

## PCB CONTAMINATION IN RELATION TO LEVELS OF THYROID HORMONES AND PROGESTERONE, AND MIGRATION PATTERNS IN POLAR BEARS (*Ursus maritimus*).

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### Introduction

The polar bear (*Ursus maritimus*) is a top predator in the Arctic food web. It is thus exposed to high levels of persistent organic pollutants (POP), in particular to polychlorinated biphenyls, (PCBs)<sup>1</sup>. Satellite tracking has shown that some individuals that den at Svalbard, annually migrate east to Frans Josef Land and Novaja Zemlja<sup>2</sup>. Ringed seals (*Phoca hispida*) are the main prey of polar bears<sup>3</sup>. Since higher PCB levels have been reported in ringed seals from the Russian Arctic than in the Svalbard ringed seals<sup>4</sup>, it is possible that migratory bears have higher PCB body burdens as compared to non-migratory "Svalbard bears".

High levels of PCBs have been linked to effects in other marine mammals and especially to reproductive failure<sup>5</sup> and endocrine disruption<sup>6,7,8</sup>. PCBs have been shown to have an effect on thyroid hormone status on laboratory rats<sup>9</sup>, and common seals (*Phoca vitulina*)<sup>5</sup>. Both studies show that PCB exposure causes a decrease in the thyroid hormone levels. Skaare *et al.*<sup>6</sup> have previously reported a negative correlation between thyroid hormone levels and  $\Sigma$ PCB in polar bears.

The aim of the present study is to reveal if migratory patterns of female polar bears that den at Svalbard affect their PCB levels and to study whether there are any relationships between PCB concentrations and plasma thyroid and progesterone levels in female polar bears from Svalbard.

### Materials and methods

Blood samples were collected from 33 female polar bears without cubs and females with 2-year old cubs at Svalbard, Hopen, Edgeøya and in the Barents Sea region. Samples were taken by standard procedures<sup>10</sup> in spring (March-May) 1995 to 1998. These females were potentially receptive for breeding and thus interesting in a reproductive perspective. Polar bears have lipid rich milk, which is an important pathway for excretion of lipophilic compounds for the females. Therefore, the amount of milk given to the cub will affect a female's PCB load. Females that are single or have 2-year old cubs give little or no milk, and therefore are likely to retain more PCB than females with newborn cubs or yearlings that receive more milk.

The bears were divided into two groups based on where they were caught: East or west of 35 °E. Concentrations of PCBs were analyzed in whole blood using gas chromatography (GC-ECD)<sup>13</sup>. The following PCB-congeners were quantified in all bears and constitute  $\Sigma$ PCB in the present

study: PCB-153, 180, 99, 194, 156, 170 (sorted by decreasing abundance).  $\Sigma$ PCB is referring to the sum of these congeners in the samples. Concentrations of progesterone and free and total thyroid hormones: Thyroxin (fT4, tT4) and tri-iodothyronine (fT3, tT3) were determined from blood plasma by radioimmunoassay (RIA), using commercially available kits: Progesterone: Spectria® Progesterone <sup>125</sup>I Coated Tube radioimmunoassay from Orion Diagnostica, Finland. Thyroid hormones: Coat-a-count®, Thyroid, from Diagnostic Products Corporation, Los Angeles. As it is the free T3 that is physiologically active, the ratio of total to free thyroid hormones was calculated and used for comparison of eastern and western areas.

Students t-test was used to test differences between areas, and Spearman correlation test was used to test relationships between variables. A p-value of less than 0.05 is defined as significant.

## Results and discussion

Mean  $\Sigma$ PCB level for all adult females was 7376 ng/g l.w. (n=33, range: 1507 - 20610 ng/g l.w.) This is consistent with earlier reported levels from Svalbard <sup>1,6</sup> and higher than levels reported in polar bears from most parts of the Canadian Arctic.

$\Sigma$ PCB in whole blood samples of the polar bears was correlated to longitude (n=33,  $r^2=0.363$ ,  $p=0.038$ ). The mean  $\Sigma$ PCBs for Svalbard bears (5544 ng/g l.w., n=16) and for bears from the Barents Sea (9101 ng/g l.w. n=17), were significantly different ( $t=-2.10$ ,  $p=0.044$ ,  $df=31$ , Fig. 1a). The difference is presumably linked to higher burdens of PCB in the polar bear's main prey, the ringed seal, in the eastern area (i.e., the Barents Sea) as compared to at Svalbard<sup>4</sup>. The results thus support our hypothesis that Svalbard polar bears migrating east to Frans Josef Land and Novaja Zemlja have higher PCB burdens than non-migratory polar bears.

As high levels of PCBs have been linked to endocrine disruption and reproductive failure<sup>6,7,8</sup>, we investigated if high levels of PCBs might affect the hormone levels of the bears, thus possibly reducing growth and metabolism and preclude successful reproduction. As shown in Fig. 1b we found significant differences in progesterone-levels between adult females from the two areas, the progesterone levels being higher in bears caught in the eastern area ( $t=-3.36$   $p=0.002$ ,  $df=27$ , Fig. 1b). We were not able to demonstrate any relationship between  $\Sigma$ PCB and progesterone levels in the polar bears.

The observed difference in progesterone levels between bears from the two areas may be a natural cause of differences in the month of sampling in the two areas. Bears from the east were caught about a month later on average than bears from the west. Progesterone levels are known to increase in pregnant females after the breeding season throughout the pre-implantation period, which will result in higher levels in the bears caught late or after the breeding season. The range of progesterone levels throughout the spring (Fig. 2a) is consistent with previously reported results from populations of polar bears in the less polluted Hudson Bay (range: 0.1 -1.6 ng/mL)<sup>10</sup>.

With respect to thyroid hormones, significant differences between female polar bears from the two areas were found for the ratio of total T3 to free T3 ( $t=3.48$ ,  $df=7$ ,  $p=0.01$ ). The ratios were highest in bears from the western area. There was on the other hand no correlation between  $\Sigma$ PCB and the tT3/fT3 ratio (n=8,  $r^2=-0.19$ ,  $p=0.651$ ).

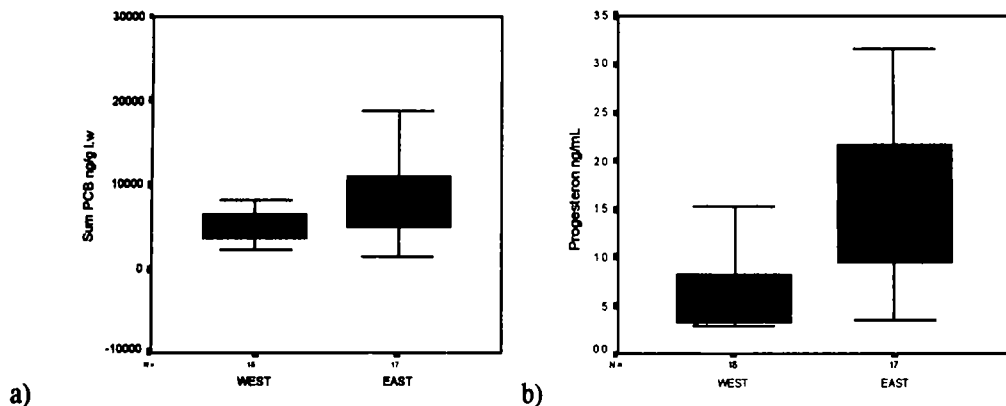


Figure 1) Boxplot showing the differences in levels of PCB (a) and progesterone (b) between female polar bears from the western and eastern areas. Median is shown as bold line in box. Outliers are not shown.

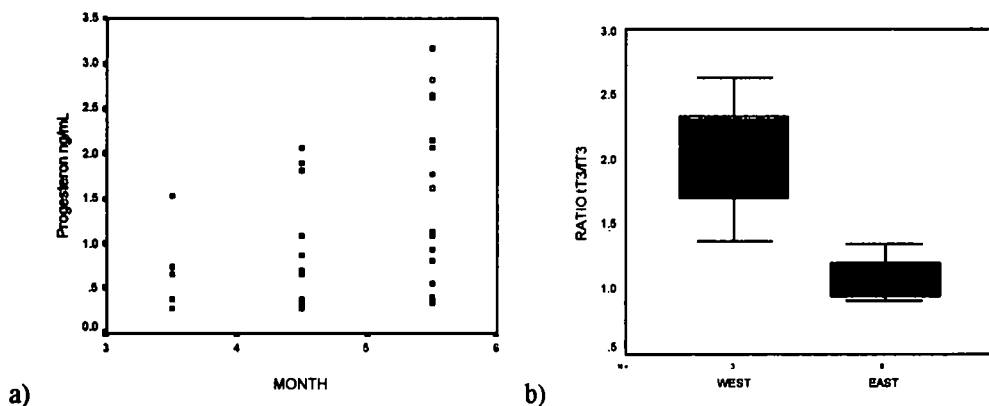


Fig.2) a) Levels of progesterone for female polar bears during the spring months. March=3, April =4, May =5. b) Boxplot showing differences in the ratio tT3 to ft3 in female polar bears from the western and eastern areas.

Since there was no relationship between  $\Sigma$ PCB and hormone concentrations, the present study does not confirm the endocrine disruptive effects of PCBs previously reported in marine mammals<sup>6, 7, 8</sup>. It is, however, possible that fine homeostatic regulations may compensate for the possible endocrine disruption caused by PCBs. Signs of disruption might thus still be present at higher regulatory levels, like in the concentrations of hypothalamic or pituitary hormones.

The lack of correlation between  $\Sigma$ PCB and the thyroid hormone ratio (tT3/ft3) opposes the results from Skaare *et al.*<sup>14</sup> who found a negative correlation between tT4/ft4 and PCB. This difference could be due to low n in our samples. Seasonal variations in thyroid hormones could also mask a possible correlation between thyroid hormones and  $\Sigma$ PCB in our samples.

In conclusion, there were differences in  $\Sigma$ PCB, tT3 and progesterone between migrating and non-migrating female polar bears. However, there were no correlation between  $\Sigma$ PCB and hormone concentrations, and possible effects of PCBs on hormone levels cannot be confirmed.

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## Litterature cited

- 1) Bernhoft, A., Wiig, O., Skaare, J. U. (1997) *Environ. Pollut.*; 95: 159-75.
- 2) Wiig, O. (1995) *J. Zool.*; Lond.; 237: 515-52.
- 3) Stirling, I, and Archibald, W., R. (1977) *J. Fish. Res. Board Can.* 34: 1126-1129.
- 4) Nakata, H., Tanabe, S., Tatsukawa, R., Koyama, Y., Miyazaki, N., Belikov, S., Boltunov, A. (1998) *Environ. Toxicol. Chem.*; 17: 1745-1755.
- 5) Reijnders, P. J. H., (1986) *Nature*; 324: 456-457.
- 6) Skaare, J. U., Bernhoft, A., Derocher, A. E., Gabrielsen, G. V., Goksoyr, A., Henriksen, E., Larsen, H. J., Lie, E., Wiig, O. (2000) *Toxicol. Letters*; 112-113: 103-109.
- 7) Safe, S. H., (1994) *Crit. Rev. Toxicol.*; 24: 87-149.
- 8) Brouwer, A., Reijnders, P.J. H., and Koeman, J. H. (1989) *Aq. Toxicol.*; 15: 99 - 106.
- 9) Brouwer, A. and Van den Berg, K. J. (1986) *Toxicol. Appl. Pharmacol.*; 85: 401 - 412.
- 10) Derocher, A. E. and Stirling, I. (1998) *J. Zool.*, Lond.; 245: 253-260.
- 11) Norstrom, R. J., Belikov, E., Born, E. W. Garner, G. W., Malone, B., Olpinski, S., Ramsay, M. A., Schliebe, S., Stirling, I., Stishov, M. S., Taylor, M. K. and Wiig, O. (1998) "Arch Environ Contam. Toxicol. 35, 354-367.
- 12) Derocher, A. E., Stirling, I., Andriashek, D. (1992) *Can. J. Zool.*; 70: 561-6.
- 13) Andersen, M., Lie, E., Belikov, S. E., Boltunov, A. N., Derocher, A. E., Garner, G. W., Wiig, O., Skaare, J. U. (In prep.).
- 14) Skaare, J. U., Bernhoft, Wiig, O., Norum, K. R., Haug, E., Eide, D. M., Derocher, A. E. (In prep).