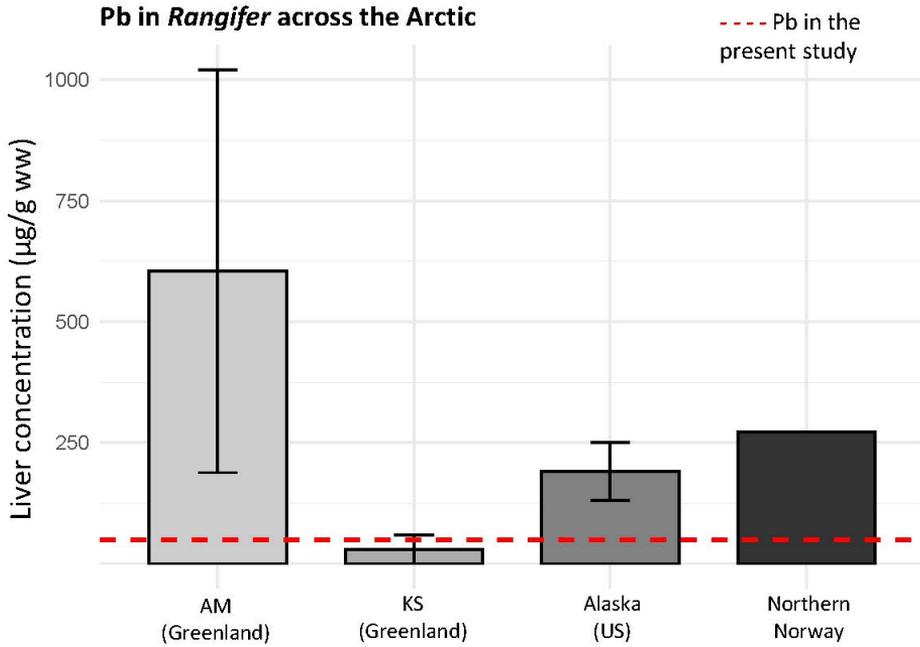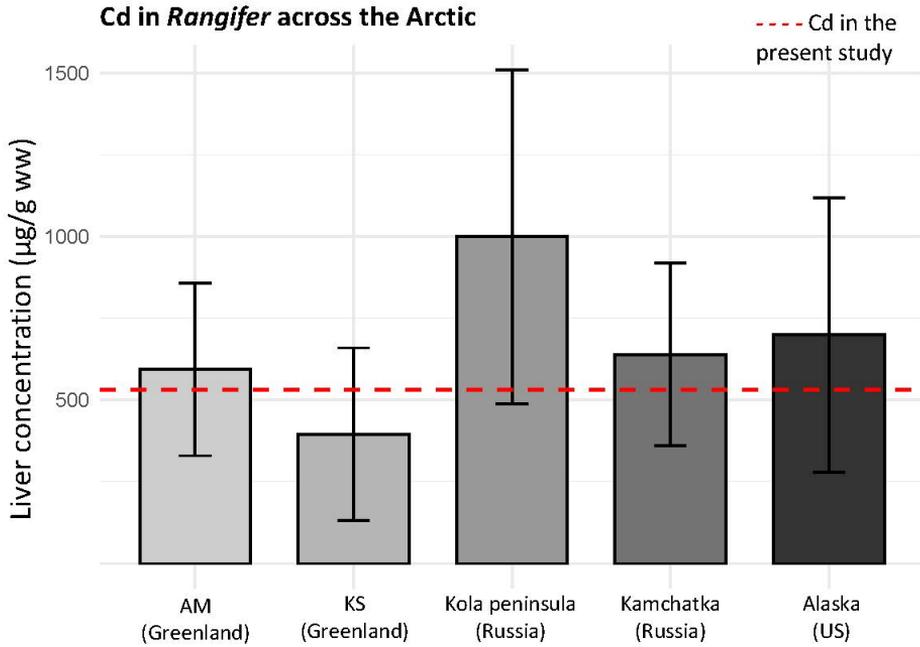
Figure S2. Hepatic Pb concentrations (ww) from various Rangifer subspecies across the Arctic. The dashed red line represents the average Pb concentration reported in individuals culled in October in the present study (49 ng/g ww). Concentrations from Akia-Maniitsoq (AM) and Kangerlussuaq-Sisimiut (KS) are reported in Gamberg et al. (2016), concentrations from Alaska derive from O'Hara et al. (2003), and the data from Northern Norway from Ali Hassan et al. (2012).

Figure S3. Hepatic Cd concentrations (ww) from various Rangifer subspecies across the Arctic. The dashed red line represents the average Cd concentration reported in individuals culled in October in the present study (532 ng/g ww). Concentrations from Akia-Maniitsoq (AM) and Kangerlussuaq-Sisimiut (KS) are reported in Gamberg et al. (2016), concentrations from Kola peninsula and Kamchatka derive from Makarov et al. (2022) and the data from Alaska from O'Hara et al. (2003).

PFHxS in *Rangifer* across the Arctic
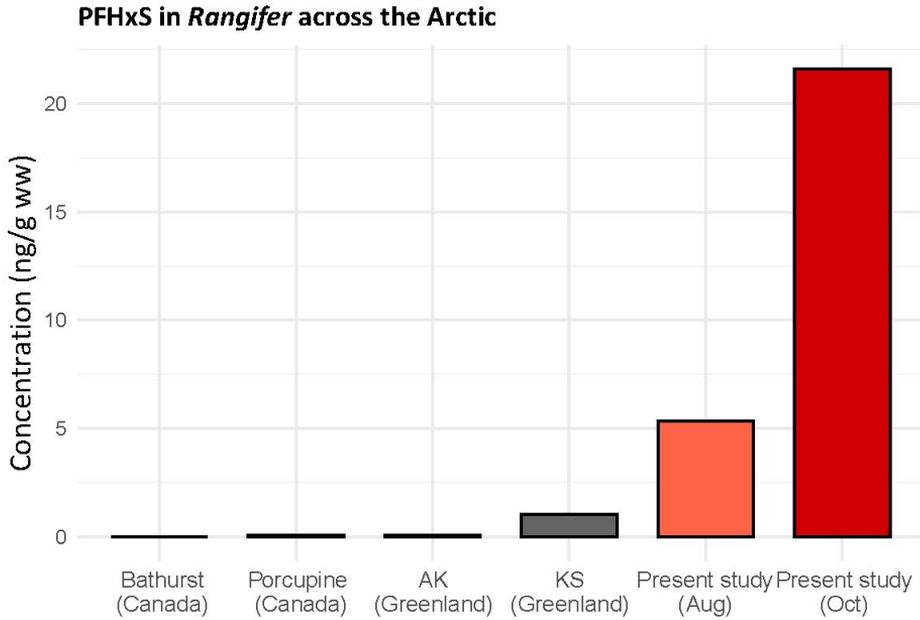
Figure S4. Hepatic PFHxS concentrations (ww) from various Rangifer subspecies across the Arctic. Concentrations from Bathurst, Porcupine, Akia-Maniitsoq (AK) and Kangerlussuaq-Sisimiut (KS) are all reported in Roos et al. (2021) and are here visualised in different shades of grey. Average concentrations from both animals culled in August and October in the present study are visualised in shades of red.
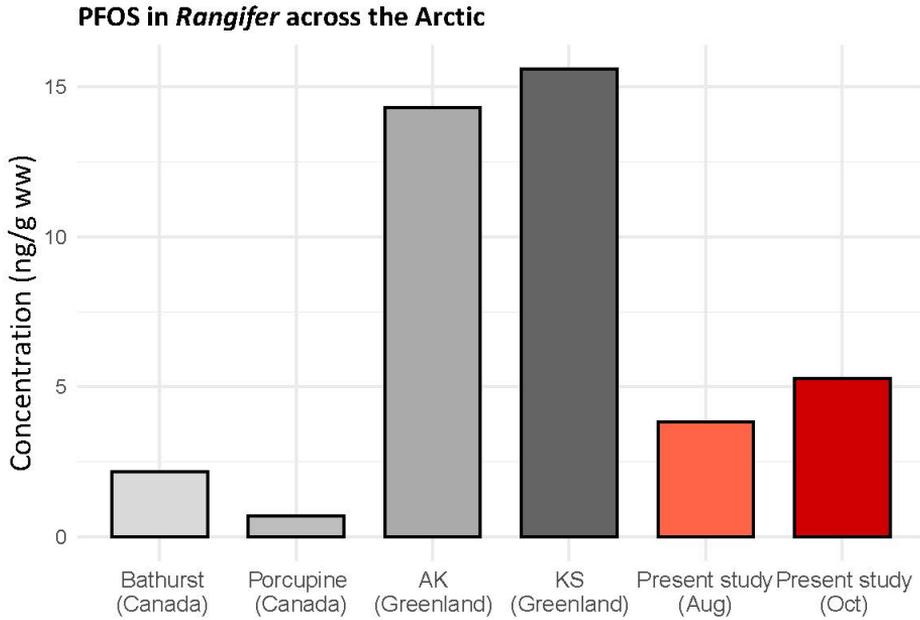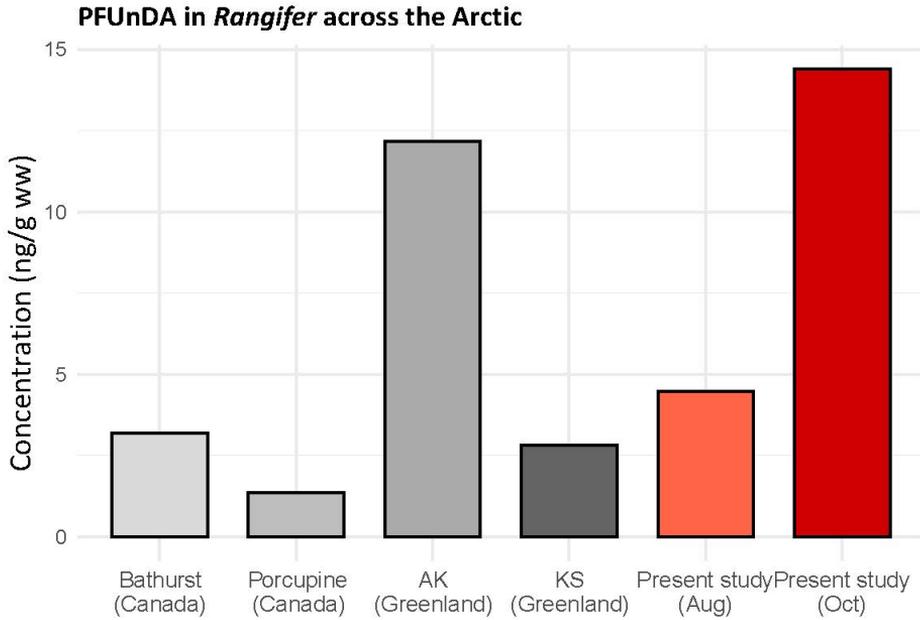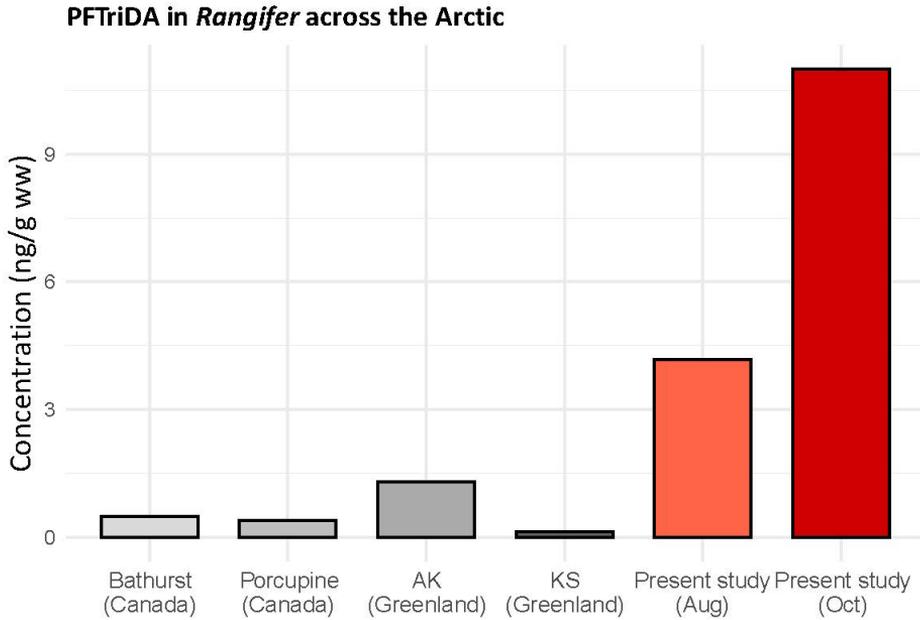
Figure S5. Hepatic PFOS concentrations (ww) from various Rangifer subspecies across the Arctic. Concentrations from Bathurst, Porcupine, Akia-Maniitsoq (AM) and Kangerlussuaq-Sisimiut (KS) are all reported in Roos et al. (2021) and are here visualised in different shades of grey. Average concentrations from both animals culled in August and October in the present study are visualised in shades of red.

Figure S6. Hepatic PFuNDA concentrations (ww) from various Rangifer subspecies across the Arctic. Concentrations from Bathurst, Porcupine, Akia-Maniitsoq (AK) and Kangerlussuaq-Sisimiut (KS) are all reported in Roos et al. (2021) and are here visualised in different shades of grey. Average concentrations from both animals culled in August and October in the present study are visualised in shades of red.

Figure S7. Hepatic PFTriDA concentrations (ww) from various *Rangifer* subspecies across the Arctic. Concentrations from Bathurst, Porcupine, Akia-Maniitsoq (AK) and Kangerlussuaq-Sisimiut (KS) are all reported in Roos et al. (2021) and are here visualised in different shades of grey. Average concentrations from both animals culled in August and October in the present study are visualised in shades of red.

REFERENCES

Ali Hassan, A., Rylander, C., Brustad, M., Sandanger, TorkjelM., 2012. Level of selected toxic elements in meat, liver, tallow and bone marrow of young semi-domesticated reindeer (*Rangifer tarandus tarandus L.*) from Northern Norway. International Journal of Circumpolar Health 71, 18187. https://doi.org/10.3402/ijch.v71i0.18187

Gamberg, M., Cuyler, C., Wang, X., 2016. Contaminants in two West Greenland caribou populations. Sci. Total Environ. 554–555, 329–336. https://doi.org/10.1016/j.scitotenv.2016.02.154

Institute of Nuclear Chemistry and Technology, 2010. Oriental Basma Tobacco Leaves (INCT-OBTL-5): Polish certified reference material for multielement trace analysis.

Institute of Nuclear Chemistry and Technology, & Gdańsk University of Technology., (n.d.). MODAS-3 Herring Tissue (M-3 HerTis): Polish certified reference material for multielement trace analysis with information values for selected radionuclides and some PCBs and PAHs.

Makarov, D.A., Ovcharenko, V.V., Nebera, E.A., Kozhushkevich, A.I., Shelepchikov, A.A., Turbabina, K.A., Kalantaenko, A.M., Bardyugov, N.S., Gergel, M.A., 2022. Geographical distribution of dioxins, cadmium, and mercury concentrations in reindeer liver, kidneys, and muscle in the Russian Far North. Environ. Sci. Pollut. Res. 29, 12176–12187. https://doi.org/10.1007/s11356-021-16310-2

O'Hara, T.M., George, J.C., Blake, J., Burek, K., Carroll, G., Dau, J., Bennett, L., McCoy, C.P., Gerard, P., Woshner, V., 2003. Investigation of Heavy Metals in a Large Mortality Event in Caribou of Northern Alaska. ARCTIC 56, 125–135. https://doi.org/10.14430/arctic608

Roos, A.M., Gamberg, M., Muir, D., Kärrman, A., Carlsson, P., Cuyler, C., Lind, Y., Bossi, R., Rigét, F., 2021. Perfluoroalkyl substances in circum-ArcticRangifer: caribou and reindeer. Environ. Sci. Pollut. Res. https://doi.org/10.1007/s11356-021-16729-7

Sait, S.T.L., Rinø, S.F., Gonzalez, S.V., Pastukhov, M.V., Poletaeva, V.I., Farkas, J., Jenssen, B.M., Ciesielski, T.M., Asimakopoulos, A.G., 2023. Occurrence and tissue distribution of 33 legacy and novel per- and polyfluoroalkyl substances (PFASs) in Baikal seals (Phoca sibirica). Sci. Total Environ. 889, 164096. https://doi.org/10.1016/j.scitotenv.2023.164096

Schuster, R.C., Gamberg, M., Dickson, C., Chan, H.M., 2011. Assessing risk of mercury exposure and nutritional benefits of consumption of caribou (Rangifer tarandus) in the Vuntut Gwitchin First Nation community of Old Crow, Yukon, Canada. Environ. Res. 111, 881–887. https://doi.org/10.1016/j.envres.2011.05.025

Paper IV

Low-Level Chronic Exposure to PFAS and Toxic Metals Induces Transcriptional Changes in Svalbard Reindeer

Malin Andersson Stavridis[1,2], Tove Petersson[1,3], Valentin Garreau[1,4],Øyvind Mikkelsen[1,5],

Vebjørn Veiberg[6], Helena C. Reinardy[1,7], Bjørn Munro Jenssen[1,2] & Tomasz Maciej

Ciesielski[1].

1. Department of Arctic Technology, University Centre in Svalbard (UNIS), PO Box 156 N-9171, Longyearbyen, Norway.

2. Department of Biology, Norwegian University of Science and Technology (NTNU), NO-7491, Trondheim, Norway.

3. Department of Ecoscience, Marine Mammal Research, Aarhus University, DK-4000, Roskilde, Denmark.

4. Department of Biology, Université Claude Bernard Lyon 1 (UCBL), FR-69622, Lyon, France.

5. Department of Chemistry, Norwegian University of Science and Technology (NTNU), NO-7491, Trondheim, Norway.

6. Land and Biodiversity, Norwegian Institute for Nature Research (NINA), NO-7485 Trondheim, Norway.

7. Scottish Association for Marine Science, Oban, PA37 1QA, United Kingdom.

Corresponding Author(s): Malin Andersson Stavridis: malins@unis.no and Tomasz Maciej Ciesielski: tomaszc@unis.no.

ABSTRACT

The Svalbard reindeer (*Rangifer tarandus platyrhynchus*) is chronically exposed to low levels

of various environmental contaminants, including toxic metals and per- and polyfluoroalkyl

substances (PFAS). However, no previous studies have assessed whether this contaminant

exposure may cause adverse biological effects in the reindeer. Therefore, the present study

examines the relationships between hepatic concentrations of mercury (Hg), cadmium (Cd),

lead (Pb), and seven PFAS compounds, and the expression of 17 genes involved in key

physiological functions in Svalbard reindeer.

Liver samples were collected from twelve female Svalbard reindeer, and hepatic mRNA was

extracted for downstream qPCR analysis using novel primers, specifically designed for this

25   study. Hepatic contaminant concentrations were previously reported for these samples

26   Andersson Stavridis et al. (2025a).

27   Using multivariate statistics, we found several genes to be downregulated with increasing

28   contaminant concentrations, primarily driven by C6 perfluorosulfonic acid (PFHxS), Cd, and

29   Hg. Notably, genes related to lipid homeostasis (e.g., *FABP3*, *PPARG*, and *THRA*) were the

30   most affected. Maintaining lipid homeostasis is vital for Arctic mammals like Svalbard

31   reindeer, which undergo large annual fluctuations in body mass and depend on lipid

32   regulation for normal physiological function. Consequently, our findings suggest that

33   contaminants may be influencing crucial physiological processes in the reindeer despite the

34   contaminant concentrations being below the current toxicity thresholds, indicating that

35   existing thresholds may underestimate the potential biological effects of contaminant

36   exposure in wildlife.

37   **Keywords**: Svalbard reindeer; Biomonitoring; qPCR; PFAS: Heavy metals

INTRODUCTION

Contaminant concentrations and effects have been extensively studied in Arctic marine

wildlife, but Arctic terrestrial species remain relatively understudied (Ankley et al. 2020;

Dietz et al. 2022). In addition, most existing studies on terrestrial wildlife focus on reporting

contaminant concentrations, rather than assessing contaminant-driven effects on biologically

relevant endpoints (Aas et al. 2014; Hallanger et al. 2019; Gamberg et al. 2020; Roos et al.

2021; Andersson Stavridis et al. 2025b). Understanding the resulting biological response of

contaminant exposure is crucial for assessing the long-term health risks to Arctic wildlife,

especially given the mixture of contaminants to which Arctic wildlife are exposed (Sonne et

al. 2021). Consequently, wildlife monitoring studies are vital, as they provide essential insight

into whether chronic, low-level contaminant exposure may result in observable effects in

wildlife (Ankley et al. 2020).

A key species in the Svalbard terrestrial ecosystem is the Svalbard reindeer. Contaminant

concentrations in the Svalbard reindeer were first reported in 1996 (Borch-Iohnsen et al.

1996) and have since been followed up in the 2020s (Roos et al. 2021; Andersson Stavridis et

al. 2025b), which included measurements of tissue concentrations of mercury (Hg), cadmium

(Cd), lead (Pb), and a range of per- and polyfluoroalkyl substances (PFAS). Based on the

findings of these studies, contaminant concentrations in Svalbard reindeer are below the

current toxicity thresholds for wildlife, which include hepatic concentrations of 4.2 µg/g (wet

weight, ww) for Hg (Dietz et al. 2022) or 655 ng/g (ww) for PFOS (Dietz et al. 2018), or renal

concentrations of 100 µg/g (ww) for Cd (Larison et al. 2000), and 15 µg/g (dw) for Pb (Ma

2011). Although these toxicity thresholds are derived from other species, they are commonly

applied to Arctic wildlife as no Arctic-specific thresholds currently exist (e.g., Dietz et al.

2022). Moreover, as such thresholds are typically based on the effects on a cellular or

62   individual level, the potential molecular responses to contaminant exposure below the toxicity

63   thresholds remain largely unexplored.

64   A common method to assess molecular effects is by measuring changes in mRNA expression.

65   This endpoint is particularly valuable in toxicological studies as it reflects changes at a low

66   level of biological organisation and may therefore be more sensitive in detecting effects even

67   before they result in, for instance, altered protein levels or enzymatic responses (Piña et al.

68   2007). Thus, it may serve as a useful early-response indicator for induced biological changes

69   (Ishibashi et al. 2008; Trego et al. 2019). Numerous studies have linked transcriptional

70   changes to contaminant exposure across Arctic wildlife, with sub-cellular effects on various

71   vital biological processes being reported (e.g., Lunardi et al. 2016; Routti et al. 2019;

72   Lühmann et al. 2020; Bjørneset et al. 2023).

73   To detect contaminant-induced changes at a low biological level in the Svalbard reindeer, the

74   present study investigated associations between hepatic contaminant concentrations and the

75   hepatic mRNA expression of 17 genes related to transcriptional regulation, xenobiotic

76   metabolism, oxidative stress, and lipid transport and metabolism. Although contaminant

77   concentrations in Svalbard reindeer generally were below established toxicity thresholds, we

78   observed contaminant-driven transcriptional changes in several key genes, especially in those

79   related to lipid homeostasis, indicating biological responses even at low exposure levels.

80   Together, these results underscore the importance of considering sublethal molecular effects

81   as early warning signals when evaluating contaminant risk in wildlife, both in the Arctic and

82   elsewhere.

MATERIALS AND METHODS

TISSUE SAMPLING

Liver and muscle samples were collected from twelve female Svalbard reindeer culled in Reindalen [77.9°N, 15.4°E] in August 2022, following the approval from the Governor of Svalbard (permission no. 16/01632–40 and 21/03815–4). Liver and muscle samples (approximately 0.5 × 1 cm) were placed in RNAlater to preserve RNA integrity for later extraction. However, due to poor RNA quality obtained from muscle samples, these were not included in downstream analysis. The age of the reindeer was determined by tooth eruption patterns or cementum annuli analysis (Veiberg et al. 2020) and ranged from 1 to 10 years old.

GENE EXPRESSION ANALYSIS

Viable hepatic RNA for downstream qPCR applications was successfully extracted from all Svalbard reindeer liver samples (n=12). Total RNA was isolated using the Qiagen RNeasy® Mini Plus Kit from samples stored in RNAlater, as described under Methods and Materials in Supplementary Information (SI). The extracted RNA was reverse transcribed into cDNA using the iScript cDNA Synthesis Kit, following the manufacturer's protocol.

A total of seventeen target genes were selected by reviewing wildlife toxicity studies that assessed the associations between mRNA expression and contaminant concentrations, with a focus on genes involved in key physiological functions (Das et al. 2017; King et al. 2023; Boyi et al. 2024). Four well-established reference genes were also included based on the stability of ruminant reference genes (Janovick-Guretzky et al. 2007; De Jonge et al. 2007; Lisowski et al. 2008). All genes are listed in Table 1.

104 **Table 1.** Target and reference [a] genes quantified in liver samples from Svalbard reindeer. Genes are categorised

105 based on physiological function into the following groups: transcriptional or nuclear regulation [b], xenobiotic

106 metabolism [c], lipid metabolism and transport [d], oxidative stress [e], and haemostatic proteins [f].

Abbreviation	Target gene	Relevant function
ACTB [a]	Actin Beta	Involved in cell structure, motility, and intracellular transport.
CYP1A2 [c]	Cytochrome P450 1A2	Involved in xenobiotic metabolism.
ECH1 [d]	Enoyl-CoA Hydratase 1	Involved in the β-oxidation of fatty acids.
FABP3 [d]	Fatty acid binding protein 3	Involved in fatty acid uptake, transport, and/or metabolism.
FADS1 [d]	Fatty acid desaturase 1	Regulates the unsaturation of polyunsaturated fatty acids.
FOS [b]	Fos proto-oncogene	Regulates cell proliferation, differentiation, and tumorigenesis.
FOXA2 [b, d]	Forkhead box A2	Involved in glucose and lipid metabolism, insulin secretion, and bile acid homeostasis.
GAPDH [a]	Glyceraldehyde-3-phosphate dehydrogenase	Involved in glycolysis.
GSR [e]	Glutathione-disulfide reductase	Involved in the cellular antioxidant system.
HAMP [f]	Hepcidin antimicrobial peptide	Regulates iron homeostasis.
JUN [b]	Jun proto-oncogene	Regulates cell proliferation, differentiation, and tumorigenesis.
MET1A [c]	Metallothionein 1A	Involved in the intracellular detoxification of toxic metals.
NR0B2 [b]	Nuclear receptor 0B2	Regulates the activity of cholesterol, bile, and fatty acids.
PPARG [b, d]	Peroxisome proliferator-activated receptor gamma	Regulates adipocyte differentiation.
POR [c]	Cytochrome P450 oxidoreductase	Electron donor protein for cytochrome P450 enzymes.
RPS9 [a]	Ribosomal protein S9	Ribosomal protein subunit.
SELENOP [e]	Selenoprotein P	Involved in selenium homeostasis and antioxidant defence.
SREBF1 [b, d]	Sterol regulatory element binding transcription factor 1	Involved in the sterol synthesis
TBP [a]	TATA-box binding protein	Part of the TFIID transcription factor, which initiates RNA transcription.
THRA [b]	Thyroid hormone receptor alpha	Receptor for thyroid hormone T3.
VLDLR [d]	Very low-density lipoprotein receptor	Involved in triglyceride metabolism.

107

108 As the Svalbard reindeer genome remains only partially annotated, with structural information

109 available but limited or no functional annotation, oligonucleotides for our selected target and

110 reference genes were designed from highly conserved regions of genes in species closely

111 related to the Svalbard reindeer. Conserved regions were determined by aligning mRNA

112 sequences from multiple mammals using the Clustal Omega Multiple Sequence Alignment

113   Tool and blasting these conserved regions against the Svalbard reindeer genome (NCBI

114   BLAST). Multiple primer pairs were generated for each gene using the primer design tool

115   from Integrated DNA Technologies (IDT), with preference for amplicon length (80-250bp),

116   G/C content (35-60%), similar annealing temperatures, and minimal secondary structures

117   (ΔG>-5).

118   Primers were tested on pooled liver cDNA (100 ng/µL) using a Mastercycler X50s PCR

119   thermocycler (Eppendorf, Hamburg, Germany), along with no-reverse transcriptase controls

120   (noRT) and no-template controls (noTC). Assays were run at various annealing temperatures

121   (50°C, 52°C, 54°C, 56°C, 58°C, and 60°C) to determine the best conditions for each primer

122   pair. Optimal temperatures were identified based on gel electrophoresis of endpoint PCR

123   products, with selection based on the presence of a single amplicon of the expected size.

124   Standard curves were produced for all primer pairs using the Applied Biosystems StepOne

125   Real-Time qPCR System instrument (Carlsbad, CA, USA) with a six-point dilution series

126   prepared from pooled liver cDNA. The primer pair with the highest R^2 value was selected for

127   downstream qPCR analysis of the Svalbard reindeer samples (Table S1). Primer specificity

128   was confirmed by observing a single, distinct peak in the generated melt curve. PCR products

129   of each primer were furthermore Sanger sequenced to confirm the primer specificity (Eurofins

130   Genomics, Germany). Additional information on the final primers used for subsequent qPCR

131   is found in Table 2.

132   **Table 2.** Forward and reverse primer sequences, amplicon length, and primer pair melting temperatures for all

133   the target and reference genes.

Gene		Sequence	Product length	Primer pair Tm
ACTB	F	CACACGGTGCCCATCTAC	245	58
	R	AGCTCGTAGCTCTTCTCCA		
CYP1A2	F	GCCATGAGTTTGTGGAATCCG	148	56
	R	TAGTGCTCCTGGACCGTTTTC		
ECH1	F	CCATCTTTATGAATCCTCCCTT	201	60
	R	AAATCCTGGAACATGAGCA		
FABP3	F	TGACCAAGCCTACCACAATC	143	56
	R	GGACTTGACCTTCCTGTCATC		
FADS1	F	GGGACAGACGAGTAACAGAAAG	208	56
	R	GCAGAGTGGTAGGAAGATGAAA		
FOS	F	CCCTCCAAGCGGAGACAGA	129	60
	R	GGATTTTGCAGGCAGGTCGG		
FOXA2	F	CCCTTCTCCATCAACAACCT	226	60
	R	AGTACACGCCCTGGTAGTA		
GADPH	F	AGTGAACAGATTCAGCCGCA	174	60
	R	CCGTTCTCTGCCTTGACTGT		
GSR	F	ACTTTGACAACACAGTTGCTATTC	129	60
	R	CGTGATGATTTGGTTCCTTTCTG		
HAMP	F	CACGACAGCTCACAGACCTC	194	60
	R	GCAGATGGGAAAGTGGGTGT		
JUN	F	CTTGAAAGCGCAGAACTCGG	119	56
	R	GCGTTAGCATGAGTTGGCAC		
MET1A	F	CTTTCACTCTCTCCTTTCTTCCC	200	60
	R	CACCAGGTCAGGTTGTATGAAT		
NR0B2	F	CCGCCCAACCATTCTGTAT	332	58
	R	CCAGCAGGATCTTCTTGAGTATG		
POR	F	GCAGAACACCTTCTACGACAT	90	60
	R	CCCTTGGTCATCAGCTTCTT		
PPARG	F	CCAGGTTTGCTGAATGTGAAG	200	60
	R	CTCATGTCCGTCTCCGTTT		
RSP9	F	ATCCCATCCTTCATCGTGCG	158	54
	R	GCTCAGTCCTCCTCTTCGTC		
SELENOP	F	CACCTGACAGTGTAAAGAAGAC	242	54
	R	AGTGTAGATTGACTTGAGGAGTA		
SREBF1	F	TGGGATCAAAGAGGAGCCAGT	136	60
	R	GTAACCCACAATCGGGGC		
THRA	F	AAGCTGATTGAGCAGAACCGA	212	60
	R	TCTGGCATGGAGACAATGGG		
TBP	F	TACACTCAGTGGTGGTAGGA	238	60
	R	CCAACTTGTCAACAGCAGTAAG		
VLDLR	F	GCCCAGGACATCATTGTCTATC	245	58
	R	CACATCCTCCATTCTCCACATC		

134   The hepatic gene expression of both target and reference genes was assessed using the

135   StepOne Real-Time qPCR System with SsoAdvanced Universal SYBR Green Supermix (Bio-

136   Rad, CA, USA). Samples were run in duplicate, and controls (both noRT and noTC) were

137   added to each well plate. Detailed primer sequences, amplification efficiencies, and melt

138   curve data are provided in the Materials and Methods in the SI, and in Table S1. The relative

139   gene expression ($2^{-\Delta Cq}$) was calculated by subtracting the mean of the reference gene Cq

140   values from the target gene expression to normalise individual variability (Schmittgen and

141   Livak 2008).

142   **CONTAMINANT CONCENTRATIONS**

143   The hepatic contaminant concentrations used in the present study were previously reported by

144   Andersson Stavridis et al. (2025a). Of these contaminants, Hg, Cd, and Pb were included

145   along with seven PFAS (Table 3), specifically perfluoroalkyl sulfonic acids (PFSAs): PFHxS,

146   PFOS, PFDS, and PFECHS, as well as perfluoroalkyl carboxylic acids (PFCAs): PFUnDA,

147   PFTriDA, and PFTDA. These PFAS were included based on a detection rate above 50 % in

148   the liver samples (Andersson Stavridis et al. 2025a), their documented toxicity, and their well-

149   documented presence in the Arctic (Muir et al. 2019; Ankley et al. 2020; Mahoney et al.

150   2022).

151   For full methodological details on analytical methods, including procedures for quality

152   control and limits of detection, see Andersson Stavridis et al. (2025a). In brief, THg was

153   determined using a Direct Mercury Analyser (DMA-80 evo, Milestone, Italy), Cd and Pb

154   were analysed using inductively coupled plasma mass spectrometry (ICP-MS, 8800 Triple

155   Quadrupole, Agilent Technologies, USA), and the PFAS analysis was performed using an

156   Xevo TQ-XS Triple Quadrupole Mass Spectrometer coupled with an ACQUITY UPLC

157   system (Waters, Milford, MA, USA).

Table 3. Mean ± SD, median, minimum, and maximum concentrations of contaminants detected in more than

50% of Svalbard reindeer liver samples, as reported in Andersson Stavridis et al. (2025a).

	Concentration (ng/g wet weight)			
	Mean ± SD	Median	Min	Max
THg	11.6±5.27	11.1	5.71	25.6
Cd	192±84.2	178	113	423
Pb	52.5±37.8	43.2	14.2	127
PFHxS	5.35±4.10	3.28	1.23	10.6
PFOS	3.83±1.32	3.84	2.16	6.60
PFDS	0.36±0.42	0.20	0.10	1.20
PFECHS	0.19±0.10	0.18	0.11	0.39
PFUnDA	4.47±3.13	4.24	1.34	12.4
PFTriDA	4.17±3.17	3.45	1.59	12.8
PFTDA	0.40±0.22	0.40	0.09	0.80

DATA ANALYSIS

Gene expression data are reported as relative fold change ($2^{-\Delta Cq}$), where ΔCq is the difference

between the target gene expression and the average reference gene expression, normalising

for individual variation (Schmittgen and Livak 2008).

Relative patterns in contaminant concentrations and gene expression (Cq-values) were

visualized in Fig. 1, where each variable was standardised using z-scores, which ensures that

each variable has a mean of zero and a standard deviation of one, allowing comparison of

relative patterns rather than absolute magnitudes across each variable. Statistical analyses

were performed using RStudio version 4.3.0 (R Core Team 2024). The relationship between

gene expression and explanatory variables (age, body mass, and contaminant concentrations)

was initially explored using Spearman rank correlations.

Subsequently, the associations between variables were analysed using orthogonal partial least

squares (O-PLS), a multivariate statistical method designed to handle datasets with few

observations and collinear predictors (Trygg and Wold 2002). Missing contaminant

concentrations were replaced by either a random value between 0 and half the LOD (for

175   concentrations <LOD) or by the mean of the respective variable (for values missing due to

176   analytical errors).

177   O-PLS model performance was assessed using explained variance (R²Y), predictive ability

178   (Q²Y), and permutation-derived p-values. Models with R²Y values >0.7, Q²Y>0.4, and p

179   ≤0.05 were considered highly robust (Sørmo et al. 2011). Variable importance in projection

180   (VIP) scores were used to assess the contribution of each explanatory variable, with VIP

181   scores greater than 1 considered indicative of a strong influence on the response variable.

182   Models were initially optimised by removing variables with VIP < 0.5. Additional variables

183   with the lowest VIP scores were sequentially removed to improve model fit until a significant

184   model with the highest possible R²Y and Q²Y scores was achieved (Ciesielski et al. 2017).

185   Each model was additionally assessed using a cross-validated ANOVA (CV-ANOVA) to

186   further confirm its significance.

187   For significant models, bootstrap resampling (with 10,000 iterations) was used to estimate the

188   variation within the regression coefficients and calculate standard deviations for each

189   explanatory variable (Ciesielski et al. 2017).

RESULTS

191   Hepatic mRNA expression levels were successfully quantified for all target genes in all

192   samples. The mRNA expression varied distinctly among individuals, with the largest

193   differences observed in genes *FABP3*, *MET1A,* and *NR0B2*, indicating relatively large

194   individual variation in gene expression (Table 4). Similarly, a relatively large variation in

195   hepatic contaminant concentrations was reported by Andersson Stavridis et al. (2025a) in

196   these samples, with several PFAS occurring in concentrations up to ten times higher in the

197   most contaminated individual, compared to the least contaminated individual (Table 3).

198 Overall, no consistent patterns in co-variation between contaminants and gene expression are

199 apparent (Fig. 1). However, certain individuals with higher-than-average contaminant

200 concentrations also show lower-than-average gene expression levels (e.g., ID 12 and 8), and

201 vice versa in individuals with relatively low contaminant levels (e.g., ID 6).

202 **Table 4.** Minimum and maximum fold change ($2^{-\Delta Cq}$) values for the 17 target genes. For each gene, the largest

203 difference between individuals was determined by dividing the maximum by the minimum fold change among

204 samples (n=12).

Gene	Min	Max	Largest fold change between individuals
FOS [b]	5.06×10^{-3}	3.43×10^{-2}	6.77
JUN [b]	7.18×10^{-2}	3.09×10^{-1}	4.30
NR0B2 [b]	1.34×10^{-5}	2.27×10^{-4}	16.9
THRA [b]	2.81×10^{-4}	1.41×10^{-3}	5.00
CYP1A2 [c]	1.62×10^{-2}	5.55×10^{-2}	3.43
POR [c]	1.38×10^{-2}	1.54×10^{-1}	11.2
MET1A [c]	4.10×10^{-4}	9.34×10^{-3}	22.8
FOXA2 [b, d]	1.07×10^{-4}	5.77×10^{-4}	5.38
PPARG [b, d]	3.26×10^{-5}	3.01×10^{-4}	9.25
SREBF1 [b, d]	1.01×10^{-4}	6.41×10^{-4}	6.38
ECH1 [d]	5.35×10^{-2}	2.74×10^{-1}	5.12
FABP3 [d]	1.29×10^{-4}	5.00×10^{-3}	38.7
FADS1 [d]	1.58×10^{-2}	1.27×10^{-1}	8.08
VLDLR [d]	1.99×10^{-4}	1.17×10^{-3}	5.87
GSR [e]	5.20×10^{-3}	2.58×10^{-2}	4.96
SELENOP [e]	2.17×10^{0}	1.42×10^{1}	6.55
HAMP [f]	4.01×10^{-2}	3.34×10^{-1}	8.35

[b] transcriptional or nuclear regulation, [c] xenobiotic metabolism,
[d] lipid metabolism and transport, [e] oxidative stress, and [f]
haemostatic proteins.

205

206
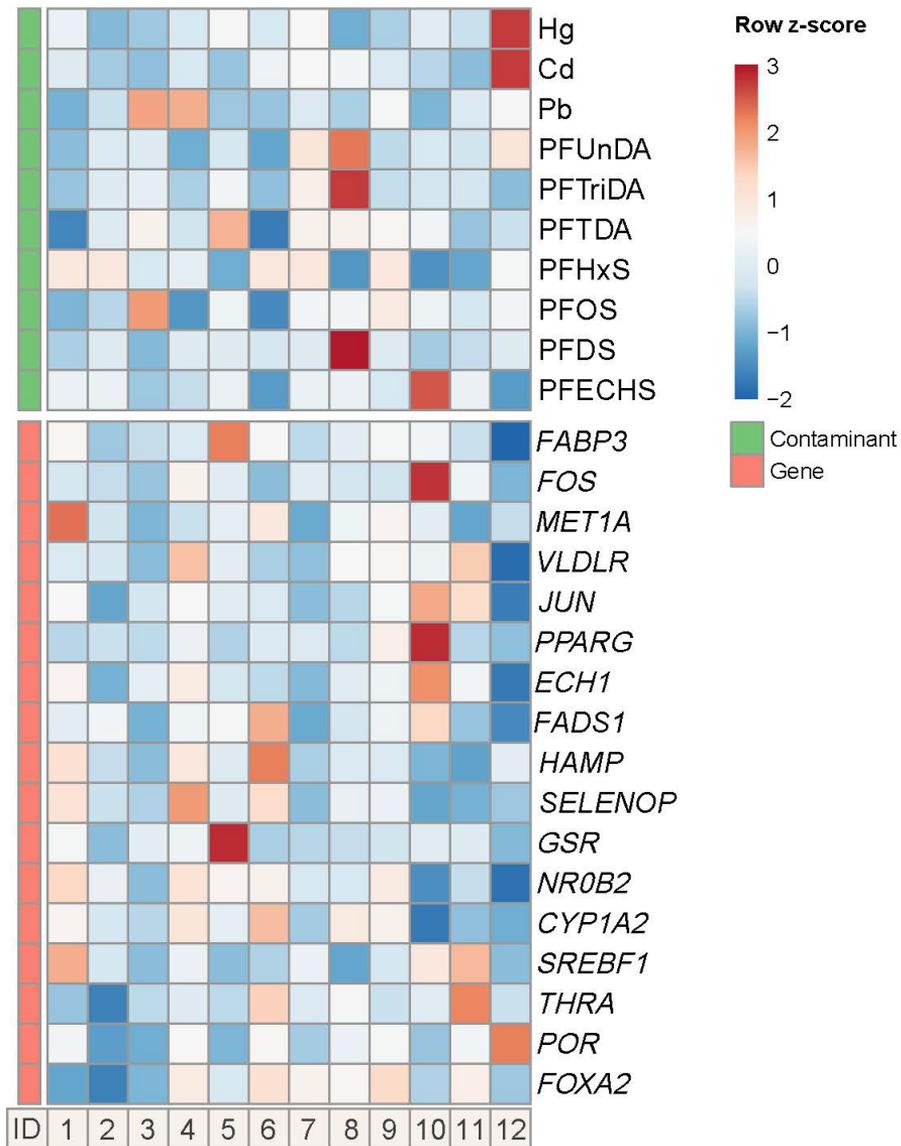
Figure 1. Relative patterns in contaminant concentrations and gene expression ($2^{-\Delta Cq}$ values) across individuals

(ID 1-12) of Svalbard reindeer (n=12). As the data was centered and scaled for each variable (z-score), the

heatmap shading is comparable across individuals for each variable, but not across variables. A blue shade

indicates values lower than the variable mean, white values indicate near the mean, while red values are higher

than the mean.

Spearman's rank correlation analysis revealed a few direct, significant relationships between

contaminant concentrations and mRNA expression levels. Pb concentrations were inversely

214   associated with *POR* and *FOXA2* (<0.05), while there were positive associations between

215   PFECHS concentrations and the expression levels of *PPARG*, *ECH1*, and *FADS1*. While the

216   expression of *HAMP* was positively related to Cd concentrations (p=0.031), it was negatively

217   associated with PFUnDA and PFTriDA concentrations (p<0.001). In addition, biometric

218   variables age and body mass were significantly positively related to several genes, such as

219   *NR0B2*, *SREBF1*, and *THRA*.

220   With contaminant concentrations having both positive and negative relationships with target

221   gene expression, and biometric variables having a significant influence on gene expression,

222   multivariate statistics were further used to better understand how the predictor variables

223   affected the transcriptional activity of the target genes.

224   Using O-PLS, significant explanatory models were obtained for the genes *JUN*, *FOXA2*,

225   *NR0B2*, *THRA*, *PPARG*, *SREBF1*, *FABP3*, and *CYP1A2* (Fig. 2). The explained variance

226   (R^2Y) ranged from 0.70 to 0.98, and predictive abilities (Q^2Y) ranged from 0.41 to 0.93

227   (Table 5). Body mass and age were strongly associated with the upregulation of genes related

228   to transcriptional and nuclear regulation (i.e., *FOXA2*, *NR0B2*, *THRA*, and *SREBF1*) and

229   xenobiotic metabolism (i.e., *CYP1A2*). In contrast, increasing body mass and age were

230   associated with a downregulation of *PPARG* gene expression. Among contaminants, Hg, Cd,

231   and PFHxS concentrations had the greatest influence on mRNA expression levels, which all

232   had strong negative relationships with *JUN*, *PPARG*, *FABP3*, and *CYP1A2* transcription

233   activity. In general, PFOS and long-chain PFCAs (PFUnDA, PFTriDA, and PFTDA) had a

234   comparatively small influence on gene expression, with VIP values ranging between 0.5-1.

235   Nonetheless, PFOS was included as a predictor variable in the models for genes *CYP1A2*,

236   *FOXA2*, *JUN*, *SREBF1*, and *THRA*, where increasing PFOS concentrations had an inverse

237   relationship with transcript levels. Long-chain PFCAs instead had both positive (e.g., FABP3,

238   JUN, PPARG) and negative (e.g., NR0B2, SREBF1) associations with gene expression.

239
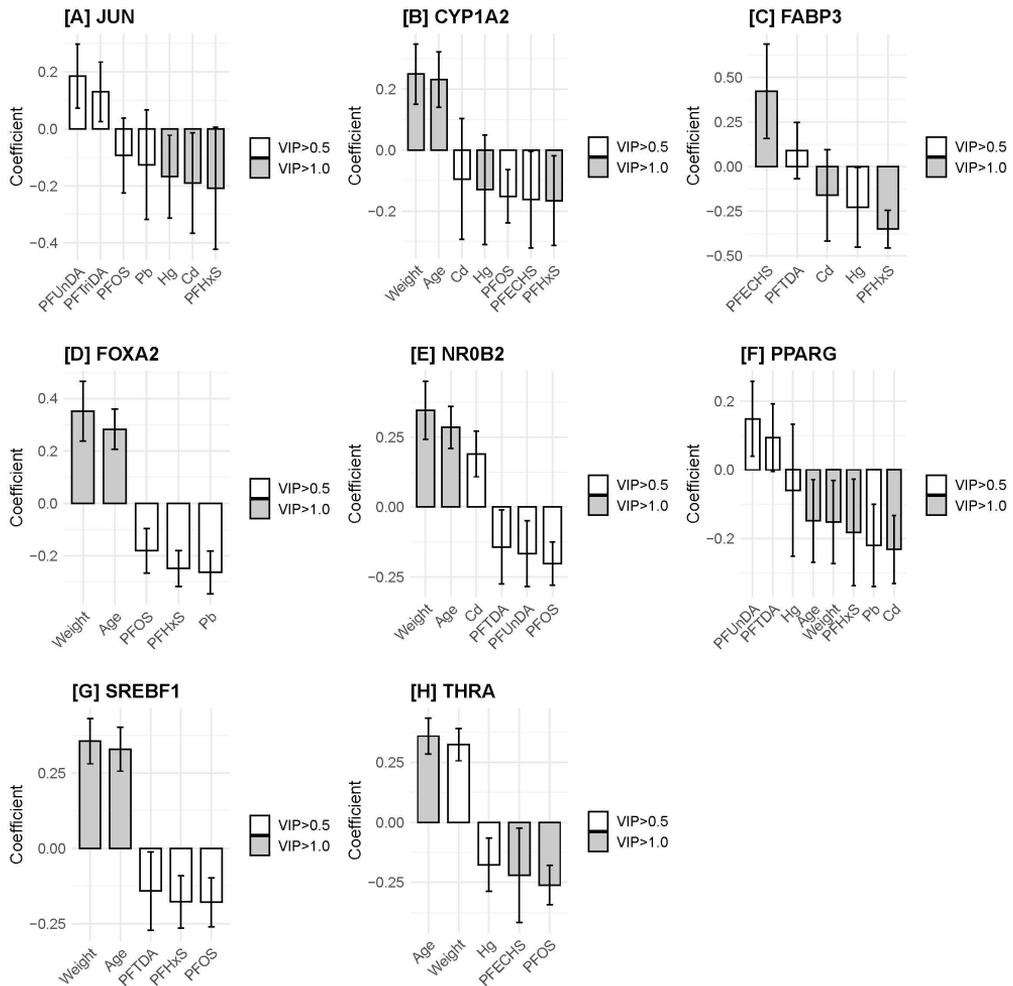
Figure 2. O-PLS regression coefficient for genes [A] *JUN*, [B] *CYP1A2*, [C] *FABP3*, [D] *FOXA2*, [E] *NR0B2*, [F] *PPARG*, [G] *SREBF1*, and [H] *THRA* with hepatic mRNA expression as a response to predictor variables: contaminant concentrations, age, and body mass (weight). The bars represent the contribution (coefficient ± standard deviation) of each predictor variable to the model. Predictor variables with VIP values greater than 1 are marked in grey.

245 **Table 5.** Target genes with significant O-PLS models. The models include the most influential predictor

246 variables (VIPs <0.5), which explain the majority of the variance in gene expression. The explained variance

247 (R^2Y) and predictive ability (Q^2Y) of the model. Predictor variables with variable importance in projection (VIP)

248 scores greater than 1 are specified in **bold**, along with the direction of the regression coefficient, indicating

249 whether the predictor is associated with gene upregulation (positive coefficient) or downregulation (negative

250 coefficient).

					Significant variables (VIPs>0.5, **VIPS >1**)	
Gene	(n)	p-value	R^2Y	Q^2Y	Downregulating	Upregulating
CYP1A2	12	<0.001	0.93	0.55	**PFHxS**, PFOS, PFECH, **Hg**, Cd	**Age**, **Body mass**
FABP3	10	<0.001	0.98	0.93	**PFHxS**, Hg, **Cd**	PFTDA, **PFECHS**
FOXA2	10	<0.001	0.90	0.76	PFHxS, PFOS, Pb	**Age**, **Body mass**
JUN	10	<0.001	0.79	0.58	**PFHxS**, PFOS, **Hg**, **Cd**, Pb	PFUnDA, PFTriDA
NR0B2	12	<0.001	0.72	0.41	PFTDA, PFUnDA, PFOS	**Age**, **Body mass**, Cd
PPARG	10	<0.001	0.85	0.44	**Age**, **Body mass**, **PFHxS**, Hg, **Cd**, Pb	PFUnDA, PFTriDA
SREBF1	10	<0.01	0.70	0.49	PFTDA, PFHxS, PFOS	**Age**, **Body mass**
THRA	12	<0.001	0.81	0.52	**PFOS**, **PFECHS**, Hg	**Age**, Body mass

DISCUSSION

252 The hepatic concentrations of contaminants in the Svalbard reindeer consistently fall far

253 below the established toxicity thresholds for wildlife (Andersson Stavridis et al. 2025a, b).

254 Still, the results of the present study suggest that a low-level chronic exposure to toxic metals

255 and PFAS may still cause biological effects in Svalbard reindeer.

256 While there is some degree of variation in individual contaminant levels and gene

257 transcription among the Svalbard reindeer (Tables 1 and 4), there are no apparent trends

258 between contaminant levels and gene transcription, as visualised in Fig. 1. This is further

259 supported by the results of the Spearman rank correlation analysis, where only a few

260 significant direct relationships between contaminant levels and transcriptional activity were

261 revealed. Instead, most of the significant models explaining the variation in gene expression

262   among individual reindeer include a combination of biometric variables and concentrations of

263   both metals and PFAS.

264   Although Pb and PFECHS were significantly associated with the mRNA expression of

265   several genes in the Spearman rank analysis (with *POR* and *FOXA2*, and *PPARG*, *ECH1*, and

266   *FADS1*, respectively), their individual contribution to explaining the variation in gene

267   expression in the O-PLS models for the same genes was small. A variable can show strong

268   individual correlation yet contribute less to the O-PLS model if it is collinear with other

269   variables or there is overlapping explanatory power between these predictor variables

270   (Eriksson et al. 2013). This may explain why Pb was included as an explanatory variable in

271   the O-PLS model for *FOXA2*, although with a VIP score <1, while PFECHS was not included

272   in the model for *PPARG* at all. Since other contaminants and biometric variables exhibited

273   higher VIP scores in the full models for these genes, contaminants such as PFHxS, PFOS, Hg,

274   and Cd in combination likely explain more of the variation in gene expression than Pb and

275   PFECHS do.

276   **INTERPRETING THE MULTIVARIATE MODELS**

277   **The impact of age and body mass**

278   The Spearman rank correlation analysis revealed significant positive relationships between

279   biometric variables: age and body mass, and the genes *NR0B2*, *SREBF1,* and *THRA*. These

280   relationships were further observed in the O-PLS models, where age and body mass were

281   among the strongest explanatory variables in this study. While age-related changes in gene

282   expression are well documented in other mammals (Kwekel et al. 2010; Glass et al. 2013),

283   body mass appears to be less frequently reported as a significant influence on gene

284   expression. However, as annual fluctuations in the body mass of Svalbard reindeer are

285   strongly related to the accumulation of fat in summer (Reimers 1984), the inclusion of body

286   mass as one of the most important explanatory variables (VIPs >1) for genes such as *FOXA2*,

287   *NR0B2, PPARG*, and *SREBF1* is biologically likely, given their roles in lipid metabolism.

288   **The impact of contaminants on lipid storage and metabolism**

289   Significant O-PLS models were primarily obtained for target genes related to lipid

290   homeostasis, and genes *FABP3* and *PPARG* were downregulated with increasing

291   concentrations of PFHxS, Cd, and Hg.

292   Svalbard reindeer experience large annual fluctuations in body mass (Albon et al. 2017), with

293   intense periods of fat accumulation in summer when forage is readily available, and heavy

294   dependence on stored fat in winter, when food availability is limited (Reimers 1984).

295   Maintaining lipid homeostasis is therefore critical, which involves the regulation of genes

296   such as *FABP3* and *PPARG*. Hence, contaminant-driven changes in the transcriptional

297   activity of these genes may disrupt vital processes in the reindeer. For example, a

298   downregulation of *PPARG* transcription levels may result in decreased PPARγ receptor

299   levels, which function as a main driver of adipogenesis (Tontonoz et al. 1994). As the

300   reindeer in the present study were culled in late summer, at their peak seasonal fattening when

301   lipid formation normally occurs, such a downregulation may impair their capacity to store

302   lipids. At the same time, hepatic accumulation of the same contaminants also appeared to

303   downregulate *FAPB3*, which functions as a lipid chaperone in the cell and enhances fatty acid

304   mobilisation and metabolism (Furuhashi and Hotamisligil 2008). Consequently, exposure to

305   these contaminants may affect pathways involved in lipid storage and lipid metabolism

306   simultaneously. We also observed a downregulation in *THRA* expression levels with

307   increasing exposure to PFOS and PFECHS. *THRA* is also involved in the regulation of fatty

308   acids (Sinha and Yen 2024), further indicating that these contaminants have an impact on

309   pathways involved in lipid homeostasis. Although the ultimate physiological outcome of these

310   changes in gene expression cannot be determined from the data presented in this study, our

18

311   findings suggest an overall disruption of various mechanisms involved in the lipid

312   homeostasis of the reindeer.

313   While heavy metals Hg, Cd, and Pb were consistently associated with a downregulation of

314   target gene expression (e.g., *FABP3*, *PPARG,* and *THRA*), we find conflicting results for

315   PFAS (Table 5). In general, long-chain PFCAs (PFUnDA, PFTriDA, and PFTDA) were

316   associated with both up- and downregulation of gene expression, while shorter-chained

317   PFSAs (PFHxS and PFOS) were consistently associated with a downregulation of target gene

318   expression. However, both chain length and functional group of PFAS can influence their

319   receptor affinity (Rosenmai et al. 2018; Houck et al. 2021; Dale et al. 2022), thereby affecting

320   their capacity to activate or inhibit upstream receptors, which in turn modulates gene

321   expression. Consequently, in addition to differences in structure-related modes of action, our

322   results may simply reflect the concentration-related differences between the subclasses of

323   PFAS. Identifying a clear mechanism behind these conflicting PFAS results or explaining

324   how Cd and Hg contribute to the observed effects, however, remains challenging. Existing

325   literature does not always provide possible mechanistic pathways, and often reports

326   inconsistencies in the direction of the effects (Lee et al. 2020; Dale et al. 2022), which is

327   attributed to species-specific responses, exposure conditions (e.g., sex), developmental stage,

328   and non-monotonic or mixture effects (Lee et al. 2020). Moreover, many receptors, such as

329   thyroid hormone receptors (e.g., THRα) or peroxisome proliferator-activated receptors (e.g.,

330   PPARγ), engage in crosstalk (Lu and Cheng 2010; Mullur et al. 2014), further confounding

331   the possible underlying mechanisms behind the observed results.

332   **Impacts of contaminants on other physiological processes**

333   In addition to genes involved in lipid homeostasis, we found significant O-PLS models for

334   genes involved in various other processes. These include early response gene *JUN*, which

335   supports cell proliferation and apoptosis (Dunn et al. 2002), and *CYP1A2*, which is involved

336   in drug and xenobiotic metabolism (Coleman 2010).

337   In the model for *CYP1A2*, several PFAS and metals were strongly associated with decreased

338   transcriptional activity, with PFHxS and Cd having the highest VIP scores. This is in line with

339   existing literature, where exposure to both PFAS and heavy metals has been linked to the

340   downregulation of *CYP1A2* gene expression (Franco et al. 2020; Alqahtani et al. 2023; Solan

341   et al. 2023) and the inhibition of CYP1A enzymatic activity (Solan and Lavado 2023; Ooka et

342   al. 2024). This process likely occurs through the interaction with ligands on the aryl

343   hydrocarbon receptor (AhR) (Alqahtani et al. 2023), which is the primary regulator of

344   *CYP1A2* transcription (Aleksunes and Klaassen 2012).

345   The variables included in the model for *JUN* are more difficult to explain. Chronic exposure

346   to metals such as Cd has previously been associated with a downregulation of *JUN* expression

347   (Sun et al. 2019), possibly explaining the inclusion of toxic metals in the O-PLS model.

348   Similar to the genes involved in lipid metabolism, we observed conflicting associations

349   between PFAS and *JUN* expression levels. While increased hepatic concentrations of PFHxS

350   and PFOS (PFSAs) were linked to a downregulation of *JUN* expression, increased PFUnDA

351   and PFTriDA (PFCAs) concentrations were linked to an upregulation. Again, this may reflect

352   structural differences among PFAS, and thus different modes of action. PFAS exposure has

353   previously been associated with an upregulation of *JUN* expression through oxidative stress-

354   modulated pathways (Sun et al. 2019), but existing literature generally offers a limited

355   mechanistic explanation for our findings.

356   Svalbard reindeer are exposed to a combination of both PFAS and toxic metals (Andersson

357   Stavridis et al. 2025a, b), both of which are known to induce oxidative stress (Balali-Mood et

358   al. 2021; Dale et al. 2022). In addition, recent studies indicate that Svalbard reindeer are also

359   exposed to PAHs (Pacyna-Kuchta et al. 2020; Na et al. 2021), which are one main

360   contaminant group that is metabolised by CYP enzymes such as CYP1A2 (Coleman 2010).

361   Genes such as *JUN*, which regulate cell regeneration or apoptosis in response to oxidative

362   damage (Meixner et al. 2010), or *CYP1A2*, which promotes xenobiotic detoxification, are

363   therefore vital for maintaining cellular defence. Consequently, a downregulation of these

364   genes may affect stress resilience and metabolic pathways in Svalbard reindeer.

365   **IMPLICATIONS AND FUTURE PERSPECTIVES**

366   Hepatic concentrations of PFHxS and Cd were among the highest contaminant concentrations

367   reported in these Svalbard reindeer (Andersson Stavridis et al. 2025a, b), which may explain

368   the consistent inclusion of PFHxS and Cd as highly influential variables in the O-PLS models

369   for many of the target genes. However, the Svalbard reindeer in the present study were culled

370   in August, whereas severalfold times higher contaminant concentrations of both metals (i.e.,

371   Hg, Cd, and Pb) and PFAS were reported for reindeer culled in October (Andersson Stavridis

372   et al. 2025a), when they are at their annual peak in body mass (Albon et al. 2017). Therefore,

373   further investigations into contaminant-modulated effects at higher contaminant body burdens

374   in autumn would be valuable to investigate. Given that the present study predominantly

375   indicates subcellular effects on pathways related to lipid homeostasis, further evaluation of

376   endpoints related to these biological processes, including assessments on higher biological

377   levels, would be especially valuable.

378   As the present study was limited to include the concentrations of metals Hg, Cd, Pb, and

379   seven PFAS, there may be contaminant-driven effects on our target genes that we cannot

380   explain with the current set of explanatory variables. Thus, it may be of interest to expand

381   potential future studies to include other groups of contaminants. For example, polychlorinated

382   biphenyls (PCBs) are known to regulate lipid homeostasis in other Arctic mammals (Tartu et

383   al. 2017; Routti et al. 2019). While concentrations of PCBs in Svalbard reindeer tissues were

384   undetectable thirty years ago (Kallenborn et al. 2011), PCBs are still present in the Svalbard

385 terrestrial environment (Aslam et al. 2019), and more current studies report detectable

386 concentrations of PCBs in Svalbard reindeer faeces (Zhu et al. 2015), indicating that Svalbard

387 reindeer are exposed to PCBs. Other contaminants of concern in the Arctic, such as

388 polybrominated diphenyl ethers (PBDEs) or polychlorinated naphthalenes (PCNs) (Sonne et

389 al. 2021), would also be of interest to include in future assessments, as no previous studies on

390 Svalbard reindeer report concentrations of these compounds.

391 Finally, our findings should be taken into consideration for future contaminant risk

392 assessments in wildlife. Toxicity thresholds for wildlife are still largely based on single-

393 contaminant exposure studies, yet wildlife across the globe is exposed to complex chemical

394 cocktails, which may lead to effects that are not predicted by the existing thresholds (Sonne et

395 al. 2021). Wildlife species with contaminant concentrations below the established toxicity

396 thresholds are often assumed to be at a low toxicological risk and are therefore rarely included

397 in studies that evaluate contaminant-driven biological effects. Instead, research on such

398 species rather reports contaminant concentrations in terms of human consumption risks (e.g.,

399 Jota Baptista et al. 2024; LaSharr et al. 2025) or assesses their use as indicators of change in

400 environmental contaminant concentrations (e.g., Gamberg et al. 2016; Andersson Stavridis et

401 al. 2024). Our results highlight that even species with low contaminant concentrations may

402 still experience mixture-driven contaminant effects, suggesting that current contaminant

403 thresholds may not fully capture the potential for biological effects in wildlife.

404 **CONCLUSIONS**

405 Although contaminant concentrations in Svalbard reindeer are well below established toxicity

406 thresholds, we observed transcriptional changes in genes involved in key physiological

407 processes, particularly those involved in lipid metabolism and transport. These findings

408 suggest that exposure to even low contaminant levels may, together, influence vital molecular

409 pathways for energy metabolism, which is crucial for Arctic mammals that rely on annual

cycles of fattening and fasting for their survival, like the Svalbard reindeer. Future studies

should assess contaminant-driven effects at higher levels of biological organisation, such as

protein levels or enzymatic activity, to better understand potential physiological consequences

of this exposure in the reindeer.

This study highlights that wildlife exposed to contaminant concentrations that are far below

the current toxicity thresholds may still experience contaminant-driven biological effects,

suggesting that the current toxicity thresholds may not fully reflect the potential for biological

effects in wildlife.

ACKNOWLEDGEMENTS

Declaration of generative AI and AI-assisted technologies in the writing process

The authors used generative AI (ChatGPT by OpenAI) to assist with language editing,

phrasing suggestions, and clarity improvements during the preparation of this manuscript. The

AI was not used to generate scientific content, perform data analysis, or interpret results. All

content was critically reviewed and edited by the authors, who take full responsibility for the

integrity and accuracy of the manuscript.

REFERENCES

431 **REFERENCES**

432 Aas CB, Fuglei E, Herzke D, et al (2014) Effect of Body Condition on Tissue Distribution of

433 Perfluoroalkyl Substances (PFASs) in Arctic Fox (*Vulpes lagopus*). Environ Sci Technol

434 48:11654–11661. https://doi.org/10.1021/es503147n

435 Albon SD, Irvine RJ, Halvorsen O, et al (2017) Contrasting effects of summer and winter

436 warming on body mass explain population dynamics in a food-limited Arctic herbivore. Glob

437 Change Biol 23:1374–1389. https://doi.org/10.1111/gcb.13435

438 Aleksunes LM, Klaassen CD (2012) Coordinated Regulation of Hepatic Phase I and II Drug-

439 Metabolizing Genes and Transporters using AhR-, CAR-, PXR-, PPARα-, and Nrf2-Null

440 Mice. Drug Metabolism and Disposition 40:1366–1379.

441 https://doi.org/10.1124/dmd.112.045112

442 Alqahtani MA, El-Ghiaty MA, El-Kadi AOS (2023) Mercury and methylmercury

443 differentially modulate hepatic cytochrome P450 1A1 and 1A2 in vivo and in vitro. J

444 Biochem & Molecular Tox 37:e23243. https://doi.org/10.1002/jbt.23243

445 Andersson Stavridis M, Peterson T, Kendir GD, et al (2025a) Seasonal Shift in Exposure and

446 Accumulation of PFAS and Heavy Metals in High Arctic Reindeer. In review with

447 Environmental Science and Technology

448 Andersson Stavridis M, Pollestad BM, Veiberg V, et al (2025b) Toxic metals and other

449 elements in Svalbard reindeer: Establishing baselines and assessing non-invasive sampling for

450 biomonitoring. Sci Total Environ 961:178226.

451 https://doi.org/10.1016/j.scitotenv.2024.178226

452 Andersson Stavridis M, Røed SB, Hansen BB, et al (2024) Tracing the footprints of Arctic

453 pollution: Spatial variations in toxic and essential elements in Svalbard reindeer (Rangifer

454   tarandus platyrhynchus) faeces. Sci Total Environ 906:167562.

455   https://doi.org/10.1016/j.scitotenv.2023.167562

456   Ankley GT, Cureton P, Hoke RA, et al (2020) Assessing the Ecological Risks of Per- and

457   Polyfluoroalkyl Substances: Current State-of-the Science and a Proposed Path Forward.

458   Environmental Toxicology and Chemistry 40:564–605. https://doi.org/10.1002/etc.4869

459   Aslam SN, Huber C, Asimakopoulos AG, et al (2019) Trace elements and polychlorinated

460   biphenyls (PCBs) in terrestrial compartments of Svalbard, Norwegian Arctic. Sci Total

461   Environ 685:1127–1138. https://doi.org/10.1016/j.scitotenv.2019.06.060

462   Balali-Mood M, Naseri K, Tahergorabi Z, et al (2021) Toxic Mechanisms of Five Heavy

463   Metals: Mercury, Lead, Chromium, Cadmium, and Arsenic. Front Pharmacol 12:643972.

464   https://doi.org/10.3389/fphar.2021.643972

465   Bjørneset J, Blévin P, Bjørnstad PM, et al (2023) Establishment of killer whale (Orcinus orca)

466   primary fibroblast cell cultures and their transcriptomic responses to pollutant exposure.

467   Environment International 174:107915. https://doi.org/10.1016/j.envint.2023.107915

468   Borch-Iohnsen B, Nilssen KJ, Norheim G (1996) Influence of season and diet on liver and

469   kidney content of essential elements and heavy metals in svalbard reindeer. Biol Trace Elem

470   Res 51:235–247. https://doi.org/10.1007/BF02784078

471   Boyi JO, Sonne C, Dietz R, et al (2024) Gene expression and trace elements in Greenlandic

472   ringed seals (Pusa hispida). Environ Res 244:117839.

473   https://doi.org/10.1016/j.envres.2023.117839

474   Ciesielski TM, Hansen IT, Bytingsvik J, et al (2017) Relationships between POPs, biometrics

475   and circulating steroids in male polar bears (Ursus maritimus) from Svalbard. Environ Pollut

476   230:598–608. https://doi.org/10.1016/j.envpol.2017.06.095

477   Coleman MD (2010) Human Drug Metabolism: An Introduction, 1st edn. Wiley

478   Dale K, Yadetie F, Horvli T, et al (2022) Single PFAS and PFAS mixtures affect nuclear

479   receptor- and oxidative stress-related pathways in precision-cut liver slices of Atlantic cod

480   (Gadus morhua). Science of The Total Environment 814:152732.

481   https://doi.org/10.1016/j.scitotenv.2021.152732

482   Das KP, Wood CR, Lin MT, et al (2017) Perfluoroalkyl acids-induced liver steatosis: Effects

483   on genes controlling lipid homeostasis. Toxicology 378:37–52.

484   https://doi.org/10.1016/j.tox.2016.12.007

485   De Jonge HJM, Fehrmann RSN, De Bont ESJM, et al (2007) Evidence Based Selection of

486   Housekeeping Genes. PLoS ONE 2:e898. https://doi.org/10.1371/journal.pone.0000898

487   Dietz R, Desforges J-P, Gustavson K, et al (2018) Immunologic, reproductive, and

488   carcinogenic risk assessment from POP exposure in East Greenland polar bears (Ursus

489   maritimus) during 1983–2013. Environment International 118:169–178.

490   https://doi.org/10.1016/j.envint.2018.05.020

491   Dietz R, Letcher RJ, Aars J, et al (2022) A risk assessment review of mercury exposure in

492   Arctic marine and terrestrial mammals. Sci Total Environ 154445.

493   https://doi.org/10.1016/j.scitotenv.2022.154445

494   Dunn C, Wiltshire C, MacLaren A, Gillespie DAF (2002) Molecular mechanism and

495   biological functions of c-Jun N-terminal kinase signalling via the c-Jun transcription factor.

496   Cellular Signalling 14:585–593. https://doi.org/10.1016/S0898-6568(01)00275-3

497   Eriksson L, Byrne T, Johansson E, et al (2013) Multi- and megavariate data analysis: basic

498   principles and applications, 3., rev. ed. MKS Umetrics, Malmö

499   Franco ME, Sutherland GE, Fernandez-Luna MT, Lavado R (2020) Altered expression and

500   activity of phase I and II biotransformation enzymes in human liver cells by

501   perfluorooctanoate (PFOA) and perfluorooctane sulfonate (PFOS). Toxicology 430:152339.

502   https://doi.org/10.1016/j.tox.2019.152339

503   Furuhashi M, Hotamisligil GS (2008) Fatty acid-binding proteins: role in metabolic diseases

504   and potential as drug targets. Nat Rev Drug Discov 7:489–503.

505   https://doi.org/10.1038/nrd2589

506   Gamberg M, Cuyler C, Wang X (2016) Contaminants in two West Greenland caribou

507   populations. Sci Total Environ 554–555:329–336.

508   https://doi.org/10.1016/j.scitotenv.2016.02.154

509   Gamberg M, Pratte I, Brammer J, et al (2020) Renal trace elements in barren-ground caribou

510   subpopulations: Temporal trends and differing effects of sex, age and season. Sci Total

511   Environ 724:138305. https://doi.org/10.1016/j.scitotenv.2020.138305

512   Glass D, Viñuela A, Davies MN, et al (2013) Gene expression changes with age in skin,

513   adipose tissue, blood and brain. Genome Biol 14:R75. https://doi.org/10.1186/gb-2013-14-7-

514   r75

515   Hallanger IG, Fuglei E, Yoccoz NG, et al (2019) Temporal trend of mercury in relation to

516   feeding habits and food availability in arctic foxes (Vulpes lagopus) from Svalbard, Norway.

517   Sci Total Environ 670:1125–1132. https://doi.org/10.1016/j.scitotenv.2019.03.239

518   Houck KA, Patlewicz G, Richard AM, et al (2021) Bioactivity profiling of per- and

519   polyfluoroalkyl substances (PFAS) identifies potential toxicity pathways related to molecular

520   structure. Toxicology 457:152789. https://doi.org/10.1016/j.tox.2021.152789

521   Ishibashi H, Iwata H, Kim E-Y, et al (2008) Contamination and Effects of Perfluorochemicals

522   in Baikal Seal (*Pusa sibirica*). 2. Molecular Characterization, Expression Level, and

523   Transcriptional Activation of Peroxisome Proliferator-Activated Receptor α. Environ Sci

524   Technol 42:2302–2308. https://doi.org/10.1021/es0720558

525   Janovick-Guretzky NA, Dann HM, Carlson DB, et al (2007) Housekeeping Gene Expression

526   in Bovine Liver is Affected by Physiological State, Feed Intake, and Dietary Treatment. JDS

527   90:2246–2252. https://doi.org/10.3168/jds.2006-640

528   Jota Baptista C, Seixas F, Gonzalo-Orden JM, et al (2024) Heavy metal and metalloid

529   concentrations in red deer (Cervus elaphus) and their human health implications from One

530   Health perspective. Environ Geochem Health 46:226. https://doi.org/10.1007/s10653-024-

531   01991-8

532   Kallenborn R, Ottesen RT, Gabrielsen GW, et al (2011) PCB ON SVALBARD REPORT

533   2011. The Governor of Svalbard

534   King MD, Su G, Crump D, et al (2023) Contaminant biomonitoring augmented with a qPCR

535   array indicates hepatic mRNA gene expression effects in wild-collected seabird embryos. Sci

536   Total Environ 904:166784. https://doi.org/10.1016/j.scitotenv.2023.166784

537   Kwekel JC, Desai VG, Moland CL, et al (2010) Age and sex dependent changes in liver gene

538   expression during the life cycle of the rat. BMC Genomics 11:675.

539   https://doi.org/10.1186/1471-2164-11-675

540   Larison JR, Likens GE, Fitzpatrick JW, Crock JG (2000) Cadmium toxicity among wildlife in

541   the Colorado Rocky Mountains. Nature 406:181–183. https://doi.org/10.1038/35018068

542   LaSharr K, Scher DP, Streets S, et al (2025) Exposure of free-ranging white-tailed deer

543   (Odocoileus virginianus) to per- and polyfluoroalkyl substances (PFAS) at PFAS-impacted

544   sites in Minnesota. Chemosphere 386:144642.

545   https://doi.org/10.1016/j.chemosphere.2025.144642

546   Lee JW, Choi K, Park K, et al (2020) Adverse effects of perfluoroalkyl acids on fish and other

547   aquatic organisms: A review. Science of The Total Environment 707:135334.

548   https://doi.org/10.1016/j.scitotenv.2019.135334

549   Lisowski P, Pierzchała M, Gościk J, et al (2008) Evaluation of reference genes for studies of

550   gene expression in the bovine liver, kidney, pituitary, and thyroid. J Appl Genet 49:367–372.

551   https://doi.org/10.1007/BF03195635

552   Lu C, Cheng S-Y (2010) Thyroid hormone receptors regulate adipogenesis and

553   carcinogenesis via crosstalk signaling with peroxisome proliferator-activated receptors.

554   Journal of Molecular Endocrinology 44:143–154. https://doi.org/10.1677/JME-09-0107

555   Lühmann K, Lille-Langøy R, Øygarden L, et al (2020) Environmental Pollutants Modulate

556   Transcriptional Activity of Nuclear Receptors of Whales *In Vitro*. Environ Sci Technol

557   54:5629–5639. https://doi.org/10.1021/acs.est.9b06952

558   Lunardi D, Abelli L, Panti C, et al (2016) Transcriptomic analysis of bottlenose dolphin

559   (Tursiops truncatus) skin biopsies to assess the effects of emerging contaminants. Marine

560   Environmental Research 114:74–79. https://doi.org/10.1016/j.marenvres.2016.01.002

561   Ma W-C (2011) Lead in Mammals. In: Environmental Contaminants in Biota, Interpreting

562   Tissue Concentrations, Second Edition. CRC Press, Taylor & Francis Group

563   Mahoney H, Xie Y, Brinkmann M, Giesy JP (2022) Next generation per- and poly-fluoroalkyl

564   substances: Status and trends, aquatic toxicity, and risk assessment. Eco-Environment &

565   Health 1:117–131. https://doi.org/10.1016/j.eehl.2022.05.002

566 Meixner A, Karreth F, Kenner L, et al (2010) Jun and JunD-dependent functions in cell

567 proliferation and stress response. Cell Death Differ 17:1409–1419.

568 https://doi.org/10.1038/cdd.2010.22

569 Muir D, Bossi R, Carlsson P, et al (2019) Levels and trends of poly- and perfluoroalkyl

570 substances in the Arctic environment – An update. Emerg Contam 5:240–271.

571 https://doi.org/10.1016/j.emcon.2019.06.002

572 Mullur R, Liu Y-Y, Brent GA (2014) Thyroid hormone regulation of metabolism. Physiol

573 Rev 94:355–382. https://doi.org/10.1152/physrev.00030.2013

574 Na G, Liang Y, Li R, et al (2021) Flux of Polynuclear Aromatic Compounds (PAHs) from the

575 Atmosphere and from Reindeer/Bird Feces to Arctic Soils in Ny-Ålesund (Svalbard). Arch

576 Environ Contam Toxicol 81:166–181. https://doi.org/10.1007/s00244-021-00851-1

577 Ooka M, Sakamuru S, Zhao J, et al (2024) Use of Tox21 screening data to profile PFAS

578 bioactivities on nuclear receptors, cellular stress pathways, and cytochrome p450 enzymes.

579 Journal of Hazardous Materials 473:134642. https://doi.org/10.1016/j.jhazmat.2024.134642

580 Pacyna-Kuchta AD, Wietrzyk-Pełka P, Węgrzyn MH, et al (2020) A screening of select toxic

581 and essential elements and persistent organic pollutants in the fur of Svalbard reindeer.

582 Chemosphere 245:125458. https://doi.org/10.1016/j.chemosphere.2019.125458

583 Piña B, Casado M, Quirós L (2007) Analysis of gene expression as a new tool in

584 ecotoxicology and environmental monitoring. TrAC Trends in Analytical Chemistry

585 26:1145–1154. https://doi.org/10.1016/j.trac.2007.09.009

586 R Core Team (2024) R: A language and environment for statistical computing

587 Reimers E (1984) Body composition and population regulation of Svalbard reindeer. Ran

588 4:16. https://doi.org/10.7557/2.4.2.499

589   Roos AM, Gamberg M, Muir D, et al (2021) Perfluoroalkyl substances in circum-

590   ArcticRangifer: caribou and reindeer. Environ Sci Pollut Res. https://doi.org/10.1007/s11356-

591   021-16729-7

592   Rosenmai AK, Ahrens L, Le Godec T, et al (2018) Relationship between peroxisome

593   proliferator-activated receptor alpha activity and cellular concentration of 14 perfluoroalkyl

594   substances in HepG2 cells. J of Applied Toxicology 38:219–226.

595   https://doi.org/10.1002/jat.3515

596   Routti H, Berg MK, Lille-Langøy R, et al (2019) Environmental contaminants modulate the

597   transcriptional activity of polar bear (Ursus maritimus) and human peroxisome proliferator-

598   activated receptor alpha (PPARA). Sci Rep 9:6918. https://doi.org/10.1038/s41598-019-

599   43337-w

600   Schmittgen TD, Livak KJ (2008) Analyzing real-time PCR data by the comparative CT

601   method. Nat Protoc 3:1101–1108. https://doi.org/10.1038/nprot.2008.73

602   Solan ME, Lavado R (2023) Effects of short-chain per- and polyfluoroalkyl substances

603   (PFAS) on human cytochrome P450 (CYP450) enzymes and human hepatocytes: An in vitro

604   study. Current Research in Toxicology 5:100116. https://doi.org/10.1016/j.crtox.2023.100116

605   Solan ME, Schackmuth B, Bruce ED, et al (2023) Effects of short-chain per- and

606   polyfluoroalkyl substances (PFAS) on toxicologically relevant gene expression profiles in a

607   liver-on-a-chip model. Environmental Pollution 337:122610.

608   https://doi.org/10.1016/j.envpol.2023.122610

609   Sonne C, Dietz R, Jenssen BM, et al (2021) Emerging contaminants and biological effects in

610   Arctic wildlife. Trends Ecol Evol 36:421–429. https://doi.org/10.1016/j.tree.2021.01.007

611   Sørmo EG, Ciesielski TM, Øverjordet IB, et al (2011) Selenium Moderates Mercury Toxicity

612   in Free-Ranging Freshwater Fish. Environ Sci Technol 45:6561–6566.

613   https://doi.org/10.1021/es200478b

614   Sun P, Gu L, Luo J, et al (2019) ROS-mediated JNK pathway critically contributes to PFOS-

615   triggered apoptosis in SH-SY5Y cells. Neurotoxicology and Teratology 75:106821.

616   https://doi.org/10.1016/j.ntt.2019.106821

617   Tartu S, Lille-Langøy R, Størseth TR, et al (2017) Multiple-stressor effects in an apex

618   predator: combined influence of pollutants and sea ice decline on lipid metabolism in polar

619   bears. Sci Rep 7:16487. https://doi.org/10.1038/s41598-017-16820-5

620   Tontonoz P, Graves RA, Budavari AI, et al (1994) Adipocyte-specific transcription factor

621   ARF6 is a heterodimeric complex of two nuclear hormone receptors, PPAR7 and RXRa. Nucl

622   Acids Res 22:5628–5634. https://doi.org/10.1093/nar/22.25.5628

623   Trego ML, Hoh E, Whitehead A, et al (2019) Contaminant Exposure Linked to Cellular and

624   Endocrine Biomarkers in Southern California Bottlenose Dolphins. Environ Sci Technol

625   53:3811–3822. https://doi.org/10.1021/acs.est.8b06487

626   Trygg J, Wold S (2002) Orthogonal projections to latent structures (O-PLS). Journal of

627   Chemometrics 16:119–128. https://doi.org/10.1002/cem.695

628   Veiberg V, Nilsen EB, Rolandsen CM, et al (2020) The accuracy and precision of age

629   determination by dental cementum annuli in four northern cervids. Eur J Wildl Res 66:91.

630   https://doi.org/10.1007/s10344-020-01431-9

631   Zhu C, Li Y, Wang P, et al (2015) Polychlorinated biphenyls (PCBs) and polybrominated

632   biphenyl ethers (PBDEs) in environmental samples from Ny-Ålesund and London Island,

633   Svalbard, the Arctic. Chemosphere 126:40–46.

634   https://doi.org/10.1016/j.chemosphere.2015.01.043

635

Low-Level Chronic Exposure to PFAS and Toxic Metals Induces Transcriptional Changes in Svalbard Reindeer

Malin Andersson Stavridis[1,2], Tove Petersson[1,3], Valentin Garreau[1,4], Øyvind Mikkelsen[1,5],

Vebjørn Veiberg[6], Helena Reinardy[1,7], Bjørn Munro Jenssen[1,2] & Tomasz Maciej Ciesielski[1].

1. Department of Arctic Technology, University Centre in Svalbard (UNIS), PO Box 156 N-9171, Longyearbyen, Norway.

2. Department of Biology, Norwegian University of Science and Technology (NTNU), NO-7491, Trondheim, Norway.

3. Department of Ecoscience, Marine Mammal Research, Aarhus University, DK-4000, Roskilde, Denmark.

4. Department of Biology, Université Claude Bernard Lyon 1 (UCBL), FR-69622, Lyon, France.

5. Department of Chemistry, Norwegian University of Science and Technology (NTNU), NO-7491, Trondheim, Norway.

6. Land and Biodiversity, Norwegian Institute for Nature Research (NINA), NO-7485 Trondheim, Norway.

7. Scottish Association for Marine Science, Oban, PA37 1QA, United Kingdom.

Corresponding Author(s): Malin Andersson Stavridis: malins@unis.no and Tomasz Maciej Ciesielski: tomaszc@unis.no.

Supplementary Information

Table of Contents

SUPPLEMENTARY METODS AND MATERIALS

RNA EXTRACTION

Approximately 20-30 mg of starting material was subsampled from liver tissues stored in

RNAlater solution. The starting material was immediately placed into 2ml Eppendorf tubes

prepared with stainless steel beads (5mm, Qiagen) and 650µl of buffer lysate (RLT plus beta-

mercaptoethanol, β-ME). Homogenisation was performed using a TissueLyser for 2×2

minutes at 25Hz. This was followed by genomic DNA elimination, RNA isolation, and

cleanup per kit instructions (Qiagen RNeasy® Plus Mini Kit (LOT 175040590). The RNA

was eluted in 50µl of nuclease-free water and aliquoted into separate vials for RNA

quantification, quality checking and cDNA synthesis. The RNA was stored at -20°C.

RNA QUALITY CHECK

RNA (3µl) from each sample was run on a Liberty 120 high-speed Pharmacia Biotech

Electrophoresis EPS 600 to assess product lengths and band strength. The purity and yield of

RNA were also evaluated using a Nanodrop 2000 (with requirements of A260/A280>2.0 and

A260/A230 ~2) and an Agilent 2200 Tape Station (RIN close to 10) according to

manufacturer instructions.

cDNA SYNTHESIS

The extracted RNA was reverse-transcribed into cDNA in 20-µl reactions using the iScript

cDNA synthesis kit (Bio-Rad) according to the manufacturer's protocol. Approximately 1000

ng of liver RNA were transcribed, based on the nucleic acid concentrations determined by the

NanoDrop™ 2000. Liver cDNA was aliquoted as undiluted, 1:3, and 1:10 dilutions at final

concentrations of 1000, 333, and 100 ng/µl, respectively. The final cDNA was stored at -20°C

until further preparations.

GENE SELECTION

FASTA mRNA sequences from several mammals were aligned using the Clustal Omega

Multiple Sequence Alignment tool to identify the conserved regions of the gene. Highly

conserved sequences with a length exceeding 200 base pairs were chosen and blasted against the Svalbard reindeer genome using the NCBI blast tool (NCBI:txid3082113). The gene of interest was considered located in the Svalbard reindeer genome if the results from the BLAST gave a single plus/minus match in the two published haplotypes of the reindeer on NCBI (with E-values below 0.05, high identity scores (>90%), and minimal gaps).

PRIMER DESIGN

Primers were generated using the primer design tool of Integrated DNA Technologies (IDT). Following the recommendations by IDT, primers with low GC content (35−60%), similar annealing temperatures for forward and reverse primers (Tm range of maximum 3°C), and secondary structures (homodimers, heterodimers and hairpin structures) with a delta G value of maximum negative five were selected. The online OligoAnalyzer Tool by IDT was used to view the delta G value of any self-dimers, hairpins, and heterodimers. The designed primers were blasted against the Svalbard reindeer genome (using NCBI blast) to ensure specificity.

PRIMER TESTING AND OPTIMISATION

Dried oligos were resuspended in nuclease-free water at pH 7.0 to a recommended concentration of 100 µM. Stocks were diluted 1:10 to achieve a working concentration of 10 µM. Primers were tested using the Applied Biosystems StepOnePlus PCR instrument on a template of pooled extracted liver concentration of 100 ng/µl (diluted 1:10 from 1000 ng/µl), no reverse transcriptase controls (noRT) prepared for two random liver samples, and no template controls (noTC). Assays were run at a range of annealing temperatures (Tm) of 50, 54, 56, 58, and 60°C. Gel electrophoresis of endpoint PCR reactions confirmed that our primers amplified a single product of the target sequence size, and optimal temperatures were selected for each primer pair.

STANDARD CURVE PREPARATIONS

Standard curves were produced for all primer pairs using the Applied Biosystems StepOne™ Real-Time qPCR Systems instrument. A six-point dilution series was prepared from pooled liver cDNA (high dilution: 100, 50, 25, 12.5, 6.25, and 3.125, low dilution: 5, 2.5, 1.25, 0.625, 0.3125, and 0.15625). The Cq values were plotted against the cDNA dilution series, and the primer pair with the highest R^2 value was chosen for each gene ($R^2 < 0.95$). Each primer pair was evaluated using qPCR at its optimal annealing temperature. NoRTs were included to ensure the absence of genomic background contamination from the RNA extraction, and noTCs (nuclease-free water) were included to ensure the absence of primer dimer formation. A minimum of five cycles between the noRT controls and the sample with the highest Cq-value was required to consider the primer pair performance acceptable. The melt curves were evaluated to ensure clean product amplification (and a lack of product formation in the noTCs).

SANGER SEQUENCING

PCR products generated with the final primer pairs were cleaned using AMPure XP beads (Beckman Coulter), followed by two 80% ethanol washes and elution using ultrapure water. Sanger sequencing was performed by Eurofins Genomics Europe Sequencing GmbH. The sequencing results confirmed that the final primer pairs amplified the correct regions of cDNA.

qPCR ANALYSIS

The gene expression of all target and reference genes was assessed in all liver samples (cDNA concentration of 100 ng/µl) in duplicate. Both noRT and noTC (ultrapure water) controls were added to each well plate. The reaction volume was 10 µL. The operating cycling conditions were as follows: 15 seconds at 98°C, followed by 40 cycles of 15 seconds at 98°C (denaturation), and 60 seconds at the optimal primer temperature (annealing and extension). This was followed by a stepwise increase in temperature (0.3°C) and fluorescence

measurements every five seconds (from 65°C to 95°C) to generate melt curves. The qPCR readings were examined to ensure amplification and confirm that the Cq values fell within the linear range of the standard curve of the assay (King et al., 2023). The repeatability of the duplicates was generally high, and intra-sample cycle differences greater than 1 Cq were rerun to generate quadruplicates. The average of the reference genes used in the assay was subtracted from the sample Cq value to obtain the ΔCq. As this study lacks controls, the 2-ΔCq method was employed to normalise the expression of the genes of interest (King et al., 2023; Schmittgen and Livak, 2008).

Table S1. Svalbard reindeer hepatic gene expression qPCR performance for standard curves, and endpoint PCR checks for assay specificity.

Gene	qPCR standard curves		qPCR gene array		Endpoint PCR	
	Efficiency (%)	R^2	Melt curve check*	Mean intra assay SD (%)	Gel electrophoresis check**	Sanger sequencing check***
ACTB	91	0.984	Y	0.18	pass	-
FOS	87	0.998	Y	0.16	pass	match
JUN	101	0.997	Y	0.10	pass	match
CYP1A2	92	0.999	Y	0.06	pass	match
ECH1	74	0.992	Y	0.13	pass	-
FABP3	95	0.980	Y	0.16	pass	match
FABS1	102	0.981	Y	0.06	pass	-
FOXA2	75	0.996	Y	0.17	pass	match
GADPH	91	0.999	Y	0.11	pass	match
GSR	114	0.999	Y	0.13	pass	match
HAMP	116	0.997	Y	0.08	pass	match
MET1A	99	0.965	Y	0.15	inconclusive	match
NR0B2	106	0.976	Y	0.29	pass	inconclusive
POR	107	0.980	Y	0.14	pass	match
PPARG	112	0.974	Y	0.30	pass	match
RSP9	88	0.995	Y	0.08	pass	-
SELENOP	90	0.959	Y	0.30	pass	-
SREBF1	96	0.998	Y	0.15	pass	match
TBP	105	0.992	Y	0.11	pass	-
THRA	89	0.999	Y	0.20	pass	match
VLDLR	101	0.953	Y	0.18	pass	-
*a single peak in the dissociation curve **a single band in the correct amplicon size; if inconclusive, a sample was sent for Sanger sequencing. ***BLAST match						

U N I S
The University Centre in Svalbard

NTNU
Norwegian University of
Science and Technology