

# Fur lice in Arctic foxes in Svalbard

Prevalence, intensity and impacts on health status and demographics



## Fur lice in Arctic foxes in Svalbard

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The Norwegian Polar Institute, a directorate under the Ministry of Climate and the Environment, carries out scientific research, mapping and environmental monitoring in the Arctic and Antarctic. The institute provides professional and strategic advice to the Norwegian authorities regarding polar issues, represents Norway internationally in several fora and is the nation's environmental authority in the Antarctic.

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### **Cover Photo**

Arctic fox in the Hornsund area, April 2024, with fur lice signs on the neck and shoulders. Photo: Ingvild  $\emptyset$ yjordet

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### **Preface**

Bloodsucking fur lice were discovered for the first time on Arctic foxes in Svalbard in 2019. Within the same year, fur lice were also described on Arctic foxes in Canada, and it turned out to be a previously undescribed lice species for Arctic foxes. Tor Slettebø, an experienced taxidermist working with skinning of Arctic foxes for many years, discovered something that he had never seen before in Arctic foxes, lice, whilst he worked with the tanning process of the pelts from Svalbard. That was the start of this project where we combine veterinary medical methods and ecology of a wild population of Arctic foxes to improve our understanding of this parasite host system.

This project made it possible to collect all Arctic fox carcasses from the 2022-2023 harvest season in order to investigate the impact of fur lice, focussing on lice prevalence, abundance and distribution of fur lice, changes in fur quality and skin pathology, develop and establish methods and guidelines for evaluation of intensity of lice infestation, and test out a method for use of camera traps on Arctic fox den sites to provide a non-invasive method for monitoring lice infections.

Special thanks go to taxidermist Tor Slettebø, Kristiansand, for his vigilance and experience which led to the discovery of fur lice in Arctic foxes in Svalbard in 2019. Thanks to Rupert Krapp and the logistic department in Longyearbyen, Norwegian Polar Institute for collecting and storing Arctic fox carcasses, to all the Arctic fox recreational trappers in the 2022-2023 season for their willingness to contribute to the project, and to the professional trapper Tommy Sandal who contributed extra information on louse prevalence and biological data of Arctic foxes from Bellsund. Thanks also to all the Svalbard people who sent pictures of Arctic foxes with potential signs of fur lice, this is still helpful for the process to understand the development and pattern of lice infestations throughout the season.

Tromsø, March 27, 2025

Eva Fuglei Researcher and project leader

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# **Summary**

In November 2019, bloodsucking lice were identified in Arctic foxes in Svalbard for the first time, coinciding with similar findings in Nunavut, Canada. Genetic analysis revealed that lice from both regions were 100 % identical, suggesting the presence of a previously undescribed species distinct from dog lice. The origin of the lice remains uncertain, but historical evidence of lice in Canadian Arctic foxes from the 1990s supports the hypothesis that they may have been introduced to Svalbard by migrating Arctic foxes.

The prevalence of lice in Arctic foxes in Svalbard has increased since the 2019–2020 trapping season, initially affecting an estimated 10 % of the population and rising to 76 % in the 2021–2022 season. Morphological and molecular analyses, along with observations of abnormal fur loss patterns in infested foxes, highlight the impact of lice infestations. These species-specific ectoparasites, which are highly dependent on their hosts, cause pruritis and fur damage, and in severe cases, can lead to anaemia.

The study aimed to collect all Arctic fox carcasses trapped during the 2022–2023 season to investigate the impact of fur lice by documenting the prevalence, abundance, and distribution of lice, changes in fur quality and skin pathology, and develop and establish methods and protocols for the evaluation of the intensity of lice infestation. The project also conducted a pilot study to test camera traps on Arctic fox den sites to provide a non-invasive method for monitoring lice infections.

During the 2022–2023 trapping season, a total of 36 Arctic foxes were captured, with fur lice detected in 16 individuals, resulting in an overall prevalence of 41.7 %. This was a decline from the previous year's prevalence of 76 % but remains higher than earlier seasons. Lice were mainly found in the neck and shoulder areas but were distributed across the entire body in heavily infested foxes. Heavier lice burdens in the pelts were related to lower body weights. Infestations were associated with significant fur damage, including discoloration and shortened fur, with infected foxes showing these changes more frequently than uninfected ones. The total lice burden ranged from 0 to 10 984 per pelt, with an average of 3 161. Egg density was higher than other life stages (eggs, larvae and adults), and the highest lice concentrations were found in the neck and shoulders. Histopathological analysis of the skin showed chronic irritation caused by lice, though there was no evidence of inflammation or immune response. Hair follicles appeared healthy and undamaged and hair loss was attributed to mechanical damage from scratching and biting, driven by tactile irritation rather than hypersensitivity.

Camera traps at five breeding dens captured over 24 000 images, with 409 showing Arctic foxes. Signs of lice infestation were evident in 279 of these images, documenting infestation at all dens. Infestation severity developed throughout the season (from January–February) and peaked in late spring (May–June). Despite this, foxes with no sign of lice on the fur were also observed on the dens. The observed seasonal trend in louse infections is noteworthy. Although the trapped fox dataset did not show a clear correlation between infection intensity and trapping dates, the trapping ends in mid-March, well before the peak seasonal changes observed in May and June through camera trap evidence. Thus, prevalence and abundance estimates based on trapped foxes may underestimate the impact of lice on the population.

Based on these findings and the literature we hypothesize that the recent introduction of fur lice to Arctic foxes in Svalbard, a previously naïve population, may lead to a gradual development of partial immunity. Over time, younger naive individuals or older foxes with weakened immune systems may become more susceptible to heavy infestations compared to healthy adults with prior exposure to lice. Immunity could also explain why some uninfected foxes coexist with infected individuals at the same breeding dens despite close physical contact. Another aspect related to the seasonal molt is the finding that seasonal shedding of winter fur in spring in other species significantly reduces louse infection intensity. Similarly, the seasonal molt of Arctic foxes from dense winter fur to summer fur in May probably plays a role in reducing louse prevalence and abundance. Hairs with attached eggs are shed during molting, decreasing the overall infection burden. So far, we have no information about fur lice in Arctic foxes during summer. Continued monitoring is recommended to better understand the dynamics of lice infestations, their health impacts, and potential development of immunity in the Arctic fox population.



Figure 1 Arctic fox infested with fur lice in Kongsfjorden, Svalbard. Photo: Arnt Rennan / Norwegian Polar Institute

## 1 Introduction

### 1.1 Background

Lice were discovered in Arctic foxes (*Vulpes lagopus*) for the first time in Svalbard in November 2019. Within the same year (May 2019) lice were also observed and documented in Arctic foxes from the mainland of Nunavut, Canada (Buhler et al. 2021). These concurrent findings resulted in close cooperation between the Norwegian Polar Institute, the Norwegian Veterinary Institute in Tromsø and the research group in Canada. Adult lice from Arctic foxes from both Nunavut and Svalbard were analyzed using molecular methods and they were 100 % genetically identical and identified as bloodsucking lice (sub order *Anoplura*) (Buhler et al. 2023). The possibility that the bloodsucking lice found in Arctic foxes could be the same species as found in dogs (*Linognathus setosus*), was investigated by collecting and analyzing two adult *L. setosus* from a dog in Saskatoon, Canada and three adult *L. setosus* from a dog from Bodø, Norway. The lice from the two host species looked morphologically similar and *L. setosus* from both dogs were 99 % genetically similar. The lice from the Arctic foxes were significantly genetically different from domestic dog lice (Figure 3 ABC, Buhler et al. 2023). These analyses suggest that Arctic fox sucking lice from Nunavut and Svalbard represent a previously undescribed bloodsucking lice species (Figure 3, Buhler et al. 2023).

The origin of the sucking louse in Nunavut and Svalbard in 2019 has so far not been established, but one earlier observation from Canada needs to be mentioned. After discovering the lice in 2019, Buhler et al. (2021) searched the Canadian Wildlife Health Cooperative database and found one unpublished case of lice and abnormal fur loss from one male Arctic fox trapped in February 1997 on the mainland of Nunavut, Canada. It was noted that the fox had thousands of ectoparasites on the fur, that were determined to be sucking lice (*Anoplura*) by the Western College of Veterinary Medicine, Saskatchewan, Canada, however, unfortunately neither morphological nor molecular methods were used, nor were any lice saved for future analyses (Buhler et al. 2021). It was also noted that the trapper had seen only one additional case of lice over several decades with trapping. After the 1990's fur trapping decreased in that area, but it was also noted that abnormal fur loss "were not unknown to trappers prior to this period" (Buhler et al. 2021). The fact that lice have been observed in Arctic foxes in Canada back in the 1990's, makes it more likely that the lice were introduced to Svalbard by an Arctic fox migrating from Canada. This assumption is consistent with previous observations of dispersal and gene flow between the circumpolar Arctic fox populations (Noren et al. 2011, Fuglei & Tarroux 2019).

In Svalbard Arctic foxes have been hunted for their fur for several hundred years (Rossnes 1993). Today the hunt is carried out by a few trappers who overwinter in isolated stations and for recreational purposes by residents of the larger settlements of Spitsbergen. Most of the harvest occurs in central Spitsbergen, Nordenskiöld Land and the hunting season lasts from 1<sup>st</sup> November to 15<sup>th</sup> March (Figure 3 a and b). The Norwegian Polar Institute has, since 1997, collected carcasses from the trappers and organized the skinning of all foxes by a professional taxidermist. The carcasses provide important data on the demographics and health of this wild population of Arctic foxes. It was taxidermist Tor Slettebø in Naturexpo, Kristiansand, that first detected and reported finding lice in foxes trapped in Svalbard in 2019 (Figure 3, picture to the left). His observations have not been reported previously in old trapping diaries (Rossnes 1993), nor have lice been described or observed in Svalbard since the recommencement of fox's carcass collection in 1997. The annual changing louse infection prevalence

subsequent to the first detection in the 2019–2020 trapping season also suggests that these ectoparasites are a recent introduction to a naive population. We do not have the full overview of the prevalence for the first year of detection but roughly estimate that 10 % of foxes trapped (N=166) were infested. After the detection of fur lice, we started to investigate each pelt from the trapped foxes more carefully and during the next trapping season (2020–2021) the estimated prevalence was 12,5 % (N=24). In the 2021–2022 season (N=208) the prevalence increased dramatically with 76 % of the foxes infected (Figure 13).



Figure 2 The underside of the Arctic fox skin, during skinning, showing multiple black spots through the skin. These were found to be adult lice at the base of the fur on the opposite side of the skin. Wood shavings are used during skinning to improve grip. Photo: Anna Galina Henriksson / Norwegian Veterinary Institute / UiT The Arctic University of Norway

Durden & Musser (1994) list over 51 *Linognathus* species and include Arctic foxes amongst the principal hosts of the dog sucking louse, *Linognathus setosus*. However further information on the geographic origin of this louse specimen in Arctic foxes is not provided. More recently only mites, ticks and fleas have been reported in Arctic foxes (Audet et al. 2002, Buhler et al. 2020), prior to the emergence of photographic evidence of bloodsucking lice in Arctic fox fur in the 1990's in Canada (Buhler et al. 2021, 2023). There also seems to be very little scientific literature regarding lice in other fox species. A number of studies have identified a low prevalence of the chewing louse *Felicola vulpis* in red foxes (*Vulpes vulpes*) (Dwuznik et al. 2020, Eren et al. 2021, Hinaidy 1971) but nothing specific about bloodsucking lice or *Linognathus* sp. was traced in red or Arctic foxes. The detection of *L. setosus* in

foxes (and wild canids) is also reported in a number of books on veterinary parasitology (Deplazes et al. 2016, Eckert et al. 1992, Georgi & Georgi 1990, Soulsby 1982, Taylor et al. 2016).

The majority of information we have regarding the impact and lifecycle of lice comes from other host species. This group of ectoparasites are largely host-specific and entirely dependent on their hosts to survive, spending all stages of their life cycle in the deep layers of the coat (Light et al. 2010). The clinical impact of lice is related to the density of infection. Heavy infestations are called "pediculosis" and the clinical signs related to damage in the skin as a result of the feeding activity of the lice (Taylor et al. 2016). The saliva produced by lice during feeding can trigger an immune response which can lead to hypersensitivity reactions (Deplazes et al. 2016). Pruritis (itchiness) dominates the clinical picture with secondary damage to the fur and skin as a result of the host scratching, rubbing or chewing affected areas. This can result in crusting in the skin and patchy hairloss (alopecia) with or without secondary infections. Very heavy infestations of bloodsucking lice, especially in young animals, can result in anaemia (Taylor et al. 2016).

Adult L. setosus lice are wingless insects 1-3 mm in length with no eyes and a dorso-ventrally flattened body and a narrow head (compared to the first thoracic segment) (Deplazes et al. 2016). The lifecycle of anopluran lice starts with the production of eggs (also called nits). The L. setosus females lay 5-10 eggs daily during their 3-6 week lifespan (Eckert et al. 1992, Deplazes et al. 2016) cementing them to the base of a hair shaft. A larva hatches within 1-2 weeks, capable of sucking blood, which undergoes development through three larval stages to become an adult (an Imago). This takes a further 2-3 weeks. All lifecycle stages, except eggs, can suck blood, with lice on average consuming 0.1 mg blood per feed (Deplazes et al. 2016). Transmission of lice between hosts generally requires direct fur to fur contact (Georgi & Georgi 1990). However, it is reported that *Linognathus* sp. can survive off the host up to 4-7 days (Deplazes et al. 2016, Eckert et al. 1992) and that L. setosus eggs can hatch over 2-3 weeks in the environment, if kept warm enough, so transmission via bedding material can occur (Taylor et al. 2016). The transmission of lice between hosts is thought to occur relatively slowly, as after a single introduction of chewing lice in sheep it took around six months (22 weeks) before more than 80 % the naive herd were infected (Deplazes et al. 2016). Lice are most commonly found around the eyes, ears and front part of the back and neck in canines but in heavy infections it can be found over much larger areas (Deplazes et al. 2016). Seasonal variations in bloodsucking louse prevalence are seen in some host species, like increased winter prevalence in cattle, but not in all host species. Lice seem to induce some degree of immunity, especially given that younger animals seem to be more commonly infected (Deplazes et al. 2016). It is thought that host immune responses impact the physiological and reproductive capabilities of lice leading to shorter lifespans and impaired egg hatching (Deplazes et al. 2016, James et al. 1998).

As part of the seasonal molt Arctic foxes start shedding their winter coat in the middle of May, with fur loss beginning on the legs, face, and sacral region (Figure 3A). They eventually lose their winter coat entirely during summer and appear brown or black depending on the color morph. This is the normal pattern of the seasonal change of the fur for this species (Buhler et al. 2021, 2023). Arctic foxes infested with sucking louse show a different and abnormal pattern of fur loss in the spring (Figure 3B-D). Arctic foxes from both Nunavut and Svalbard infested with sucking louse had similar patterns of such

abnormal fur loss occurring on the dorsum of the neck, shoulders and back in the spring (<u>Figure 3</u> B-D, and picture to the right).



Figure 3A) An Arctic fox displaying normal fur loss on the rump and limbs consistent with the first stages of molting in spring, B) a sedated Arctic fox with uncharacteristic fur loss across the neck and shoulders, C) another sedated Arctic fox in with uncharacteristic fur loss on the neck and shoulders, and (D) the same fox depicted in (B) before capture and sedation. All pictures from Canadian Arctic foxes from Buhler et al. (2021). Picture to the right shows an Arctic fox in Svalbard with uncharacteristic fur loss across the neck and shoulder in May 2021. Photo: Automatic camera and Eva Fuglei / Norwegian Polar Institute

There is an important management responsibility for the Arctic fox population in Svalbard which is knowledge-based and founded on monitoring and research (MOSJ, COAT). Information about the population size of the Arctic fox is used by the Governor of Svalbard for making decisions about sustainable harvesting. The Arctic fox breeding population is monitored by the Norwegian Polar Institute annually in two areas on Spitsbergen, one on West Spitsbergen and one on Nordenskiöld Land and the population has been relatively stable over time with some year-to-year variations (Eide et al. 2012, Fuglei et al. 2003, Nater et al. 2021, MOSJ, COAT). It is still not known to what extent sucking louse infections affect the general health status and demography of Arctic fox individuals, and the population dynamics over time.

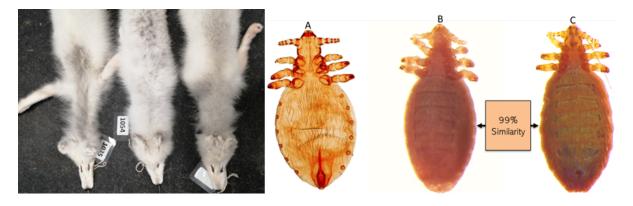


Figure 4 The picture on the left is of Arctic fox winter fur from Svalbard infested with lice (to the right and left) whilst the one in the middle is uninfected (Photo Tor Slettebø). On the right are the images of three Anopluran lice (ventral view): from a dog after clearing in lactophenol (A); from Arctic foxes in Svalbard (B) and Nunavut, Canada (C) both not cleared, from Buhler et al. 2023.

### 1.2 Objectives

The primary objective of this project was to document the prevalence, abundance and distribution of bloodsucking louse infections in Arctic foxes in Svalbard.

The project will ensure that a limited number of Arctic foxes are harvested and collected from a certain number of locations in Spitsbergen for the trapping season 2022-2023. The foxes will be collected and delivered to the project with the fur still on.

The two secondary objectives are to:

- (1) Secure the continuation of the monitoring of the bloodsucking louse infection in Arctic foxes in Svalbard for the 2022–2023 trapping season so as to document the prevalence of lice, increase the material to document changes in fur quality and skin pathology and describe the impact the lice are having on the fur quality and any associated dermatology.
- (2) Develop and establish methods and guidelines for qualitative evaluation of intensity of the sucking louse on the fur in trapped foxes and prevalence of heavy infections in phototrapped foxes at den sites.

### 2 Method

### 2.1. Study species and area

The Arctic fox is endemic to the Arctic tundra, and in Svalbard it is the apex terrestrial predator and scavenger with no natural enemies or competitors. It is abundant and functionally important because it affects both terrestrial and marine prey species. Due to lack of cyclically fluctuating small arctic rodents (like lemmings) the population dynamic is more stable compared to most other tundra ecosystems. The population dynamics have largely been driven by access to reindeer carcasses that are affected by rain-on-snow events (Eide et al. 2012, Fuglei et al. 2003, Hansen et al. 2012). Despite dramatic climate warming, the population dynamic is relatively stable and balanced by the exploitation of resources from different ecosystems, that buffers Arctic foxes against climate change (Nater et al. 2021).

Arctic foxes have been hunted and trapped in Svalbard for their highly insulative fur for more than a hundred years (Rossnes 1993). Since World War II, trapping has declined and now takes please as so-called recreational hunting, reserved exclusively for residents in Svalbard. <u>Trapping is allowed</u> between 1<sup>st</sup> November and 15<sup>th</sup> March in 25 restricted trapping terrains (<u>Figure 5</u>), and for some commercially operated trapping stations.

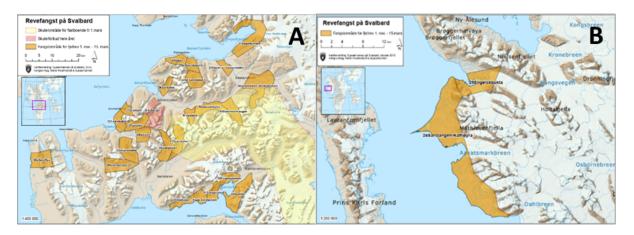


Figure 5 Maps showing the 25 Arctic fox (*Vulpes lagopus*) trapping terrains in Svalbard (from the Governor of Svalbard). Map A) showing the 23 terrains at Nordenskiöld Land and map B) the two terrains south of Ny-Ålesund.

Trapping is regulated by the Svalbard Environmental Protection Act, that states "All harvesting of species in Svalbard shall be done such that the natural productivity and diversity of species are preserved and that the composition and development of populations are not significantly altered" (Forskrift om høsting på Svalbard – Lovdata, Fuglei et al. 2013). Because the prevalence of fur lice in the Arctic fox population in Svalbard had such a dramatic increase over the trapping season 2021–2022 with 76 % infested and the uncertainty around how this could impact the population, we arranged a user involvement meeting before the start of the trapping season in 2022. This meeting consisted of a

representative from the hunting organization "Longyearbyen Jeger og Fisk forening" (LJFF), the Governor of Svalbard, the Norwegian Veterinary Institute in Tromsø (VI) and the Norwegian Polar Institute (NPI) to discuss the Arctic fox situation and the question whether trapping should be conducted in the 2022–2023 trapping season or not. The den monitoring data from the monitoring areas conducted by NPI from summer 2022, showed reproduction rates as above average, indicating no signs of any sudden drops in population. Based on the discussions, the user involvement meeting decided to arrange Arctic fox trapping for the 2022–2023 season, but with a restricted bag size and with a system to follow up the trapping terrains over the season to make sure that at least some foxes were collected from as many areas as possible. The Norwegian Polar Institute have a long-established system for collecting fox carcasses from the trappers by use of a Zarges box (Revekassa) that makes continuous follow up of every trapper and the number of foxes delivered from each trapping area possible. All the collected trapped foxes were transferred to a minus 80-degree freezer for 7 days, as a treatment to kill the eggs of the zoonotic parasite *Echinococcus multilocularis*, and after that all foxes were stored in Longyearbyen in minus 20-degree freezers until skinning could be carried out.

Thirty-six Arctic foxes were trapped during the 2022–2023 season. These were collected from trappers after a payment of a compensation for each carcass delivered to the project with the fur on. Such financial compensation is not something that is regularly paid but was introduced specifically for this specific funded project to ensure that foxes with poor quality fur were submitted.

# 2.2. Collection of Arctic fox carcasses in Longyearbyen 2022–2023 – prevalence of fur lice

Most of the recreational trappers do not have professional skills for taking off the fur from the trapped carcasses. Therefore, a professional taxidermist skinned all the foxes in Longyearbyen between  $23^{rd} - 30^{th}$  of April 2023. We systematically examined each fox without magnification, but under bright light conditions, for the presence of lice. This was done by two researchers and a student. Any changes in fur color, length and density were noted using a standardized registration form (Appendix 6.1). Standardized lateral and dorsoventral photographs (Figure 6) were taken of each fox regardless of infection status. All carcass and the furs got an individual ID number, and the weight and sex of each individual were recorded, a tooth was taken for aging as well as visually estimating (the age) and body condition (fat index) (Grue & Jensen 1976, Prestrud & Nilssen 1992).

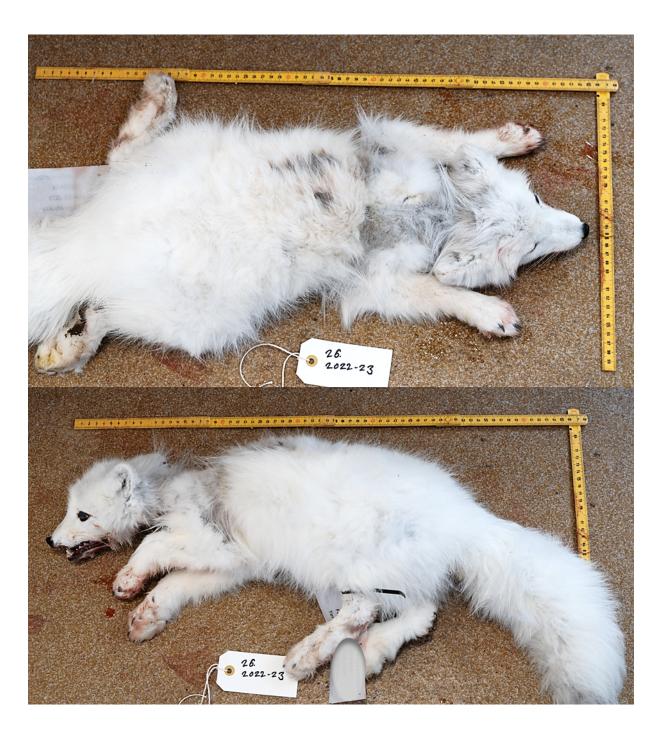


Figure 6 Standardized dorsoventral (top) and lateral (bottom) photographs were taken of each Arctic fox prior to skinning. The label shows the individual id number and trapping year of the fox. Photo: Anna Galina Henriksson / Norwegian Veterinary Institute / UiT The Arctic University of Norway

The fur was examined by using curved anatomical forceps to part the hair to visualize down to the skin along the length of the upper and lower eyelids as well as the upper lip across both sides of the nose. The examination then continued using fingers to part the hair and visualize the skin on the top of the head between the ears, continuing dorsally along the neck, shoulders, thorax, pelvis and tail (Figure 7).

If no lice were detected, then the examination was completed at this point, and the results recorded as no lice detected. However, if lice were detected then the lateral and ventral sides of the fox, including the limbs, were also examined for the presence of nits, larvae and/or adults. Areas that had black dust or discoloration in the fur close to the skin were examined extra carefully for the presence of lice or eggs. The dense fur made it sometimes difficult to visualize the skin, in which case the forceps were used to help part the fur.



Figure 7 Panel of images showing the investigation of Arctic foxes for lice: parting the fur over the shoulders down to the skin showing no evidence of infestation (a), evidence of infestation (b); and the head over the nose (c) and around the eyes with multiple adult lice visible between the forceps (d). Photo: Anna Galina Henriksson / Norwegian Veterinary Institute / UiT The Arctic University of Norway

An initial estimation of louse burden was based on the subjective evaluation of the number of lice seen and in how many locations they were detected.

- 1. Low intensity only a few individual lice seen in one or two locations
- 2. Moderate intensity two individuals seen per field of view, per location, in more than two locations
- 3. High intensity multiple lice (more than 4) seen in each field at multiple locations.

Seventeen frozen pelts: 15 positive; one uncertain; and one negative were transported directly from Longyearbyen, Svalbard to NVI Tromsø for further analysis on 30<sup>th</sup> April 2023 (import licence 202/23 Mattilsynet, journal no.: 703/23 Oslo Gardermoen airport border control Official Veterinarian). The

remaining uninfected pelts were returned to the trappers for their own use. All skinned fox carcasses were later transported in freezers from Longyearbyen to Tromsø where *post mortem* and further analyses at the Norwegian Veterinary Institute in Tromsø were conducted by researchers from VI and NPI.

# 2.3. Evaluation of the pelt in Tromsø – abundance and distribution of lice, changes of pelt quality and skin pathology

The 17 pelts from trapped Arctic foxes underwent further *post-mortem* analysis at the Norwegian Veterinary Institute in Tromsø in 12<sup>th</sup> -14<sup>th</sup> June 2023 for the presence of lice.

Each pelt was spread out, fur side up, and new photos taken for comparison with those taken prior to skinning. The length of the fur, both the insulating undercoat and longer guard hairs, were measured, using a 15 cm ruler, over the shoulders and in the lumbar region. A template was used to cut out standardized skin samples over the neck and shoulders and, when the pelt was large enough also from the pelvic lumbar region (21.5 x 21.5 cm), for further study into the thermal properties of the different furs with varying degrees of damage from lice (Henriksson 2023), as well as louse abundance counts. We also took smaller skin samples (5 x 5 cm) at seven standardized locations: between the ears, under the chin, on the thorax between the front legs, on the belly between the hind limbs and, across the pelvis cranial to the base of the tail, forelimb above the elbow and hindlimb above the stifle for louse abundance counts (Figure 8).

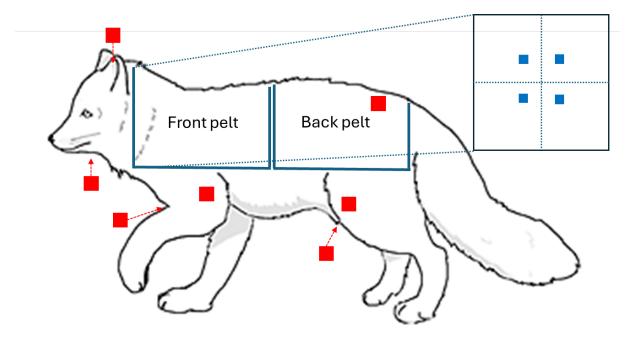


Figure 8 Showing the locations of the different skin samples for thermal testing (front pelt and back pelt if the skin was large enough to take both  $(21.5 \times 21.5 \text{ cm})$ ) as well as the locations of the smaller  $(5 \times 5 \text{ cm})$  in red;  $3 \times 3 \text{ cm}$  in blue) skin samples taken to estimate louse abundance. The sample taken over the lumbar region was dependent on the size of the pelt. If large enough thermal testing was carried out, if not then a smaller skin sample  $(5 \times 5 \text{ cm})$  was taken to estimate louse abundance only.

The larger pieces front and back pelt were refrozen and transported to UiT The Arctic University of Norway for thermal testing as described in Henriksson (2023). Four smaller (3 x 3 cm) skin samples were taken from these pelts after thermal testing was completed and analyzed for the louse abundance.

The louse abundance on each of the skin samples was estimated by placing the skin samples fur side down into a graded square petri dish (36 (8.8 x 8.8 mm) grided squares) containing 20 mL of 10 % potassium hydroxide (KOH). The samples were fully submerged in the KOH to remove any trapped air from the fur and left for 24 hours in a fume cupboard at room temperature (20 °C) to ensure that the fur and superficial surface of the skin had dissolved. The samples were agitated by swirling once during each 24-hour period. The skin was then removed and placed right side up (skin surface facing up) on the inside of the lid of the squared dish whilst the contents of the liquid were examined using a stereomicroscope at 10 x magnification and the number of eggs (with larva in them), larvae and adults counted. The skin sample was also examined under the stereomicroscope using overhead lighting in the stereomicroscope and any lice and eggs remaining attached to the skin also counted. The louse burden from the analysis of the front and back pelts was based on the count of the four subsamples taken from the larger pelt used for thermal testing.

We developed a protocol for a semi-qualitative scoring of the degree of lice infestation based on the photos of the pelts in addition to the initial visual estimation of louse burden during skinning. We visually graded the degree of fur loss and damage to estimate a local damage score which focused on the worst score visible on each pelt (Table 1, Figure 9 and Figure 10)

Table 1 The fur damage score evaluation table with scoring from 0 (no damage and in pristine condition) with increasing severity of damage up to 5 (extensive damage).

Fur damage score	0	1	2	3	4	5
Fur loss	No change	Slight thinning	Noticeable fur loss/thinning	Prominent fur loss	Fur less than half normal depth for winter coats	Fur less than half normal depth for winter coats, patchy alopecia
Fur damage	None	Very little damage	Hairs broken and thinning	Thinned undercoat, sheared appearance	Sheared and deteriorated	Ragged
Colour change	None	None to very faint	Yes – no longer snowy white	Yes - evident	Yes – highly noticeable	Yes – highly noticeable
Guard hairs	Normal length	Normal	Even but thinning	Missing or sparsely distributed, uneven coverage	Scarcely distributed or missing	Scarcely distributed or missing

Fur damage score	0	1	2	3	4	5
Insulation hairs	Dense and fluffy	Dense and fluffy	Thinning but still present	Less dense	Downy undercoat exposed, damage and reduced	Downy undercoat rough condition or missing
Skin visible under the fur	Not visible	Not visible	Not visible	Almost possible to see down to base undercoat	Skin is just visible	Skin exposed

The area of damage was also estimated given that the fur damage was not evenly distributed (<u>Figure 9</u>). This was done by estimating the area affected using photo editing software in Adobe photoshop (v.23.5.1.) using the analysis tools outlining the irregular shape to estimate the size of the area ( $cm^2$ ) impacted by the fur damage/loss.



Figure 9 The dotted black and white line marks the outline of the area of damaged fur and the program then provides an estimate of the total area within that outline. This fur has a local damage score of 4. Photo: Anna Galina Henriksson / Norwegian Veterinary Institute / UiT The Arctic University of Norway

The overall damage score was estimated by multiplying the local damage score and area affected together.

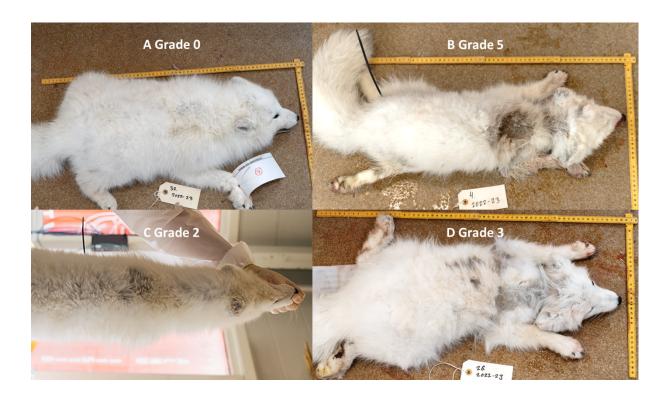


Figure 10 Arctic foxes with different levels of fur damage graded using the scale described in Table 1 from Grade 0 (A) with pristine fur coat to Grade 5 (B) with extensive fur loss, visible skin and ragged appearance. Grade 2 (C) shows slight thinning of the fur at the neck and some faint discoloration over the neck and shoulders whilst Grade 3 (D) shows more extensive fur damage, obviously different coat color over the neck and shoulders but no skin is visible. Photo: Anna Galina Henriksson / Norwegian Veterinary Institute / UiT The Arctic University of Norway

### 2.4. Histological investigation of the skin

Three skin and fur tissue samples were taken from each of the 17 Arctic fox furs. A biopsy was taken from the neck and nose of all the foxes whilst the third sample was either from the pelvic lumbar region ((N=10), or from the limbs (N=7), shoulder, forelimb or thigh). These were fixed in 4 % formalin and embedded in paraffin wax (FFPE) stained and examined with haematoxylin and eosin (H&E) according to standard histological techniques for routine histological examination (Bancroft & Layton 2019). Whole-slide images were obtained (Nano Zoomer S210, Hamamatsu Photonics).

# 2.5. Establishment of camera traps on Arctic fox breeding dens to evaluate lice on free living foxes

Camera traps at dens have become an international methodological standard in the monitoring of Arctic fox populations (Eide et al. 2012, Ehrich et al. 2017). As heavy lice infections appear to be clearly visible from camera trap photos (see <u>Figure 1</u>; Buhler et al. 2021, 2023), such monitoring may provide a non-invasive, internationally widely applicable method for monitoring lice infections. NPI have conducted camera trapping in their long-term annual den monitoring study of Arctic foxes and hence, photos from this camera trap based den monitoring during the winter 2024 were used to assess to what extent camera trap photos found similar fur loss/damage patterns as was seen in the *post mortem* foxes. Since the fur trapping ends on March 15 (Arctic fox fur trapping lasts from November 1<sup>st</sup> to March

15<sup>th</sup>), the camera trap method will provide data on potential signs of fur lice over a longer time scale, from January to June, than data from the trapped foxes.

Camera traps were placed out in Adventdalen and Sassendalen in Svalbard by use of snow mobiles on five different Arctic fox den localities between January 21<sup>st</sup> and 22<sup>nd</sup>, 2024. Breeding den localities are "not for public release" (unntatt offentlighet) and is therefore not made visible on any maps in the present report. The five automatic movement-triggered cameras (Reconyx PC850/PC800; Reconyx Inc., Holmen, WI, USA) were fixed on aluminum poles 30–60 cm above ground at approximately 2–8 m from den entrances in a position providing a good overview of the den. The cameras were set to be triggered by movements and programmed to use a motion sensor set to "high sensitivity" taking five pictures for each trigger on picture interval "rapid fire" and with no "quiet period". Memory cards and 12 internal lithium batteries were changed between April 30<sup>th</sup> and May 2<sup>nd</sup>, 2024. On all but one den, the cameras stood out until memory cards and batteries were changed again between June 23<sup>rd</sup> and June 27<sup>th</sup>, 2024. On the one den that had shorter time with the camera trap in place the camera stood out until May 2<sup>nd</sup>, 2024.

All pictures obtained from the cameras were visually evaluated by a single researcher for signs of fur lice on Arctic foxes, like abnormal patterns of fur loss, previously described by Buhler et al. (2021, 2023) (see Figure 1 and Figure 3). To evaluate the visual signs of fur lice from the pictures taken, we defined different classifications of sucking louse infestations when going through the pictures of foxes from each of the five dens. The classifications were based on subjective evaluations of each picture and distributed as follows: "no sign", "mild signs", "medium signs", "extensive signs" and "summer fur" (Appendix Table 1). The classification "summer fur" was included because it appeared to be more and more difficult to evaluate the lice classification when more and more of the winter fur disappeared, and the foxes turned into summer fur during its seasonal molt (Figure 3 A). Since the camera traps were placed on a breeding den, the same foxes were expected to be seen on the pictures over time. That made it possible to follow the development of louse infestation over time. Attempts were made to assess and record one fox per day when it was possible to separate the individuals. It must be noted that separating individuals was difficult specifically early in the season, when infestations were difficult to see on the winter fur, and late in the season (May-June), on foxes that were completely in summer fur. The results are expressed as the "total number of foxes with no sign of fur lice" and the "total number of foxes with signs of fur lice" after combining all the defined classifications per month for all five dens. Results were also expressed based on the defined classifications ("no sign", "mild signs", "medium signs", "extensive signs" and "summer fur") summed up for every month (January, February, March, April, May and June; see Appendix Table 1).

### 2.6. Statistical analyses

The data set regarding the initial examination during skinning and the laboratory analysis of the pelts was analyzed using JMP statistical software (14.0.0 SAS Institute Inc) to compare prevalence and abundance data by sex, age, body weight, body fat index and trapping location of the foxes using contingency and bivariate analysis. The two methods of evaluating louse burdens, the initial visual assessment and the laboratory quantification of louse abundance were compared using kappa analysis (Dohoo et al. 2003). A linear mixed model was also used to assess relationships between the fur damage score and the log transformed louse abundance (log(total lice/cm²)) using the lme4 package in R. A significance level of p<0.05 was selected for all analyses.

### 3 Results and discussion

#### 3.1. Prevalence of fur lice 2022–2023

A total of 36 Arctic foxes were trapped during the trapping season 2022–2023 (Figure 13) and foxes were taken in 14 of 25 trapping areas (Figure 12). Lice, and/or eggs, were detected in 16 of the 36 pelts during the initial evaluation prior to skinning (Table 2; Figure 11), albeit one pelt with an uncertain status. Initial evaluation of the louse burden gave slightly varying results than the final evaluation based on the total numbers of lice (Table 2). The uncertain individual and the negative control pelt had no lice whilst the remaining louse burden was evenly split with five individuals categorised at low, moderate and high based on total louse counts (Table 4). This gives an overall louse prevalence of 41.7 % [95 % confidence interval 27.1-57.8] for the 2022–2023 trapping season (N=36, (Table 2) A total of 15 females and 21 males were trapped. The age of the foxes, based on tooth analysis, ranged from 1 to 11 years old (mean age 3.1 years [95 % confidence interval (Cl) 2.2-4.1], median 2 years). The body weight range of the foxes was from 2.7-5.5 kg (mean 3.6 [3.4-3.9 95 % Cl], median 3.5). There were no significant trends relating to louse prevalence and the age (p=0.16), sex (p=0.61), body weight (p=0.15), body fat index (p=0.08) or trapping location (p=0.13) of the Arctic foxes.



Figure 11 An adult louse in stark contrast to the white of the Arctic fox fur. Photo: Eva Fuglei / Norwegian Polar Institute

Lice were detected in foxes from the following locations in the 2022–2023 trapping season (in alphabetic order): Bellsund (number of positives 2), Colesbukta (2), Engelskbukta (3), Grumant (2), Isfjordflya (2), Mälardalen (1), Svea (2), and Utløp av Sassendalen (1).

Table 2 The results from the initial fur investigation to detect lice prior to skinning, shown by the sex, of the Arctic foxes trapped during the 2022–2023 trapping season in Svalbard (N=36). \*This number is for one fox that had an uncertain positive status, given that only one suspected egg was found under initial examination. This individual was considered positive after the initial examination but negative for the final result when no lice or eggs were detected after louse burden analysis.

Sex	Lice detected	No lice detected
Female	7 (+1*)	8
Male	8	13
Total (final result)	15	21*

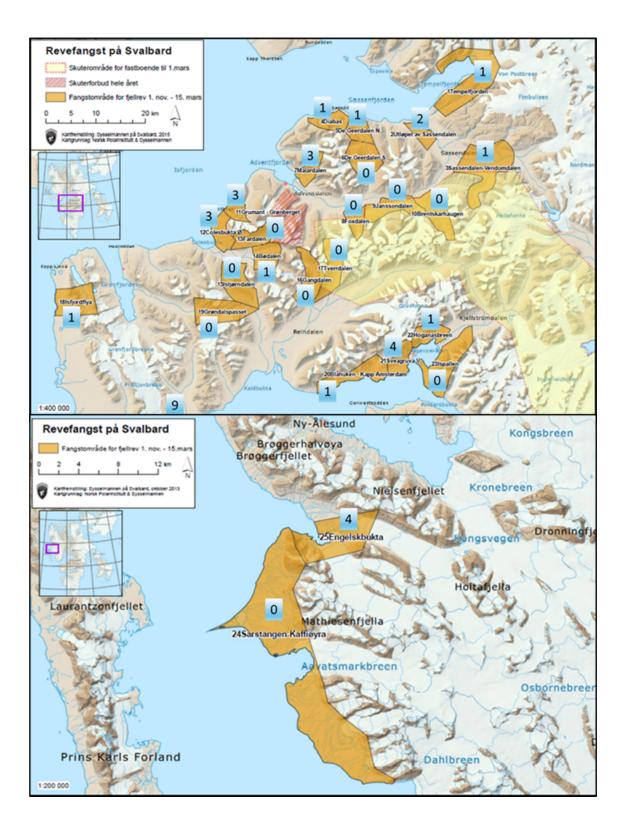


Figure 12 Maps showing the total number of foxes (blue squares) trapped from each of the Arctic fox trapping areas at Nordenskiöld Land (1 to 23) and south of Ny-Ålesund (24 to 25) in 2022–2023. Nine foxes were also trapped in the Bellsund area by a commercially trapper indicated on the upper map. Maps: <u>The Governor of Svalbard</u>

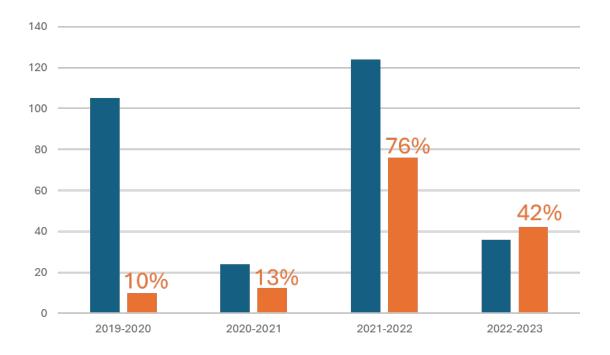


Figure 13 Total number of Arctic foxes trapped each season (blue bars; 2029-2020, 2020-2021, 2021-2022 and 2022-2023) and the fur lice prevalence (yellow bars) also indicated in %.

# 3.2 Evaluation of the pelt – abundance and distribution of bloodsucking lice, changes of pelt quality and skin pathology

#### 3.2.1 Initial visual assessment of louse distribution

All the positive foxes had adult lice, as well as larvae and egg stages, on their neck and shoulder region. Lice were found not only on the neck and shoulders but also on the face, head, legs, along the back to the tail and on the abdomen. During the initial examination nine of the infected foxes had lice in all four primary areas examined, two had lice in three of the four areas, three had lice in only two areas whilst only one individual only had lice over the shoulders. Fur changes (colour and/or shortened fur length) were detected in 17 pelts, of which 14 also had lice. All but one of the foxes with lice exhibited fur changes. There were significantly more foxes with fur changes in the group that had lice than in the uninfected group (p<0.001).

#### 3.2.2 Detailed assessment of louse burden

A total of eight different areas of the body were sampled for estimating the louse burden (<u>Figure 8</u>). The total number of lice found on the 17 pelts examined in greater detail varied from none to 10 984 which includes adults, larvae and eggs containing larvae. Mean abundance was 3 161 whilst the median was 1 405. In addition, one two-year-old male fox, from Grumant with high louse burdens, was also coinfected with bird fleas (Insecta: Siphonaptera). We detected lice in all eight regions on the pelt for individuals with moderate to high burdens. Individuals with low burdens had lice at only five or six of these locations, not all. The majority of pelts (12/15) had a higher proportion of eggs than the other two lifecycle stages (larvae and adults).

Comparison of the louse burden between the different areas of the body is based on the density (lice/cm<sup>2</sup>), rather than the number of counted lice at each location as the area analysed varied from 25-36 cm<sup>2</sup>. The density of lice varied depending on the stage of lice and the area of the body examined. However, there were no significant differences between the proportion of the different lifecycle stages at each site, so further analysis is based on the total louse burden (TLB) or the mean density of lice/cm<sup>2</sup>.

The neck and shoulder area had significantly higher louse density than the legs (elbow and stifle), breast, chin and abdomen (<u>Table 3</u>). The highest mean density of lice was found on the neck and shoulders and the lowest under the chin (Figure 14).

Table 3 An overview of the mean total louse burden/cm<sup>2</sup> at the different locations on the fur that had quantitative counts carried out after skin digestion. The 95 % confidence interval for the mean and the median result are also shown (N=15).

Location	Mean Total louse burden/cm²	95 % CI for the mean	Median
Neck and shoulder	42.2	18.6-65.9	28.1
Between the ears	19.2	2.4-35.9	5.0
Pelvis-lumbar region	18.3	-0.4-37.0	0.75
Elbow	8.7	1.7-15.8	2.0
Stifle	6.3	-1.6-14.1	0.0
Chest between the front legs	6.2	1.0-11.3	0.9
Ventral abdomen	6.2	1.0-11.3	0.9
Chin	1.6	0.0-3.1	0.3

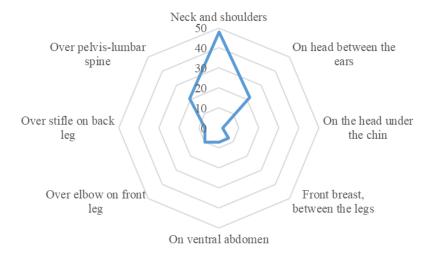


Figure 14 A radar chart showing the mean total density of all louse lifecycle stages (total louse burden (TLB: the total number of adults, nymphs and eggs)/cm2) at the eight different sites on the infected Arctic fox furs.

Bivariate analysis of the density of lice at the different locations compared to the total louse burden found the highest correlation with the density of lice at the shoulders: total louse burden (total number of all lifecycle stages found in the eight areas of the body analysed) = 53.85+76.15\*Density of lice at neck and shoulder (all lifecycle stages) (p<0.0001; Rsquared adjusted 0.79). There was one outlier, a male that had a surprisingly low density of lice at the neck and shoulders compared to the total number of lice detected (Figure 15). Similar analysis at the other seven sites also found significant correlation between the total louse burden and the density of lice at each site, however the Rsquared adjusted values were lower.

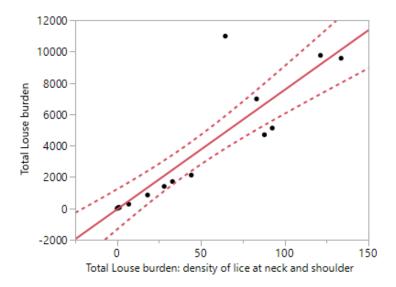


Figure 15 A bivariate fit of the total louse burden (the number of all louse lifecycle stages counted at eight different locations on the body) of Arctic foxes compared to the density of all louse lifecycle stages counted at the samples from the neck and shoulders (36 cm²). Total louse burden= 53.85+76.15\*density of total louse burden at the neck and shoulders. The dotted line shows the 95 % confidence interval. The outlier at the top with nearly 11 000 total louse burden had a surprisingly low density of lice at the neck and shoulders but very high densities at all other locations.

It is possible that in long-standing infections in which there has been considerable pruritis and alopecia that the burden of lice in the most affected areas is lower given the local loss of fur in which the lice would normally lay their eggs. The one 11-year-old male fox which had low louse density over the shoulders and neck, but otherwise the highest total louse burden of all the foxes, would seem to support this hypothesis. However, the degree of fur damage over the neck and shoulders, in this individual was mild, so loss of hair at this site does not explain the low louse burden.

#### 3.2.3 Louse abundance and trends

The comparison of the final louse burden estimate (none, low, moderate or high) to age (p=0.46), sex (p=0.47), fat index (p=0.55) and body weight (p=0.08) and trapping location (p=0.52), by contingency analysis only found a slight trend with body weight (Figure 16). Foxes classified as having a high louse burden, had significantly lower body weight compared to those with a low or no louse burden (p=0.04 and p=0.05 respectively). There were no significant body weight differences between those with high

and moderate burdens, or between those with a low burden and no burden. Extensive louse infestations, in other species, are reported to result in weight loss as well as reduced live weight gains in young animals (Deplazes et al. 2016, Zachary 2016) and drops in milk yields. If this holds true in foxes, something the trend of lower weights in Arctic foxes with high louse burdens would seem to support, then this may have knock on impacts at a time of year when vixens have especially high energy requirements due to reproduction and lactation. Fox pups from extensively infested females may therefore not only be exposed to high louse infection pressure from birth, risking anaemia, but may also get a suboptimal start if the vixen has poorer milk production. These concerns highlight the importance of further monitoring of this population for demographic and louse infection prevalence and abundance changes.

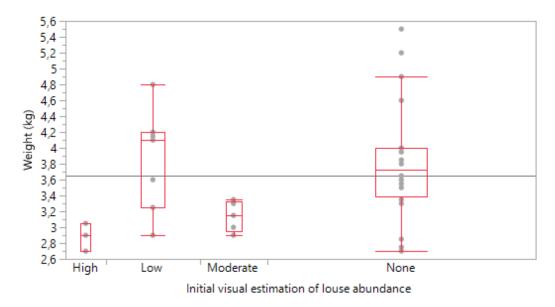


Figure 16 A quantile box plot of the body weight (kg) of Arctic foxes trapped for their fur in Svalbard, during the 2022-2023 trapping season (N=36), by final estimated louse burden using either visual assessment only (N=20) and skin digestions to count lice at eight locations on the body (N=16).

Comparison of the initial visual louse burden assessment in Longyearbyen before the skinning and the results from the laboratory analysis of pelts to quantify the number of lice showed substantial agreement (kappa score 0.73 [CI 0.55-0.91]; p<0.01; <u>Table 4</u>)).

Table 4 A comparison of the two methods used to evaluate louse burden from none, low, moderate or high using either visual assessment or skin digestions and quantification of total louse burden (all lifecycle stages: adults, larvae and eggs containing larva). The one fox with a low initial analysis was flagged as uncertain as only 1 egg was seen. Subsequent laboratory analysis found no lice in this individual. Overall, there was good agreement (cells marked green) between the two methods for low or no infections. Differentiating between moderate and high levels of infection was not as clear-cut.

Louse burden criteria	Skin digestions to count total louse burden (TLB) in the laboratory (adults, larvae and eggs)					
Initial visual assessment	None	Low <500	Moderate 250-2500	High >2500		
None	1					
Low (One or two adults per field of view in 2 locations or fewer)	1	5	1			
Moderate (more than 2 adults per field of view in more than 2 locations)			2	2		
High (several adult lice in each field of view at multiple locations)			2	3		
Final total in abundance categories	2	5	5	5		

A negative trend in fur length, both for the dense undercoat and guard hairs, compared to TLB (p=0.2 and p=0.4 for the two hair types respectively) as well as louse density at the shoulders (p=0.10 and p=0.37) was seen, but the significance level was not reached given only 14 fox furs had fur depth measurements for comparison. The comparison of damage score and log transformed louse burden also surprisingly found no significant trends in the number of lice and the degree of damage to the pelts even though the front pelts (N=16, p=0.2), had significantly higher lice densities (p<0.01) as well as having significantly higher damage scores than the back pelts (N=5, p=0.03).

### 3.3 Histological investigation

There was a mild to moderate increase of thickened laminated keratin with intervening clefts on the skin surface. There were hairshafts in nearly all the follicles and they were predominately in late anagen phase. We were able to fixate the lice on the slides that were close to the skin in most of the animals. There is no visible infiltration of inflammatory cells in the dermis. In one animal a *Trichinella* nurse cell was identified in the *panniculus carnousus* (subcutaneous muscle layer) (Figure 17). Therefore, the morphological diagnosis is set to mild to moderate, chronic, laminar orthokeratotic hyperkeratosis. The disease is called pediculosis caused by Anopluran lice.

The cause of the pruritus is not known but is thought to be a result of more than mechanical irritation alone (Zachary 2016). No visible inflammation in the skin suggests that the intense pruritus resulted from tactile skin irritations from the bloodsucking lice at this stage of infestation in the Arctic foxes. In all individuals investigated we found undamaged hair follicles in a healthy and growing state (anagen). Thus, the hairs did not fall out from the skin and the hair loss was not due to damage or visible disease

under the skin surface. We suggest that the hair loss at this stage is caused by mechanical breakage of hair shafts from scratching. The macroscopic lesions seen are mostly secondary to scratching, rubbing, or biting. Photographs of infested Arctic foxes taken much later in the winter, after the closure of the trapping season in mid-March, show considerably greater macroscopic fur and skin damage than was seen in the histological investigations of foxes (Figure 18). Histopathological examination of the skin in these individuals would most likely have shown a very different picture than that found in the foxes earlier in the year from the trapping period. Gross lesions of heavy infestations usually consist of inflammation in the skin with papules, crusts, excoriations, and self-induced damage to hairs.

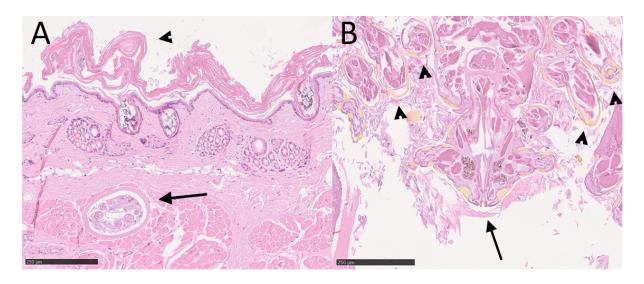


Figure 17 Histological image of a hematoxylin and eosin-stained section of the superficial skin of a fox infested with lice (A). The laminated hyperkeratosis on the surface of the skin can be seen as a thickened layer of keratin (arrowhead) and a Trichinella nurse cell is visible in the most superficial muscular layer of the skin (arrow). We were able to fixate lice in some of the sections and here the anterior parts of a louse can be studied (B). The mouth part is visible (arrow) and its legs are cross-sectioned (arrowheads). The yellow stain seen in this slide is the integument of the parasite.



Figure 18 Arctic fox in Bjørndalen May 6, 2024. The fox in the picture is showing extensive fur damage and skin changes over the neck and shoulders as well as around the eyes suggestive of severe lice infestation. Photo: Harald Berger

# 3.4 Camera traps on Arctic fox breeding dens to evaluate lice on free living foxes

A total number of 24282 pictures were taken on the automatic camera traps from January to June in 2024 (<u>Table 5</u>). Of this a total of 409 Arctic fox pictures were classifiable (as described in 2.5) and 279 of these showed signs of fur lice (mild, medium or extensive). The large total number of pictures were due to pictures of species other than Arctic foxes, specifically the Svalbard reindeer (*Rangifer tarandus platyrhynchus*), but also Svalbard rock ptarmigan (*Lagopus muta hyperborea*) and snow bunting (*Plectrophenax nivalis*).

Table 5 The total number of pictures, the number of pictures of Arctic foxes and the number of pictures with signs of fur lice on the five different camera traps on five breeding dens from January to June 2024.

Breeding den	Total # of pictures	# pictures with foxes	# pictures sign of fur lice
Den 13	7366	20	11
Den 24	3160	102	69
Den 34	1030	5	1
Den 150	705	26	13
Den 4	12021	256	185
SUM	24282	409	279

When combining all the defined classifications of signs of fur lice "mild+medium+extensive" referred to as "Fur lice" and summing up the total number of pictures of foxes taken at the five dens, the total number of foxes with "fur lice" per month increased from January and February to March (<u>Figure 19</u>). The highest number of foxes with signs suggestive of fur lice was seen in April.

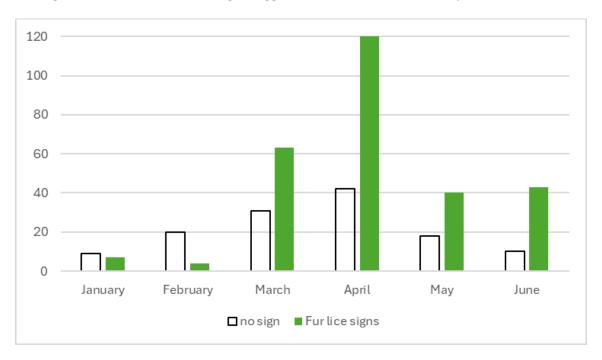


Figure 19 A histogram showing the monthly total number of pictures of foxes which were classified as having "mild+medium+extensive" fur lice signs or with no signs from all cameras on dens.

For a more detailed assessment of the seasonal pattern of how the more severe louse classification than "mild" development on the individual foxes, we plotted the combination of "medium+extensive" classifications (Figure 20).

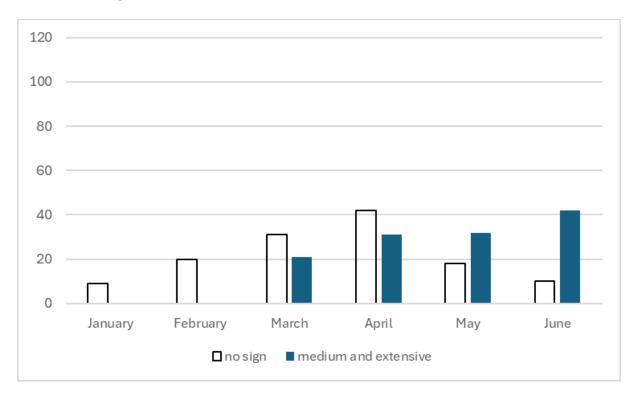


Figure 20 A histogram showing the total number of pictures of foxes with the fur louse classification no signs, and the combination of "medium+extensive" signs of infestation per month from all cameras on dens.

This gave more or less the same seasonal pattern for "medium+extensive" as for the total classifications that also included "mild", an increase in signs of infestation over the season. However, it also indicates a fairly clear tendency towards a more delayed development of "medium+extensive" infestations from April onwards, which suggests that foxes with a faint classification in the winter (January and February) had more intense infections/worsening clinical signs (more strongly affected) in the spring as infections become more established in each individual. The development of the "medium+extensive" infestation over the season, can be seen even more clearly when expressed as the "medium+extensive" signs in percent of the total classification (mild+medium+extensive, (Figure 21). The small dip in the proportion of pictures of medium and extensive in April is difficult to explain, but the numbers of dens are small with relatively low numbers of individuals, so it could be a coincidence. (Figure 21) also shows that the development of the proportion of "medium+extensive" signs on the foxes over the season showed the highest proportions in May and June.

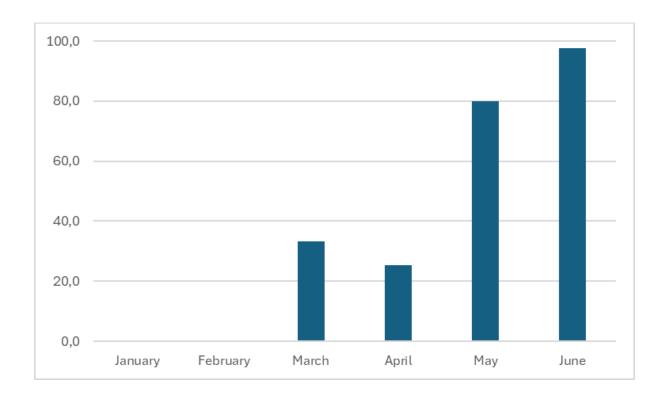


Figure 21 A histogram showing the proportion of "medium+extensive" classifications of louse infestation signs in foxes as a percentage (%) of the total classification "mild+medium+extensive" for signs of louse infestation per month from all cameras on dens.

An overview of the observed infections of sucking lice in Arctic foxes, classified from the pictures related to how severe lice infested the foxes were ("no sign", "mild signs", "medium signs", "extensive signs" and "summer fur"), are given in <u>Appendix Table 1</u> per breeding den and per month (January, February, March, April, May and June).

The lice infestation classification from all dens over time showed that foxes infested with fur lice were located on all five dens (<u>Figure 22</u>, <u>Appendix Figure 1</u>). There were also individuals with no signs of fur lice at dens.



Figure 22 An image of an Arctic fox, classified as having extensive signs of louse infestation, taken on 30<sup>th</sup> April 2024. Photo: Reconyx camera

### 4 Conclusion

The prevalence of louse in trapped Arctic foxes in 2022–2023 was 41.7 %. This is significantly less than the prevalence reported in the previous trapping season (76 %) despite increased efforts to detect the lice in the fur in this project, but higher than the two first trapping seasons, 10 % and 12.5 % respectively (Figure 13). This is promising for the Arctic fox population, but further monitoring is needed to ensure that the trend of reduced prevalence continues. Not surprisingly, albeit still worryingly, heavy lice burdens were associated with lower body weights. Further monitoring of louse burden in addition to prevalence is still therefore needed. The project collaborators (Norwegian Polar Institute and Norwegian Veterinary Institute) in discussion with Longyearbyen Jeger- og Fiskerforbund and the Governor of Svalbard have asked that the hunting bag in each trapping area continue to be limited to a maximum of five foxes during the current season (2023–2024) until more data on the impact of lice becomes available. The hunting bag size will be tabled for further discussions once the results from this year's trapping and den monitoring seasons are finalised.

The burden of lice over the shoulders was significantly higher than in other areas of the body. Foxes with lower total lice burdens also had a lower louse prevalence at other areas on the body than the shoulders. The best sites to investigate infection prevalence, and to a degree the abundance of lice, remain therefore on the neck and the shoulders. The initial analysis of louse burden had to be reassessed, after the results from the laboratory analysis, in a few individuals. Overall, the initial visual assessment was able to distinguish between low, and moderate or high levels of infection. This will allow future studies to carry out an initial visual assessment (none, low or high burdens) during skinning, followed by digestion of a skin biopsy from between the shoulders to estimate total louse burdens.

The estimation of fur damage from photos (both from camera traps of free-living foxes and of pelts in the laboratory), as well as measuring fur length showed greater and more frequent, damage around the neck and shoulders compared to other areas of the body. However, changes in colouration and damage to the fur were also seen in some of the uninfected individuals so photographic images can of course only be suggestive of louse infestation and would require subsequent confirmation by close visual and/or laboratory assessment of the skin. Surprisingly the fur damage score was not significantly correlated with the abundance of lice. However, given that we don't know when each fox was infected, we cannot estimate how long the fox has been itching and damaging their fur and skin.

This small study showed no age-related effects in louse prevalence or abundance. This requires further monitoring. If, as we suspect, the recent detection of this ectoparasite represents an introduction into a previously naïve population partial immunity may develop over time and, in the future, younger naïve or very old foxes, with weakened immune systems, might be more susceptible to mass infestation than health adults with previous louse exposure. The development of immunity could also explain the presence of uninfected foxes at breeding dens with infected individuals despite the extremely close contact between animals sharing a den. The absence of an inflammatory/immunological response in

the skin histopathology suggests that the pruritic response is directly related to the mechanical damage and irritation from the biting (bloodsucking) lice rather than a hypersensitivity or inflammatory reaction in the skin. Further investigation is needed to explore the immunological response to pediculosis in wild canines like Arctic foxes.

The seasonal trend in infection is particularly interesting and although in the trapped fox dataset we were unable to see a trend between infection intensity and trapping date, the sample size was relatively small, and the trapping season closes 15<sup>th</sup> of March well before the peak changes seen in the den photographic study in May and June and in photos provided by Svalbard people. Therefore, louse prevalence and abundance estimates based on findings in trapped foxes probably underestimate the true prevalence, impact and abundance of lice in this population.

In cattle it is reported that seasonal hair growth in spring with the loss of the thicker winter hair, reduces infection intensities dramatically (Deplazes et al. 2016). Seasonal molting to summer fur with the loss of the dense winter fur may therefore help regulate louse infection prevalence and abundance in Arctic foxes. Hairs with eggs attached will be shed and therefore the burden of infection will significantly be reduced. This could be one explanation as to why the dramatic clinical signs seen in winter and spring are not reported during the summer. However, it must be noted that we do not have data on signs of lice in summer because classifying the degree of fur damage, suggestive of pediculosis, was very difficult once the summer seasonal molt had started.

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# **6** Appendix

# **6.1.** Description of method for sucking louse evaluation in Longyearbyen before the Arctic fox was skinned

### Method for evaluating louse prevalence and burden in Arctic fox fur

Arctic foxes that had been trapped in Svalbard during the 2023–2024 trapping season were submitted to the Norwegian Polar Institute for skinning. All the carcases were frozen to -80 °C upon receipt for seven days to inactivate any potential infections with *Echinococcus multilocularis*. The carcasses were then stored at -20 °C until skinning was carried out. The carcasses were removed from the freezer 48 hours prior to skinning to allow them to defrost thoroughly.

The foxes were weighed (using a spring gauge weight hanging from the ceiling) and their gender and age and ID number were recorded.

The fur was then systematically manually inspected starting at the eyes under bright light conditions but without magnification.

- 1. Using a curved anatomical forceps the hairs were parted to visualise the skin along the length of the upper and lower eyelids.
- 2. The same was done along the rostral half of the upper lip on both sides and across the bridge of the nose.
- 3. The examination then continued using fingers to part the hair and visualise the skin on the top of the head between the ears and then continuing dorsally along the neck, shoulders, thorax, pelvis and base of the tail. If no lice were found the examination was stopped at this point and the animal recorded as having no lice detected.
- 4. If lice were found the examination was continued to look at the lateral aspect of all four legs and the entire ventral side of the fox was also examined for the presence or absence of lice. Findings were recorded on a paper record form, along with drawings showing the extent of the presence of the lice.

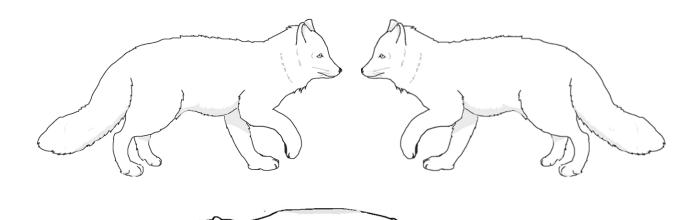
An evaluation of the number of lice was made: as low (only a few individuals seen in a couple of locations); moderate (more than two adult individuals seen per field of view per location in more than 2 locations); high (multiple lice (>4) seen in each field of view at multiple locations). Sometimes it was difficult to see the skin given the fur density. Areas in which black dust was present in the lower layers of the fur were examined extra carefully for the presence of lice or louse eggs.

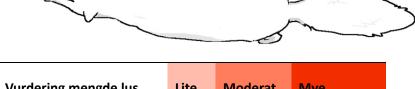
After skinning their body condition was evaluated using a scale of 1-4. The upper jaw was also removed just posterior to the first molar for more accurate age assessment at a later date.

### Arctic Lus – Fangstsesong

Vurdering av pelskvalitet og lusforekomst – ID nr \_\_\_\_\_

Lokalitet	Fangstdato	Kjønn	Alder (est.)	Hold vurdering	Vekt (kg)
Lus		Mengder lus		Grad pels- og hud	forandringer
Ikke påvist		Ingen		Ingen	
Påvist		Lave		Farge	$\square$ se tegning
Øyne og nese		Moderate		Hårlengde	$\square$ se tegning
Hodet – me ørene	lom 🗆	Mye		Pelstykkelse	$\square$ se tegning
Nakken Bak skuldrene		Bilder tatt		Håravfall Hudbetennelse	<ul><li>☐ se tegning</li><li>☐ se tegning</li></ul>





Vurdering mengde lus	Lite	Moderat	Mye
Antall plasser med lus	1-2	>2	Flere steder
Mengder/synsfelt/plass	<2	>2	>4

Kommentarer
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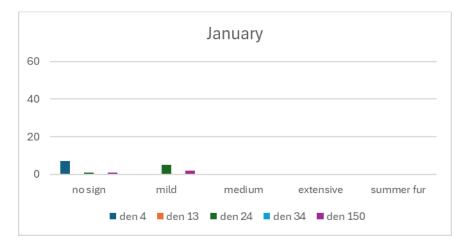
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### 6.2. Seasonal development of signs of fur lice in Arctic foxes

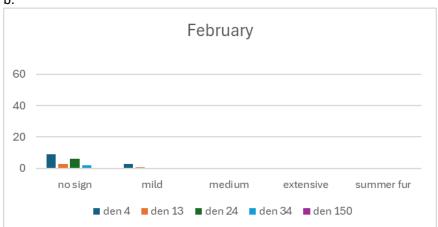
**Appendix Table 1**. Observed infection of sucking louse on pictures of Arctic foxes at five breeding dens (#4, 13, 24, 34 and 150) from January to June 2024. Observations were characterized in five categories (no sign, mild, medium, extensive) and a notion about when foxes were seen in summer fur on the pictures.

Months/den#	Observed	Den 4	Den 13	Den 24	Den 34	Den 150	TOTAL
January	no sign	7	0	1	0	1	9
	mild	0	0	5	0	2	7
	medium	0	0	0	0	0	0
	extensive	0	0	0	0	0	0
	summer fur	0	0	0	0	0	0
February	no sign	9	3	6	2	0	20
	mild	3	1	0	0	0	4
	medium	0	0	0	0	0	0
	extensive	0	0	0	0	0	0
	summer fur	0	0	0	0	0	0
March	no sign	16	5	10	0	0	31
	mild	27	2	8	0	5	42
	medium	16	2	0	0	0	18
	extensive	0	3	0	0	0	3
	summer fur	0	0	0	0	0	0
April	no sign	27	0	12	2	1	42
	mild	75	1	11	1	3	91
	medium	25	0	1	0	1	27
	extensive	0	2	1	0	1	4
	summer fur	0	0	0	0	0	0
May	no sign	12	0	4	NA	2	18
	mild	7	0	1	NA	0	8
	medium	19	0	5	NA	0	24
	extensive	1	0	7	NA	0	8
	summer fur	11	0	0	NA	2	13
June	no sign	0	1	0	NA	9	10
	mild	0	0	0	NA	1	1
	medium	10	0	13	NA	0	23
	extensive	2	0	17	NA	0	19
	summer fur	31	11	2	NA	10	54

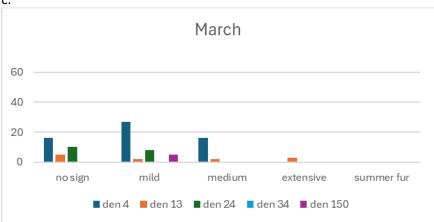
a.



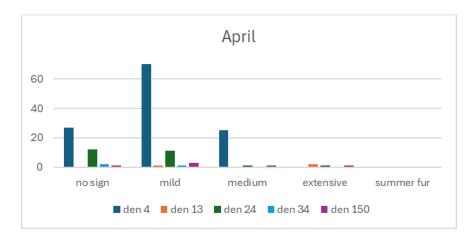
b.

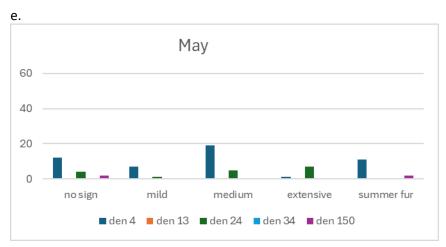


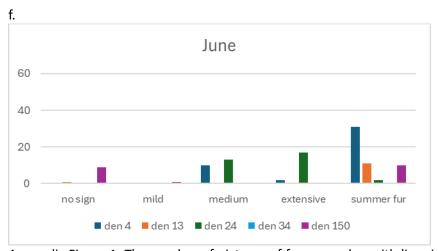
c.



d.







Appendix Figure 1. The number of pictures of foxes per den with lice signs classified as no sign, mild, medium and extensive signs, in addition to when summer fur for the first time appeared on foxes per month. a: January, b: February, c: March, d: April, e: May, f: June.