Ecotoxicological and pathological studies of common guillemots *Uria aalge* beached on the Belgian coast during six successive wintering periods (1989-90 to 1994-95)

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ABSTRACT: During 6 successive wintering periods, 727 common guillemots *Uria aalge* were recovered from Belgian beaches. One-third of the birds were already dead; the rest passed through rehabilitation centres where they eventually died. All birds were monitored for general condition (body mass, fat reserves), eventual status of oiling and pathological changes (cachexia, acute hemorrhagic gastroenteropathy); 339 birds were sampled for trace metals (total and organic Hg, Cu, Zn, Fe, Cd) and PCB (polychlorinated biphenyl) analysis. Oiling is still a major cause of death for wintering pelagic seabirds: half of the birds showed signs of external or internal oiling, probably a still greater number of oiled birds never reach the shores. Although a low body mass can be considered a normal winter condition for wintering guillemots, pathology results showed that three-quarters of the studied animals were in a state of cachexia with emaciated pectoral muscle and lowered muscle lipid content. Elevated levels of Cu, Zn, Hg and PCBs were linked to the state of cachexia and may well represent an additional stress factor leading to the debilitation and death of part of the wintering guillemot population.

KEY WORDS: Heavy metals · PCBs · Cachexia · Guillemots · Belgian coast

INTRODUCTION

Although a relatively small ecosystem, the North Sea is known for its high fish productivity and catches. However, oil refineries, steelworks, metallurgy, chemical and paper industries form a dense network in the adjacent countries with subsequent busy shipping routes. During the past few decades, offshore gas and oil industries have developed rapidly.

To assess the human impact on this complex ecosystem, the Belgian authorities, within the frame of the 3rd European North Sea Conference, promoted a programme to monitor the health and causes of death of seabirds and marine mammals. Emphasis was placed

particularly on seabirds, which are found dead or dying on beaches in far larger numbers than are marine mammals. With a population of about 10 million wintering birds, the North Sea is one of the world's major areas for sea, shore and water birds (Birkhead 1974, Mead 1974, Bourne & Vauk 1988, Dunnet et al. 1990, North Sea Task Force Report 1993a, b). Pelagic species—petrels, auks and gannets—are particularly sensitive to ecosystem alteration such as depletion of fish stocks, oil spills, breeding site destruction, and chronic or acute organochlorine and heavy metals pollution (Bourne & Vauk 1988, Dunnet et al. 1990, Carter et al. 1993). In particular, oiling is known to be a severe threat to wintering seabirds (Mead & Baillie 1981, Stowe & Underwood 1984, Camphuysen & van Franeker 1992, Carter et al. 1993, Dahlmann et al. 1994). The common guillemot *Uria aalge* outnumbers

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by far all other wintering species. As a consequence, it became the focus of this study.

Seabird mortality and, in particular, winter strandings have been carefully reported and monitored along the Belgian coast (Kuyken 1978) and coasts of the neighbouring countries (e.g. Camphuysen & van Franeker 1992). Camphuysen & Leopold (1994) estimated the number of wintering guillemots in the 130 000 km² southern North Sea area at about 235 000 individuals for the 1984 and 1987 October–November peak period. A decline in density occurs around February–March as the birds move back towards the breeding grounds. To what extent birds dying at sea contribute to this decline is unclear, as the percentage of these birds finally reaching the shores is unknown.

Seabirds are likely candidates to accumulate toxic pollutants (organochlorines and heavy metals) and have been widely used as bioindicators (Muirhead & Furness 1988, Ohlendorf & Fleming 1988, Thompson 1990, Walsh 1990, Elliot et al. 1992, Thompson et al. 1992, Stewart et al. 1994, Burger & Gochfeld 1995, Wenzel & Gabrielsen 1995). Long-term chronic effects of contaminants may have severe consequences on reproduction, disease, stress susceptibility (immunosuppression) and behaviour patterns (Scheuhammer 1987, Peakall 1992). Despite extensive information on heavy metal and organochlorine levels in seabirds, few papers have considered the possible links to pathological findings. The aim of this paper is to combine ecotoxicological data and the most severe pathological ones (cachexia, acute and hemorrhagic gastro-enteropathy) in order to evaluate the possible causes of death of wintering common guillemots. Preliminary results concerning the mean heavy metal content of the birds collected between 1990 and 1993 suggested high levels of Cu, Zn and Hg (Bouquegneau et al. 1994).

MATERIAL AND METHODS

Collection and storage. A regular and systematic collection of stranded seabirds was organised along the 67 km of the Belgian shore during 6 successive winters (1989–90 to 1994–95). A total of 251 dead guillemots were collected from the beaches; an additional 476 live guillemots were collected and went through rehabilitation centres where they eventually died. Putrified specimens were discarded. Collected carcasses were kept frozen until necropsy was performed at the Pathology Department of the Veterinary College, Liège University, using a consistent protocol (Dorrestein & van der Hage 1993). They were weighed, and oil contamination on plumage and/or in intestinal tract and lesions were noted. Nutritional state, absence

of subcutaneous fat, and light to severe atrophy of pectoral muscle (visible signs of cachexia) were evaluated on a range from 0 to 3; specifically: 0, presence of subcutaneous fat, normal pectoral muscle; 1, absence of fat and slight pectoral muscle atrophy; 2, moderate pectoral muscle atrophy; 3, severe pectoral muscle atrophy. For statistical purposes, group 0 was tested against groups 1, 2, and 3 to compare normal versus cachectic birds. Necropsy technique involved opening of body cavities, dissection of the digestive tract, and examination of the respiratory, urinary and genital systems (Jauniaux et al. 1996). Intestinal serosal surface congestion, hyperaemic and thickened intestinal wall, and hemorrhagic content were used as parameters for acute and hemorrhagic gastro-enteropathy diagnosis (Dorrestein & van der Hage 1997). Parasites were identified on 248 guillemots and have been previously reported by Brosens et al. (1996). Respiratory tract mycetes (*Aspergillus* spp.) have been identified on 7 guillemots out of 198 (Jauniaux & Coignoul 1994). For bacteriology, all 727 birds were evaluated for evidence of intestinal salmonellosis following a classic isolation procedure reported elsewhere (Jauniaux et al. 1996). Three birds were positive for *Salmonella* (2 cases of *S. enteritidis* and 1 case of undetermined *Salmonella* spp.). Histopathology was restricted to lesions observed at necropsy. Most lesions were histolytic and bore freezing artifacts. The only significant lesions seen in guillemots were in relation with infectious agents such as *Aspergillus* spp. (see above). No test was used for virus isolation. Two age classes were considered based on the presence of cloacal *bursa fabricii* (Camphuysen & van Franeker 1992): class I comprising juvenile (1st winter) and immature (2nd and 3rd winter) birds; class II (4th winter and thereafter), consisting of mature but not necessarily breeding birds. Of a total of 727 birds, 339 (170 beached dead, 169 from rehabilitation centres) were dissected and samples of liver, kidney and pectoral muscle were collected for analysis of total Hg, organic Hg, polar lipids and PCBs (Laboratory for Ecotoxicology and Polar Ecology of Brussels Free University) and heavy metals, metallothioneins and total lipids (Oceanology Department of Liège University).

Total mercury and organic mercury analyses. Total mercury analyses were performed by specific atomic absorption spectrometry using a Perkin-Elmer MAS-50 Mercury analyser after the method described by Hatch & Ott (1968), modified by Bouquegneau (1973).

Organic mercury (MeHg) concentrations were measured by ECD semi-capillary gas chromatography on a Packard 437 following a toluene (Merck 8389) 3-step extraction (Uthe et al. 1972). Fresh weight/dry weight ratio was determined by lyophilising. Mercury concentrations were expressed as $\mu\text{g g}^{-1}$ dry weight (DW).

Quality control measurements for both total and organic mercury included replicate analysis resulting in coefficients of variation <10% and analysis of certified reference material (DORM-1, NRC, Canada) with a variation in the measurement up to 10% at the most. Limits of detection were 0.01 μg and 0.02 ng respectively, corresponding to 0.01 and 0.02 $\mu\text{g g}^{-1}$ DW for an average 1 g sample.

Other trace element analysis. Atomic absorption spectrophotometry (ARL 3510) was used to determine heavy metal concentrations (Cu, Zn, Cd, Fe). Pb, Ni, Cr and Ti contents were also determined but the results most often were below the detection limits and will not be discussed. After being weighed and dried for 48 h at 110°C, samples were digested with a mixed solution of chloric (Merck 317) and nitric (Merck 456) acids (1:3, v:v) and slowly heated to 100°C until complete digestion. The samples were then diluted, filtered and analysed. Parallel to the samples, a set of certified material samples (CRM 278 Community Bureau of Reference, Commission of the European Communities) was also analysed to ensure the method's sensitivity. Recoveries ranged from 92 to 102% for Cu, Zn and Fe and 80% for Cd. Limits of detection were 0.01 $\mu\text{g g}^{-1}$ DW for Cu, 0.33 for Zn and 0.22 for Cd. Concentrations are expressed as $\mu\text{g g}^{-1}$ DW.

PCB analysis. PCB (polychlorinated biphenyl) residues were determined by ECD-gas chromatography on a Shimadzu GC14A using a 30 m fused silica CPSil 8CB capillary column following a hexane extraction (Jansen 26.836.64) and florasil (Macherey-Nagel 81571) clean-up. PCBs were identified using a congener mixture including International Union of Pure and Applied Chemistry (IUPAC) congeners 28, 52, 101, 118, 138, 153, 156, 170, 180 and 194. Results were expressed as $\mu\text{g g}^{-1}$ DW. Since the sample PCB patterns did not sufficiently coincide with Aroclor 1254 or 1260 patterns, results were expressed as ΣPCB , or the sum of the 10 individually identified congeners, which represents $\pm 35\%$ of the total PCB load.

Sample preparation and lipids analysis. The method used for the total lipids extraction was described by Barnes & Blackstock (1973). The polar lipid content was determined gravimetrically after lipid hexane extraction included in the PCB procedure. Total and polar lipids are expressed as g g^{-1} DW.

Statistical analysis. All statistical tests were performed using Statistica® for Windows 5.1 computer programme. Tissue concentrations for each metal were tested to fit a normal distribution using Kolmogorov-Smirnov 1-sample tests. In the case of normal distribution, data were analysed using a *t*-test. When data significantly differed from a normal distribution, a non-parametric test (Mann-Whitney *U*-test) was used. Differences were considered significant when $p < 0.01$.

RESULTS AND DISCUSSION

None of the birds recovered in the present study were ringed, so that no information was available on their origin and/or their wanderings prior to death. This situation most probably reflects the fact that only a small proportion of birds are ringed and that not all dying birds are washed ashore (Pionneau 1987, Camphuysen & van Franeker 1992). Nevertheless, a small number of ringed guillemots ($n = 27$) found in Belgium during the 1980s and 1990s were mainly of Scottish origin (17/27); only a minor fraction came from Germany, Sweden, The Netherlands, the South of England and Ireland (W. Roggeman pers. comm.). Recoveries of guillemots during the 1980s in The Netherlands revealed that a majority of birds had been ringed in Scotland (Camphuysen & van Franeker 1992). With a necessary word of caution based on the fact that ringing efforts are not the same in all countries, it still seems reasonable to assume that most of the guillemots collected during the past 6 yr originated from the Scottish area. Several studies show that guillemots have no clear migration pattern, but rather disperse at sea, and that immature individuals are likely to show a higher mortality rate than adult birds (Birkhead 1974, Mead 1974, Nettleship & Evans 1985, Lloyd et al. 1991). Both Landsborough (1953) and Mead (1974) showed that guillemots ringed at colonies on the eastern coasts of England and Scotland had moved through the English Channel and the southern part of the North Sea. Aerial and ship surveys in the southern North Sea clearly indicate that large numbers of guillemots enter this area by October–November and move out again by February–March (Camphuysen & Leopold 1994).

A sample of 339 guillemots was fully investigated. During the 6 winters included in the study period, 89% of the birds were collected from January to March (Fig. 1). Peak densities (number of guillemots per km^2 sea surface) in the southern North Sea were recorded from October to January (Camphuysen & Leopold 1994). High densities, probably combined with severe environmental constraints such as low temperatures, storms and starvation, provoke an important mortality during the second half of the wintering period. A large proportion of the birds were oiled, either externally or both externally and internally, or showed clear signs of exhaustion, with emaciated pectoral muscle and very little or absence of abdominal and subcutaneous fat, 2 distinctive features of cachexia, a long and chronic condition (Table 1).

Significant differences appeared for Zn, Fe, total Hg, organic Hg and PCBs between dead birds from the beach and those provided by rehabilitation centres (Table 2). These high levels of pollutants in rehabili-

Fig. 1 *Uria aalge*. Overall stranding (percentage of total number) of guillemots (■, this work) compared to their densities (□, number per km²; Camphuysen & Leopold 1994) in the southern North Sea

tated birds are not likely to result from a decrease of body mass but probably instead from dietary changes. [This is indeed confirmed by the fact that $\delta^{13}\text{C}$ content of the tissues was lower in rehabilitated guillemots (S. Caulle, P. Dauby & V. Debacker unpubl. results).] For this reason, from the third winter on, we decided to focus only on individuals found dead, considering that birds that passed through rehabilitation centres could be an important bias. The following discussion therefore only refers to individuals washed ashore dead. Nevertheless, this sample is not necessarily fully representative of the 'natural' population.

Most of the birds were oiled (55%) and cachectic (76%) (Table 1). Sixty-one percent had developed acute hemorrhagic gastro-enteropathy. Thirty-one percent were oiled externally and internally; 24% showed only external traces; 45% showed no signs of oiling. Oiling is known to be a major cause of death for wintering guillemots entering the fairly polluted southern North Sea (Stowe & Underwood 1984, Camphuysen & Leopold 1994, Dahlmann et al. 1994, Camphuysen 1995). Partial or extensive oiling necessarily leads to starvation, debilitation and subsequent death, and eventual stranding.

We systematically examined the influence of age, sex, the most frequent lesions (cachexia, acute hemorrhagic gastro-enteropathy), and stable pollutant levels (heavy metals and PCBs) on the contamination levels of the tissues (Table 3). No clear-cut differences appeared between class I (juvenile and immature) versus class II (adult) birds, nor between male and female birds, except for cadmium concentrations which were twice as high in adult kidney ($p < 0.01$). The 2 groups displayed median Cd concentrations of 4.9 and 9.2 $\mu\text{g g}^{-1}$ DW, with different distribution patterns for age class I and class II (Fig. 2). Variations in kidney Cd levels are likely to reflect both dietary differences and age accumulation effects. Cd concentrations in the kidney has been shown to correlate with age in several seabird species (Thompson 1990, Lock et al. 1992).

One might expect a general increase of pollutant levels in the case of cachexia. Apart from a general decrease of subcutaneous fat, the total weight loss in the case of cachectic birds (708 ± 116 g, non-cachectic 781 ± 140 g) was linked to a general decrease in

Table 1. *Uria aalge*. Percentages of class I (juvenile and immature) and class II (adult), male and female, non-cachectic (–) and cachectic (+), acute hemorrhagic gastro-enteropathy negative (–) and positive (+), oiling: no oiling, external only (E) and external and internal oiling (E + I), of guillemots collected either directly from the beach (Beached) or after a stay in a rehabilitation centre (Centre)

	n	Age class I Juv. + imm.	Age class II Adult	Sex Male Female	Cachexia – +	Gastro-enteropathy – +	Oiling No E E + I
Beached							
Winter 1989-90	48	77	23	62 38	23 77	54 46	0 11 89
Winter 1990-91	31	45	55	60 40	18 82	54 46	9 0 91
Winter 1991-92	12	67	33	42 58	30 70	60 40	33 0 67
Winter 1992-93	75	54	46	49 51	12 88	34 66	56 24 20
Winter 1993-94	74	70	30	57 43	30 70	36 64	63 31 6
Winter 1994-95	11	70	30	80 20	27 73	9 91	36 67 0
All	251	65	35	56 44	24 76	39 61	45 24 31
Centre							
Winter 1989-90	83	67	33	69 31	13 87	58 42	0 31 69
Winter 1990-91	122	63	38	50 50	40 60	52 48	0 12 88
Winter 1991-92	64	83	17	65 35	37 63	60 40	12 23 65
Winter 1992-93	116	81	19	50 50	9 91	43 57	34 20 46
Winter 1993-94	76	67	33	63 37	18 82	43 57	49 42 9
Winter 1994-95	15	60	40	64 36	40 60	47 53	53 47 0
All	476	75	25	61 39	26 74	55 45	13 23 64

Table 2. *Uria aalge*. Body mass (g) and trace elements concentrations ($\mu\text{g g}^{-1}$ DW) in liver, kidney and muscle of guillemots, collected either directly from the beach (Beach) or after a stay in a rehabilitation centre (Centre), expressed as a mean $\pm$ standard deviation, median, range of concentrations (minimum – maximum), and number of samples (n); nd: non determined, <dl: below detection limit, ns: not significant. Total and polar lipids are expressed as g g^{-1} DW. Statistical significant differences at $p < 0.01$ are given in bold

	Body mass								
	Beach	Centre	p						
	725 $\pm$ 125 700 440–1180 (168)	634 $\pm$ 102 600 465–1280 (164)	<0.01						
	Liver			Kidney			Muscle		
	Beach	Centre	p	Beach	Centre	p	Beach	Centre	p
Cu	52 $\pm$ 17 52 14 – 100 (144)	51 $\pm$ 26 50 10 – 152 (104)	ns	28 $\pm$ 12 27 1.1 – 76.3 (110)	31 $\pm$ 13 31 2 – 74 (53)	ns	18 $\pm$ 6 18 9 – 53 (145)	20 $\pm$ 11 18 3 – 90 (107)	ns
Zn	145 $\pm$ 39 138 66 – 328 (144)	168 $\pm$ 46 158 84 – 413 (104)	<0.01	169 $\pm$ 41 168 41 – 284 (111)	173 $\pm$ 46 176 37 – 286 (54)	ns	60 $\pm$ 14 59 31 – 131 (145)	73 $\pm$ 31 67 36 – 235 (107)	<0.01
Fe	2549 $\pm$ 1354 2274 393 – 5928 (144)	3557 $\pm$ 1564 3468 775 – 7946 (104)	<0.01	613 $\pm$ 294 569 122 – 2376 (111)	700 $\pm$ 257 668 367 – 1759 (53)	<0.05	669 $\pm$ 241 641 337 – 2428 (145)	903 $\pm$ 784 732 78 – 5724 (107)	<0.01
Cd	2.4 $\pm$ 1.6 2.1 <dl – 10.1 (144)	2.5 $\pm$ 1.9 2.1 <dl – 9.7 (104)	ns	7.8 $\pm$ 6.5 6.4 <dl – 39.9 (111)	6.2 $\pm$ 5.3 4.7 <dl – 30.2 (54)	ns	<dl <dl	<dl <dl	
Total Hg	5.9 $\pm$ 2.9 5.4 0.8 – 20.7 (156)	7.9 $\pm$ 6.3 6.0 1.2 – 35.8 (125)	<0.05	4.6 $\pm$ 3.0 4.0 1.0 – 23.8 (90)	5.7 $\pm$ 2.4 4.5 4.4 – 9.3 (4)	ns	2.1 $\pm$ 1.1 1.8 0.3 – 6.7 (163)	3.8 $\pm$ 3.4 2.8 0.4 – 23.2 (139)	<0.01
Org. Hg	4.6 $\pm$ 2.2 4.1 0.8 – 14.1 (138)	6.6 $\pm$ 5.4 5.0 1.3 – 32.3 (105)	<0.01	3.3 $\pm$ 1.5 3.0 1.0 – 6.9 (55)	5.2 $\pm$ 1.8 4.6 3.6 – 7.8 (4)	<0.05	1.6 $\pm$ 0.8 1.4 0.3 – 4.9 (136)	3.1 $\pm$ 2.8 2.2 0.4 – 17.8 (114)	<0.01
Inorg. Hg	1.1 $\pm$ 1.1 0.9 0.0 – 6.5 (135)	1.3 $\pm$ 1.6 0.9 0.0 – 10.8 (105)	ns	0.9 $\pm$ 0.7 0.7 0.0 – 2.6 (54)	0.6 $\pm$ 0.7 0.5 0.0 – 1.5 (4)	ns	0.4 $\pm$ 0.4 0.3 0.0 – 1.9 (136)	0.6 $\pm$ 0.8 0.4 0.0 – 5.3 (113)	ns
Sum PCB	5.7 $\pm$ 6.0 3.5 0.3 – 27.2 (130)	11.7 $\pm$ 13.0 8.7 1.0 – 60.4 (68)	<0.01	3.4 $\pm$ 2.8 2.6 0.1 – 12.8 (88)	2.6 $\pm$ 1.6 2.2 1.1 – 4.7 (4)	ns	2.1 $\pm$ 1.8 1.6 0.1 – 10.5 (130)	5.4 $\pm$ 10.0 3.0 0.2 – 81.9 (77)	<0.01
Total lipids	0.18 $\pm$ 0.07 0.17 0.03 – 0.60 (120)	0.16 $\pm$ 0.07 0.15 0.06 – 0.33 (32)	ns	nd nd	nd nd		0.10 $\pm$ 0.08 0.08 0.01 – 0.63 (119)	0.14 $\pm$ 0.12 0.10 0.05 – 0.65 (31)	<0.01
Polar lipids	0.11 $\pm$ 0.03 0.11 0.04 – 0.29 (130)	0.11 $\pm$ 0.04 0.10 0.07 – 0.27 (68)	ns	0.12 $\pm$ 0.02 0.12 0.03 – 0.17 (88)	0.07 $\pm$ 0.04 0.08 0.01 – 0.11 (4)	<0.05	0.04 $\pm$ 0.03 0.03 0.01 – 0.16 (130)	0.05 $\pm$ 0.04 0.04 0.01 – 0.19 (77)	ns

muscle lipid content. Elevated liver levels for PCBs in the case of cachectic birds might indicate a remobilization after depletion of fat deposits (Fig. 3). It is also worth noting that the highest levels for PCBs, particularly in the liver, were always found in cachectic animals. For all tissues, significantly higher levels of Zn were also linked to the status of cachexia.

Acute hemorrhagic gastro-enteropathy showed no clear relation with levels of stable contaminants, except in the case of organic Hg, which was found in higher concentrations in the kidney of animals which had developed acute hemorrhagic gastro-enteropathy (Table 3b). The inflammatory nature of the intestinal lesion could not be conclusively assessed, due to the

Table 3. *Uria aalge*. Body mass (g) and trace elements concentrations ($\mu\text{g g}^{-1}$ DW) in (a) liver, (b) kidney and (c) muscle of guillemots expressed as a mean $\pm$ standard deviation, median and number of samples (n) in individuals found dead on the shores (n = 170): class I (juvenile and immature) and class II (adult), male and female, non-cachectic (–) and cachectic (+), acute hemorrhagic gastro-enteropathy negative (–) and positive (+), oiling: no oiling, external and internal oiling (E + I) and external only (E). Total and polar lipid content expressed as g g^{-1} DW. Statistical significant differences at $p < 0.01$ are given in bold

	Age class I Juv. + imm.	Age class II Adult	Sex		Cachexia		Gastro-enteropathy			Oiling	
			Male	Female	–	+	–	+	No	E + I	E
Body mass	707 $\pm$ 109 688 (92)	761 $\pm$ 133 725 (49)	725 $\pm$ 123 700 (86)	717 $\pm$ 117 700 (65)	781 $\pm$ 140 760 (40)	708 $\pm$ 116 680 (126)	748 $\pm$ 132 707 (64)	711 $\pm$ 120 687 (102)	698 $\pm$ 116 680 (75)	745 $\pm$ 123 715.0 (51)	747 $\pm$ 137 700.0 (41)
(a) Liver											
Cu	51 $\pm$ 17 51 (75)	52 $\pm$ 17 53 (42)	53 $\pm$ 17 53 (74)	51 $\pm$ 16 51 (54)	48 $\pm$ 20 45 (36)	53 $\pm$ 16 52 (108)	52 $\pm$ 17 57 (55)	52 $\pm$ 17 52 (89)	51 $\pm$ 16 52 (66)	52 $\pm$ 16 50 (39)	53 $\pm$ 21 55 (38)
Zn	145 $\pm$ 39 137 (75)	150 $\pm$ 38 147 (42)	147 $\pm$ 46 134 (74)	145 $\pm$ 28 143 (54)	129 $\pm$ 33 125 (36)	151 $\pm$ 40 143 (108)	148 $\pm$ 47 130 (55)	144 $\pm$ 33 140 (89)	142 $\pm$ 34 136 (66)	150 $\pm$ 30 145 (39)	147 $\pm$ 54 132 (38)
Fe	2325 $\pm$ 1202 2178 (75)	2867 $\pm$ 1438 2554 (42)	2353 $\pm$ 1087 2231 (74)	2758 $\pm$ 1603 2372 (54)	2528 $\pm$ 1391 2130 (36)	2557 $\pm$ 1347 2320 (108)	2701 $\pm$ 1484 2337 (55)	2455 $\pm$ 1266 2260 (89)	2509 $\pm$ 1424 2318 (66)	2499 $\pm$ 1335 2189 (39)	2679 $\pm$ 1288 2362 (38)
Cd	2.2 $\pm$ 1.6 1.9 (75)	2.7 $\pm$ 1.4 2.3 (42)	2.4 $\pm$ 1.5 2.2 (74)	2.7 $\pm$ 1.8 2.2 (54)	2.5 $\pm$ 2.3 2.0 (36)	2.4 $\pm$ 1.3 2.2 (108)	2.7 $\pm$ 2.1 2.3 (55)	2.2 $\pm$ 1.3 2.0 (89)	2.7 $\pm$ 2.1 2.1 (66)	2.2 $\pm$ 1.2 2.0 (39)	2.2 $\pm$ 1.2 2.1 (38)
Total Hg	6.4 $\pm$ 3.2 5.7 (87)	5.1 $\pm$ 2.4 4.7 (45)	4.3 $\pm$ 2.1 4.1 (20)	5.9 $\pm$ 2.4 6.0 (24)	5.3 $\pm$ 2.3 4.9 (37)	6.1 $\pm$ 3.1 5.5 (118)	5.8 $\pm$ 2.8 5.5 (57)	6.0 $\pm$ 3.1 5.3 (98)	5.3 $\pm$ 1.9 5.3 (71)	6.9 $\pm$ 3.9 6.9 (43)	5.7 $\pm$ 3.0 5.5 (41)
Org. Hg	4.9 $\pm$ 2.4 4.2 (75)	4.2 $\pm$ 2.1 3.9 (41)	3.5 $\pm$ 1.8 3.0 (17)	4.9 $\pm$ 2.1 4.8 (23)	4.3 $\pm$ 2.0 4.3 (34)	4.7 $\pm$ 2.3 4.0 (103)	4.6 $\pm$ 2.3 4.2 (51)	4.6 $\pm$ 2.3 3.9 (86)	4.2 $\pm$ 1.3 4.1 (63)	5.1 $\pm$ 2.8 4.3 (37)	4.6 $\pm$ 2.7 3.9 (37)
Inorg. Hg	1.2 $\pm$ 1.3 0.9 (74)	1.0 $\pm$ 1.1 0.7 (40)	1.0 $\pm$ 1.0 0.7 (17)	1.1 $\pm$ 1.2 0.5 (22)	0.8 $\pm$ 0.9 0.6 (33)	1.2 $\pm$ 1.2 0.9 (101)	1.1 $\pm$ 1.0 0.8 (49)	1.2 $\pm$ 1.2 0.9 (85)	1.0 $\pm$ 0.8 0.9 (63)	1.4 $\pm$ 1.5 0.8 (34)	1.2 $\pm$ 1.2 0.7 (37)
Sum PCB	5.2 $\pm$ 5.8 3.3 (68)	7.5 $\pm$ 6.8 5.5 (40)	4.2 $\pm$ 2.6 4.1 (17)	10.4 $\pm$ 7.8 9.8 (22)	2.8 $\pm$ 2.9 1.7 (31)	6.6 $\pm$ 6.4 4.2 (99)	6.0 $\pm$ 6.4 3.5 (46)	5.6 $\pm$ 5.8 3.5 (84)	4.8 $\pm$ 4.8 3.3 (68)	8.1 $\pm$ 7.1 5.0 (22)	6.0 $\pm$ 6.9 3.5 (39)
Total lipids	0.17 $\pm$ 0.08 0.17 (58)	0.18 $\pm$ 0.05 0.18 (35)	0.17 $\pm$ 0.08 0.17 (60)	0.18 $\pm$ 0.05 0.18 (44)	0.18 $\pm$ 0.07 0.18 (32)	0.18 $\pm$ 0.07 0.17 (88)	0.19 $\pm$ 0.05 0.18 (43)	0.17 $\pm$ 0.07 0.17 (77)	0.18 $\pm$ 0.07 0.17 (66)	0.18 $\pm$ 0.06 0.19 (17)	0.18 $\pm$ 0.06 0.17 (36)
Polar lipids	0.11 $\pm$ 0.03 0.11 (68)	0.11 $\pm$ 0.04 0.10 (40)	0.12 $\pm$ 0.04 0.12 (17)	0.10 $\pm$ 0.02 0.1 (22)	0.11 $\pm$ 0.05 0.1 (31)	0.11 $\pm$ 0.03 0.11 (99)	0.11 $\pm$ 0.03 0.10 (46)	0.11 $\pm$ 0.03 0.11 (84)	0.11 $\pm$ 0.03 0.11 (68)	0.09 $\pm$ 0.02 0.09 (22)	0.12 $\pm$ 0.04 0.11 (39)
(b) Kidney											
Cu	28 $\pm$ 14 27 (53)	29 $\pm$ 11 29 (33)	28 $\pm$ 13 27 (56)	29 $\pm$ 13 29 (40)	22 $\pm$ 12 18 (27)	30 $\pm$ 12 29 (83)	27 $\pm$ 13 27 (38)	28 $\pm$ 12 27 (72)	29 $\pm$ 11 28 (61)	25 $\pm$ 13 25 (12)	27 $\pm$ 14 27 (36)
Zn	170 $\pm$ 42 170 (53)	175 $\pm$ 40 182 (33)	169 $\pm$ 42 164 (56)	177 $\pm$ 39 187 (40)	150 $\pm$ 46 147 (28)	176 $\pm$ 37 177 (83)	161 $\pm$ 45 160 (37)	174 $\pm$ 38 173 (73)	174 $\pm$ 41 176 (61)	159 $\pm$ 35 155 (12)	164 $\pm$ 42 167 (37)
Fe	597 $\pm$ 320 563 (53)	622 $\pm$ 259 629 (33)	610 $\pm$ 216 594 (56)	592 $\pm$ 358 559 (40)	748 $\pm$ 278 694 (28)	567 $\pm$ 287 540 (83)	617 $\pm$ 270 604 (37)	611 $\pm$ 307 563 (73)	542 $\pm$ 225 529 (61)	688 $\pm$ 259 689 (12)	702 $\pm$ 375 650 (37)
Cd	5.9 $\pm$ 4.0 4.9 (53)	10.4 $\pm$ 6.8 9.2 (33)	7.2 $\pm$ 6.2 6.2 (56)	9.4 $\pm$ 7.3 7.1 (40)	9.2 $\pm$ 10.0 5.9 (28)	7.3 $\pm$ 4.9 6.5 (83)	8.6 $\pm$ 8.1 6.5 (38)	7.3 $\pm$ 5.6 6.3 (73)	8.3 $\pm$ 7.5 6.3 (61)	4.4 $\pm$ 2.5 3.8 (12)	7.9 $\pm$ 5.6 6.6 (37)
Total Hg	4.9 $\pm$ 3.5 4.0 (43)	4.3 $\pm$ 2.6 3.5 (24)	3.9 $\pm$ 2.1 3.6 (12)	5.1 $\pm$ 3.0 3.8 (11)	4.0 $\pm$ 1.9 3.5 (26)	4.8 $\pm$ 3.2 4.1 (64)	4.0 $\pm$ 2.2 3.5 (32)	4.9 $\pm$ 3.2 4.2 (58)	4.0 $\pm$ 1.5 4.0 (49)	8.7 $\pm$ 6.9 7.2 (17)	4.5 $\pm$ 2.6 3.5 (33)
Org. Hg	3.6 $\pm$ 1.5 3.1 (25)	2.9 $\pm$ 1.6 2.5 (16)	2.9 $\pm$ 1.3 2.5 (9)	3.1 $\pm$ 2.0 2.5 (6)	2.9 $\pm$ 1.5 2.4 (19)	3.5 $\pm$ 1.4 3.3 (36)	2.6 $\pm$ 0.9 2.4 (18)	3.6 $\pm$ 1.6 3.7 (37)	3.0 $\pm$ 1.0 3.0 (33)	4.9 $\pm$ 1.7 5.9 (3)	3.5 $\pm$ 1.8 3.9 (18)
Inorg. Hg	0.8 $\pm$ 0.7 0.7 (25)	0.9 $\pm$ 0.8 0.6 (15)	0.8 $\pm$ 0.9 0.5 (8)	1.1 $\pm$ 0.7 1.0 (6)	0.8 $\pm$ 0.6 0.7 (18)	0.9 $\pm$ 0.7 0.8 (36)	0.8 $\pm$ 0.6 0.8 (18)	0.9 $\pm$ 0.7 0.7 (36)	0.8 $\pm$ 0.6 0.8 (32)	1.2 $\pm$ 0.8 0.9 (3)	0.9 $\pm$ 0.8 0.7 (18)
Sum PCB	3.0 $\pm$ 2.4 2.4 (43)	3.3 $\pm$ 3.1 2.8 (25)	2.6 $\pm$ 1.9 2.8 (13)	4.5 $\pm$ 3.9 3.4 (11)	2.9 $\pm$ 2.8 2.1 (27)	3.6 $\pm$ 2.8 2.8 (61)	3.6 $\pm$ 3.1 2.9 (32)	3.3 $\pm$ 2.7 2.6 (56)	3.2 $\pm$ 2.7 2.5 (46)	4.9 $\pm$ 4.4 2.8 (7)	3.2 $\pm$ 2.7 2.8 (34)
Total lipids	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd	nd
Polar lipids	0.12 $\pm$ 0.02 0.12 (43)	0.12 $\pm$ 0.03 0.13 (25)	0.12 $\pm$ 0.03 0.13 (13)	0.11 $\pm$ 0.03 0.11 (11)	0.11 $\pm$ 0.02 0.11 (27)	0.12 $\pm$ 0.02 0.12 (61)	0.11 $\pm$ 0.02 0.12 (32)	0.12 $\pm$ 0.02 0.12 (56)	0.12 $\pm$ 0.2 0.12 (46)	0.11 $\pm$ 0.03 0.11 (7)	0.12 $\pm$ 0.02 0.12 (34)

Table 3 (continued)

	Age class I Juv. + imm.	Age class II Adult	Sex		Cachexia		Gastro-enteropathy		No	Oiling E + I	E
			Male	Female	-	+	-	+			
(c) Muscle											
Cu	18 ± 6 18 (77)	16 ± 4 16 (41)	18 ± 6 18 (76)	18 ± 5 17 (53)	18 ± 5 18 (37)	18 ± 6 18 (108)	17 ± 5 17 (56)	19 ± 6 18 (89)	20 ± 7 19 (65)	16 ± 4 16 (41)	18 ± 5 18 (38)
Zn	61 ± 15 59 (77)	58 ± 11 57 (41)	58 ± 16 55 (76)	61 ± 11 61 (53)	53 ± 12 52 (37)	62 ± 14 62 (108)	57 ± 13 55 (56)	62 ± 15 61 (89)	63 ± 15 63 (65)	56 ± 11 54 (41)	59 ± 15 59 (38)
Fe	693 ± 292 643 (77)	612 ± 130 584 (41)	688 ± 297 641 (76)	649 ± 131 640 (53)	586 ± 145 580 (37)	697 ± 260 645 (108)	646 ± 238 590 (56)	683 ± 242 663 (89)	711 ± 264 663 (65)	601 ± 107 586 (41)	674 ± 289 648 (38)
Cd	<dl	<dl	<dl	<dl	<dl	<dl	<dl	<dl	<dl	<dl	<dl
Total Hg	2.2 ± 1.1 1.9 (89)	2.0 ± 1.2 1.7 (49)	1.6 ± 0.9 1.4 (20)	2.3 ± 1.4 1.9 (28)	2.0 ± 0.8 2.1 (39)	2.1 ± 1.2 1.7 (122)	2.2 ± 1.3 2.0 (62)	2.0 ± 1.0 1.7 (99)	1.9 ± 0.8 1.8 (72)	2.5 ± 1.3 2.3 (49)	2.0 ± 1.3 1.5 (41)
Org. Hg	1.7 ± 0.8 1.6 (77)	1.5 ± 0.9 1.3 (39)	1.2 ± 0.8 1.0 (15)	1.7 ± 0.9 1.4 (23)	1.5 ± 0.7 1.5 (33)	1.7 ± 0.9 1.4 (101)	1.7 ± 1.0 1.4 (49)	1.6 ± 0.8 1.4 (85)	1.5 ± 0.6 1.4 (64)	1.8 ± 0.9 1.5 (35)	1.6 ± 1.1 1.3 (36)
Inorg. Hg	0.4 ± 0.4 0.3 (77)	0.4 ± 0.4 0.2 (39)	0.3 ± 0.3 0.2 (15)	0.4 ± 0.5 0.3 (23)	0.4 ± 0.4 0.3 (33)	0.4 ± 0.4 0.3 (101)	0.4 ± 0.4 0.3 (49)	0.4 ± 0.4 0.3 (85)	0.3 ± 0.4 0.2 (64)	0.6 ± 0.4 0.5 (35)	0.3 ± 0.4 0.2 (36)
Sum PCB	1.8 ± 1.4 1.3 (68)	2.6 ± 2.1 2.1 (40)	1.9 ± 0.9 1.6 (17)	3.1 ± 2.5 2.5 (22)	2.5 ± 2.3 2.0 (31)	2.0 ± 1.5 1.6 (99)	2.4 ± 1.9 1.9 (46)	2.0 ± 1.7 1.6 (64)	1.7 ± 1.4 1.3 (38)	2.9 ± 2.5 2.0 (22)	2.4 ± 1.7 2.0 (39)
Total lipids	0.08 ± 0.04 0.08 (59)	0.11 ± 0.08 0.08 (33)	0.11 ± 0.09 0.08 (61)	0.09 ± 0.05 0.08 (42)	0.13 ± 0.11 0.11 (32)	0.09 ± 0.05 0.08 (87)	0.12 ± 0.11 0.08 (43)	0.09 ± 0.04 0.08 (76)	0.09 ± 0.05 0.08 (65)	0.09 ± 0.05 0.07 (17)	0.13 ± 0.11 0.10 (36)
Polar lipids	0.04 ± 0.02 0.03 (68)	0.05 ± 0.04 0.04 (40)	0.06 ± 0.033 0.04 (17)	0.04 ± 0.03 0.03 (22)	0.07 ± 0.04 0.05 (31)	0.04 ± 0.01 0.03 (99)	0.05 ± 0.04 0.04 (46)	0.04 ± 0.02 0.03 (84)	0.03 ± 0.01 0.03 (68)	0.05 ± 0.03 0.04 (22)	0.05 ± 0.04 0.04 (39)

poor quality of the material for histopathology. However, we felt the lesion was worth mentioning, since it affected 61 % of the birds and had no clear correlation with decay. Previous reports mentioned a hemorrhagic gastro-enteritis as a terminal lesion, related to stress, in marine birds (Dorrestein & van der Hage 1993, Leighton 1993). In addition, parasitological and bacteriological examinations failed to isolate a likely infectious cause for that lesion (Jauniaux & Coignoul 1994,

Brosens et al. 1996, Jauniaux et al. 1996). No significant overall trend could be linked to oiling status when comparing non-oiled and externally oiled birds, which could partially be explained by the fact that external oiling may have occurred as a postmortem artifact. However, significant differences in metal content appeared at different levels in comparisons of non-oiled guillemots with individuals which were oiled both externally and internally; it is still unclear whether or not these differences can be linked to changes in the metabolism of the metals involved in response to oiling.

Compared to guillemots captured in the northern Norway area (Wenzel & Gabrielsen 1995) and to those shot in northwest Scotland (Stewart et al. 1994), the individuals collected on the Belgian coast were heavily contaminated with Cu, Zn and Hg (Table 4). Similarly high Cu and Zn levels for *Uria aalge* and for other species from the Belgian coast (*Larus ridibundus*, *Rissa tridactyla*, *Melanitta nigra*) were described by Antoine et al. (1992) and Bouqueneau et al. (1994). Moreover, a previous study on the speciation of metals in the cytosol of the liver and kidney of *U. aalge* stranded along the Belgian coast showed that the birds failed to maintain constant Cu, Zn and Cd levels in the high molecular weight soluble proteins in both organs; only a small part of the excess metal was found to be detoxified by metallothioneins (Bouqueneau et al. 1996).

Fig. 2. *Uria aalge*. Relative distribution of Cd concentration for age class I (juvenile and immature) and age class II (adult) in kidneys of guillemots found dead on the Belgian coast

Fig. 3. *Uria aalge*. Relative distribution of PCB concentrations for non-cachectic (Cach. -) and cachectic (Cach. +) birds in (a) liver and (b) kidney of guillemots found dead on the Belgian coast

CONCLUSIONS

Oiling is a major cause of death for wintering guillemots in the southern North Sea: 55% of guillemots found on the Belgian shores showed evidence of external or internal oiling. However, a large majority of birds (76%) were in a state of cachexia, probably due to shortage of food, bad weather conditions and natural disease. On the other hand, high levels of Cu, Zn, Hg and PCBs were clearly linked to cachexia, which can be considered as favourable to the development of lethal, acute, hemorrhagic gastro-enteropathy. None of these pollutants can be considered as the unique and direct cause of death, but might be an additional source of physiological stress, leading to debilitation and death. Further research is needed to determine the actual effects of stable pollutants on the health status of guillemots. The beaching of birds can be considered as a multifactorial response to numerous natural phenomena and a series of anthropogenic threats.

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Table 4. *Uria aalge*. Comparison of trace element concentrations ($\mu\text{g g}^{-1}$ DW), expressed as a range of mean values or as a mean $\pm$ standard deviation, in guillemots of different origins. Data from the Belgian coast given in bold. nd: non determined, <dl: below detection limit

Time	Place	Cu	Zn	Cd	Total Hg	Source
Liver						
n = 51 1970 to 1981	Belgian coast	nd	nd	nd	7.2 ± 2.4	Delbeke et al. (1984)
n = 83 Apr to Nov 1988	Northwest Scotland	12.9 – 16.1	58.4 – 69.7	1.4 – 2.5	0.9 – 3.7	Stewart et al. (1994)
n = 10 Summer 1992 and 1993	Hornoya, North Norway	20.0 ± 2.9	86.7 ± 14.9	3.1 ± 1.1	1.9 ± 0.4	Wenzel & Gabrielsen (1995)
n = 143 Winter 1990 to 1995	Belgian coast	52 ± 17	145 ± 39	2.4 ± 1.6	6.1 ± 3.4	This study
Kidney						
n = 9 1970 to 1981	Belgian coast	nd	nd	nd	4.4 ± 1.7	Delbeke et al. (1984)
n = 10 Summer 1992 and 1993	Hornoya, North Norway	14.4 ± 1.9	114 ± 13	24.1 ± 7.5	1.5 ± 0.2	Wenzel & Gabrielsen (1995)
n = 83 Apr to Nov 1988	Northwest Scotland	12.3 – 15.2	59.3 – 74.1	1.6 – 11.7	0.8 – 3.9	Stewart et al. (1994)
n = 143 Winter 1990 to 1995	Belgian coast	28 ± 12	169 ± 41	7.8 ± 6.6	4.6 ± 2.9	This study
Muscle						
n = 24 Apr to Nov 1988	Northwest Scotland	10.2 – 14.0	20.9 – 26.0	nd	0.5 – 1.8	Stewart et al. (1994)
n = 10 Summer 1992 and 1993	Hornoya, North Norway	19.2 ± 0.9	49.3 ± 3.3	0.2 ± 0.1	0.4 ± 0.1	Wenzel & Gabrielsen (1995)
n = 143 Winter 1990 to 1995	Belgian coast	18 ± 6	60 ± 14	<dl	2.1 ± 1.2	This study

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