the decrease of the porpoise around 1945 in such a large area. Disposal of war chemicals ?

It is stated several times that observations on living porpoises indicate a second decrease in number from 1960 on. Your reviewer, who can not find the relevant base for this conclusion in the paper, is content to follow the author. Though this statement, combined with a reference to the (multi-interpretable) stranding data is a poor introduction, the following discussion on the effects of pollution by the discharge of poisonous chemicals contains many veritable facts, the summary of which may be quite valuable to many a reader.

A number of Appendices contain the details about the observations on the presence of *Tursiops* and *Phocoena* in the Marsdiep area and annotated literature of food, reproduction and growth of these species.

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POLYCHLORINATED HYDROCARBONS AND HEAVY METALS IN HARBOUR PORPOISE *(PHOCOENA PHOCOENA)* AND WHITEBEAKED DOLPHIN *(LAGENORHYNCHUS ALBIROSTRIS)* FROM DANISH WATERS

by S. H. Andersen and A. Rebsdorff**.*

Introduction

The purpose of the present study was to investigate the level of persistent chlorinated hydrocarbons and heavy metals in different tissues of four harbour porpoises caught at Danish coasts and of one beached Whitenbeaked dolphin, and to compare the results with the levels in adjacent geographical regions. The investigations were also performed in a search for a possible explanation for the change in geographical distribution of the harbour porpoise in the North Sea area during the last 3 - 4 decades in connection with the many diseased animals taken in pond nets (ANDERSEN, 1972 & 1974).

Material

The samples originate from 5 male and 2 female harbour porpoises which had trapped themselves in pond nets in inner Danish waters in the period 1972—1973.
As for the trapped specimens, judging from their length and weight they have been

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agegrouped as follows : 1 year old : 1 specimen, 2 years old : 3 specimens and 3 years old : 3 specimens. Five of the animals died during transport or a few days after and 2 have been in captivity for 1 month and 1 year, respectively, and living on a diet of herring. Autopsies have been performed on 6 of them revealing fatal verminious bronchitis with 3 cases of dermatomycosis and from other material from inner Danish waters, which was put at our disposal by the Zoological Museum of Copenhagen.

The whitebeaked dolphin was included in the material for camparison with a more pelagic species. It was found beached in Kolding Fjord, December 1972, euthanized and brought to our laboratory. It was a female of 2.40 meter, weighing approximately 250 kg. The only abnormality noted during autopsy was a healed peritonitis.

Methods

A. Polychlorinated hydrocarbons.

After transport to the laboratory tissue samples were frozen at -18°C until required for analysis. The samples were ground with purified sea sand and anhydrous sodium sulphate and were then extracted in Soxhlet apparatus with petroleum ether for 4 hours. The crude extract was cleaned up by a n-hexane - dimethylformamide differential extraction. Further clean-up was accomplished by column chromatography on standardized aluminium oxide containing 5% water.

Separation between PCB and DDE on one hand was performed according to instructions from National Food Institute which were almost idenitcal to the procedure described by HOLDEN (1973). The separated eluates I and II were analyzed on a Varian 1800 gaschromatograph with a dual column and electron capture detectors. In eluate I the PCB's and most of the DDE (as a rule around 75% of the total amount) were determined, and in eluate II dieldrin, DDD, DDT and the remaining part of DDE were determined. The columns used were a 10% DC-200, and 1 : 1 mixture of 10% DC-200 and 15% QF-1, both on Chromosorb G. The oven temperature was 200°C, and usually 5 μl extract or diluted extract with added heptachlorepoxide as an internal standard was injected.

B. Total mercury.

In order to avoid loss of mercury the destruction of organic matter of the samples was made on native samples. The destruction was furthermore performed in closed teflon containers with an outer steel container with a screw cap able to withstand several atmospheres pressure. Usually 0.5 g sample was taken for oxidation with 5 ml concentrated nitric acid for 1 hour at 150°C. After cooling the clear remnant was transferred into a 50 ml volumetric flask and diluted to the mark with distilled water. An aliquot of diluted remnant was reduced with stannochloride, and the reduced mercury was transferred by a stream of air into a 20 cm cell with quartz windows, and the atomic absorption of the cold mercury vapours was determined at 253.7 nm using a Beckmann DU spectrophotometer (HOLAK et al. 1972, HOLDEN, 1973).

C. Other heavy metals.

The destruction of organic matter prior to analysis of the other metals was made on samples dried at 103°C. 2.5 dried sample was oxodized with 50 ml of a 50% v/v nitric acid for 4 hours in Erlenmeyer flasks placed on a sand bath. Evaporated water during the oxidation was replaced by adding water. The diluted destruates were analyzed by conventional atomic absorption spectrophotometry (Unicam SP 90).

Results

The results of the analyses are shown in tables 1-3 (Polychlorinated hydrocarbons) and 4-6 (Heavy metals), and in table 7 the total mercury concentration in museum samples is shown. High concentrations of polychlorinated hydrocarbons were found in blubber samples in both harbour porpoises and the whitebeaked dolphin, the maximum values being 81 ppm wet weight of total DDT residues and 125 ppm of PCB's (Table 1). In the whitebeaked dolphins higher concentrations were found in the fascial fat than in the blubber (Table 2). In the liver samples the higest values were 5.2 ppm total DDT residues and 11 ppm PCB's, respectively, in a harbour porpoise (Table 1).

TABLE 1 : Polychlorinated hydrocarbons in tissues of harbour porpoises, **Phocoena phocoena** from Danish waters — Average concentrations (ranges in brackets)

Tissue	Number of samples	p.p.m. wet weight						% Dry matter
		Dieldrin	p , p'-DDE	p , p'-DDD	Σ p , p'-DDT	p , p'-DDT	PCB	
Blubber 1)	4	8.1 (0.32-9.8)	16 (5.4-26)	15 (3.6-24)	18 (3.9-27)	53 (14-81)	78 (28-125)	74 (66-89)
Liver	4	0.36 (<0.02-0.71)	0.99 (0.17-2.0)	0.93 (0.20-1.8)	0.55 (<0.04-1.1)	2.5 (0.49-5.2)	5.4 (0.7-11)	26 (22-29)
Muscle	4	0.09 (<0.02-0.21)	0.26 (0.06-0.42	0.26 (0.05-0.48)	0.06 (0.04-0.09)	0.47 (0.16-1.0)	1.3 (0.3-2-1)	28 (26-30)

1) with adherent skin

TABLE 2 : Polychlorinated hydrocarbons in tissues of a whitebeaked dolphin, **Lagenorhynchus albirostris**, beached in Denmark.

Tissue	p.p.m. wet weight.						% Dry matter
	Dieldrin	DDE	DDD	DDT	Σ DDT	PCB	
Blubber 1)	7.7	19	6.5	12	40	82	67
Fascial fat	13	62	14	19	104	119	82
Liver	0.7	2.3	1.1	0.2	4.0	3.8	33
Muscle	0.09	0.36	0.13	0.17	0.72	2.0	27

1) with adherent skin

TABLE 3 : Polychlorinated hydrocarbons in tissues of dolphins from Danish waters.

Tissue		p.p.m. wet weight					% Dry matter
		Dieldrin	P , -DDE	p , p-DDD	p , p-DDT	PCB	
Brain	1)	0.13-0.29	0.36-0.40	0.08-0.36	0.34-0.45	0.85-1.2	27
Testicles	2)	0.05-0.15	0.20-0.24	0.16-0.25	<0.04-0.13		
Pancreas	3)	0.46	1.1	1.3	0.57	4.3	26
Kidney	3)	0.27	0.33	0.30	0.15	1.3	20
Lungs	3)	0.23	0.53	0.50	0.50	2.0	20
Spleen	1)	< 0.02-0.1	<0.05-0.29	<0.02-0.15	<0.04-0.04	0.5-4.3	25
Adrenal gland	3)	< 0.02	3.1	< 0.02	< 0.04	3.1	
Shoulder blade	3)	< 0.02	0.18	< 0.02	< 0.04	0.4	
Intestinal lymph glands	3)	< 0.1	9.3	< 0.1	< 0.2	11	
Pulmonary lymph glands	3)	< 0.2	27			38	
Ovary	4)	0.05	0.14	0.07	0.04	0.7	

1) One harbour porpoise and one whitebeaked dolphin.
2) Two harbour porpoises.
3) One harbour porpoise.
4) One whitebeaked dolphin.

TABLE 4 : Heavy metals in tissues of harbour porpoises, **Phocoena phocoena** from Danish waters. Means and ranges (ranges in brackets).

n = number of samples

Tissue	p.p.m. wet weight					% Dry matter
	n	Hg	Pb	Cu	Zn	
Blubber 1)	4	0.7 (0.5-0.9)	6.0 (<1.5-12)	1.5 (1.0-2.7)	290 (150-600)	74 (66-88)
Liver	4	22 (1.5-69)	3.5 (1.9-5.3)	4.5 (2.6-8.3)	59 (45-72)	26 (22-29)
Muscle	4	1.9 (0.8-3.2)	3.3 (1.6-4.7)		19 (18-21)	28 (26-30)

1) without skin

TABLE 5 : Heavy metals in tissues of whitebeaked dolphin, **Lagenorchynchus albirostris,** beached in Denmak.

Tissue	p.p.m. wet weight				% Dry matter
	Hg	Pb	Cu	Zn	
Fascial fat	1.3	<1.5	<2	5	82
Blubber 1)	0.9	5.4	<2	130	67
Liver	19	4.5	6.4	24	33
Muscle	2.0	2.2	1.4	13	27

1) without skin

Table 3 shows the measured concentrations of polychlorinated hydrocarbons in tissues which are more rarely measured. It may be noted that rather high concentrations of DDE and PCB's were found in intestinal and pulmonary lymph glands, although the highest concentrations were found in the blubber samples.

The concentrations of the metals were generally lower or within the ranges believed to be natural in the tissues under consideration. Only two liver samples showed high concentrations of total mercury, one of them belonged to a harbour porpoise (69 ppm) and the other to the whitebeaked dolphin (19 ppm), see tables 4 and 5. Table 6 shows the measured concentrations of the heavy metals in different tissues others than those most often taken for analysis. It may be seen from table 6 and 4, that the mercury concentration is highest in the liver, and the lead concentration is highest in liver and blubber while the zinc concentration in highest in the blubber.

TABLE 6 : Heavy metals in tissues of dolphins from Danish waters.

Tissue	p.p.m. wet weight				% Dry matter.
	Hg	Pb	Cu	Zn	
	1)	2)	1)	1)	
Testicles	0.7	1.8-3.0	0.9	25	
Pancreas 1)		1.1	1.6	35	26
Kidney 1)	1.6	2.0	2.8	21	20
Lungs 1)		1.7	1.4	20	20
Spleen 3)	1.2	1.4	0.7	18	25
Brain	3.0 3)	4.0 1) 710 3)	1.1 3)	13 3)	27

1) One harbour porpoise
2) Two harbour porpoises
3) One whitebeaked dolphin. The animal was euthanized with a shotgun, which explains the high value of lead.

TABLE 7 : Mercury analysis of skin and blubber from 3 museum specimens and one recent harbour porpoise, **Phocoena phocoena.**

	Year of capture	p.p.m. total Hg		% Dry matter	
		Blubber	Skin		Skin
Embryo, 6 months 1)	1943	0.02	0.51	64	35
Embryo, 6 months 1)	1941	0.50 2)		0.20 2)	
Adult 1)	1936	0.56	12.9	50	52
Adult	1973	0.19	0.54	89	42

1) Preserved in 70% ethanol -
2) Mixed sample of blubber and skin.

Discussion

The concentrations of polychlorinated hydrocarbons measured in the blubber of harbour porpoises and a whitebeaked dolphin (Table 1 and 2) from Danish waters are almost identical to those measured by HOLDEN & MARSDEN (1967) in harbour porpoises from East Scotland, and to the results of KOEMAN et al. (1972) in harbour porpoises from the North Sea, while the concentrations found by GASKIN et al. (1971) in porpoises from the Bay of Fundy Region were approximately six times higher for total DDT residues. On the other hand, porpoises from South-West Greenland (CLAUSEN et al. 1973), and a porpoise from the Orkney Islands (HOLDEN & MARSDEN 1967) are about ten times less contaminated with polychlorinated hydrocarbons than the porpoises from the Baltic Sea and the North Sea.

The highest accumulation of mercury in the tissues analyzed was found in liver samples (Table 4 and 5). The concentrations found were, for harbour porpoises, in ppm weight : 7.5 - 69 - 1.5 - 11. (Table 4) These figurs are similar to the concentrations in liver from harbour porpoises from the Bay of Fundy Region Region (GASKIN et al. 1972), while the concentrations in liver from three harbour porpoises analyzet bij KOEMAN et al. (1972) ranged from 5.7 to 192 with an average of 70 ppm. The liver of one porpoise from the French coast was analyzed by THIBAUD & DUGUY (1973), and it showed a total mercury concentration of 61 ppm.
The results in table 7 indicate a possible route of elimination of mercury in dolphins. Mercury content was determined separately in skin and blubber and the skin showed considerably higher concentration than the blubber. As shown by HARRISON and THURLEY (1974) the epidermis of dolphins has a fast turnover rate and this could contribute to the elimination of mercury and thus show a parallel to the relatively high concentration of mercury in mammalian hair. For future research in this field it should be clearly stated whether mercury determination is performed on pure skin, pure blubber or a mixture of both.

From the whitebeaked dolphin samples were taken from both blubber and the underlying fat in the loose connective tissue, fascial fat. This was done because the fascial fat in dolphins is a fat store available for the metabolism while the blubber obviously only has an insulating function. The latter keeps its thickness even in extreme emaciated individuals (harbour porpoises) while the fascial fat changes with the animals nutritional state (ANDERSEN, unpublished). It seems therefore more realistic to measure concentrations of a pollutant in the fascial fat than in the blubber. The polychlorinated hydrocarbons in the fascial fat showed considerably higher values than in the blubber whereas only Zn among the mesaured heavy metals showed there also substantial higher concentrations. Future investigations should also include samples of fascial fat.

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