Supplementary file for: Mosbacher J.B., Desforges J.-P., Michelsen A., Stelvig M., Eulaers I., Sonne C., Dietz R., Jenssen B.M., Ciesielski T.M., Lierhagen S., Flaten T.P., Aggerbeck M.R. & Schmidt N.M. 2022. Hair mineral levels as indicator of wildlife demographics?—a pilot study of muskoxen. *Polar Research 41*. Correspondence: Niels Martin Schmidt. Department of Ecoscience, Aarhus University, Frederiksborgvej 399, DK-4000 Roskilde, Denmark. E-mail: <u>nms@ecos.au.dk</u>.

Abbreviations

DOY: day of year NDVI: normalized difference vegetation index PCA: principal component analysis

Chemical analyses of hair samples

Prior to chemical analysis, guard hairs and large external contaminants, such as soil and vegetation, were removed from the wool using tweezers. The samples were washed twice in 96% ethanol and MO water to remove further contamination, before being stored in clean paper bags and oven-dried at 50°C for 48 hours. For samples covering 2000-2015, approximately 150 mg (108-180 mg) of uncut muskox wool was added to 5 mL HNO₃ (50:50; v:v) and digested using a high-pressure microwave reactor (UltraCLAVE, Milestone, Leutkirch, Germany). The temperature was gradually increased from room temperature to 250°C over one hour, before gradually cooled down to room temperature again over the next hour. After digestion, the decomposed samples were diluted to 50 mL (50.0 g \pm 3 g) in a Teflon bottle with ultrapure water, before being transferred to 15 mL polypropylene vials. The hair concentration of 10 elements was analysed using high-resolution inductively coupled plasma mass spectrometry (Thermo Finnigan Element 2, Bremen, Germany) at the Norwegian University of Science and Technology, Trondheim. Quality Assurance was confirmed by analysing Polish Virginia Tobacco Leaves (INCT-PVTL-6; Warsaw, Poland) as certified reference material. For samples from 2016 and 2017, approximately 150 mg (134-160 mg) of uncut muskox wool was placed in acid cleaned digitubes (SCP Sciences 010-500-263) together with 3 mL of ultrapure nitric acid (HNO3; ca. 67-69%, OptimaTM Grade, Fisher), closed with airtight caps, and digested overnight at 90 °C. After being cooled down to room temperature the digestions were diluted in two steps; first the digest was transferred to 15 mL Falcon tubes and diluted with mQ H2O (3:7 digest:mQ H2O) to create a mother solution (MS) to be stored in dark conditions at ca. 6°C until further processing. Two aliquot of the mother solution was taken and further diluted to 10ml with MQ water and analysed with a High Resolution Inductively Coupled Plasma Mass Spectrometer (Thermo-Fisher Element-XR, OMP-GET, Toulouse, France) at the CNRS EcoLab in Toulouse, France. The accuracy of digestion and analysis protocol was checked using certified standards; PRON-1 (River prawn/Crevette, NRC Canada), IAEA-336 (Lichen) and NIST 1515 (Apple leaves). For all samples, blanks were negligible, and the concentrations

measured were within the certified values for all trace elements. No samples were below limits of detection.

Muskox calf recruitment rates

The muskox population has been monitored within a designated area of approximately 47 km² every summer since 1996 to provide data on muskox densities and sex and age composition of groups (Schmidt et al. 2015). As descriptors of vital population demographics in the muskox population over the study period, we calculated calf recruitment: the number of calves per 100 adult females during July and August, as defined by Schmidt et al. (2015).

Local environmental conditions

As a general descriptor of the inter-annual variation in ambient environmental conditions, we used data on snow depth (m) and air temperature (°C) recorded at an automatic weather station located centrally in the Zackenberg valley (Hansen et al. 2008). The NDVI as an indicator of vegetation biomass and quality (Hogrefe, Patil, Ruthrauff, Meixell, Budde et al. 2017) was measured using a handheld CropCircle system on a weekly basis during the snow-free periods inside permanent monitoring plots in the Zackenberg valley lowland (Schmidt et al. 2014), located in areas utilized by muskoxen (Kristensen et al.2011).

We used these data to calculate a suite of environmental variables, describing the ambient conditions experienced by muskoxen while their wool was being formed (i.e., the year before the wool is shed and collected, and hence also the year before calving). *Onset of snow season* was defined as the DOY_{*t*-1} when snow depth was consistently above 10 cm, while *End of snow season* was defined as the DOY_{*t*} when snow depth was consistently below 10 cm. *Duration of snow season* was the number of days between onset in year_{*t*-1} to end of snow season in year_{*t*}, whereas *Duration of snow-free season* was the number of days between end_{*t*-1} and onset of snow season_{*t*-1}. Similarly, we calculated the *mean summer NDVI* (June_{*t*-1} through August_{*t*-1}) as an indicator of plant forage in summer, and the *DOY of maximum NDVI in year*_{*t*}. Finally, we calculated *Mean summer air temperature* the year before wool formation (June_{*t*-1} through August_{*t*-1}).

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	Year	n 1	Calf recruitment	Cu	Se	Мо	Co	Zn	Na	Mn	Mg	Fe	Cr
High calf recruitment	2000	2	54	5.16 ± 0.38	0.53 ± 0.21	0.05 ± 0.02	0.12 ± 0.05	52.09 ± 17.3	107.20 ± 60.90	6.60 ± 1.30	127.76 ± 61.20	235.32 ± 7.24	0.85 ± 0.09
	2004	5	61	5.88 ± 0.80	0.36 ± 0.06	0.10 ± 0.03	0.39 ± 0.03	86.85 ± 2.62	199.49 ± 50.83	14.34 ± 1.31	346.87 ± 35.78	893.36 ± 67.14	2.04 ± 0.08
	2006	9	54	5.93 ± 1.46	0.35 ± 0.05	0.06 ± 0.01	0.32 ± 0.04	78.07 ± 3.80	174.28 ± 30.86	16.41 ± 2.98	308.06 ± 49.64	643.70 ± 82.02	1.80 ± 0.37
	2007	3	49	7.72 ± 2.11	0.22 ± 0.04	0.14 ± 0.07	0.48 ± 0.10	92.18 ± 7.45	200.07 ± 73.16	21.30 ± 5.30	412.35 ± 99.41	1268.9 ± 213.21	3.33 ± 0.91
	2013	5	68	5.36 ± 0.85	0.41 ± 0.20	0.06 ± 0.01	0.32 ± 0.06	83.00 ± 2.26	188.53 ± 69.18	27.79 ± 12.45	226.95 ± 33.07	747.97 ± 127.11	1.64 ± 0.27
	2017	10	53	4.92 ± 0.08	0.48 ± 0.02	0.08 ± 0.02	0.26 ± 0.11	60.98 ± 1.41	74.00 ± 7.57	21.85 ± 14.76	170.50 ± 17.42	294.95 ± 72.47	0.61 ± 0.21
Mean			56	5.83 ± 0.16	0.39 ± 0.02	$\boldsymbol{0.08 \pm 0.01}$	0.32 ± 0.02	75.53 ± 2.45	157.26 ± 8.37	18.05 ± 1.15	265.42 ± 17.07	680.70 ± 60.40	1.71 ± 0.15
Low calf recruitment	2003	10	33	5.40 ± 0.40	0.25 ± 0.04	0.08 ± 0.01	0.59 ± 0.12	68.95 ± 4.85	167.89 ± 34.47	24.72 ± 3.49	442.55 ± 54.42	1158.84 ± 216.85	2.06 ± 0.33
	2009	9	8	4.72 ± 0.23	0.31 ± 0.07	0.05 ± 0.01	0.44 ± 0.12	90.82 ± 2.50	177.64 ± 47.50	29.68 ± 14.88	405.49 ± 76.63	944.56 ± 232.94	1.63 ± 0.41
	2010	8	13	6.28 ± 1.07	0.22 ± 0.02	0.06 ± 0.01	0.48 ± 0.09	79.79 ± 5.63	216.00 ± 68.94	26.50 ± 4.19	329.99 ± 44.78	1052.12 ± 177.78	1.92 ± 0.50
	2012	10	8	4.75 ± 0.90	0.28 ± 0.04	0.05 ± 0.01	0.39 ± 0.13	82.21 ± 8.41	332.12 ± 251.7	22.91 ± 11.10	275.30 ± 40.90	749.17 ± 180.50	1.01 ± 0.19
	2014	10	1	3.80 ± 0.18	0.27 ± 0.02	0.06 ± 0.01	0.30 ± 0.07	61.62 ± 3.23	97.23 ± 15.43	15.23 ± 3.96	172.53 ± 20.48	668.13 ± 165.30	1.04 ± 0.11
	2015	10	3	4.26 ± 0.21	0.35 ± 0.03	0.04 ± 0.01	0.25 ± 0.04	83.50 ± 2.10	139.29 ± 20.49	12.82 ± 2.43	287.16 ± 37.78	597.07 ± 98.63	1.35 ± 0.21
	2016	10	16	3.75 ± 0.09	0.33 ± 0.03	0.03 ± 0.02	0.08 ± 0.02	62.39 ± 2.37	61.36 ± 2.35	3.72 ± 1.15	119.56 ± 13.51	142.34 ± 29.95	0.31 ± 0.05
Mean			11	$\textbf{4.71} \pm \textbf{0.10}$	0.29 ± 0.01	0.05 ± 0.01	0.37 ± 0.02	75.29 ± 1.27	169.49 ± 9.99	19.85 ± 1.00	293.49 ± 13.19	772.45 ± 37.85	1.34 ± 0.07

Supplementary Table S1. Sampling years, samples size, calf recruitment and mean mineral levels ($\mu g/g \pm SE$) in muskox wool collected over 13 years at Zackenberg in north-east Greenland. Boldface indicates significant difference between recruitment groups (*t*-tests; *p* < 0.10).

Supplementary Table S2. Bray-Curtis dissimilarities between mineral levels and environmental parameters in high versus low calf recruitment years. Significant differences (p < 0.10) are shown in boldface.

Variable	Average	Standard deviation	Ratio	Average high	Average low	р
Minerals						
Fe	0.169800	0.128800	1.318	680.7036	772.4498	0.56
Mg	0.051630	0.039500	1.307	265.4154	293.4895	0.89
Na	0.033020	0.028590	1.155	157.2626	169.4902	0.85
Zn	0.006318	0.005141	1.229	75.52746	75.586	0.67
Mn	0.003989	0.003802	1.049	18.04674	19.8454	0.81
Cu	0.000575	0.000424	1.357	5.82888	4.7075	0.07
Cr	0.000360	0.000281	1.285	1.71133	1.3396	0.57
Co	0.000069	0.000057	1.216	0.31572	0.3676	0.79
Se	0.000058	0.000050	1.166	0.38926	0.2889	0.02
Мо	0.000014	0.000012	1.198	0.08248	0.0507	0.05
Environmental parameters						
Onset of snow season	0.024300	0.016020	1.517	355	297.1429	0.04
Duration of winter	0.024330	0.018510	1.314	223.1667	191.5714	0.64
End of snow season	0.005477	0.004201	1.304	175.3333	168.1429	0.61
Duration of snow-free season Mean summer air	0.020970	0.014520	1.444	179.6667	129	0.05
temperature	0.000368	0.000282	1.307	3.8171	3.728	0.51
Mean summer NDVI	0.000020	0.000012	1.688	0.4408	0.3954	0.03

Supplementary Table S3. Correlation coefficients and, in brackets, the associated p-values for the correlations between the minerals and environmental parameters included in the PCA. Significant differences (p < 0.10) are shown in boldface.

	Duration of snow-free season	Onset of snow season	Mean summer NDVI
Se	0.16 (0.604)	0.25 (0.403)	0.37 (0.220)
Cu	0.51 (0.075)	0.45 (0.122)	0.44 (0.134)
Mo	0.47 (0.109)	0.42 (0.149)	0.40 (0.170)