Pattern of mercury accumulation in the Antarctic minke whale and its prey based on JARPAII data

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ABSTRACT
To examine the features of accumulation and yearly changes of total Hg in Antarctic Ocean Areas IV and V, total Hg levels in samples from Antarctic krill (*Euphausia superba*), and Antarctic minke whales (*Balaenoptera bonaerensis*), were measured. Total Hg levels in Antarctic krill from Areas IV and V were in the range 0.006-0.026 and 0.003-0.052 ppm dry wt., respectively. There were no significant differences between Areas. No yearly changes of total Hg levels in Antarctic krill were observed in samples from both Areas in the period from 2005 to 2011. Hg levels in liver of Antarctic minke whales from Areas IV and V were in the range 0.003-0.13 and <0.001-0.25 ppm wet wt., respectively. Hepatic Hg levels of minke whales of all age groups in Area IV decreased significantly with research years and that of 15-26 year olds in Area V increased significantly.

KEYWORDS: ANTARCTIC MINKE WHALE; TRACE ELEMENT; BIOACCUMULATION, AGE TREND, FOOD AVAILABILITY

INTRODUCTION

In 1994, the International Whaling Commission (IWC) decided to establish a regular agenda item for research on the effects of environmental change on cetaceans in the Scientific Committee (SC) (IWC, 1995). At the IWC/SC meeting in 1994, the Committee addressed: (1) global warming; (2) ozone depletion; (3) pollution; (4) direct and indirect fisheries; (5) noise; and (6) other human activities (e.g. tourism, coastal development) (IWC, 1995). However, because of the difficulty to examine all topics simultaneously, it agreed to initially focus on two specialized items; the relationships between chemical pollutants and cetaceans, and the potential ecological effects of climate change and ozone depletion on cetaceans (IWC, 1999). The SC decided to hold two workshops (IWC, 1999). The JARPA was designed to cover these items by collecting related samples and data from the beginning of the program, in order to respond to a growing concern over environmental changes. Some results on the effects of environmental changes on cetaceans in JARPA surveys were reported in the JARPA review meeting in 1997 (Government of Japan, 1995; Fujise *et al.*, 1997).

Fujise *et al.* (1997), Watanabe *et al.* (1998) and Honda *et al.* (2006) reported that 'yearly changes of age-accumulation patterns of hepatic Hg in Antarctic minke whales (*Balaenoptera bonaerensis*) suggest possible environment changes such as changes of food availability.' This hypothesis was also supported by the studies on biological parameters (Zenitani *et al.*, 1997), body fatness and blubber thickness (Osumi *et al.*, 1997; Konishi *et al.*, 2008) of Antarctic minke whales. At the 2006 JARPA review meeting, Yasunaga *et al.* (2006) reported that Hg intake in young minke whales (1-15 years) significantly decreased with sampling years, while that of individuals over 16 years old was stable. These results can be interpreted as changes of the feeding environment of Antarctic minke whales in the later years of the JARPA.

The objective of the pollution studies in the JARPAII is to continuously monitor levels and behavior of Hg in minke whales and their food item, Antarctic krill (*Euphausia superba*). However, most of the samples for this study since 2005/2006 were lost after the 2011 earthquake and tsunami (see IWC, 2012). Therefore, the limited number of samples from 2007/2008 and 2010/2011 that were not lost in the disaster were compared with the previous JARPA data from 1988/1989 and 2004/2005, so that the recent trend of Hg levels in minke whales could be examined.

MATERIALS AND METHODS

Materials

Antarctic krill samples were obtained from stomach contents of a total of 30 Antarctic minke whales sampled from Areas IV (70-130E) in 2005/2006, 2007/2008 and 2009/2010, and V (130E-170W) in 2006/2007, 2008/2009 and 2010/2011 in JARPAII (Table 1, Fig. 1). All samples were stored in 10% formalin solution until analysis. Hg was measured from liver samples of 31 (11 males and 20 females) minke whales taken from Areas IV in 2007/2008 and those of 51 males taken from Area V in 2010/2011. Because of the absence of sex difference of hepatic Hg in minke whales (Fujise *et al.*, 1997; Yasunaga *et al.*, 2006), liver sample of males and females were combined in 2007/2008 (Table 1). All cryopreserved samples were stored in polyethylene bags at -20°C until analysis.

Laboratory analysis

In the laboratory the whole body of the krill were rinsed with milli-Q water, weighted, dried for 12 hr. at 80°C and reweighted. The samples homogenized using an agate mortar were measured twice. The liver samples were stripped externally to avoid contamination and were collected from three places. The samples were measured three times. For Hg analysis, the samples (ca. 100 mg) were set on a ceramic boat and were subjected to cold-vapor atomic absorption spectrometry with heat-vaporization, gold amalgamation method (Nippon Instruments Co., MA-3000). Accuracy and precision of the methods were confirmed using standard materials, DORM-3 (NRCC: muscle of dogfish).

Statistical analysis

Differences of Hg concentration in Antarctic krill among the samples from both areas were assessed by Kruscal-Wallis test, and those between Area IV and V were assessed by Mann-Whitney U test (Zar, 1999). Difference of Hg concentrations in the livers of minke whales between Areas IV and V was also assessed by Mann-Whitney U test (Zar, 1999). Temporal trends of hepatic Hg levels in minke whales were assessed by simple linear regression analysis (Zar, 1999). In the regression analysis, hepatic Hg levels in minke whales were logarithmically converted, because the distribution of persistent toxic metals, such as Hg, levels in biota are skewed to higher order. A p value of less than 0.05 was used as the criterion of statistical significance. These statistical analyses were executed by PASW Statics 17.0 for Windows (SPSS Co. Ltd.).

RESULTS

Antarctic krill

Hg levels in Antarctic krill form Areas IV and V were in the range 0.006-0.026 and 0.003-0.052 ppm dry wt., respectively (Table 2). Plots of Hg levels in krill during 2005/2006 and 2009/2010 in Area IV, during 2006/2007 and 2010/2011 in Area V against their sampling years are shown in Fig. 2. No significant differences were observed in Hg concentrations in krill among the research years in both areas.

Antarctic minke whale

Table 3 shows Hg levels in liver of minke whales form Area IV in 2007/2008 and Area V in 2010/2011, and those of JARPA data in 1988/89 and 2004/2005 (Yasunaga *et al.*, 2006). Total Hg levels in liver of minke whales from Areas IV in 2007/2008 and V in 2010/2011 were in the range 0.003-0.13 and <0.001-0.25 ppm wet wt., respectively. Plots of hepatic Hg levels in minke whales of 3 age groups (1-15, 16-25 and over 26 years old) in Area IV during a period from 1989/1990 to 2007/2008 and in Area V in the period from 1988/1989 to 2010/2011 are shown in Fig. 3a and 3b.

Hepatic Hg levels in Area V in 2010/2011 were significantly higher than those in Area IV in 2007/2008 in all age groups with Mann-Whitney U test ($p<0.05$). Significant linear regression equation were observed in in all year groups in Area IV ($p<0.05$, inclination: -) and 16-25 years old in Area V ($p<0.05$, inclination: +).

DISCUSSION

Hg levels in Antarctic krill

It is known that the minke whale is a useful species for examining Hg levels given its high position on the food chain and long life span (Honda *et al.*, 1987; Yasunaga *et al.*, 2009). Furthermore, Antarctic minke whales in the Antarctic Ocean mainly feed on the Antarctic krill (Ichii and Kato, 1991; Ichii et al., 1998).

It is important that temporal Hg levels of Antarctic krill in the research area are revealed for understanding the accumulation of Hg in minke whales. Yasunaga *et al.* (2009) reported that temporal trends of Hg concentrations in Antarctic krill were not observed during years from 1989 to 1999 using cryopreserved samples. We analyzed 30 formalin-stored krill samples during the period from 2005/2006 to 2010/2011 in both areas because cryopreserved samples for this monitoring study were lost after the 2011 earthquake and tsunami (see IWC, 2012). Formalin-stored samples are not necessary appropriate for Hg monitoring in biota, because mercury in the human organs immersed in 10% formalin solution is released out of the organs (Miyama and Suzuki, 1971). Accordingly, Hg levels in the formalin solution stored in 05/06-AM507, 07/08-AM307 and 09/10-AM464 were measured, and were 0.003, 0.001 and 0.002 ppm, respectively. This result suggests that the influence of the release of Hg is likely small for assessing spatial trends within a short period. Consequently, the results of no temporal and no regional trend of Hg in Antarctic krill in the period from 2005/2006 to 2010/2011 in both areas would be appropriate.

Spatial and temporal trend of hepatic Hg

No spatial significant difference of hepatic Hg levels between Area IV in 2007/08 and Area V in 2010/11 was observed. The results indicated that hepatic Hg levels of all age groups in Area IV (Fig. 3a) were significantly decreased with research years and that of 15-26 years old in Area V (Fig. 3b) were significantly increased.

Assuming a constant level of Hg in a food item, Hg levels in minke whales indicate food intake over a long period (Honda *et al.*, 1987; Fujise *et al.*, 1997). Honda *et al.* (1987) found no significant linear correlation between Hg in minke whales and ages, suggesting the possibility of the increased food intake after the 1980's. Fujise *et al.* (1997) reported hepatic Hg levels in younger minke whales were lower in the 1994/95 than those in 1980's, suggesting possibility that the increased food intake has begun to decrease in 1990's. Whether or not the increasing in hepatic Hg for age group 15-26 years old in Area V is related to a increased food intake is unknown at present.

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Table 1. Samples of Antarctic minke whales and Antarctic krill taken from the stomach contents examined in the present study.

Species (*Scientific name*)	Year	Area	*n* sex	Body length (m) Ave. (min.-max.)
Antarctic krill (*Euphausia superba*)	2005/2006	Area IV	5	
	2006/2007	Area V	5	
	2007/2008	Area IV	5	
	2008/2009	Area V	5	
	2009/2010	Area IV	5	
	2010/2011	Area V	5	
Antarctic minke whale (*Balaenoptera bonaerensis*)	2007/2008	Area IV	*11* males	7.95 (5.53-9.58)
			20 females	8.15 (6.79-9.07)
	2010/2011	Area V	*51* males	8.46 (5.31-9.67)

Table 2. Total Hg concentrations (μg/g dry wt.) of Antarctic krill taken from stomach of Antarctic minke whales in JARPAII surveys.

	research year	2005/2006	2007/2008	2009/2010	total
Area-IV	average±SD	0.019 ± 0.004	0.011 ± 0.004	0.013 ± 0.008	0.014 ± 0.006
	range	(0.012 - 0.022)	(0.006 - 0.016)	(0.007 - 0.026)	(0.006 - 0.026)
	n	5	5	5	5
	research year	2006/2007	2008/2009	2010/2011	total
Area-V	average±SD	0.036 ± 0.017	0.016 ± 0.012	0.017 ± 0.007	0.024 ± 0.015
	range	(0.017 - 0.052)	(0.003 - 0.031)	(0.011 - 0.028)	(0.003 - 0.052)
	n	5	5	5	5

Table 3. Total mercury concentrations in liver of Antarctic minke whales taken from Areas IV and V in JARPA and JARPAII surveys.

Area IV				
Age Class		1-15 years old	16-25 years old	Ovre 26 years old
89/90	Ave±SD	0.078 ± 0.036	0.078 ± 0.034	0.088 ± 0.041
	range	(0.027 - 0.166)	(0.033 - 0.14)	(0.021 - 0.16)
	n	41	32	27
91/92	Ave±SD	0.078 ± 0.037	0.070 ± 0.032	0.071 ± 0.029
	range	(0.036 - 0.15)	(0.037 - 0.14)	(0.045 - 0.13)
	n	15	10	11
93/94	Ave±SD	0.038 ± 0.019	0.069 ± 0.046	0.065 ± 0.057
	range	(0.012 - 0.080)	(0.024 - 0.16)	(0.014 - 0.20)
	n	16	8	12
95/96	Ave±SD	0.048 ± 0.028	0.061 ± 0.017	0.060 ± 0.024
	range	(0.012 - 0.15)	(0.039 - 0.086)	(0.035 - 0.15)
	n	30	13	27
97/98	Ave±SD	0.041 ± 0.027	0.086 ± 0.030	0.089 ± 0.056
	range	(0.004 - 0.098)	(0.049 - 0.13)	(0.038 - 0.16)
	n	13	6	6
99/00	Ave±SD	0.040 ± 0.024	0.067 ± 0.015	0.070 ± 0.041
	range	(0.007 - 0.076)	(0.044 - 0.084)	(0.029 - 0.13)
	n	10	5	5
01/02	Ave±SD	0.033 ± 0.018	0.054 ± 0.023	0.075 ± 0.017
	range	(0.008 - 0.064)	(0.031 - 0.086)	(0.056 - 0.092)
	n	10	5	5
03/04	Ave±SD	0.033 ± 0.028	0.060 ± 0.023	0.050 ± 0.034
	range	(0.004 - 0.096)	(0.034 - 0.087)	(0.019 - 0.11)
	n	10	5	5
07/08	Ave±SD	**0.037 ± 0.032**	**0.044 ± 0.027**	**0.038 ± 0.026**
	range	(**<0.001 - 0.13**)	(**0.018 - 0.082**)	(**0.003 - 0.066**)
	n	**23**	**4**	**4**

Area V				
Age Class		1-15 years old	16-25 years old	Ovre 26 years old
88/89	Ave±SD	0.061 ± 0.031	0.079 ± 0.033	0.095 ± 0.072
	range	(0.006 - 0.14)	(0.030 - 0.13)	(0.023 - 0.23)
	n	36	12	7
90/91	Ave±SD	0.067 ± 0.023	0.074 ± 0.030	0.088 ± 0.031
	range	(0.038 - 0.11)	(0.036 - 0.12)	(0.056 - 0.16)
	n	14	10	12
92/93	Ave±SD	0.082 ± 0.034	0.064 ± 0.037	0.084 ± 0.044
	range	(0.041 - 0.15)	(0.029 - 0.14)	(0.027 - 0.18)
	n	11	9	16
94/95	Ave±SD	0.067 ± 0.036	0.086 ± 0.041	0.12 ± 0.049
	range	(0.015 - 0.15)	(0.046 - 0.18)	(0.052 - 0.24)
	n	18	16	15
96/97	Ave±SD	0.069 ± 0.074	0.068 ± 0.032	0.095 ± 0.042
	range	(0.007 - 0.27)	(0.028 - 0.12)	(0.047 - 0.20)
	n	11	5	12
98/99	Ave±SD	0.050 ± 0.028	0.10 ± 0.049	0.13 ± 0.11
	range	(0.008 - 0.11)	(0.054 - 0.19)	(0.039 - 0.43)
	n	27	11	14
00/01	Ave±SD	0.051 ± 0.020	0.067 ± 0.030	0.13 ± 0.074
	range	(0.027 - 0.090)	(0.044 - 0.12)	(0.054 - 0.24)
	n	10	5	5
02/03	Ave±SD	0.044 ± 0.029	0.13 ± 0.066	0.13 ± 0.084
	range	(0.006 - 0.095)	(0.063 - 0.24)	(0.060 - 0.27)
	n	10	5	5
04/05	Ave±SD	0.048 ± 0.025	0.078 ± 0.037	0.075 ± 0.020
	range	(0.019 - 0.086)	(0.047 - 0.14)	(0.058 - 0.11)
	n	10	5	5
10/11	Ave±SD	**0.066 ± 0.040**	**0.11 ± 0.048**	**0.089 ± 0.042**
	range	(**<0.001 - 0.15**)	(**0.036 - 0.21**)	(**0.022 - 0.15**)
	n	**27**	**15**	**9**

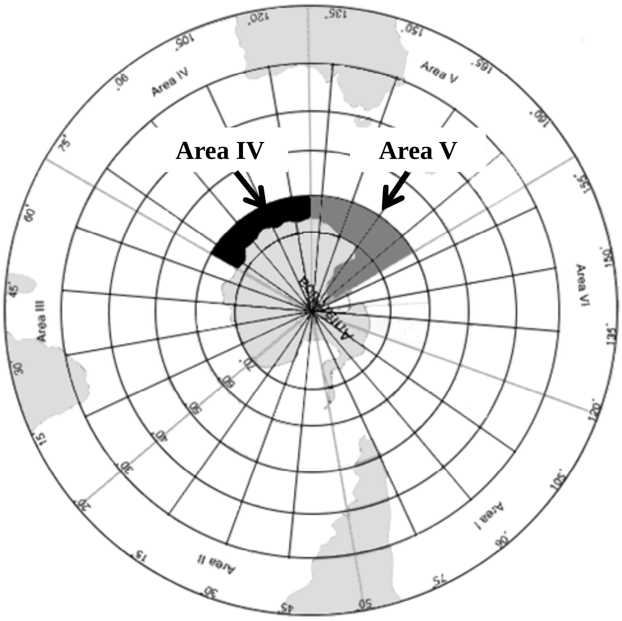

Figure 1. Sampling area of Antarctic minke whales in JARPAII surveys.

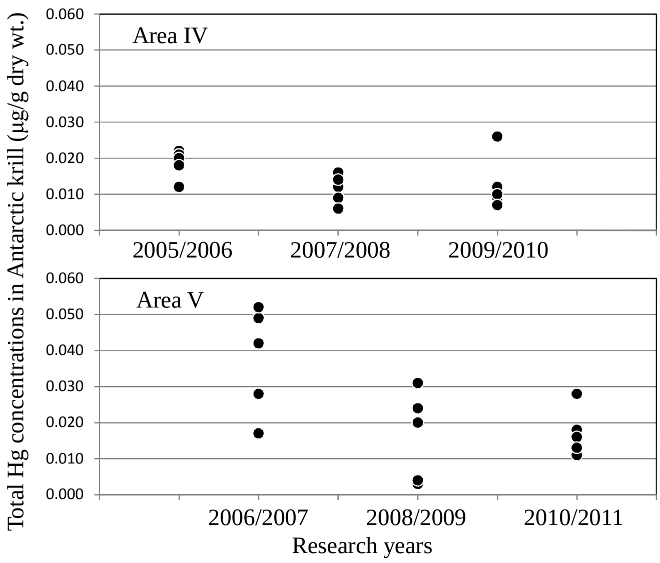

Figure 2. Total mercury concentrations (µg/g dry wt.) in Antarctic krill
taken from sotmach content of Antarctic minke whales in JARPAII surveys.

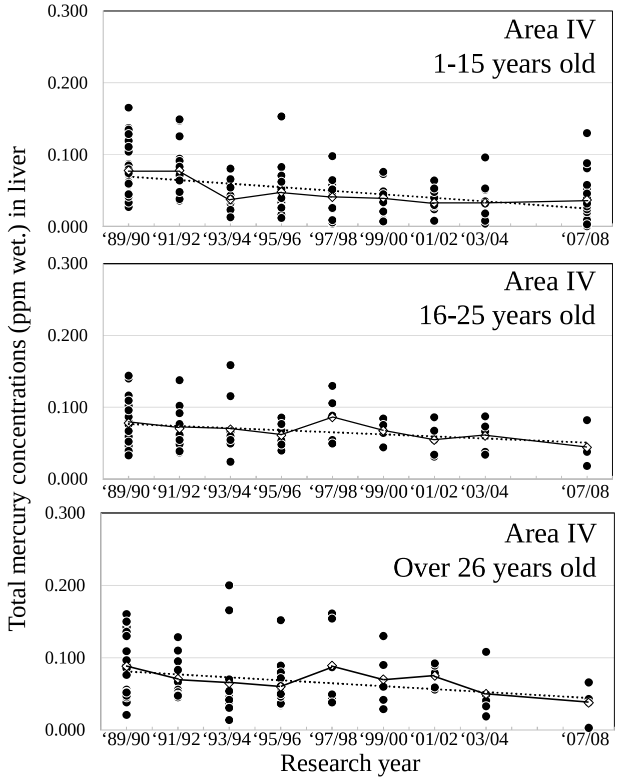

Figure 3a. Total mercury concentrations (ppm wet wt.) in liver of Antarctic minke whales in Area IV in the JARPA and JARPAII surveys.
●: Hg concentrations in individuals, ———: simple linear regression line
—◇—: Average of Hg concentrations in whales in each research year.

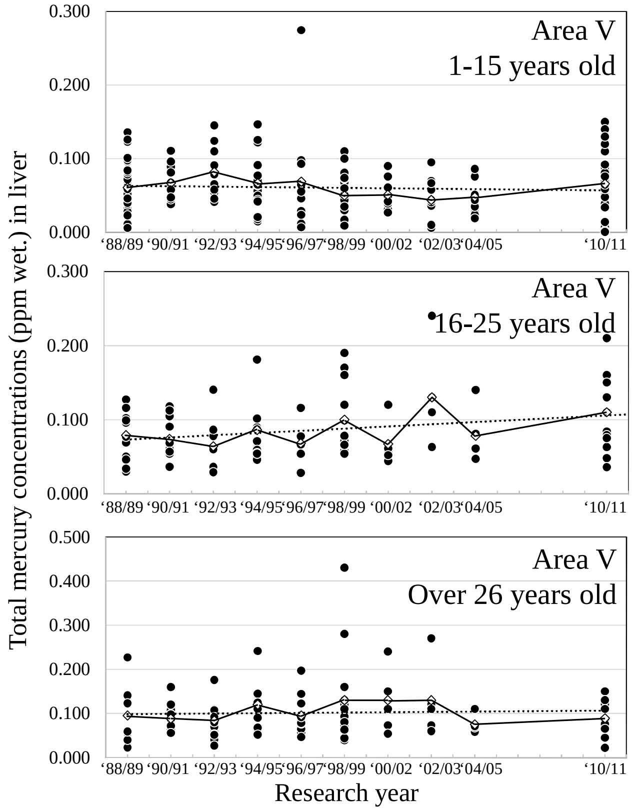

Figure 3b. Total mercury concentrations (ppm wet wt.) in liver of Antarctic minke whales in Area V in the JARPA and JARPAII surveys.
●: Hg concentrations in individuals, ———: simple linear regression line
—◇—: Average of Hg concentrations in whales in each research year.