### POLYCHLORINATED BIPHENYLS IN MOTHER/OFFSPRING PAIRS OF POLAR BEARS (Ursus maritimus) AT SVALBARD

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#### Introduction

Biomagnifications of lipophilic and persistent organochlorines (OCs) in the arctic marine ecosystem has resulted in high levels of OCs in polar bears<sup>1,2,3,4</sup>. Particularly high levels of polychlorinated biphenyls (PCB) have been found in polar bears at Svalbard, Norway in the western Barents Sea<sup>1,2,4</sup>. Adverse effects related to exposure to such levels have been reported in marine mammals living in polluted waters<sup>3,5,6,7</sup>. PCBs have been associated with detrimental effects including neuro-, reproductive- and immuno-toxic effects<sup>8,9,10,11</sup>.

Female polar bears are sexually mature at 4-6 yr. Females have a three years reproductive cycle and mate during spring<sup>12</sup>. Pregnant female enters den in September-October. About the same time that the fertilized eggs implant<sup>12</sup>. Species with delayed implantation appear especially susceptible to deleterious reproductive effects from OC exposure<sup>13</sup>. The cubs-of-the-year are born around Christmas and the female emerges from the den usually with two cubs in March-April. She has then fasted for about six months. Starvation of the female during gestation and initial nursing may create a high risk for toxic effects due to their body burden of OCs. High fat content of polar bear milk<sup>14</sup> makes OC excretion into milk important for this species. The cubs have been completely dependent on mother's milk for nutrition since birth. At birth the cubs weigh around 0.6 kg. The weight of the cubs when emerging the den is around 10 kg. The cubs nurse to some degree for about 2.5 years.

The present study is a part of a project on levels, tissue distribution and possible biological/toxic effects of OCs in polar bears at Svalbard. The project is a collaboration between the Norwegian Polar Institute, the Universities of Oslo and Trondheim, Norway, National Wildlife Research Centre, Environment Canada, the Norwegian School of Veterinary Science and the National Veterinary Institute, Norway. Studies of the possible effects, particularly on the immune system and reproduction, of the very high OCs levels in polar bears are currently being investigated. Indications of possible immunotoxic effect<sup>15</sup>, and negative association between OCs and retinol and thyroid hormones<sup>16</sup> have been found, and low cub survival in the Svalbard population is reported<sup>17</sup>. Furthermore, immunization studies, which include studies of specific antibody response, immunoglobulin concentrations and occurrence of natural antibodies in polar bears from areas with high and low PCB exposure, to reveal possible effects of PCBs on the immune system are in progress. This study will focus on PCB contamination in polar bear young (cubs and yearlings) and their mothers at Svalbard.

#### Materials and methods

Sampling of 35 mother/cubs pairs of polar bears was conducted during late March to mid May in 1995 to 1998 from the Svalbard area. Polar bears (> 1 year) were captured by remote injection of a drug (Zoletil®) filled dart (Palmer Cap-Chur Equipment, Douglasville, Georgia) fired from a helicopter<sup>18</sup>. A vestigial premolar tooth was extracted from all bears (> 1 year old) for age

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determination<sup>19</sup>. Blood samples were collected from the femoral vein. After centrifuging the plasma was transferred and kept frozen in polypropylene containers at -20°C until analysis. The bears were measured and their physical condition assessed.

The PCB-analyses of the plasma samples were done at the Environmental Toxicology Laboratory at the Norwegian School of Veterinary Science, Oslo. The laboratory is accredited for these analyses according to the requirements of NS-EN45001 and ISO/IEC Guide 25. Lipids were extracted from the blood with cyclohexane and acetone. Cleanup was done with ultra-pure sulphuric acid. Details of the analytical method are described elsewhere<sup>20</sup>. The concentrations of PCBs were quantified by GC-ECD. In this study, sum PCB (sPCB) refers to six individual PCB congeners (IUPAC nos.<sup>21</sup>): 99, 118, 153, 156, 180, 194. On average these six congeners made up 78% of total PCB in polar bears (sum of 28 congeners). Quantification was performed using PCB 29, 112 and 207 as internal standards in each sample. Percent recoveries and CV of PCBs in spiked blood varied from 83 to 115% and 4.8 to 7.8%, respectively. Detection limits for the PCBs were 0.01 - 0.06 ng/g wet weight.

The samples from polar bears were grouped according to age and reproduction status: cubs (cubsof-the-year), females with cubs, yearlings and females with yearlings. Statistical analyses were performed using SAS<sup>22</sup>. The PCB and age data derivate from normal distribution (Shapiro-Wilk W test), and were  $log_{10}$  transformed to restore normality. Differences in PCB levels between cubs, yearlings, females with cubs, and females with yearlings were investigated using analysis of variance (ANOVA). To determine which PCB concentrations were significantly different, the Tukey t-test was used. Statistical significant level was set at  $p \le 0.05$ .

#### **Results and Discussion**

Figure 1 presents the mean levels and range of sPCB (log-transformed ng/g lipid weight) in cubs, yearlings, females with cubs, and females with yearlings. Sum PCB levels in the four groups of the bears were significantly different (ANOVA, p < 0.05). Sum PCB levels (geometric means (with ranges)) of cubs (12301 (3483-28984) ng/g lipid weight, n=33) were significantly higher (Tukey-test, p < 0.05) compared to sum PCB in females with cubs (5821 (1591-11584) ng/g lipid weight, n=25), and yearlings (6815 (1528-23485, n=12) ng/g lipid weight). Females with yearlings had significantly lower (Tukey-test, p < 0.05) sPCB levels (2945 (1056-7522) ng/g lipid weight, n=10). Mean percent extractable lipid of blood plasma from cubs, yearlings, females with cubs, and females with yearlings were 1.3%, 1.6%, 1.2%, 1.3%, respectively.

Levels of PCB exposure in blood plasma from three to four month old cubs at Svalbard have not been presented in earlier studies. The levels found in blood plasma from older bears were comparable with levels reported in other studies of polar bears at Svalbard<sup>2,15</sup>. Within each female group, sPCB concentrations were not significantly correlated with age, which is in accordance with earlier findings<sup>2</sup>. Geometric mean age (and range) of females with cubs was 8 yr (5-21 yr) and 12 yr (8-20 yr) for females with yearlings.

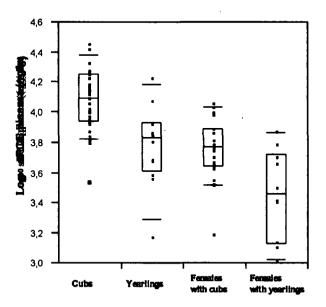


Figure 1:  $Log_{10}$  sPCB (ng/g lipid weight) in blood plasma of polar bears: cubs, yearlings, females with cubs, and females with yearlings.

The results indicate that in polar bears at Svalbard, cubs have higher PCB levels in blood plasma than older bears. Higher PCB concentrations in blood plasma from cubs compared to older bears were also found in a study of polar bears from a less exposed area<sup>23</sup>. High levels of OCs have been detected in polar bear milk and cub tissues<sup>2,23</sup>. Lactation is an important excretion route for lipophilic compounds<sup>24</sup>. An efficient generation transfer of OCs has been found in studies of seals<sup>25,26</sup> and of bears<sup>2,23</sup>. The generation transfer of OC residues seemed to be selective with respect to OC-groups and certain PCBs<sup>25</sup>. The actual OC pattern in offspring is of great importance for the toxicity assessment.

From birth to den emergence, the cubs are at critical stages at development and maturation of vital organs and functions, and are particularly sensitive to adverse effects of OCs. Effects may include immuno-, neuro-, developmental and reproductive toxicity. Adverse effects of environmental OC pollution have been demonstrated in marine mammals from particularly polluted waters<sup>6,7,27,28</sup>. The cub survival in the Svalbard population is low compared to other populations but the cause is unknown<sup>17</sup>. High PCB exposure or high population density could however explain such low survival.

The present study demonstrates a significantly higher PCB exposure in blood plasma in three to four month old polar bear cubs than in older bears at Svalbard, which may indicate a high risk for toxic effects for newborns. Studies are in progress to investigate disposition of PCB congeners and different pesticides in mother and offspring.

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