Diet and metabolic state are the main factors determining concentrations of perfluoroalkyl substances in female polar bears from Svalbard

Sabrina Tartu[1]; Sophie Bourgeon[1,2]; Jon Aars[1]; Magnus Andersen[1]; Karen Lone[1]; Bjørn Munro Jenssen[3]; Anuschka Polder[4]; Gregory W. Thiemann[5]; Vidar Torget[3]; Jeffrey M. Welker[6,7] and Heli Routti[1]*

[1] Norwegian Polar Institute, Fram Centre, Tromsø, Norway

[2] UiT-The Arctic University of Norway, Department of Arctic and Marine Biology, Tromsø, Norway

[3] Norwegian University of Science and Technology, Department of Biology, Trondheim, Norway

[4] Norwegian University of Life Science, Campus Adamstua, Oslo, Norway

[5] York University, Faculty of Environmental Studies, Toronto, ON, Canada

[6] University of Alaska Anchorage, Department of Biological Sciences, Anchorage, AK, USA

[7] University Center in Svalbard, Longyearbyen, Svalbard, Norway

Corresponding author: Sabrina Tartu, tel. +47 777 50 544, e-mail: tartu.sabrina@gmail.com

ABSTRACT

Perfluoroalkyl substances (PFASs) have been detected in organisms worldwide, including Polar Regions. The polar bear (*Ursus maritimus*), the top predator of Arctic marine ecosystems, accumulates high concentrations of PFASs, which may be harmful to their health. The aim of this study was to investigate which factors (habitat quality, season, year, diet, metabolic state [i.e. feeding/fasting], breeding status and age) predict PFAS concentrations in female polar bears captured on Svalbard (Norway). We analyzed two perfluoroalkyl sulfonates (PFSAs: PFHxS and PFOS) and C_8-C_{13} perfluoroalkyl carboxylates (PFCAs) in 112 plasma samples obtained in April and September 2012-2013. Nitrogen and carbon stable isotope ratios ($\delta^{15}N$, $\delta^{13}C$) in red blood cells and plasma, and fatty acid profiles in adipose tissue were used as proxies for diet. We determined habitat quality based on movement patterns, capture position and resource selection functions, which are models that predict the probability of use of a resource unit. Plasma urea to creatinine ratios were used as proxies for metabolic state (i.e. feeding or fasting state). Results were obtained from a conditional model averaging of 42 general linear mixed models. Diet was the most important predictor of PFAS concentrations. PFAS concentrations were positively related to trophic level and marine diet input. High PFAS concentrations in females feeding on the eastern part of Svalbard, where the habitat quality was higher than on the western coast, were likely related to diet and possibly to abiotic factors. Concentrations of PFSAs and C_8-C_{10} PFCAs were higher in fasting than in feeding polar bears and PFOS was higher in females with cubs of the year than in solitary females. Our findings suggest that female polar bears that are exposed to the highest levels of PFAS are those 1) feeding on high trophic level sea ice-associated prey, 2) fasting and 3) with small cubs.

Capsule: PFAS concentrations are driven by diet and metabolic state (feeding/fasting) in female polar bears; decreasing sea ice extent is likely to modify PFAS exposure in polar bears.

Keywords: Ursus maritimus; PFAS; breeding status; habitat quality; fasting; stable isotope

INTRODUCTION

Perfluoroalkyl substances (PFASs) are a group of anthropogenic chemicals that have been manufactured for more than 50 years. PFASs are commonly used in the production of stain repelling agents, fluoropolymers, pesticides, lubricants, paints, medicines and fire-fighting foams due to their ability to repel both water and oils (Key et al., 1997; Prevedouros et al., 2006). PFAS are thermally and chemically stable, have no route of degradation and cannot be metabolized under normal environmental conditions, which makes them extremely persistent in the environment (Muir and de Wit, 2010). PFASs have been detected in blood and tissues of wildlife and humans worldwide, including remote regions such as the Arctic (Haukås et al., 2007; Houde et al., 2011; Lau et al., 2007; Martin et al., 2004).

In contrast to persistent lipophilic pollutants, such as polychlorinated biphenyls (PCBs), PFASs have a high affinity towards plasma proteins, in particular albumin, and tend to accumulate in protein-rich compartments such as blood, liver and kidneys (Buck et al., 2011). Retention of PFASs in these organs and tissues may be toxicologically significant. In laboratory mammals, the effects of PFAS include disrupted steroid hormone and lipid homeostasis, reduced body weight, increased liver weight and a steep dose–response curve for mortality (Guruge et al., 2006; Jensen and Leffers, 2008; Lau et al., 2007).

The degree of bioaccumulation of PFASs generally increases with chain length (Martin et al., 2003a, 2003b). For instance, perfluorooctanesulfonic acid (PFOS) and C_9-C_{13} perfluoroalkyl carboxylate (PFCA, C_n refers to the carbon chain length) concentrations increase with trophic position thus, several PFASs can reach very high levels in top predators (Martin et al., 2004; Tomy et al., 2009; Van de Vijver et al., 2003). In addition, PFAS are transported by air and ocean currents to remote Arctic regions (Armitage et al., 2009; Shoeib et al., 2006; Wania,

2007). Polar bears (*Ursus maritimus*), as Arctic top predators are therefore highly exposed to PFASs (Kelly et al., 2009; Tomy et al., 2004).

Polar bears are among the most polluted species in the Arctic (Letcher et al., 2010). Quantitatively, PFAS is the most important contaminant group found in polar bear blood in wet weight concentrations (Bytingsvik et al., 2012a, 2012b). Among polar bears subpopulations, the concentrations of both lipophilic and proteinophilic pollutants are higher in the Barents Sea (i.e. Svalbard) than in most other subpopulations (McKinney et al., 2011; Smithwick et al., 2005a). Polar bears are seasonal feeders, their preferred prey being ringed (*Pusa hispida)* and bearded seals (*Erignathus barbatus*) especially in spring and early summer. Polar bears also feed opportunistically on a large range of land-based and marine species (Iversen et al., 2013; Tartu et al., 2016; Thiemann et al., 2008). Because of bioaccumulation up the food chain, bears feeding on seals may have higher pollutant concentrations than bears that feed on species lower in the food web. Moreover, pollutant exposure may also be affected by life history traits, during prolonged fasts, which can last up to 6-8 months for pregnant females (Andersen et al., 2012; Ramsay and Stirling, 1988) polar bears can lose over 40% of their body mass and the energy is drawn primarily from fat tissue (Atkinson and Ramsay, 1995).

Information on the effects of PFAS in polar bears is scarce. Modelling and correlative field studies suggest that concentrations of PFASs in polar bears are associated with increased steroid hormone concentrations in the brain, impaired reproduction and immunity (Dietz et al., 2015; Pedersen et al., 2016). There is currently little knowledge of the intrinsic or extrinsic factors that determine individual variation in PFAS concentrations in Arctic wildlife. For example, trophic level is a likely factor to influence PFAS exposure in marine mammals (Van de Vijver et al., 2003). Furthermore, PFAS concentrations have been related to body condition in Arctic

92 foxes (*Vulpes lagopus*), harbor seals (*Phoca vitulina*) and Arctic breeding black-legged
93 kittiwakes (*Rissa tridactyla*) (Aas et al., 2014; Tartu et al., 2014; Van de Vijver et al., 2003).

94 Breeding status in mammals may also be a source of variation as PFAS can be transferred from
95 mother to young during pregnancy and lactation. Placental transfer is the dominant pathway
96 for PFASs in hooded seals (*Cystophora cristata*) and polar bears (Bytingsvik et al., 2012b;
97 Grønnestad et al., 2016). In polar bears, maternal transfer of PFASs is relatively low
98 (Bytingsvik et al., 2012b). Finally, space-use patterns may also influence exposure to PFAS
99 and other contaminants in polar bears through abiotic or biotic factors (Olsen et al., 2003; van
100 Beest et al., 2015). The aim of this study was to investigate which factors (habitat quality,
101 season, year, diet, metabolic state [i.e. feeding/fasting], breeding status and age) predict PFAS
102 concentrations in female polar bears from Svalbard. This information is highly valuable for
103 management to identify which individuals are the most vulnerable to PFAS exposure and how
104 ongoing climate change might alter PFAS exposure in polar bears.

105 **MATERIAL AND METHODS**

106 FIELD SAMPLING

107 Adult female polar bears (age 4-28 years) from the Barents Sea subpopulation were captured
108 non-selectively throughout Svalbard in April and September 2012 and 2013. The 112 samples
109 collected (April 2012, n=33, age: 12.9 ± 1.1 years (mean ± standard deviation), September
110 2012, n=24, 13.2 ± 1.4 years, April 2013, n=29, 13.4 ± 1.0 years and September 2013, n=26,
111 12.8 ± 1.2 years) represented 78 females. Twenty-six females were captured more than once,
112 specifically, we captured 19 females twice, six females three times and one female four times.
113 However, females were not recaptured within the same fieldwork season.

Females were immobilized by remote injection of tiletamine hydrochloride and zolazepam hydrochloride (Zoletil Forte Vet ®; Virbac, France), delivered by a dart fired from a helicopter (Eurocopter AS350 Ecureuil). We collected 50-100 ml of blood from the femoral vein using vacutainers (9-10 ml) with Lithium-Heparine to avoid clotting. We kept samples cool and out of sunlight until centrifuged within 10 h (3500 rpm, 10 minutes). Red blood cells and plasma were transferred to two separate cryotubes and frozen at -20ºC. Adipose tissue samples were collected using an 8 mm biopsy punch taken approximately 15 cm lateral to the base of the tail. In the field, adipose tissue samples were stored in a dry-shipper then kept at -80 ºC until analyses. Immobilization and handling procedures followed standard protocols (Derocher and Wiig, 2002; Stirling et al., 1989), and were approved by the National Animal Research Authority (Norwegian Animal Health Authority, P.O. Box 8147 Dep., N-0033 Oslo, Norway).

Females were classified in three groups according to their breeding status: solitary (i.e., alone or together with a male in spring), with 1 or 2 cubs of the year (COYs; cubs younger than 1 year old) or with 1 or 2 yearlings (cubs aged between 1 and 2 years). No females with older cubs were captured as part of the current project. Female polar bears were aged using a vestigial premolar tooth (P1) following a method described previously (Calvert and Ramsay, 1998). The age of the females was not significantly different between groups ($p>0.25$). Body condition index (BCI) was calculated as described for polar bears (Cattet et al., 2002) based on body mass (BM) and straight-line body length (SL): $BCI=(\ln BM-3.07 \times \ln SL+10.76) / (0.17+0.009 \times \ln SL)$.

ANALYSIS OF PFASs

Plasma samples (n=112) were analysed for PFASs at the Laboratory of Environmental Toxicology at the Norwegian University of Life Sciences (NMBU), Oslo, Norway. The plasma

137 samples were analysed for six perfluoroalkyl carboxylic acids (PFCAs: perfluorooctanoate
138 PFOA, perfluorononanoate PFNA, perfluorodecanoate PFDA, perfluoroundecanoate
139 PFUnDA, perfluorododecanoate PFDoDA and perfluorotridecanoate PFTrDA) and two
140 perfluoroalkyl sulfonic acids (PFSAs: perfluorohexane sulfonate PFHxS and PFOS). The
141 methods were described in another study (Grønnestad et al., 2016).

142 Plasma samples (1 ml) were weighed in 15 ml Falcon centrifuge tubes (VWR International,
143 LLC Radnor, USA). All tubes and pipettes used were made of plastic. Internal standards (^{13}C-
144 labeled equivalents, 20 ng/ml) and 5 ml methanol (Rathburn chemicals, Walkerburn, Scotland)
145 were added to the samples. The samples were mixed for 10 seconds on a Whirlymixer (MS2
146 Minishaker, IKA®, MA, USA) followed by 30 minutes of mixing in a Vibrax machine (Vibrax
147 VXR, IKA®, MA, USA). The samples were centrifuged at 3000 rpm for 10 minutes (Allegra®
148 X-12R, Beckman Coulter, CA, USA). The supernatant was extracted and transferred it to new
149 Falcon tubes. The extraction was repeated with 3 ml methanol. The supernatant was evaporated
150 to a volume of 2 ml using a zymark instrument (TurboVap® LV, Zymark Corporation
151 Hopkinton, MA, USA) with water bath (40°C) and a gentle flow of nitrogen gas (N_2) (Purity:
152 99.6%, Aga AS, Oslo, Norway). The samples were cleaned-up by adding approximately 0.2 -
153 0.3 g active coal (ENVI-Carb™, Sigma-Aldrich, Oslo, Norway) to each sample. The samples
154 were mixed on the Whirlymixer (10 seconds) and then centrifuged (3000 rpm, 10 minutes). The
155 supernatant was transferred quantitatively to new Falcon tubes calibrated to 0.5 ml. The extract
156 was evaporated to a final volume of 0.5 ml and the samples were centrifuged (3000 rpm, 10
157 minutes) and transferred to vials with plastic inserts (200 µl).

158 The final extracts were separated on a high-performance liquid chromatograph (HPLC) with a
159 Discovery C18 column (15 cm × 2.1 mm × 5 µm, Supelco, Sigma-Aldrich, Oslo, Norway),

connected to a pre-column; Supelguard Discovery C18 column (2 cm × 2.1 mm × 5 µm, Supelco, Sigma-Aldrich, Oslo, Norway). Detection and quantification was accomplished with a tandem mass spectrometry (MS-MS) system (API 3000, LC/MS/MS System). The injected volume was 5 µl. Calculation was performed using MassHunter Quantitative analysis Version B.05.02 (Agilent Technologies). LOD were three times signal to noise ratio found in the samples and are given in **Table 1**.

For each series of approximately 30 samples, three procedural blank without matrix, one blind and two recovery samples were analysed. The relative recovery rate ranged from 86% to 103% for the PFCAs and 99% to 110% for the PFSAs. The results were corrected for recoveries. The laboratory participates in several international ring tests per year, one of the series included three samples of human serum as part of the ring test by Arctic Monitoring and Assessment Program (www.amap.no) and the results were satisfactory.

STABLE ISOTOPES IN PLASMA AND FATTY ACIDS IN ADIPOSE TISSUE

Determination methods of δ^{15}N and δ^{13}C in red blood cells and plasma (n=112) and fatty acids (FA) composition in adipose tissue (n=83) have been previously described (Tartu et al., 2016). Briefly, δ^{15}N values change in a predictable fashion between trophic levels and thus reflect trophic position of the individual polar bears (Hobson, 1999; Hobson et al., 1996). In contrast, δ^{13}C remains little changed according to trophic position and thus can indicate sources of primary productivity for example marine vs. terrestrial, pelagic vs. benthic, inshore vs. offshore (Hobson, 1999; Hobson et al., 1996). Therefore, stable isotopes can be used as proxies for diet. In polar bear red blood cells, half-life for δ^{13}C is ~1.5 months whereas half-life for δ^{15}N is at least twice as long (Rode et al., 2016). In polar bear plasma, half-lives for δ^{13}C and δ^{15}N are 10 and 18 days, respectively (Rode et al., 2016). Thus, once acquired, polar bear red blood cells

183 and plasma can provide a retrospective record of diet sources over months to days' time periods,
184 respectively (Rogers et al., 2015; Tartu et al., 2016).

185 Dietary FAs are predictably incorporated into a consumer's tissues and can thus provide insight
186 into an organism's diet over the preceding weeks to months (Iverson et al., 2004), and perhaps
187 longer in some species (Budge et al., 2006). Seventy-five different FAs were determined in the
188 fat samples. As suggested by Budge et al. (2012), for further analyses we selected 33 FAs that
189 were ≥ 0.2% of total FAs and collectively accounted for 96.9% of total FAs. FA data were
190 transformed by calculating the log of the ratio of each FA to c18:0 prior to principal component
191 analysis (PCA) (Budge et al., 2006). Since the log of 0 cannot be taken, 0 values were replaced
192 with a small constant (0.005%) prior to transformation. The 32 FAs (without 18:0) used in the
193 present study included iso-14:0, 14:0, 14:1n-5, 15:0, 16:0, 16:1n-11, 16:1n-9, 16:1n-7, 16:1n-
194 5, iso-17:0, 16:2n-4, c17:0, 18:1n-11, 18:1n-9, 18:1n-7, 18:1n-5, 18:2n-6, 18:3n-4, 18:3n-3,
195 18:4n-3, 20:1n-11, 20:1n-9, 20:1n-7, 20:2n-6, 20:4n-6, 20:4n-3, 20:5n-3, 22:1n-11, 22:1n-9,
196 21:5n-3, 22:5n-3 and 22:6n-3. We generated FA principal components (PCs) for further
197 analysis from the first, second and third axis of the PCA (projected inertia: PC1: 31.6, PC2:
198 16.7, PC3:12.7%, respectively). Using PCA scores enables to summarize FA composition into
199 three continuous variables. The three first axes accounted for 61.0% of the total variance of the
200 data cloud. Individual FAs that contributed most (>5%) to PC1 were: 15:0, 16:1n-11, 16:1n-7,
201 16:1n-5, 16:2n-4, 18:4n-3, 20:1n-11, 20:1n-9, 20:5n-3, 22:1n-9; to PC2: 16:0, iso-17:0, 17:0,
202 18:1n-7, 18:3n-4, 20:1n-9, 20:4n-6, 22:1n-11 and to PC3: iso-14:0, 14:0, 14:1n-5, 16:1n-9,
203 18:1n-7, 22:1n-11, 21:5n-3, 22:5n-3.

204 METABOLIC STATE DETERMINATION

The ratio of urea to creatinine (urea:creatinine) is indicative of the metabolic state (feeding/fasting state) of polar bears, low values indicating a fasting state (Derocher et al., 1990; Nelson et al., 1984). Molar concentrations of urea and creatinine were analysed in plasma (n=111), the samples were stored at -20 ºC for 1-2 years and thawed before being analysed in autumn 2014. The analyses were performed using a "dry" clinical-chemical analyzer, Reflotron® (Model IV, Boehringer-Mannheim GmhB, Mannheim, Germany). The system is composed of a reagent carrier (test strip) and a microprocessor controlled reflectance photometer. The system uses individual strips for each parameter, and each strip uses a specific reaction to produce a dye that is measured and evaluated by the reflectance photometer. All samples were analysed in duplicates, if high variation was observed between the duplicates, an additional replicate was analysed. Limits of detection (LOD) are given in **Table 1**. Previous studies have reported a threshold value of urea:creatinine <10 to report a fasting state (Cherry et al., 2009; Nelson et al., 1984). This calculation was performed on urea and creatinine concentrations in mg/dl, if converted to molar concentrations as used in the present study we obtain a threshold value of 47.5. We therefore considered that females with urea:creatinine ≤ 47.5 were in a fasting state.

HABITAT QUALITY

Polar bears movements follow a circannual pattern with season-specific area fidelity (Mauritzen et al., 2001), for example female polar bears show fidelity to denning and spring feeding areas (Mauritzen et al. 2001; Lone, Aars & Ims 2012). We categorized the quality of habitat available to bears based on their movement patterns or capture positions. A resource selection function (RSF) for bears in the Barents Sea subpopulation (Lone et al., under review) was used to predict the distribution of high quality habitat during four periods preceding each capture effort

(September 2011-March 2012, April 2012-August 2012, September 2012-March 2013, April 2013-August 2013). The seasonal RSF models, which are based on telemetry data from 224 females between 1991 and 2015, predict the probability of use of a habitat based on sea ice concentration, distance to the ice edge and ocean depth (Lone et al., under review). Daily predictions were classified as habitat or non-habitat using a cut-off corresponding to 70% of all polar bear positions occurring in pixels classified as habitat, and these daily maps were summed across each period of interest. According to these maps produced using RSF, the western coast of Svalbard has fewer habitat days in all four periods compared to the eastern side (**Figure S1**). Therefore, we divided Svalbard into two relative habitat categories with the western side considered as a poor quality habitat and the eastern side as a good quality habitat (**Figure S1**). Among the 78 individual bears used in this study, 59 were equipped with satellite telemetry collars during the study period or previous years. For these bears, we used location data to determine whether they used the good or poor habitats (**Figure S1**). For the bears without collars, we used the capture position during the study period to determine if they were using good or poor habitats. Seventy-nine females were assigned to the "eastern good quality habitat" and 33 to the "western poor quality habitat".

STATISTICS

PFAS concentrations were log transformed (*ln*) because of left-skewed distributions, and continuous predictor variables such as stable isotopes in plasma and red blood cells, urea:creatinine, BCI, age and FA PCs were standardized (mean = 0, SD = 1) before analysis to facilitate the comparison of effect sizes (Gelman and Hill, 2006). Values below LOD were replaced by ½ LOD. Creatinine was above LOD in all samples, whereas urea values were below LOD in 26 samples (**Table 1**). Except for PFDoDA, PFASs in the 112 samples were above

LOD (**Table 1**). We conducted statistical analyses using R version 3.2.1 (R Core Team, 2016). We used generalized linear mixed models (GLMMs; R-package *nlme* version 3.1-121, Pinheiro *et al.*, 2015) with female identity (female ID) as a random factor to test whether plasma concentrations of PFASs were affected by individual characteristics and environmental factors. To do so, we selected 42 biologically relevant models (**Table S1**). We used an information-theoretic approach (Burnham and Anderson, 2004) based on Akaike's information criterion corrected for small sample size (AICc, R package *MuMIn*, Barton, 2016). We calculated the number of parameters (K), the difference in AICc values between the "best" model and the model at hand (ΔAICc) and a normalized weight of evidence in favor of the specific model, relative to the whole set of candidate models, derived by $e^{(-0.5(\Delta AICc))}$ (AICc weights). We used model averaging to make inference from all the models. This method produces averaged estimates of all predictor variables in the candidate model list (**Table S1**), weighted using the AICc weights (Burnham and Anderson, 2003; Lukacs et al., 2009). From this, we obtained conditional parameter-averaged estimates (β) and 95% confidence intervals (CIs) for all the predictors included in the models. We used 95% CI of the model averaged estimates to determine if parameters were significantly different from 0 at the 5% level, 95% CI provide information about a range in which the true value lies with a certain degree of probability, as well as about the direction and strength of the demonstrated effect (du Prel et al., 2009). If the 95% CI does not include the value of zero effect, it can be assumed that there is a statistically significant result. We used Redundancy analysis (RDA, R-package *ade4* version 1.7-4, Dray and Dufour, 2007) to illustrate the relationship between response variables (individual PFASs) and predictors (stable isotopes in plasma and red blood cells, urea:creatinine, BCI, age and FA PCs). RDA is a method to extract and summarize the variation in a set of response variables (PFAS concentrations) that can be explained by a set of explanatory variables (Legendre and

Anderson, 1999; Ramette, 2007). More specifically, it summarizes the response variables' variance explained by a set of explanatory variables using linear relationships (Legendre and Anderson, 1999; Ramette, 2007). To investigate the overlapping effect of habitat quality and season on PFAS concentrations we used least squares means method (LSM, R-package *lsmeans*, Lenth and Hervé, 2015).

RESULTS AND DISCUSSION

PFAS concentrations

On average, PFOS accounted for 67.6% of total plasma PFAS concentration in female polar bears of the present study. Then followed by decreasing order: 9.9% PFNA, 8.8% PFHxS, 6.4% PFUnDA, 3.1% PFDA, 2.1% PFTrDA, 1.3% PFOA and 0.8% PFDoDA. PFAS concentrations (ng/g wet weight) are presented in **Table 1**.

In the same females, plasma concentrations of PCBs and their metabolites (OH-PCBs) were recently reported (Tartu et al., 2017). In comparison, plasma PFOS concentrations were 5.8- and 3.6-fold higher than plasma Σ_{16}PCBs and Σ_{8}OH-PCBs (39.9 and 56.1 ng/g ww, respectively; see Tartu et al. (2017) for a detailed list of the congeners included in the sums above). In most polar bear subpopulations, including the Barents Sea, previous studies have reported the dominance of PFOS among PFASs (Bytingsvik et al., 2012b; Smithwick et al., 2005a). Svalbard female polar bears sampled in 2008 had similar proportions of PFASs (PFOS > PFNA > PFHxS > PFUnDA > PFDA > PFTrDA > PFOA > PFDoDA) (Bytingsvik et al., 2012b).

PFASs increase with trophic level and proportion of marine diet

296 Concentrations of all PFASs increased with the trophic level of the female polar bears, as
297 inferred from δ^{15}N values in red blood cells and plasma (**Figure 1A-2, Table 2**). This is in
298 accordance with previous studies showing biomagnification of PFASs in Arctic marine
299 ecosystems (Haukås et al., 2007; Kelly et al., 2009; Tomy et al., 2004). In our study, PFAS
300 concentrations were positively related to sources of primary productivity (i.e. δ^{13}C values) in
301 red blood cells but not in plasma (**Figure 1A-2, Table 2**). In polar bears the half-lives of δ^{15}N
302 and δ^{13}C in red blood cells and plasma δ^{15}N (weeks to months) are longer than those of plasma
303 δ^{13}C (~10 days) (Rode et al., 2016). Considering that, our results indicate that plasma PFAS
304 concentrations in polar bears reflect exposure over the past weeks/months rather than over the
305 past days. The higher PFAS exposure in female polar bears from Svalbard having a more
306 marine based diet is in accordance with previous studies that report higher PFAS concentrations
307 in marine than in terrestrial prey (Kelly et al., 2009; Müller et al., 2011). Prior to capture, several
308 of the females from the present study were observed feeding on whale carcasses, walruses,
309 seabirds, geese or reindeers (Tartu et al., 2016), which is also in accordance with previous
310 studies showing the opportunistic and highly variable diet of polar bears (Aars et al., 2015;
311 Dyck and Romberg, 2007; Gormezano and Rockwell, 2015, 2013; Iversen et al., 2013; Iverson
312 et al., 2014; Prop et al., 2015, 2013; Rogers et al., 2015; Smith et al., 2010; Stempniewicz, 2006,
313 1993; Stempniewicz et al., 2014).

314 The FA composition as inferred by FA PC2 and FA PC3, correlated positively and negatively,
315 respectively with PFUnDA, PFDoDA and PFTrDA (**Figure 3, Table 2**). These results were
316 expected as FA PC2 and FA PC3 respectively correlated positively and negatively with stable
317 isotopes (**Table S2**). Yet, for FA PC2, the significance of the relationships with PFCAs were
318 driven by two outliers, and the correlation was not significant when the outliers were removed

(PFUnDA: β=0.28, 95% CI [-0.09, 0.66], PFDoDA: β=0.33, 95% CI [-0.05, 0.70] and PFTrDA: β=0.27, 95% CI [-0.08, 20.62]). We should therefore remain cautious when interpreting the FA PC2 results. The individual FAs that contributed most to FA PC3 were mainly saturated and monounsaturated FAs (see list in the method section). This suggests that C_{11}-C_{13} PFASs could be in lower proportion in prey with larger proportions of saturated and monounsaturated FAs. In polar bears, FA composition is a product of both diet and lipid metabolism so both factors may affect PFASs (Iverson et al., 2004). In addition, longer chained PFCAs possess higher log K_{ow} (octanol-water partition coefficient) values, indicating that they have an increased solubility in lipid-soluble environments (Greaves et al., 2012). It has been suggested that long-chain PFCAs greatly resemble saturated FAs (Greaves et al., 2012), this could explain the relationships between PFASs and FA composition.

There is a great uncertainty regarding future temporal trends in polar bears PFAS concentrations in relation to the ongoing climate change and its effects on polar bears diet and energetic metabolism. While a diet shift towards more land-based lower trophic level (Gormezano and Rockwell, 2013; Prop et al., 2015) is likely to diminish their exposure to PFAS, the increasing proportion of fasting bears due to melting sea ice (Cherry et al., 2009) may lead to increased PFAS concentrations in plasma.

High PFAS concentrations in fasting polar bears

In this study, urea:creatinine ratios were negatively related to plasma PFHxS, PFOS, PFOA, PFNA and PFDA concentrations (**Figure 4, Table 2**). Low values of urea:creatinine indicate a fasting state (i.e. urea:creatinine ≤ 47.5 using molar concentrations), females in a fasting state had PFAS concentrations that were 1.18-1.47 fold higher than in feeding females (urea:creatinine > 47.5) . When fasting, bears can conserve their protein pool by recycling urea

342 nitrogen into plasma proteins (Nelson et al., 1975) and previous studies in polar bears have
343 reported increased concentrations of β-globulins in plasma of fasting polar bears (Cattet, 2000).
344 Considering that PFAS bind to proteins, an increased proportion of proteins in blood could
345 explain the higher PFHxS, PFOS, PFOA, PFNA and PFDA concentrations in fasting female
346 polar bears. Yet, in hooded seals and human, protein concentrations in plasma were not an
347 explanatory factor for PFAS variation (Butenhoff et al., 2012; Grønnestad et al., 2016). An
348 alternative explanation could be that metabolic rate and contaminant excretion are reduced in
349 fasting animals (Aas et al., 2014). In female polar bears we observed no significant relationships
350 between BCI and PFASs although BCI was selected among the best models (ΔAICc<2) for
351 PFHxS, PFOA, PFDoDA and PFTrDA (**Table S3**). While body condition was a stronger
352 predictor than diet for the concentrations of lipophilic pollutants (Tartu et al., 2017), PFAS
353 concentrations were not affected by body condition. Noticeably, BCI was not related to
354 urea:creatinine (β=0.38, 95% CI [-2.95, 2.19]) which could result from a mismatch between
355 blood parameters (e.g. urea and creatinine) and the lag for adipose tissue accumulation. Indeed,
356 in spring, polar bears are on average thinner after a winter period with low prey availability but
357 they are also feeding as ringed seal pups are abundant (Cattet, 2000; Derocher et al., 1990;
358 Lønø, 1970). In contrast, in autumn, polar bears still have large fat reserves after the intensive
359 feeding period in spring and early summer, but a larger proportion of individuals may be fasting
360 due to the absence of sea ice and thus less access to seals (Cattet, 2000; Derocher et al., 1990;
361 Lønø, 1970). These seasonal variations in body fat and metabolic state were also observed in
362 the females from the present study; females were fatter and a larger proportion were fasting in
363 September compared to April (BCI: β= 0.55, 95%CI [0.32; 0.79] and urea:creatinine: β= -0.45,
364 95%CI [-0.75; -0.15]).

365 ***PFASs in relation to sea ice condition***

366 Concentrations of PFOS and C_9-C_{13} PFCAs were higher in polar bears from eastern part of
367 Svalbard, where habitat quality was higher than in females from the western part of Svalbard,
368 where the habitat quality was poorer (**Figure 1B-S1, Table 2**). These results support previous
369 findings showing that PFAS concentrations in polar bears increased as home ranges covered
370 areas more eastwards of Svalbard (van Beest et al., 2015). This pattern could result from the
371 combination of differences in energy need, prey availability and abiotic factors affecting PFAS
372 cycling (e.g. sea ice extent). The eastern coast of Svalbard experiences large amplitude of sea
373 ice retreat during summer in comparison to the western coast that is often ice free year-long
374 (Hop et al., 2000; Pavlova et al., 2014; Vinje and Kvambekk, 1991). The home range size might
375 also influence contaminant intake, as a larger home range requires greater energy expenditure
376 and thus higher food intake leading to a higher total intake of contaminants (Mauritzen et al.,
377 2001; Olsen et al., 2003).

378 Diet variation in polar bears from different areas from Svalbard may also affect their PFAS
379 uptake. In a previous study using the same data set (Tartu et al., 2016), the authors divided
380 captured females into three groups according to the geographical area they were captured in
381 (see Figure 1 in Tartu et al., 2016). In Svalbard, large variations in sea-ice cover occur between
382 the north-west (poor sea-ice cover) and the south-east (large amplitude of sea-ice cover),
383 whereas sea ice around Nordauslandet and south Spitsbergen is extended and stable. Variations
384 in diet proxies according to the three geographical areas in Svalbard have been described in
385 details previously (Tartu et al., 2016). In this study, we used habitat quality based on RSF to
386 divide geographically the captured females (**Figure S1**). Our results indicate that females using
387 the eastern, high quality habitat had higher δ^{15}N values in red blood cells (LSM, β=0.51, 95%CI

388 [0.17; 0.85]) and were in better body condition (LSM, β=0.47, 95%CI [0.18; 0.76]) than females
389 using the western, lower quality habitat. Hence, our findings indicate that females using the
390 eastern habitat could have access to a higher quantity of preferred prey such as ringed and
391 bearded seals. Ringed and bearded seals are more contaminated than terrestrial prey thought to
392 be consumed in larger proportions by females using the poorer quality western habitat (Müller
393 et al., 2011; Tartu et al., 2016). An access to different type of prey between females using
394 different habitats could explain the higher concentration of more bioaccumulative PFASs such
395 as PFOS and C_9-C_{13} PFCAs in eastern females (Kelly et al., 2009). These geographic
396 differences present an ecological and physiological conundrum: bears that choose to use regions
397 where the prey base is of higher quality-seals in eastern Svalbard, are inadvertently assimilating
398 prey that are highly contaminated; thus, although they are fatter, their vulnerability to being
399 contaminated is dramatically different.

400 Interestingly, we observed higher concentrations of PFNA and PFDA in autumn compared to
401 spring (**Table 2**). Yet, this result only appeared significant in females captured in the eastern
402 habitat (LSM, PFNA: β= -0.27, 95%CI [-0.45; -0.09] and PFDA: β= -0.23, 95%CI [-0.41; -
403 0.06], **Figure 5A**) and not in females captured in the western habitat (LSM, PFNA: β= -0.01,
404 95%CI [-0.27; 0.25] and PFDA: β= 0.10, 95%CI [-0.14; 0.33], **Figure 5A**). Although δ^{15}N
405 values in red blood cells were not season dependent in females from the eastern habitat (LSM,
406 β= 0.08, 95%CI [-0.26; 0.41]), plasma δ^{15}N and δ^{13}C values were higher in spring compared to
407 autumn (LSM, β= 0.60, 95%CI [0.22; 0.98] and β=1.32, 95%CI [0.94; 1.69], respectively,
408 **Figure 5B**). Consequently, in summer, female polar bears from eastern Svalbard could ingest
409 a larger proportion of lipid rich terrestrial food source such as waterfowl eggs (Tartu et al.,
410 2016). Feeding on terrestrial species would result in a decrease in δ^{13}C values and a

411 modification of PFAS composition as PFAS proportions in terrestrial prey (e.g waterfowl eggs)
412 are likely different from those in seal species (Eriksson et al., 2016; Tomy et al., 2004).

413 In female polar bears, relationships between PFAS concentrations, habitat quality and season
414 may also be influenced by other abiotic factors. PFASs are generally more concentrated in
415 surface snow than in seawater, due to a dilution effect (Kwok et al., 2013). In addition, the
416 surface load (ng/m^2) of C_6-C_8 PFSAs and C_{10}-C_{12} PFCAs increases in the snowpack during snow
417 melting (Codling et al., 2014). In areas where sea ice cover is more extended, PFASs and their
418 precursors that are transported in the atmosphere are deposited on the sea ice from which they
419 are released into the seawater during melting periods. Pollutants released in seawater are then
420 assimilated by the food web. The sea ice melt is followed by a sharp increase in phytoplankton
421 biomass. Once the pollutants are assimilated by phytoplankton, the latter are consumed by the
422 copepod *Calanus glacialis*, a key Arctic planktonic herbivore, which is an important food item
423 for higher trophic levels (Leu et al., 2011; Søreide et al., 2010). In contrast, in areas with less
424 or no sea ice, PFAS deposition will more rapidly be diluted into seawater. Concentrations of
425 several PFAS are therefore expected to be higher in food webs from areas where sea ice extent
426 is subjected to a larger amplitude, such as the eastern habitat. This could also contribute to the
427 observed seasonal variation in PFAS concentrations between females using the eastern versus
428 those using the western habitats.

429 ***PFOS and breeding status***

430 Breeding status predicted PFOS concentrations in plasma (**Table 2**). We observed higher PFOS
431 concentrations in females with cubs of the year (COYs) than in solitary females (**Table 2**).
432 Although the other PFASs did not vary between breeding statuses, C_{10}-C_{13} PFCAs tended to be

higher in females with COYs than in solitary ones. PFAS concentrations in females with yearlings were not different from any of the latter two groups (**Table 2**).

The high plasma PFOS concentrations in females with COYs could be related to an increased protein synthesis for milk production coupled to a low metabolic state. Indeed, female polar bears produce large quantities of milk for COYs (Arnould and Ramsay, 1994) and during lactation, the activity of some lipoproteins, such as the lipoprotein lipase (LPL) increases (Iverson et al., 1995; McBride and Korn, 1963; Mellish et al., 1999). LPL is critical for the uptake and secretion of FA in milk (Hamosh et al., 1970). We therefore postulate that increased lipoprotein synthesis related to lactation will increase the protein pool in females' body, which will result in a higher proportion of PFOS bound proteins in plasma. In addition, females with COYs may have been fasting for up to 6 months when they emerge from their dens in March to April (Andersen et al., 2012) and as observed from the present results, a fasting state is related to higher PFAS concentrations (**Table 2**). The high PFOS concentrations in females with COYs are unlikely related to differences in feeding patterns between the females of different breeding statuses. Indeed, females with COYs rather feed at a lower trophic level than solitary females, although results were not statistically significant (Tartu et al., 2016). According to numerous studies on murine and simian models, PFOS is highly toxic to mammals (Lau et al., 2004). Consequently, females with COYs could be more at risk considering they have an increased energy demand and are in poorer body condition compared to solitary females (data not shown).

PFOA and age

In the present study, the age of female polar bears predicted a decrease of 1.14 ng/g ww per year in PFOA concentrations whereas other PFASs were not related to age (**Table 2**). The relationships between PFAS and age are inconsistent across wildlife. In polar bears from four

other subpopulations, hepatic concentrations of PFOS and C_8-C_{14} PFCAs increased with age (Smithwick et al., 2005a, 2005b), blood PFOA concentrations and age were not related in southern Beaufort Sea polar bears (Bentzen et al., 2008) and blood PFSA increased with increasing age in East Greenland polar bears (Greaves et al., 2012). In other marine mammals, such as ringed seals and beluga whales (*Delphinapterus leucas*), plasma PFAS concentrations were not related to age (Butt et al., 2008; Kelly et al., 2009; Routti et al., 2016), whereas in bottlenose dolphins (*Tursiops truncatus*), plasma concentrations of PFSAs decreased with age (Fair et al., 2012). The reason for age-PFOA relationships in polar bears is unclear and may be related to other confounding factors (e.g. age-related hormonal changes) not taken into account in this study.

CONCLUSIONS

Considering all the potential health effects of PFAS, it is important to increase knowledge on the underlying drivers of PFAS concentrations in polar bears. This study demonstrates that diet is the strongest predictor for circulating PFAS concentrations in Svalbard female polar bears, with individuals feeding at a higher trophic level and more marine prey being more exposed to PFASs. Diet is also a likely factor explaining seasonal and spatial differences in plasma PFAS concentrations in polar bears from Svalbard. PFAS concentrations were higher in fasting than in feeding female polar bears. The higher PFOS levels in females with COYs are likely related to both metabolic state and milk production. In conclusion, our findings suggest that feeding on high trophic level marine prey, fasting and having COYs are all factors that may lead to high PFAS exposure among adult female polar bears. The health effects of PFAS are numerous, but considering their disruption potential on lipid metabolism and the importance of storage and utilization of lipids in Arctic wildlife, further studies should focus on the relationships between

PFAS and energetic metabolism of polar bears and whether climate changes reinforces or not these relationships.

ACKNOWLEDGEMENT

We thank Rupert Krapp (NPI, Longyearbyen), the helicopter (Airlift) and Lance crews for logistic help. We acknowledge Mahin Karimi (NMBU, Oslo), Martin S. Andersen (NPI, Tromsø) and Astrid Kolind Riemer (NTNU, Trondheim) for their help in pollutant determination. We also would like to thank the two anonymous reviewers for their careful reading of our manuscript and their insightful comments. This study was financed by the Research Council of Norway (grant 216568/E10 to H.R) and the Norwegian Polar Institute. The fieldwork was also supported by the Centre for Ice, Climate and Ecosystems (ICE) at the Norwegian Polar Institute and World Wildlife Fund. J. M. Welker's participation was supported in part by his Norwegian Fulbright Distinguished US Arctic Chairship.

REFERENCES

Aars, J., Andersen, M., Brenière, A., Blanc, S., 2015. White-beaked dolphins trapped in the ice and eaten by polar bears. Polar Res. 34, 26612. doi:10.3402/polar.v34.26612

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. doi:10.1021/es503147n

Andersen, M., Derocher, A.E., Wiig, Ø., Aars, J., 2012. Polar bear (Ursus maritimus) maternity den distribution in Svalbard, Norway. Polar Biol. 35, 499–508. doi:10.1007/s00300-011-1094-y

Armitage, J.M., Schenker, U., Scheringer, M., Martin, J.W., MacLeod, M., Cousins, I.T., 2009. Modeling the Global Fate and Transport of Perfluorooctane Sulfonate (PFOS) and Precursor Compounds in Relation to Temporal Trends in Wildlife Exposure. Environ. Sci. Technol. 43, 9274–9280. doi:10.1021/es901448p

Arnould, J.P.Y., Ramsay, M.A., 1994. Milk production and milk consumption in polar bears during the ice-free period in western Hudson Bay. Can. J. Zool. 72, 1365–1370. doi:10.1139/z94-180

Atkinson, S.N., Ramsay, M.A., 1995. The Effects of Prolonged Fasting of the Body Composition and Reproductive Success of Female Polar Bears (Ursus maritimus). Funct. Ecol. 9, 559–567. doi:10.2307/2390145

Barton, K., 2016. MuMIn: Multi-Model Inference.

Bentzen, T.W., Muir, D.C.G., Amstrup, S.C., O'Hara, T.M., 2008. Organohalogen concentrations in blood and adipose tissue of Southern Beaufort Sea polar bears. Sci. Total Environ. 406, 352–367. doi:10.1016/j.scitotenv.2008.07.030

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. Integr. Environ. Assess. Manag. 7, 513–541. doi:10.1002/ieam.258

Budge, S.M., Iverson, S.J., Koopman, H.N., 2006. Studying Trophic Ecology in Marine Ecosystems Using Fatty Acids: A Primer on Analysis and Interpretation. Mar. Mammal Sci. 22, 759–801. doi:10.1111/j.1748-7692.2006.00079.x

Budge, S.M., Penney, S.N., Lall, S.P., 2012. Estimating diets of Atlantic salmon (Salmo salar) using fatty acid signature analyses; validation with controlled feeding studies. Can. J. Fish. Aquat. Sci. 69, 1033–1046. doi:10.1139/f2012-039

Burnham, K.P., Anderson, D.R. (Eds.), 2004. Model Selection and Multimodel Inference. Springer New York, New York, NY.

Burnham, K.P., Anderson, D.R., 2003. Model Selection and Multimodel Inference: A Practical Information-Theoretic Approach, 2nd ed. Springer Science & Business Media, New York, NY.

Butenhoff, J.L., Pieterman, E., Ehresman, D.J., Gorman, G.S., Olsen, G.W., Chang, S.-C., Princen, H.M.G., 2012. Distribution of perfluorooctanesulfonate and perfluorooctanoate into human plasma lipoprotein fractions. Toxicol. Lett. 210, 360–365. doi:10.1016/j.toxlet.2012.02.013

Butt, C.M., Mabury, S.A., Kwan, M., Wang, X., Muir, D.C.G., 2008. Spatial trends of perfluoroalkyl compounds in ringed seals (Phoca hispida) from the Canadian Arctic. Environ. Toxicol. Chem. 27, 542–553. doi:10.1897/07-428.1

Bytingsvik, J., Lie, E., Aars, J., Derocher, A.E., Wiig, Ø., Jenssen, B.M., 2012a. PCBs and OH-PCBs in polar bear mother–cub pairs: A comparative study based on plasma levels in 1998 and 2008. Sci. Total Environ. 417–418, 117–128. doi:10.1016/j.scitotenv.2011.12.033

Bytingsvik, J., van Leeuwen, S.P.J., Hamers, T., Swart, K., Aars, J., Lie, E., Nilsen, E.M.E., Wiig, Ø., Derocher, A.E., Jenssen, B.M., 2012b. Perfluoroalkyl substances in polar bear mother–cub

pairs: A comparative study based on plasma levels from 1998 and 2008. Environ. Int. 49, 92–99. doi:10.1016/j.envint.2012.08.004
Calvert, W., Ramsay, M.A., 1998. Evaluation of age determination of polar bears by counts of cementum growth layer groups. Ursus 449–453.
Cattet, M., 2000. Biochemical and physiological aspects of obesity, high fat diet, and prolonged fasting in free-ranging polar bears (PhD dissertation). University of Saskatchewan, Saskatchewan.
Cattet, M.R.., Caulkett, N.A., Obbard, M.E., Stenhouse, G.B., 2002. A body-condition index for ursids. Can. J. Zool. 80, 1156–1161. doi:10.1139/z02-103
Cherry, S.G., Derocher, A.E., Stirling, I., Richardson, E.S., 2009. Fasting physiology of polar bears in relation to environmental change and breeding behavior in the Beaufort Sea. Polar Biol. 32, 383–391. doi:10.1007/s00300-008-0530-0
Codling, G., Halsall, C., Ahrens, L., Del Vento, S., Wiberg, K., Bergknut, M., Laudon, H., Ebinghaus, R., 2014. The fate of per- and polyfluoroalkyl substances within a melting snowpack of a boreal forest. Environ. Pollut. 191, 190–198. doi:10.1016/j.envpol.2014.04.032
Derocher, A.E., Nelson, R.A., Stirling, I., Ramsay, M.A., 1990. Effects of Fasting and Feeding on Serum Urea and Serum Creatinine Levels in Polar Bears. Mar. Mammal Sci. 6, 196–203. doi:10.1111/j.1748-7692.1990.tb00243.x
Derocher, A.E., Wiig, Ø., 2002. Postnatal growth in body length and mass of polar bears (Ursus maritimus) at Svalbard. J. Zool. 256, 343–349. doi:10.1017/S0952836902000377
Dietz, R., Gustavson, K., Sonne, C., Desforges, J.-P., Rigét, F.F., Pavlova, V., McKinney, M.A., Letcher, R.J., 2015. Physiologically-based pharmacokinetic modelling of immune, reproductive and carcinogenic effects from contaminant exposure in polar bears (Ursus maritimus) across the Arctic. Environ. Res. 140, 45–55. doi:10.1016/j.envres.2015.03.011
Dray, A.B., Dufour, S., 2007. The ade4 package: implementing the duality diagram for ecologists. J. Stat. Softw. 22, 1–20.
du Prel, J.-B., Hommel, G., Röhrig, B., Blettner, M., 2009. Confidence Interval or P-Value? Dtsch. Ärztebl. Int. 106, 335–339. doi:10.3238/arztebl.2009.0335
Dyck, M.G., Romberg, S., 2007. Observations of a wild polar bear (Ursus maritimus) successfully fishing Arctic charr (Salvelinus alpinus) and Fourhorn sculpin (Myoxocephalus quadricornis). Polar Biol. 30, 1625–1628.
Eriksson, U., Roos, A., Lind, Y., Hope, K., Ekblad, A., Kärrman, A., 2016. Comparison of PFASs contamination in the freshwater and terrestrial environments by analysis of eggs from osprey (Pandion haliaetus), tawny owl (Strix aluco), and common kestrel (Falco tinnunculus). Environ. Res. 149, 40–47. doi:10.1016/j.envres.2016.04.038
Fair, P.A., Houde, M., Hulsey, T.C., Bossart, G.D., Adams, J., Balthis, L., Muir, D.C.G., 2012. Assessment of perfluorinated compounds (PFCs) in plasma of bottlenose dolphins from two southeast US estuarine areas: Relationship with age, sex and geographic locations. Mar. Pollut. Bull. 64, 66–74. doi:10.1016/j.marpolbul.2011.10.022
Gelman, A., Hill, J., 2006. Data Analysis Using Regression and Multilevel/Hierarchical Models. Cambridge University Press.
Gormezano, L.J., Rockwell, R.F., 2015. The Energetic Value of Land-Based Foods in Western Hudson Bay and Their Potential to Alleviate Energy Deficits of Starving Adult Male Polar Bears. PLoS ONE 10, e0128520. doi:10.1371/journal.pone.0128520
Gormezano, L.J., Rockwell, R.F., 2013. What to eat now? Shifts in polar bear diet during the ice-free season in western Hudson Bay. Ecol. Evol. 3, 3509–3523. doi:10.1002/ece3.740

586 Greaves, A.K., Letcher, R.J., Sonne, C., Dietz, R., Born, E.W., 2012. Tissue-Specific Concentrations and
587 Patterns of Perfluoroalkyl Carboxylates and Sulfonates in East Greenland Polar Bears.
588 Environ. Sci. Technol. 46, 11575–11583. doi:10.1021/es303400f
589 Grønnestad, R., Villanger, G.D., Polder, A., Kovacs, K.M., Lydersen, C., Jenssen, B.M., Borgå, K., 2016.
590 Maternal transfer of perfluoroalkyl substances in hooded seals. Environ. Toxicol. Chem. 36,
591 763–770. doi:10.1002/etc.3623
592 Guruge, K.S., Yeung, L.W.Y., Yamanaka, N., Miyazaki, S., Lam, P.K.S., Giesy, J.P., Jones, P.D.,
593 Yamashita, N., 2006. Gene Expression Profiles in Rat Liver Treated With Perfluorooctanoic
594 Acid (PFOA). Toxicol. Sci. 89, 93–107. doi:10.1093/toxsci/kfj011
595 Hamosh, M., Clary, T.R., Chernick, S.S., Scow, R.O., 1970. Lipoprotein lipase activity of adipose and
596 mammary tissue and plasma triglyceride in pregnant and lactating rats. Biochim. Biophys.
597 Acta BBA - Lipids Lipid Metab. 210, 473–482. doi:10.1016/0005-2760(70)90044-5
598 Haukås, M., Berger, U., Hop, H., Gulliksen, B., Gabrielsen, G.W., 2007. Bioaccumulation of per- and
599 polyfluorinated alkyl substances (PFAS) in selected species from the Barents Sea food web.
600 Environ. Pollut. 148, 360–371. doi:10.1016/j.envpol.2006.09.021
601 Hobson, K.A., 1999. Tracing origins and migration of wildlife using stable isotopes: a review.
602 Oecologia 120, 314–326. doi:10.1007/s004420050865
603 Hobson, K.A., Schell, D.M., Renouf, D., Noseworthy, E., 1996. Stable carbon and nitrogen isotopic
604 fractionation between diet and tissues of captive seals: implications for dietary
605 reconstructions involving marine mammals. Can. J. Fish. Aquat. Sci. 53, 528–533.
606 Hop, H., Poltermann, M., Lønne, O.J., Falk-Petersen, S., Korsnes, R., Budgell, W.P., 2000. Ice
607 amphipod distribution relative to ice density and under-ice topography in the northern
608 Barents Sea. Polar Biol. 23, 357–367. doi:10.1007/s003000050456
609 Houde, M., De Silva, A.O., Muir, D.C.G., Letcher, R.J., 2011. Monitoring of Perfluorinated Compounds
610 in Aquatic Biota: An Updated Review. Environ. Sci. Technol. 45, 7962–7973.
611 doi:10.1021/es104326w
612 Iversen, M., Aars, J., Haug, T., Alsos, I.G., Lydersen, C., Bachmann, L., Kovacs, K.M., 2013. The diet of
613 polar bears (Ursus maritimus) from Svalbard, Norway, inferred from scat analysis. Polar Biol.
614 36, 561–571. doi:10.1007/s00300-012-1284-2
615 Iverson, S.A., Gilchrist, H.G., Smith, P.A., Gaston, A.J., Forbes, M.R., 2014. Longer ice-free seasons
616 increase the risk of nest depredation by polar bears for colonial breeding birds in the
617 Canadian Arctic. Proc. R. Soc. Lond. B Biol. Sci. 281, 20133128. doi:10.1098/rspb.2013.3128
618 Iverson, S.J., Field, C., Don Bowen, W., Blanchard, W., 2004. Quantitative fatty acid signature analysis:
619 a new method of estimating predator diets. Ecol. Monogr. 74, 211–235. doi:10.1890/02-4105
620 Iverson, S.J., Hamosh, M., Bowen, W.D., 1995. Lipoprotein lipase activity and its relationship to high
621 milk fat transfer during lactation in grey seals. J. Comp. Physiol. B 165, 384–395.
622 doi:10.1007/BF00387309
623 Jensen, A.A., Leffers, H., 2008. Emerging endocrine disrupters: perfluoroalkylated substances. Int. J.
624 Androl. 31, 161–169. doi:10.1111/j.1365-2605.2008.00870.x
625 Kelly, B.C., Ikonomou, M.G., Blair, J.D., Surridge, B., Hoover, D., Grace, R., Gobas, F.A.P.C., 2009.
626 Perfluoroalkyl Contaminants in an Arctic Marine Food Web: Trophic Magnification and
627 Wildlife Exposure. Environ. Sci. Technol. 43, 4037–4043. doi:10.1021/es9003894
628 Key, B.D., Howell, R.D., Criddle, C.S., 1997. Fluorinated Organics in the Biosphere. Environ. Sci.
629 Technol. 31, 2445–2454. doi:10.1021/es961007c
630 Kwok, K.Y., Yamazaki, E., Yamashita, N., Taniyasu, S., Murphy, M.B., Horii, Y., Petrick, G., Kallerborn,
631 R., Kannan, K., Murano, K., Lam, P.K.S., 2013. Transport of Perfluoroalkyl substances (PFAS)
632 from an arctic glacier to downstream locations: Implications for sources. Sci. Total Environ.
633 447, 46–55. doi:10.1016/j.scitotenv.2012.10.091

634 Lau, C., Anitole, K., Hodes, C., Lai, D., Pfahles-Hutchens, A., Seed, J., 2007. Perfluoroalkyl Acids: A
635 Review of Monitoring and Toxicological Findings. Toxicol. Sci. 99, 366–394.
636 doi:10.1093/toxsci/kfm128
637 Lau, C., Butenhoff, J.L., Rogers, J.M., 2004. The developmental toxicity of perfluoroalkyl acids and
638 their derivatives. Toxicol. Appl. Pharmacol. 198, 231–241. doi:10.1016/j.taap.2003.11.031
639 Legendre, P., Anderson, M.J., 1999. Distance-Based Redundancy Analysis: Testing Multispecies
640 Responses in Multifactorial Ecological Experiments. Ecol. Monogr. 69, 1–24.
641 doi:10.1890/0012-9615(1999)069[0001:DBRATM]2.0.CO;2
642 Lenth, R.V., Hervé, M., 2015. lsmeans: Least-Squares Means.
643 Letcher, R.J., Bustnes, J.O., Dietz, R., Jenssen, B.M., Jørgensen, E.H., Sonne, C., Verreault, J., Vijayan,
644 M.M., Gabrielsen, G.W., 2010. Exposure and effects assessment of persistent organohalogen
645 contaminants in arctic wildlife and fish. Sci. Total Environ., Levels, trends and effects of
646 legacy and new persistent organic pollutants in the Arctic: An AMAP Assessment 408, 2995–
647 3043. doi:10.1016/j.scitotenv.2009.10.038
648 Leu, E., Søreide, J.E., Hessen, D.O., Falk-Petersen, S., Berge, J., 2011. Consequences of changing sea-
649 ice cover for primary and secondary producers in the European Arctic shelf seas: Timing,
650 quantity, and quality. Prog. Oceanogr., Arctic Marine Ecosystems in an Era of Rapid Climate
651 Change 90, 18–32. doi:10.1016/j.pocean.2011.02.004
652 Lone, K., Aars, J., Ims, R.A., 2012. Site fidelity of Svalbard polar bears revealed by mark-recapture
653 positions. Polar Biol. 36, 27–39. doi:10.1007/s00300-012-1235-y
654 Lønø, O., 1970. The Polar bear (Ursus maritimus Phipps) in the Svalbard area. 118.
655 Lukacs, P.M., Burnham, K.P., Anderson, D.R., 2009. Model selection bias and Freedman's paradox.
656 Ann. Inst. Stat. Math. 62, 117–125. doi:10.1007/s10463-009-0234-4
657 Martin, J.W., Mabury, S.A., Solomon, K.R., Muir, D.C.G., 2003a. Dietary accumulation of
658 perfluorinated acids in juvenile rainbow trout (Oncorhynchus mykiss). Environ. Toxicol.
659 Chem. 22, 189–195. doi:10.1002/etc.5620220125
660 Martin, J.W., Mabury, S.A., Solomon, K.R., Muir, D.C.G., 2003b. Bioconcentration and tissue
661 distribution of perfluorinated acids in rainbow trout (Oncorhynchus mykiss). Environ. Toxicol.
662 Chem. 22, 196–204. doi:10.1002/etc.5620220126
663 Martin, J.W., Smithwick, M.M., Braune, B.M., Hoekstra, P.F., Muir, D.C.G., Mabury, S.A., 2004.
664 Identification of Long-Chain Perfluorinated Acids in Biota from the Canadian Arctic. Environ.
665 Sci. Technol. 38, 373–380. doi:10.1021/es034727+
666 Mauritzen, M., Derocher, A.E., Wiig, Ø., 2001. Space-use strategies of female polar bears in a
667 dynamic sea ice habitat. Can. J. Zool. 79, 1704–1713. doi:10.1139/z01-126
668 McBride, O.W., Korn, E.D., 1963. The lipoprotein lipase of mammary gland and the correlation of its
669 activity to lactation. J. Lipid Res. 4, 17–20.
670 McKinney, M.A., Letcher, R.J., Aars, J., Born, E.W., Branigan, M., Dietz, R., Evans, T.J., Gabrielsen,
671 G.W., Muir, D.C.G., Peacock, E., Sonne, C., 2011. Regional Contamination versus Regional
672 Dietary Differences: Understanding Geographic Variation in Brominated and Chlorinated
673 Contaminant Levels in Polar Bears. Environ. Sci. Technol. 45, 896–902.
674 doi:10.1021/es102781b
675 Mellish, J.E., Iverson, S.J., Bowen, W.D., Hammill, M.O., 1999. Fat transfer and energetics during
676 lactation in the hooded seal: the roles of tissue lipoprotein lipase in milk fat secretion and
677 pup blubber deposition. J. Comp. Physiol. B 169, 377–390. doi:10.1007/s003600050234
678 Muir, D.C.G., de Wit, C.A., 2010. Trends of legacy and new persistent organic pollutants in the
679 circumpolar arctic: Overview, conclusions, and recommendations. Sci. Total Environ., Levels,
680 trends and effects of legacy and new persistent organic pollutants in the Arctic: An AMAP
681 Assessment 408, 3044–3051. doi:10.1016/j.scitotenv.2009.11.032

682 Müller, C.E., De Silva, A.O., Small, J., Williamson, M., Wang, X., Morris, A., Katz, S., Gamberg, M.,
683 Muir, D.C.G., 2011. Biomagnification of Perfluorinated Compounds in a Remote Terrestrial
684 Food Chain: Lichen–Caribou–Wolf. Environ. Sci. Technol. 45, 8665–8673.
685 doi:10.1021/es201353v
686 Nelson, R., Jones, J., Wahner, H., McGill, D., Code, C., 1975. Nitrogen metabolism in bears: urea
687 metabolism in summer starvation and in winter sleep and role of urinary bladder in water
688 and nitrogen conservation. Mayo Clin. Proc. 50, 141–146.
689 Nelson, R.A., Beck, T., Steiger, D.L., 1984. Ratio of serum urea to serum creatinine in wild black bears
690 | Science 226, 841–842.
691 Olsen, G.H., Mauritzen, M., Derocher, A.E., Sørmo, E.G., Skaare, J.U., Wiig, Ø., Jenssen, B.M., 2003.
692 Space-Use Strategy Is an Important Determinant of PCB Concentrations in Female Polar
693 Bears in the Barents Sea. Environ. Sci. Technol. 37, 4919–4924. doi:10.1021/es034380a
694 Pavlova, O., Pavlov, V., Gerland, S., 2014. The impact of winds and sea surface temperatures on the
695 Barents Sea ice extent, a statistical approach. J. Mar. Syst. 130, 248–255.
696 doi:10.1016/j.jmarsys.2013.02.011
697 Pedersen, K.E., Letcher, R.J., Sonne, C., Dietz, R., Styrishave, B., 2016. Per- and polyfluoroalkyl
698 substances (PFASs) – New endocrine disruptors in polar bears (Ursus maritimus)? Environ.
699 Int. 96, 180–189. doi:10.1016/j.envint.2016.07.015
700 Pinheiro, J., Bates, D., Debroy, S., Sarkar, D., R core team, 2015. nlme: Linear and Nonlinear Mixed
701 Effects Models.
702 Prevedouros, K., Cousins, I.T., Buck, R.C., Korzeniowski, S.H., 2006. Sources, Fate and Transport of
703 Perfluorocarboxylates. Environ. Sci. Technol. 40, 32–44. doi:10.1021/es0512475
704 Prop, J., Aars, J., Bårdsen, B.-J., Hanssen, S.A., Bech, C., Bourgeon, S., de Fouw, J., Gabrielsen, G.W.,
705 Lang, J., Noreen, E., Oudman, T., Sittler, B., Stempniewicz, L., Tombre, I., Wolters, E., Moe, B.,
706 2015. Climate change and the increasing impact of polar bears on bird populations. Front.
707 Ecol. Evol. 3, 1–12. doi:10.3389/fevo.2015.00033
708 Prop, J., Oudman, T., Spanje, T.M. van, Wolters, E.H., 2013. Patterns of predation of Pink-footed
709 Goose nests by polar bear. Ornis Nor. 36, 38–46.
710 R Core Team, 2016. R: A Language and Environment for Statistical Computing, R Foundation for
711 Statistical Computing. Vienna, Austria.
712 Ramette, A., 2007. Multivariate analyses in microbial ecology. FEMS Microbiol. Ecol. 62, 142–160.
713 doi:10.1111/j.1574-6941.2007.00375.x
714 Ramsay, M., Stirling, I., 1988. Reproductive biology and ecology of female polar bears (Ursus
715 maritimus). J. Zool. 214, 601–633.
716 Rode, K.D., Stricker, C.A., Erlenbach, J., Robbins, C.T., Cherry, S.G., Newsome, S.D., Cutting, A.,
717 Jensen, S., Stenhouse, G., Brooks, M., Hash, A., Nicassio, N., 2016. Isotopic Incorporation and
718 the Effects of Fasting and Dietary Lipid Content on Isotopic Discrimination in Large
719 Carnivorous Mammals. Physiol. Biochem. Zool. 89, 182–197. doi:10.1086/686490
720 Rogers, M.C., Peacock, E., Simac, K., O'Dell, M.B., Welker, J.M., 2015. Diet of female polar bears in
721 the southern Beaufort Sea of Alaska: evidence for an emerging alternative foraging strategy
722 in response to environmental change. Polar Biol. 38, 1035–1047. doi:10.1007/s00300-015-
723 1665-4
724 Routti, H., Gabrielsen, G.W., Herzke, D., Kovacs, K.M., Lydersen, C., 2016. Spatial and temporal trends
725 in perfluoroalkyl substances (PFASs) in ringed seals (Pusa hispida) from Svalbard. Environ.
726 Pollut. 214, 230–238. doi:10.1016/j.envpol.2016.04.016
727 Shoeib, M., Harner, T., Vlahos, P., 2006. Perfluorinated Chemicals in the Arctic Atmosphere. Environ.
728 Sci. Technol. 40, 7577–7583. doi:10.1021/es0618999

729 Smith, P.A., Elliott, K.H., Gaston, A.J., Gilchrist, H.G., 2010. Has early ice clearance increased
730 predation on breeding birds by polar bears? Polar Biol. 33, 1149–1153. doi:10.1007/s00300-
731 010-0791-2
732 Smithwick, M., Mabury, S.A., Solomon, K.R., Sonne, C., Martin, J.W., Born, E.W., Dietz, R., Derocher,
733 A.E., Letcher, R.J., Evans, T.J., Gabrielsen, G.W., Nagy, J., Stirling, I., Taylor, M.K., Muir, D.C.G.,
734 2005a. Circumpolar Study of Perfluoroalkyl Contaminants in Polar Bears (Ursus maritimus).
735 Environ. Sci. Technol. 39, 5517–5523. doi:10.1021/es048309w
736 Smithwick, M., Muir, D.C.G., Mabury, S.A., Solomon, K.R., Martin, J.W., Sonne, C., Born, E.W.,
737 Letcher, R.J., Dietz, R., 2005b. Perflouroalkyl contaminants in liver tissue from East Greenland
738 polar bears (Ursus maritimus). Environ. Toxicol. Chem. 24, 981–986. doi:10.1897/04-258R.1
739 Sonne, C., 2010. Health effects from long-range transported contaminants in Arctic top predators: An
740 integrated review based on studies of polar bears and relevant model species. Environ. Int.
741 36, 461–491. doi:10.1016/j.envint.2010.03.002
742 Sonne, C., Bossi, R., Dietz, R., Leifsson, P.S., Rigét, F.F., Born, E.W., 2008. Potential correlation
743 between perfluorinated acids and liver morphology in East Greenland polar bears (Ursus
744 maritimus). Toxicol. Environ. Chem. 90, 275–283. doi:10.1080/02772240701391629
745 Sonne, C., Letcher, R.J., Bechshøft, T.Ø., Rigét, F.F., Muir, D.C.G., Leifsson, P.S., Born, E.W., Hyldstrup,
746 L., Basu, N., Kirkegaard, M., Dietz, R., 2012. Two decades of biomonitoring polar bear health
747 in Greenland: a review. Acta Vet. Scand. 54, 1–7. doi:10.1186/1751-0147-54-S1-S15
748 Søreide, J.E., Leu, E., Berge, J., Graeve, M., Falk-Petersen, S., 2010. Timing of blooms, algal food
749 quality and Calanus glacialis reproduction and growth in a changing Arctic. Glob. Change Biol.
750 16, 3154–3163. doi:10.1111/j.1365-2486.2010.02175.x
751 Stempniewicz, L., 2006. Polar Bear Predatory Behaviour toward Molting Barnacle Geese and Nesting
752 Glaucous Gulls on Spitsbergen. Arctic 59, 247–251.
753 Stempniewicz, L., 1993. The polar bear Ursus maritimus feeding in a seabird colony in Frans Josef
754 Land. Polar Res 12, 33–36.
755 Stempniewicz, L., Kidawa, D., Barcikowski, M., Iliszko, L., 2014. Unusual hunting and feeding
756 behaviour of polar bears on Spitsbergen. Polar Rec. 50, 216–219.
757 Stirling, I., Spencer, C., Andriashek, D., 1989. Immobilization of polar bears (Ursus maritimus) with
758 Telazol® in the Canadian Arctic. J. Wildl. Dis. 25, 159–168.
759 Tartu, S., Bourgeon, S., Aars, J., Andersen, M., Ehrich, D., Thiemann, G.W., Welker, J.M., Routti, H.,
760 2016. Geographical Area and Life History Traits Influence Diet in an Arctic Marine Predator.
761 PLOS ONE 11, e0155980. doi:10.1371/journal.pone.0155980
762 Tartu, S., Bourgeon, S., Aars, J., Andersen, M., Polder, A., Thiemann, G.W., Welker, J.M., Routti, H.,
763 2017. Sea ice-associated decline in body condition leads to increased concentrations of
764 lipophilic pollutants in polar bears (Ursus maritimus) from Svalbard, Norway. Sci. Total
765 Environ. 576, 409–419. doi:10.1016/j.scitotenv.2016.10.132
766 Tartu, S., Gabrielsen, G.W., Blévin, P., Ellis, H., Bustnes, J.O., Herzke, D., Chastel, O., 2014. Endocrine
767 and Fitness Correlates of Long-Chain Perfluorinated Carboxylates Exposure in Arctic Breeding
768 Black-Legged Kittiwakes. Environ. Sci. Technol. 48, 13504–13510. doi:10.1021/es503297n
769 Thiemann, G.W., Iverson, S.J., Stirling, I., 2008. Polar bear diets and arctic marine food webs: insights
770 from fatty acid analysis. Ecol. Monogr. 78, 591–613. doi:10.1890/07-1050.1
771 Tomy, G.T., Budakowski, W., Halldorson, T., Helm, P.A., Stern, G.A., Friesen, K., Pepper, K., Tittlemier,
772 S.A., Fisk, A.T., 2004. Fluorinated Organic Compounds in an Eastern Arctic Marine Food Web.
773 Environ. Sci. Technol. 38, 6475–6481. doi:10.1021/es049620g
774 Tomy, G.T., Pleskach, K., Ferguson, S.H., Hare, J., Stern, G., MacInnis, G., Marvin, C.H., Loseto, L.,
775 2009. Trophodynamics of Some PFCs and BFRs in a Western Canadian Arctic Marine Food
776 Web. Environ. Sci. Technol. 43, 4076–4081. doi:10.1021/es900162n

van Beest, F.M. van, Aars, J., Routti, H., Lie, E., Andersen, M., Pavlova, V., Sonne, C., Nabe-Nielsen, J., Dietz, R., 2015. Spatiotemporal variation in home range size of female polar bears and correlations with individual contaminant load. Polar Biol. 1–11. doi:10.1007/s00300-015-1876-8

Van de Vijver, K.I., Hoff, P.T., Das, K., Van Dongen, W., Esmans, E.L., Jauniaux, T., Bouquegneau, J.-M., Blust, R., De Coen, W., 2003. Perfluorinated Chemicals Infiltrate Ocean Waters: Link between Exposure Levels and Stable Isotope Ratios in Marine Mammals. Environ. Sci. Technol. 37, 5545–5550. doi:10.1021/es0345975

Vinje, T., Kvambekk, Å.S., 1991. Barents Sea drift ice characteristics. Polar Res. 10, 59–68. doi:10.1111/j.1751-8369.1991.tb00635.x

Wania, F., 2007. A Global Mass Balance Analysis of the Source of Perfluorocarboxylic Acids in the Arctic Ocean. Environ. Sci. Technol. 41, 4529–4535. doi:10.1021/es070124c

Table 1: Biological parameters, PFAS concentrations, proxies for diet and metabolic state in 112 female polar bears representing 78 individuals sampled in Svalbard (2012-2013). We show averages and median values followed by the range (min-max). Limits of detection (LOD) and the number of samples for which values were below LOD (n<LOD) are given for PFASs, urea and creatinine. PFASs' abbreviations are followed by their carbon chain length. Metabolic state proxies were measured in 111 females representing 77 individuals. [a]The ratio is in molar concentration, ratios ≤ 47.5 correspond to fasting individuals.

	Spring		Autumn		LOD (n<LOD)
	n	average/median (min; max)	n	average/median (min; max)	
Age	62	13.2/12 (4;28)	50	12.7/10.5 (5;28)	
Body condition index	62	-1.46/-1.42 (-3.09;0.08)	50	-1.17/-0.93 (-2.61;-0.03)	
PFSAs (ng/g wet weight)					
PFHxS (C_6)	62	28.6/27.6 (5.5;65.3)	50	32.4/31.3 (11.0;70.7)	0.200 (0)
PFOS (C_8)	62	221/196.8 (54;593.2)	50	248.7/243.6 (40.1;622.2)	0.200 (0)
PFCAs (ng/g wet weight)					
PFOA (C_8)	62	4.6/4.2 (1;12.4)	50	4.8/4.4 (0.8;13)	0.050 (0)
PFNA (C_9)	62	30.4/27.2 (10;78.8)	50	38.8/35.3 (9.3;90.5)	0.160 (0)
PFDA (C_{10})	62	9.7/8.5 (2.8;25.9)	50	12.2/10.8 (2.2;31.3)	0.200 (0)
PFUnDA (C_{11})	62	20.8/18.4 (7;51.8)	50	24.1/23.6 (3.4;58.1)	0.250 (0)
PFDoDA (C_{12})	62	2.6/2.5 (0.9;6.3)	50	2.8/3 (LOD;7.2)	0.400 (1)
PFTrDA (C_{13})	62	6.9/5.8 (2.2;23.2)	50	7.4/7 (1.3;17.4)	0.500 (0)
Diet proxies					
$\delta^{15}N$ plasma	62	17.9/18 (15.1;19.2)	50	16.7/16.9 (12.4;20.1)	
$\delta^{13}C$ plasma	62	-20.3/-20.2 (-22.3;-19)	50	-21.2/-21 (-23.9;-17.6)	
$\delta^{15}N$ rbc	62	16.0/16.3 (12.7;18.3)	50	15.5/15.7 (12.2;17.6)	
$\delta^{13}C$ rbc	62	-20.0/-19.8 (-22.3;-19)	50	-20.0/-19.9 (-21.8;-18.9)	
Metabolic state					
Urea (mmol/l)	61	6.7/6.5 (LOD; 16.4)	50	5.0/4.12 (LOD;18.7)	3.33 (26)
Creatinine (µmol/l)	61	86.5/82.3 (54.9;159.0)	50	135.2/131.0 (59.3;221.0)	44.50 (0)
Urea:Creatinine[a]	61	83.4/76.3 (11.4;241.3)	50	43.5/27.7 (8.4;145.6)	

Table 2: Relationships between PFAS concentrations and diet proxies as stable nitrogen and carbon isotopes (δ^{15}N and δ^{13}C, respectively) in plasma and red blood cells (rbc), metabolic state (urea:creatinine), habitat quality, sampling season and year, body condition, age and breeding status (defined as solitary females (solitary), females with cubs of the year (with COYs) or females with yearlings (YRLs)). Adult female polar bears were capture in Svalbard (2012-2013). As urea and creatinine concentrations were not available for one female, conditional model averaging analyses were run on 111 samples from 77 individuals except in models including fatty acid principal components (FA PCs: 82 samples from 63 individuals). Values are parameter estimates and 95% confidence intervals derived from conditional model averaging of general linear mixed models that included female identity as a random factor. Values in bold are significantly different from 0 at the 5% level. COYs: cubs of the year.

	PFHxS	PFOS	PFOA	PFNA	PFDA	PFUnDA	PFDoDA	PFTrDA
Intercept (n=111)	**3.30 [3.2; 3.41]**	**5.38 [5.27; 5.49]**	**1.41 [1.32; 1.5]**	**3.45 [3.36; 3.54]**	**2.31 [2.22; 2.41]**	**3.02 [2.91; 3.13]**	**0.91 [0.8; 1.02]**	**1.83 [1.72; 1.94]**
δ^{15}N plasma	**0.11 [0.02; 0.19]**	**0.11 [0.03; 0.19]**	**0.20 [0.10; 0.29]**	0.08 [0; 0.16]	**0.12 [0.04; 0.20]**	**0.22 [0.13; 0.3]**	**0.27 [0.18; 0.36]**	**0.31 [0.22; 0.4]**
δ^{15}N rbc	**0.15 [0.05; 0.24]**	**0.17 [0.08; 0.27]**	**0.26 [0.17; 0.35]**	**0.13 [0.05; 0.22]**	**0.18 [0.09; 0.27]**	**0.28 [0.18; 0.37]**	**0.32 [0.22; 0.42]**	**0.37 [0.27; 0.47]**
δ^{13}C plasma	0.01 [-0.08; 0.10]	-0.03 [-0.11; 0.06]	0 [-0.11; 0.11]	-0.05 [-0.13; 0.04]	-0.03 [-0.12; 0.05]	-0.04 [-0.14; 0.06]	0.01 [-0.09; 0.11]	0.04 [-0.07; 0.15]
δ^{13}C rbc	**0.15 [0.06; 0.25]**	**0.14 [0.05; 0.22]**	**0.23 [0.14; 0.32]**	**0.16 [0.08; 0.24]**	**0.18 [0.10; 0.26]**	**0.20 [0.10; 0.30]**	**0.22 [0.12; 0.32]**	**0.25 [0.14; 0.36]**
Urea:Creatinine	**-0.10 [-0.18; -0.02]**	**-0.11 [-0.18; -0.04]**	**-0.12 [-0.21; -0.04]**	**-0.14 [-0.21; -0.06]**	**-0.12 [-0.19; -0.04]**	-0.07 [-0.15; 0.01]	-0.03 [-0.11; 0.06]	-0.03 [-0.11; 0.06]
Habitat (West vs East)	-0.08 [-0.34; 0.18]	**-0.41 [-0.67; -0.14]**	-0.15 [-0.41; 0.12]	**-0.30 [-0.53; -0.06]**	**-0.38 [-0.65; -0.1]**	**-0.36 [-0.63; -0.09]**	**-0.47 [-0.78; -0.16]**	**-0.51 [-0.86; -0.16]**
Season (Autumn vs Spring)	0.13 [-0.09; 0.34]	0.05 [-0.13; 0.22]	0.15 [-0.21; 0.51]	**0.28 [0.07; 0.49]**	**0.22 [0.02; 0.41]**	0.13 [-0.05; 0.31]	-0.05 [-0.26; 0.17]	-0.04 [-0.26; 0.19]
Year (2013 vs 2012)	0.09 [-0.13; 0.32]	-0.01 [-0.19; 0.17]	**0.22 [0.01; 0.44]**	**0.27 [0.06; 0.48]**	0.15 [-0.06; 0.36]	0.10 [-0.13; 0.34]	0.08 [-0.2; 0.35]	0.19 [-0.07; 0.46]
Body condition index	-0.06 [-0.14; 0.02]	-0.01 [-0.09; 0.06]	-0.03 [-0.11; 0.06]	0.03 [-0.04; 0.11]	0.02 [-0.06; 0.10]	-0.02 [-0.09; 0.06]	-0.04 [-0.13; 0.04]	-0.03 [-0.12; 0.05]
Age	-0.05 [-0.16; 0.05]	0.06 [-0.05; 0.18]	**-0.13 [-0.22; -0.04]**	0.01 [-0.08; 0.11]	0.04 [-0.06; 0.14]	0.05 [-0.06; 0.16]	0.04 [-0.07; 0.16]	0.01 [-0.11; 0.13]
Breeding status (with COYs vs solitary)	0.10 [-0.09; 0.30]	**0.20 [0.03; 0.38]**	0.16 [-0.27; 0.58]	0.09 [-0.11; 0.30]	0.13 [-0.05; 0.32]	0.17 [-0.03; 0.36]	**0.22 [0.005; 0.428]**	0.22 [-0.001; 0.441]
Breeding status (with YRLs vs solitary)	-0.07 [-0.32; 0.18]	0.06 [-0.16; 0.28]	-0.02 [-0.40; 0.36]	0.09 [-0.16; 0.34]	0.10 [-0.12; 0.33]	0.09 [-0.15; 0.33]	0.11 [-0.14; 0.37]	0.08 [-0.19; 0.36]
Intercept (n=82)	**3.29 [3.15; 3.42]**	**5.35 [5.22; 5.49]**	**1.40 [1.27; 1.53]**	**3.42 [3.31; 3.54]**	**2.27 [2.15; 2.4]**	**2.98 [2.85; 3.11]**	**0.87 [0.74; 1]**	**1.8 [1.67; 1.93]**
FA PC1	0.01 [-0.07; 0.09]	-0.02 [-0.1; 0.07]	0.01 [-0.11; 0.13]	-0.08 [-0.18; 0.03]	-0.05 [-0.15; 0.05]	0 [-0.11; 0.10]	0.03 [-0.08; 0.15]	0.03 [-0.08; 0.15]
FA PC2	-0.01 [-0.11; 0.09]	0.04 [-0.07; 0.15]	0.10 [-0.02; 0.23]	0.03 [-0.08; 0.15]	0.03 [-0.08; 0.15]	**0.12 [0.003; 0.239]**	**0.17 [0.05; 0.29]**	**0.18 [0.06; 0.3]**
FA PC3	-0.05 [-0.15; 0.05]	-0.06 [-0.17; 0.04]	-0.09 [-0.22; 0.03]	-0.06 [-0.17; 0.05]	-0.11 [-0.22; 0]	**-0.20 [-0.31; -0.08]**	**-0.23 [-0.35; -0.12]**	**-0.26 [-0.38; -0.15]**

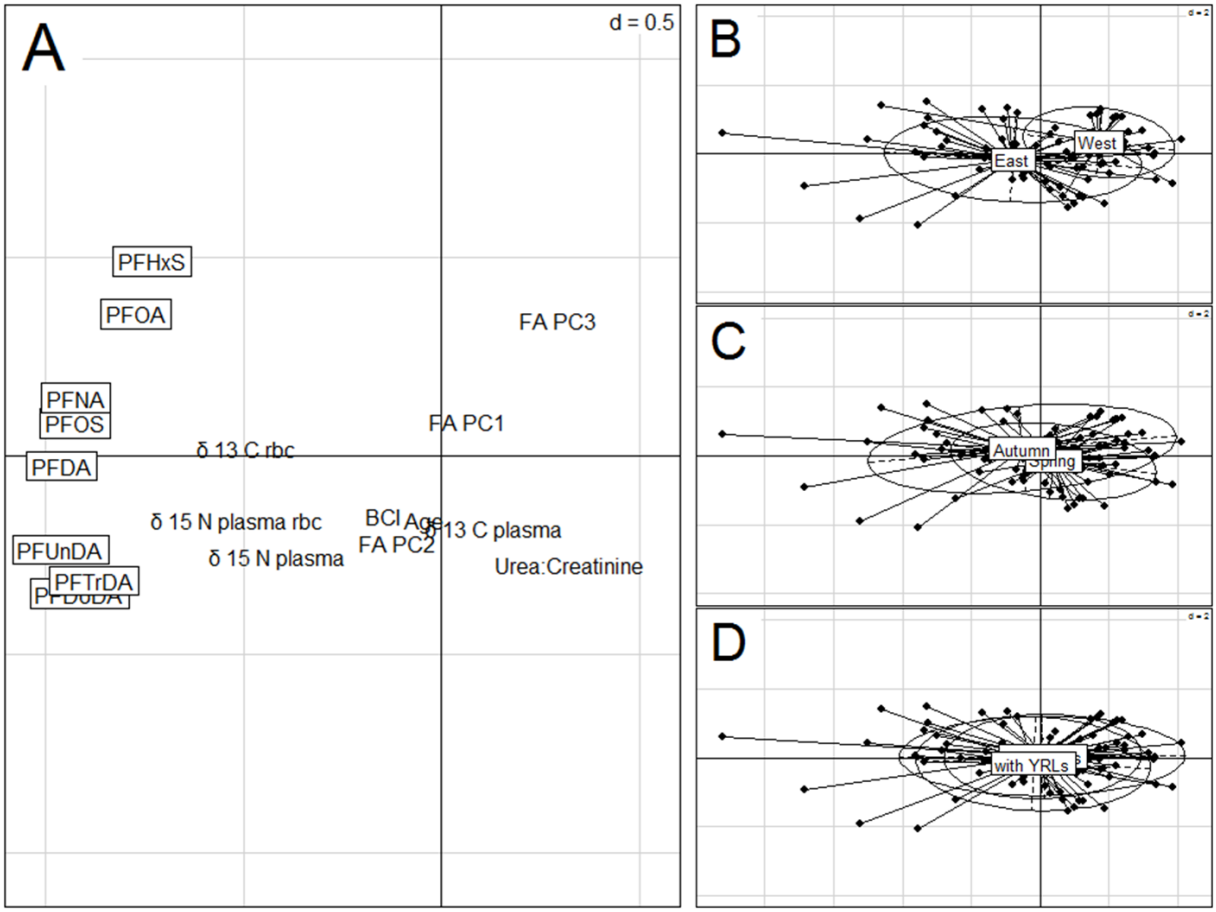


Figure 1: A-Correlation plot from redundancy analysis (RDA) illustrating relationships between plasma concentrations of PFAS, proxies for diet, body condition, metabolic state and age in female polar bears sampled in Svalbard in spring and autumn 2012-2013 (n=82). Boxed labels (PFAS) represent response variables and unboxed labels explanatory variables. In ordination plots grouped by B- habitat, C- season and D- breeding status. Each dot represents an individual. Individuals with similar PFAS concentrations are near each other and individuals with dissimilar PFAS concentrations are farther from each other. Explanatory variables are age; body condition index (BCI); diet proxies defined as nitrogen (δ^{15}N) and carbon (δ^{13}C) stable isotope values in plasma and red blood cells (rbc), fatty acid principal component scores (FA PC1, 2 and 3) in adipose tissue; metabolic state proxy is defined as urea to creatinine ratio (Urea:Creatinine).

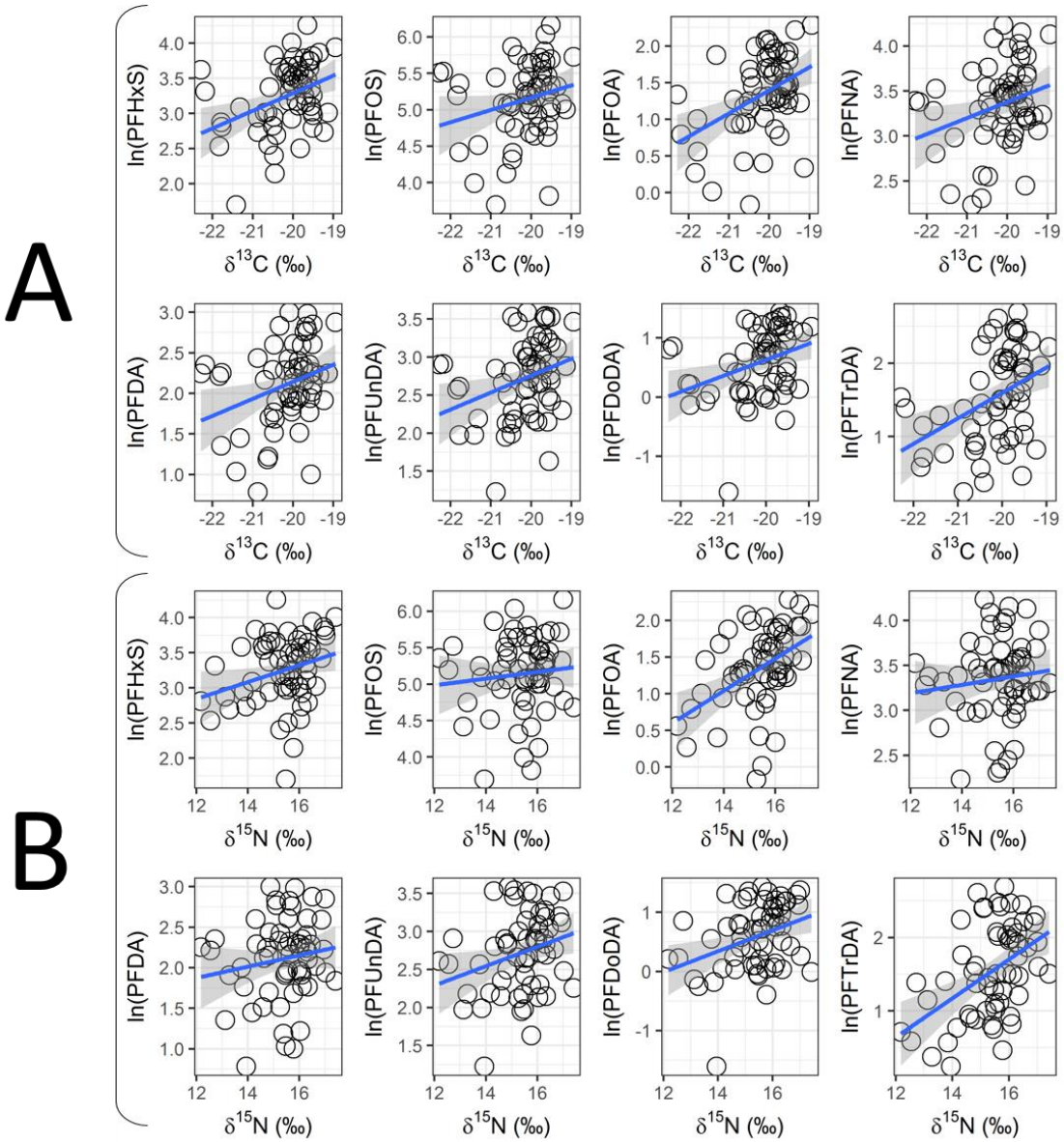


Figure 2: Relationship between PFAS (ng/g wet weight) in plasma and A- carbon ($\delta^{13}C$) and B- nitrogen ($\delta^{15}N$) stable isotope values (‰) in red blood cells. Female polar bears were sampled in Svalbard in 2012-2013. Plots show individuals (n=112), regression lines and shaded area 95% confidence interval.

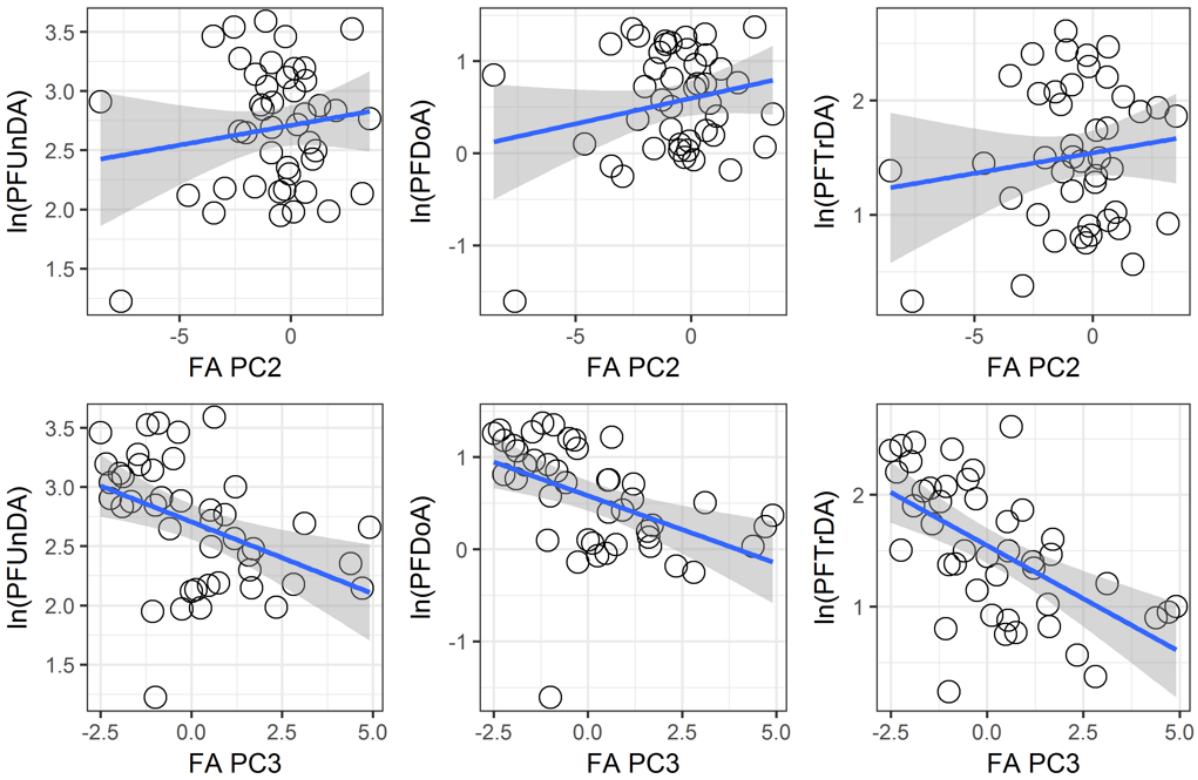


Figure 3: Relationships between PFAS in plasma (ng/g ww) and the fatty acid principal component scores 2 and 3 (FA PC2, FA PC3). Fatty acids were measured in adipose tissue, female polar bears were sampled in Svalbard in 2012-2013. Plots show individuals (n=83), regression lines and shaded area 95% confidence interval.

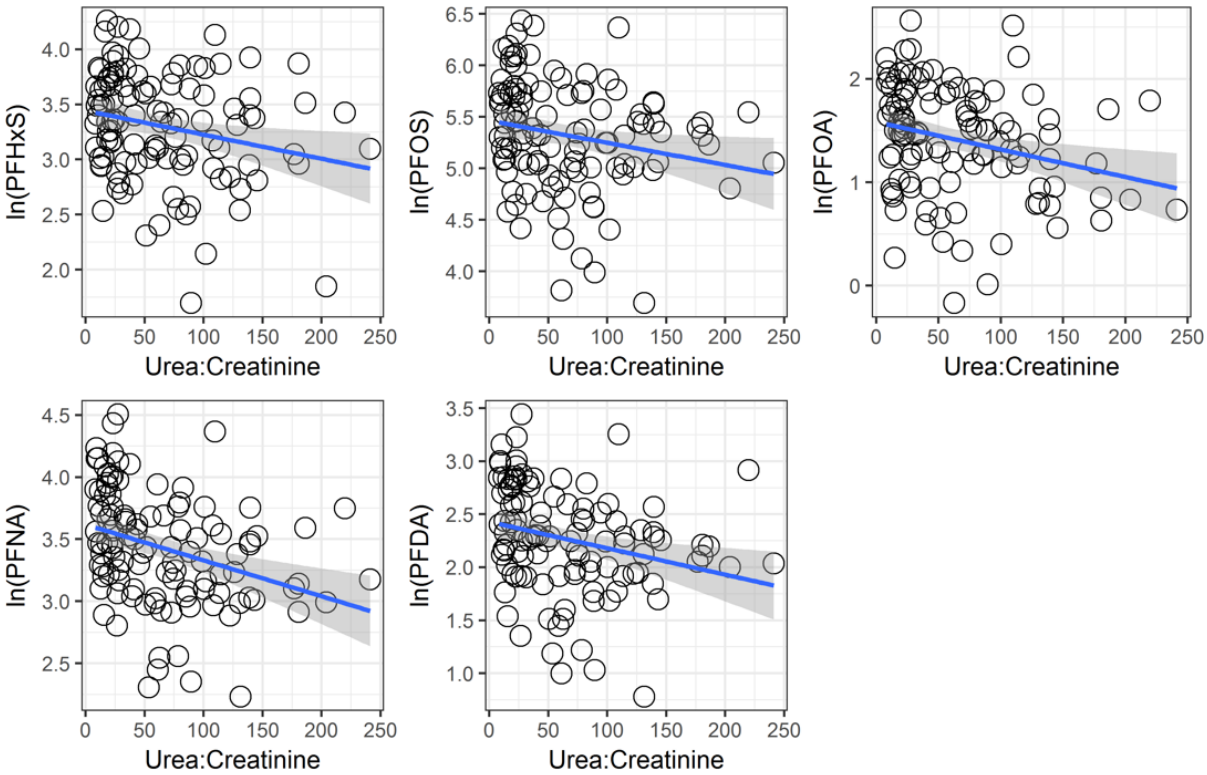
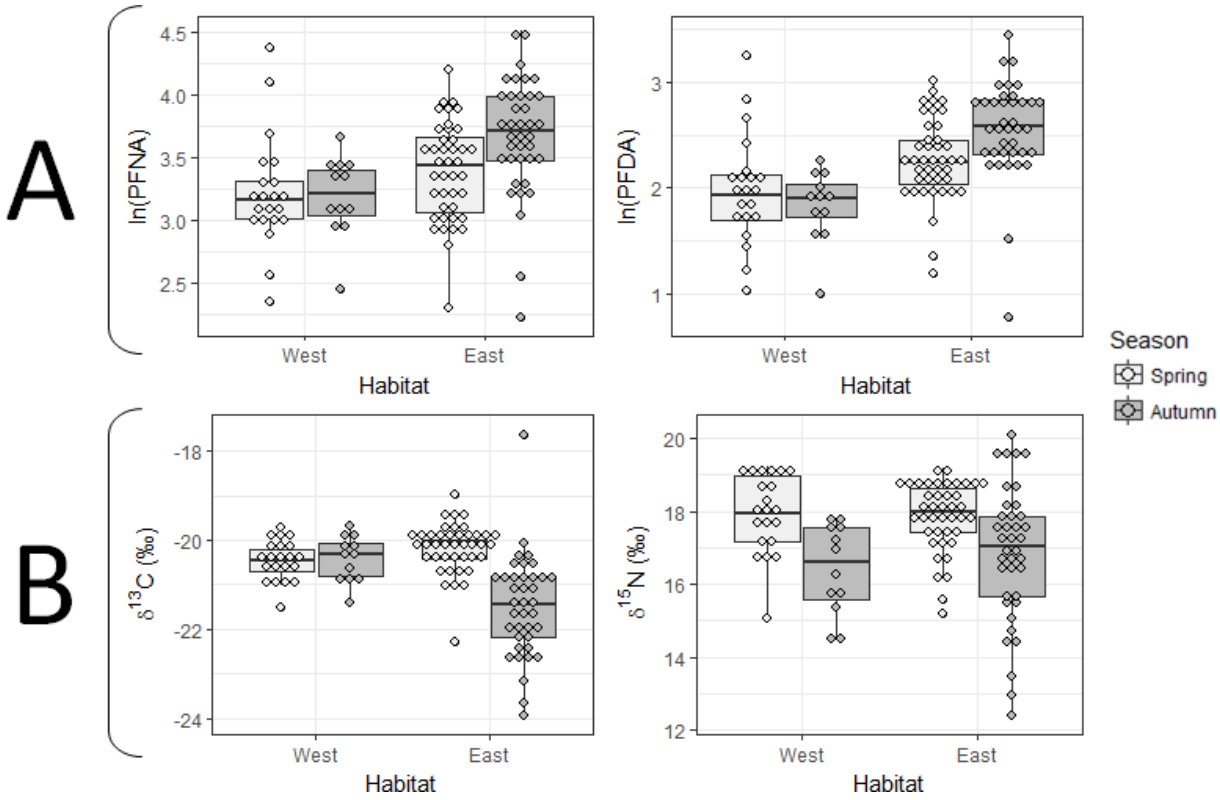


Figure 4: Relationship between plasma PFAS (ng/g wet weight) and plasma urea to creatinine ratio (urea:creatinine). Female polar bears were sampled in Svalbard in 2012-2013. Plots show individuals (n=111), regression lines and shaded area 95% confidence interval.



27

28 **Figure 5:** Plasma concentrations of A- PFNA and PFDA and B- stable isotope values

29 according to season and habitat. PFAS are in ng/g wet weight, female polar bears (n=112)

30 were sampled in Svalbard in 2012-2013. Light grey boxes and dots represent females

31 captured in spring (April) and dark grey boxes and dots represent females captured in autumn

32 (September).

33

SUPPORTING INFORMATION

Diet and metabolic state are the main factors determining concentrations of perfluoroalkyl substances in female polar bears from Svalbard

Sabrina Tartu[1]; Sophie Bourgeon[1,2]; Jon Aars[1]; Magnus Andersen[1]; Karen Lone[1]; Bjørn Munro Jenssen[3]; Anuschka Polder[4]; Gregory W. Thiemann[5]; Vidar Torget[3]; Jeffrey M. Welker[6,7] and Heli Routti[1]*

[1] Norwegian Polar Institute, Fram Centre, Tromsø, Norway

[2] UiT-The Arctic University of Norway, Department of Arctic and Marine Biology, Tromsø, Norway

[3] Norwegian University of Science and Technology, Department of Biology, Trondheim, Norway

[4] Norwegian University of Life Science, Campus Adamstua, Oslo, Norway

[5] York University, Faculty of Environmental Studies, Toronto, ON, Canada

[6] University of Alaska Anchorage, Department of Biological Sciences, Anchorage, AK, USA

[7] University Center in Svalbard, Longyearbyen, Svalbard, Norway

Corresponding author: Sabrina Tartu, tel. +47 777 50 538, e-mail: tartu.sabrina@gmail.com

Table S1: List of candidate models to explain PFASs variations in Svalbard female polar bears in 2012-2013. Except for models with fatty acids* (n=82), 111 females were included in the models.

Candidate models	Variables
1	Habitat
2	Season
3	Year
4	Status
5	Habitat + Season
6	Habitat + Status
7	Habitat + Year
8	Season + Status
9	Body condition
10	δ^{15}N plasma
11	δ^{13}C plasma
12	δ^{15}N rbc
13	δ^{13}C rbc
14	Body condition + δ^{15}N plasma
15	Body condition + δ^{13}C plasma
16	Body condition + δ^{15}N rbc
17	Body condition + δ^{13}C rbc
18	Age
19	Age + Body condition
20	Age + δ^{15}N plasma
21	Age + δ^{13}C plasma
22	Age + δ^{15}N rbc
23	Age + δ^{13}C rbc
24	Age + δ^{15}N plasma + Body condition
25	Age + δ^{13}C plasma + Body condition
26	Age + δ^{15}N rbc + Body condition
27	Age + δ^{13}C rbc + Body condition
28	Urea:Creatinine
29	Urea:Creatinine + Body condition
30	Urea:Creatinine + Age
31	Urea:Creatinine + δ^{15}N plasma
32	Urea:Creatinine + δ^{13}C plasma
33	Urea:Creatinine + δ^{15}N rbc
34	Urea:Creatinine + δ^{13}C rbc
35	Age + Urea:Creatinine + Body condition + δ^{15}N plasma
36	Age + Urea:Creatinine + Body condition + δ^{13}C plasma
37	Age + Urea:Creatinine + Body condition + δ^{15}N rbc
38	Age + Urea:Creatinine + Body condition + δ^{13}C rbc
39	Fatty acids PC1*
40	Fatty acids PC2*
41	Fatty acids PC3*
42	Null model

Table S2: Relationships between the fatty acid principal components scores (FA PCs) and stable isotope in plasma and red blood cells in female polar bears from Svalbard (2012-2013). Values are parameter estimates and 95% confidence intervals derived from conditional model averaging of general linear mixed models that included female identity as a random factor. Values in bold are significantly different from 0 at the 5% level.

	$\delta^{15}N$		$\delta^{13}C$	
	Plasma	Red blood cells	Plasma	Red blood cells
FA PC1	-0.01 [-0.24; 0.22]	0.11 [-0.16; 0.38]	0.05 [-0.2; 0.3]	0.18 [-0.09; 0.46]
FA PC2	**0.66 [0.45; 0.88]**	**0.53 [0.31; 0.75]**	0.18 [-0.08; 0.45]	**0.33 [0.08; 0.58]**
FA PC3	**-0.45 [-0.7; -0.21]**	-0.45 [-0.69; -0.22]	0.12 [-0.15; 0.38]	-0.1 [-0.37; 0.16]

Table S3: Variables included (×**)** in the five models with the lowest AICc explaining the concentration of individual PFAS compounds in plasma. All models (linear mixed models) include female identity as a random factor. "ΔAICc" is the difference in AICc between each candidate model and the model with the lowest AICc and "AIC wt" the Akaike weights.

	Explanatory variables						AICc	ΔAICc	AICc wt
Response variables	Age	BCI	δ^{13}C rbc	δ^{15}N rbc	δ^{15}N plasma	Urea: Creatinine			
PFHxS			×			×	130.24	0	0.25
	×	×	×			×	130.62	0.38	0.21
				×		×	130.63	0.4	0.21
	×	×		×		×	132.78	2.54	0.07
					×	×	133.42	3.18	0.05
PFOS				×		×	126.92	0	0.62
	×	×		×		×	130.08	3.17	0.13
					×	×	131.32	4.41	0.07
			×			×	131.37	4.45	0.07
	×	×	×			×	133.34	6.42	0.02
PFOA	×	×		×		×	140.57	0	0.76
				×		×	144.31	3.74	0.12
	×			×			145.42	4.84	0.07
	×	×		×			147.39	6.81	0.03
	×	×	×			×	149.12	8.54	0.01
PFNA			×			×	110.63	0	0.8
				×		×	114.38	3.75	0.12
							116.18	5.55	0.05
	×	×		×		×	118.51	7.88	0.02
			×				121.88	11.25	0
PFDA			×			×	117.68	0	0.65
				×		×	120.55	2.87	0.15
	×	×	×			×	121.47	3.79	0.1
	×	×		×		×	123.19	5.51	0.04
			×				125.08	7.4	0.02
PFUnDA				×		×	131.52	0	0.42
				×			132.88	1.36	0.21
	×			×			134.27	2.75	0.11
		×		×			134.95	3.43	0.08
	×	×		×		×	135.2	3.67	0.07
PFDoDA				×			141.54	0	0.33
		×		×			142.77	1.23	0.18
	×			×			143.28	1.74	0.14
				×		×	143.35	1.81	0.13
	×	×		×			144.52	2.98	0.07
PFTrDA				×			147.32	0	0.36
		×		×			148.91	1.59	0.16
				×		×	149.24	1.92	0.14
	×			×			149.52	2.19	0.12
	×	×		×			151.14	3.82	0.05

69

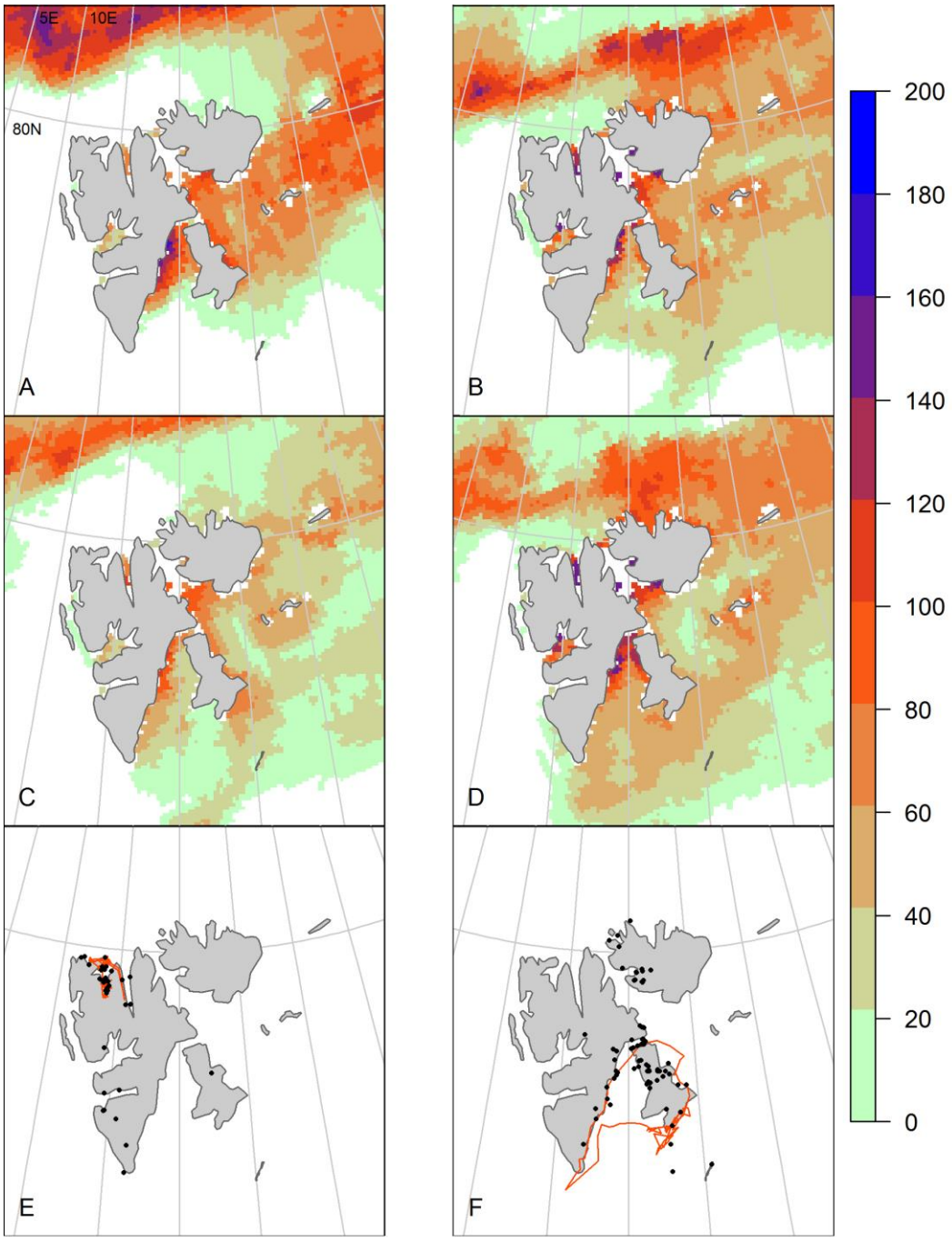


Figure S1: Days with polar bear habitat in the period preceding the four sampling periods, predicted from a resource selection function. The panels for the April sampling periods (A- April 2012, C- April 2013) sum up the days of habitat during the preceding 7 months (September-March), while the panels for the September sampling periods (B- September 2012 and D- September 2013) cover the preceding 5 months (April-August). Triangles represent location of captured females using E- the Western habitat and F- the Eastern habitat, red lines represent telemetry tracks for E- one female using the Western habitat and F- one female using the Eastern habitat. The color scale represents the number of days with optimal polar bear habitat (from 0-20 in green to 180-200 in blue).