Miscellaneous skin lesions of unknown aetiology in cetaceans from South America

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ABSTRACT

We report on miscellaneous skin diseases or syndromes of unknown aetiology including whitish, velvety lesions (WVL, often associated with unrelated skin injuries, scars and tooth rakes), large, rounded lesions (LRL, large to very large lesions with an orange or dark outline and a light inner colour) and vesicular skin disease (VSD, small to medium vesicles) in *Megaptera novaeangliae*, *Cephalorhynchus commersonii*, *C. eutropia*, *Pseudorca crassidens*, *Sotalia guianensis* and *Tursiops truncatus* from marine waters of Argentina, Brazil, Chile, Ecuador, Peru and the Antarctic. No biopsy samples have been available yet for histopathology. WVL are now commonly recorded opportunistically through photo-identification studies in several coastal species and populations from South America. Mortality rates, if any, associated with these skin diseases is unknown. Though sometimes extensive and ulcerated WVL do not seem life-threatening and, at least in some individuals, may eventually heal. A calf *C. eutropia* with LRL died some weeks after being first sighted. While unknown bacteria or fungi superinfecting miscellaneous skin traumata and poxvirus tattoos are thought to cause WVL and LRL, vesiviruses are suspected as the aetiological agents of VSD. Importantly, all lesions were primarily seen in coastal cetaceans living in biologically or chemically contaminated waters. These various skin conditions may be indicative of a deteriorating coastal water environment and should be systematically monitored. Collection of biopsies or fresh samples for histopathology and microbiological analysis is urgently needed.

INTRODUCTION

We recently reported on skin diseases of various aetiologies in small cetaceans from South America (Van Bressem *et al.*, 2007). Poxviruses and *Lacazia loboi* cause tattoo skin disease and lobomycosis in cetaceans, respectively. Lobomycosis, lobomycosis-like (LLD) disease, large rounded lesions (LRL, large to very large lesions with an orange or dark outline and a lightly coloured inner core) as well as whitish, velvety lesions (WVL, often associated with unrelated skin injuries, scars and tooth rakes) and vesicular skin disease (VSD, small to medium vesicles) may constitute severe skin conditions in dolphins and whales (Reif *et al.*, 2006; Van Bressem *et al.*, 2007). There is important circumstantial evidence that biological and chemical pollution are linked to the emergence and severity of many skin diseases in cetaceans (Wilson *et al.*, 1999; Reif *et al.*, 2006; Van Bressem *et al.*, 2003; 2007; 2008a,b). Here we report on the clinical and epidemiological characteristics of WVL, LRL and VSV in cetaceans from South America as well as on the possible impact of cutaneous diseases on the survival of populations and the causative or synergistic roles of harmful environmental factors.

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MATERIAL AND METHODS

Free-ranging cetaceans

The presence and evolution of WVL and LRL was studied in a minimum of 926 free-ranging Delphinidae belonging to five species (Table) mostly from photo-identification images taken during surveys from small boats or from shore in the period 1991-2008 (Reyes *et al.*, 2002; Viddi *et al.*, 2005, Flach, 2006). Dolphins were individually identified from natural marks (Würsig and Jefferson, 1990). The maturity class (calf, juvenile, adult) was estimated from the relative body size, behavioural clues (Wells *et al.*, 1980; Shane, 1990; Santos and Rosso, 2007, 2008) or from the period of residence. Dedicated surveys to assess the presence of skin conditions and traumata were conducted in common bottlenose dolphins (*Tursiops truncatus*) in Paracas Bay (Peru), Guiana dolphins (*Sotalia guianensis*) in Sepetiba Bay and Lagamar estuary, Peale's dolphins (*Lagenorhynchus australis*), Chilean dolphins (*Cephalorhynchus eutropia*) and *T. truncatus* from Chile's southern fjords and in Commerson's dolphins (*Cephalorhynchus commersonii*) from the Northern Patagonian Sea, Argentina, allowing a fairly precise estimation of prevalence (Reyes *et al.*, 2002; Flach, 2006; Viddi *et al.*, 2005; Van Bressem *et al.*, 2007; 2008a; Klaich *et al.*, 2008). Photographs of inshore *T. truncatus* were taken opportunistically from both shore and boats in Salinas, Ecuador in 2005-2007. Besides, we also included a humpback whale (*Megaptera novaeangliae*) photographed in Admiralty Bay, King George Island, Antarctic, in 1994.

Recently dead specimens

Carcases of 502 small cetaceans from Peru, Ecuador and Brazil were examined, predominantly under field conditions (*e.g.* fish markets and beaches), for cutaneous diseases in 1992-1994 and 2005-2007 (Table). Sexual maturity was determined, in females, from the presence of at least one corpus luteum or corpus albicans in one of the ovaries, or evidence of lactation or pregnancy; in males, if seminal fluid was detected macroscopically in at least one freshly cut epididymis. When sexual maturity status could not be determined directly, it was inferred based on an approximation for the mean standard body length (SL) at sexual maturation for these populations (Van Waerebeek *et al.*, 1990; Van Waerebeek, 1992; Reyes and Van Waerebeek, 1995; Felix *et al.*, 1992; Van Bressem *et al.*, 2006).

RESULTS

Whitish, velvety lesions (WVL)

General clinical features

The whitish, velvety, sometimes ulcerated lesions were characteristic of the acute stage of the disease. During the healing period the lesions cicatrised, lost their velvety aspect and turned progressively light and dark gray (regressing form). Normal skin pigmentation eventually returned though in some animals light blotches remained. When present in a population their prevalence varied between 0.7% (Chubut River estuary, Argentina) and 22.2% (Salinas, Ecuador) (Table).

Case study 1: Southern coast of Peru

Prevalence of the disease was 7.1% in 70 coastal T. truncatus from the Paracas Bay in 2004-2007. Affected dolphins included subadult and adult specimens. In two photo-identified T. truncatus (PBD-15 and -29), dorsal fin skin lesions were ulcerated when first observed, exposing the underlying connective tissues (Fig. 1a). They subsequently healed over a period of 2-6 months in 2004-2005 (Fig. 1b). Recurrence occurred in PBD-15 in July 2007 though to a lesser grade (Fig. 1c). The lesions did not ulcerate in PBD-55 and faded over the years. A fourth dolphin (PBD-60), possibly transient in the Bay had extensive lesions on the back and was not seen after 2006. Notably, no skin lesions were observed in a group of 15 coastal T. truncatus resident in Supay-La Mina within the Reserve, a cleaner area, 6km south of the Paracas Bay, away from fishmeal factories and contaminated freshwater run-off. The WVL were associated with tooth rakes and other minor skin wounds in at least three of the five affected dolphins. Though beaches were surveyed in the area in 2004-2008, no coastal T. truncatus were seen stranded. Besides, there were no observations or reports of die-offs of this species along the central and southern coast of Peru during that period. Altogether these data suggest that although what appears to be a chronic disease and likely affects the fitness of the animals to a varying degree WVL disease probably does not lead to death, and dolphins may eventually clear it. Recurrence is apparently associated with contamination of the area by run-off waters from the fishmeal factories (Echegaray and Reves, pers. observations). The lesions were not seen in 15 coastal and offshore dolphins caught in fisheries in 1993-1994 (Table).

Case study 2: Sepetiba Bay, Brazil.

Prevalence of WVL (including the active and regressive forms) was seen in 4.2% of 168 adult *S. guianensis* from Sepetiba Bay in 2005-2008 and in 4.4% of 45 dead specimens in 2005-2007. Five dolphins had active

infection: SEP-24, -33 and -40 were free-ranging while the two others, both males (SEP-3 and -17) had been by-caught in 2006-2007. In SEP-33 the sores were associated with a deep scar in front of the dorsal fin, possibly inflicted by fishing gear (Flach *et al.*, 2007), and with tooth rakes on the animal's left side (Fig. 2a). First observed in July 2005, the WVL lesions faded away over a six-month period (Fig. 2b,c). In SEP-40, some lesions were associated with scars, and though quite impressive resolved in four and a half months leaving only grayish blemishes (Fig. 3a,b). Remains of WVL were seen on the dorsal fin and back of four other dolphins.

Case study 3: Lagamar Estuary, Brazil.

Prevalence of WVL disease was 1% among 103 *S. guianensis* photo-identified from the Paraná estuary in 2006-2008. Adult dolphin 'S' showed healing WVL (Fig. 4a), unassociated with skin traumas, in March 2007. The marks had disappeared early August 2007 (Fig. 4b). None of the 200 dolphins inhabiting the cleaner Cananéia estuary had WVL but this difference was not statistically significant with Lagamar Estuary ($\chi^2 = 2.2$, df =1, P = 0.16).

Case study 4: Chubut River estuary, Argentina.

A *C. commersonii* had a medium-size ulcerated WVL on its dorsal fin. The lesion was not associated with a scar but may have developed on a tattoo. The dolphin presented six other skin blemishes that may have been tattoos on the dosal fin (Fig. 5). Prevalence of this condition was 0.7%.

Case study 5: Chanduy, Ecuador.

A false killer whale (*Pseudorca crassidens*) of a group of about 60 individuals stranded in November 1992¹ had a large, whitish, velvety mark on the anterior side of the tail (Fig. 6). A cursory laboratory analysis pointed to a bacterial infection. Prevalence of this condition in the 28 examined specimens is 3.6%.

Case study 6: Salinas, Ecuador.

A least two of the nine (22.2%) *T. truncatus* photo-identified in 2005-2007 had lesions on the tip of their dorsal fin that evoked WVL. Both were apparently adults. Smaller individuals swimming close to them were not affected.

Large, rounded cutaneous lesions (LRL)

LRL were observed in a *C. commersonii* from Puerto Deseado, Argentina, in October 2001 (Fig. 7a), a *C. eutropia* calf from Reñihue fjord, northern Patagonia, Chile, in January 2003 (Fig. 7b) and a humpback whale from Admiralty Bay, Antarctic, in 1994. Their irregular rounded shape evoked superinfected tattoo lesions as seen in a calf *T. truncatus* from the Sado Estuary, Portugal (Van Bressem *et al.*, 2003) but their aetiology is unknown. Poxvirus lesions may have facilitated the entry of super-infecting micro-organisms as described in an Atlantic white-sided dolphin (*Lagenorhynchus acutus*) and a pygmy sperm whale (*Kogia breviceps*) stranded along the northeast coast of the USA in 1991 (Frasca *et al.*, 1996). The affected area was extensive in the two dolphins, and the calf *C. eutropia* apparently died six weeks after the sores were first noted. The *C. commersonii* from Argentine was not seen again. No other cases were reported to us.

Vesicular skin disease (VSD)

Generalized vesicular lesions possibly caused by vesiviruses of the Family *Caliciviridae* were seen in a transient adult female *T. truncatus* from Northern Patagonia, Chile, photo-identified in January 2004 (Table; Fig. 8a). No other free-ranging and by-caught small cetaceans displayed such lesions (Table).

Some vesicles were also observed on the body of 1 of 83 *S. guianensis* (1.2%) specimens incidentally captured off the coast of Amapá state, northern Brazil, in November 2007 (Fig. 8b). The dolphin also had numerous black, rounded lesions over its entire body similar to those described as caused by a herpes-like virus in Peruvian dusky dolphins (*Lagenorhynchus obscurus*) (Van Bressem *et al.*, 1994). This population is genetically isolated from the *S. guianensis* from southern Brazil (Sholl and Siciliano, unpublished data).

DISCUSSION

WVL, LRL and VSD were mostly observed in coastal dolphins living in biologically and chemically contaminated waters (Table). The affected *T. truncatus* population in Peru ranged from Paracas Bay to Tambo de Mora; both localities that are heavily contaminated by organic material (phosphorus and organic nitrogen) released by fish-meal factories and surrounding towns, resulting in severe water eutrophication (PNUMA/CONAM, 2006). The factories also release caustic soda used to clean the machines (CPPS, 1998). In addition, ballast water from cargo ships entering the Paracas Bay to dock either at the Camisea gas platform or

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¹ Only 28 specimens could be examined in details.

the San Martín harbour may introduce alien micro-organisms that could cause significant damage (Ruiz *et al.*, 2000; PNUMA/CONAM, 2006; Drake *et al.*, 2007). The development of more tourist facilities along the shores of the Paracas Bay, and the ongoing expansion of the Camisea LNG gas plant (with the subsequent increase of ship traffic) represent new challenges for the inshore bottlenose dolphin communities in the departamento of Ica, Peru.

Chile's southern fjords are home to large numbers of salmon fish farms that continue to expand, heavily use prophylactic antibiotics that select antibiotic resistance in pathogenic bacteria, and release biological and chemical contaminants directly into the ocean (Moore and Wieting, 1999; Kemper *et al.*, 2003; Cabello, 2004; 2006). A large port, small villages with up to 2,000 inhabitants and an illegal shrimp farm that operated for a short time in 2006-2007 (Santos, pers. observations) likely biologically and chemically contaminate the Parana Estuary. Sepetiba Bay is home to two large ports and is characterized by chemical and organic pollution as well as water eutrophication (Copeland *et al.*, 2003; Molisani *et al.*, 2004). Puerto Deseado, Argentina, presents a medium level of contamination by hydrocarbons (Commendatore *et al.*, 2000). In Ecuador, Salinas is a major coastal tourist resort characterized by high levels of microbiological and chemical pollution (DIGEIM, 2007), heavy maritime traffic and an important port harbouring artisanal fishing boats, trawlers and purse-seine vessels as well as tankers transporting petroleum to a refinery in the nearby city of La Libertad.

Skin traumata and poxvirus infection, common conditions in cetaceans, may represent routes of entry for bacteria and fungi that are thought to cause WVL and LRL. *Vibrio* spp. and Aeromonadaceae have been isolated from miscellaneous skin diseases in cetaceans and could play a role in the aetiology of WVL and LRL (Pereira *et al.*, 2007, 2008). These bacteria thrive in the aquatic environment and some including *V. cholerae* are frequently transported by ships to estuaries with commercial ports (Ruiz *et al.*, 2000). Chemical pollution may favour super-infection by lowering the immune response of dolphins (see e.g. Aguilar and Borrell, 1994; Ross, 2002; Jepson *et al.*, 2005; Hall *et al.*, 2006a; Reif *et al.*, 2006). Though the WVL were extensive and in some cases invasive, most of the dolphins recovered over a six-month period. Recurrence of the disease occurred in at least one dolphin. Though likely affecting overall fitness to some degree, especially in severe cases, WVL seems to be a chronic self-limiting cutaneous disease for which there are no indications that it may cause mortality. To the contrary, LRL seem to be potentially lethal in calves and, thus might have an impact on populations. The immune system of calves is still incompletely developed making them more vulnerable to 'normal' viral infection as tattoo skin disease and super-infections. Besides, surviving first-born calves that have received a high PCB load through lactation (Aguilar *et al.*, 1999; Hall *et al.*, 2006b) may have a deficient immune function and be more at risk of developing serious infectious diseases.

We conclude that the various skin conditions described here may be indicative of a deteriorating coastal water environment and should be closely monitored. Collection of biopsies or fresh samples for histopathology and microbiological analysis is urgently needed.

ACKNOWLEDGEMENTS

We thank Dr E. Secchi and Martin Frixione for kindly providing pictures and data on a humpback whale and a *C. commersonii* from Puerto Deseado, repectively The work of M-F Van Bressem was funded by WDCS and CSI. ACOREMA received support from Gesellschaft zur Rettung der Delphine (GRD -Germany) and CSI. Dolphin research in the Paracas National Reserve was carried out under permits issued by the Instituto Nacional de Recursos Naturales (INRENA). The work of L. Flach was supported by Companhia Vale do Rio Doce. CEPEC research has benefitted from sustained support by IFAW. Van Waerebeek attends the IWC/60 meeting as head of scientific delegation for the Federal Public Service, Public health, Food Chain security and Environment, Brussels, Belgium. S. Siciliano is supported by fellowships provided by FIOCRUZ/FAPERJ and PROJETO PIATAM OCEANO – CENPES/PETROBRAS, UFF, UFPA and MPEG.

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FIGURES



Figure 1a. Active, ulcerated, whitish, velvety lesion (WVL) in *Tursiops truncatus* PBD015 from Paracas Bay, Peru, on 10 November 2004.



Figure 1b. Healed WVL in *Tursiops truncatus* PBD015, from Paracas Bay, Peru, on 18 January 2005.



Figure 1c. Recurring WVL in Tursiops truncatus PBD015, from Paracas Bay, Peru, in July 2007.



Figure 2a. Active WVL in Sotalia guianensis SEP-33, from Sepetiba Bay, Brazil, on First July 2005.



Figure 2b. Fading WVL in Sotalia guianensis SEP-33 from Sepetiba Bay, Brazil, on 29 December 2005.



Figure 2c. Healed WVL in Sotalia guianensis SEP-33 from Sepetiba Bay, Brazil, on 4 January 2006.



Figure 3a. Active WVL in Sotalia guianensis, Sepetiba Bay, Brazil, on 28 October 2007.



Figure 3b. Faded WVL in Sotalia guianensis from Sepetiba Bay, Brazil, on 4 March 2008.



Figure 4a. Fading WVL in Sotalia guianensis 'S' from the Parana Estuary, Brazil, on 27 March 2007.



Figure 4b. The WVL have disappeared in Sotalia guianensis 'S' from the Parana estuary, Brazil, on 2 August 2007.



Figure 5. Active, ulcerated, WVL and possible regressing tattoos on the dorsal fin of a *Cephalorhynchus commersonii* from the Chubut River, Argentina in October 2007.



Figure 6. WVL on the tail of a *Pseudorca crasssidens* stranded in Chanduy, Ecuador in November 1992.



Figure 7a. Large rounded lesions (LRL) in a *Cephalorhynchus commersonii* from Puerto Deseado, Argentina in October 2001.



Figure 7b. LRL in a Cephalorhynchus eutropia calf from northern Patagonia, Chile, in January 2003.



Figure 8a. Vesicular disease in an adult, female Tursiops truncatus from Palena, Chile.



Figure 8b. Vesicles on the body of a Sotalia guianensis captured off the coast of Amapá state, Brazil in November 2008.

Country &	Habitat	Region	Sampling	Specimens	Skin diseases			
species			period			Velvety lesions	Rounded lesions	Vesicular lesions
					N	Prev	Prev	Prev
Argentina								
Cephalorhynchus commersonii	Inshore/estuarine	Chubut river estuary	2006-2008	Free-ranging	147	0,7%	0%	0%
Cephalorhynchus commersonii	Inshore/estuarine	Puerto Deseado	2001	Free-ranging	1	-	-	-
Brazil								
Sotalia guianensis	Inshore/estuarine	Sepetiba Bay	2005-2008	Free-ranging	168	4.2%	0%	0%
Sotalia guianensis	Inshore/estuarine	Sepetiba Bay	2005-2007	By-caught	45	4.4%		
Sotalia guianensis	Inshore/estuarine	Parana estuary	2006-2008	Free-ranging	103	1%	0%	0%
Sotalia guianensis	Inshore/estuarine	Cananeia estuary	2006-2008	Free-ranging	200	0%	0%	0%
Sotalia guianensis	Inshore/estuarine	Amapá state	2007	By-caught	87	-	-	1.2%
Tursiops truncatus	Inshore/estuarine	Tramandaí Estuary	1991-2007	Free-ranging	10	0%	0%	0%
Tursiops truncatus	Inshore/estuarine	Mampituba Estuary	2003-2004	Free-ranging	3	0%	0%	0%
Ecuador								
Pseudorca crassidens	Offshore/pelagic	Santa Elena Peninsula	1992	Stranded	28	3.6%	0%	0%
Tursiops truncatus	Inshore	Santa Elena Peninsula		Free-ranging	9	22.2%	0%	0%
Chile								
Cephalorhynchus eutropia	inshore	Pumalin	2002-2004	Free-ranging	33	0%	3%	0%
Lagenorhynchus australis	Inshore/estuarine	Pumalin	2002-2004	Free-ranging	41	0%	0%	0%
Tursiops truncatus	unknown	Palena	2002-2004	Free-ranging	2	0%	0%	50%
Cephalorhynchus eutropia	inshore	Guaitecas	2007-2008	Free-ranging	42	0%	0%	0%
Lagenorhynchus australis	Inshore/estuarine	Guaitecas	2007-2008	Free-ranging	59	0%	0%	0%
Tursiops truncatus	unknown	Guaitecas	2008	Free-ranging	23	0%	0%	0%
D.								
Peru	0.004			-		0.04	001	
Delphinus capensis	Offshore/neritic	Central coast	1993-1994	By-caught	54	0%	0%	0%
Lagenorhynchus obscurus	Offshore/neritic	Central coast	1993-1994	By-caught	196	0%	0%	0%
Phocoena spinipinnis	Inshore/neritic	Central coast	1993-1994	By-caught	77	0%	0%	0%
Tursiops truncatus	Offshore/pelagic	Central coast	1993-1994	By-caught	12	0%	0%	0%
Tursiops truncatus	Inshore/neritic	Central coast	1993-1994	By-caught	3	0%	0%	0%
Tursiops truncatus	Inshore/neritic	Paracas Bay	2004-2007	Free-ranging	70	7.1%	0%	0%
Tursiops truncatus	Inshore/neritic	Supay-La Mina	2004-2006	Free-ranging	15	0%	0%	0%

Table. Prevalence of skin diseases in odontocetes from South America. N = total number of specimens examined, Prev= prevalence.