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SHORT COMMUNICATIONS

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Rise and Fall over 26 Years of a Marine Epizootic in Hawaiian Green Sea Turtles

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ABSTRACT: Estimates of chronic disease prevalence are needed to improve our understanding of marine disease epizootiology, which is poorly known for marine megafauna such as marine turtles. An emerging worldwide threat to green sea turtles (*Chelonia mydas*) is fibropapillomatosis (FP), which is a pandemic tumor-forming disease associated with herpesviruses. We report on a 26-yr FP epidemic in the Hawaiian Archipelago and show that apparent disease prevalence in the world's main endemic hot spot increased rapidly following a late 1980s outbreak, peaked during the mid-1990s, and then declined steadily ever since. While this disease is a major cause of sea turtle stranding in Hawaiian waters and can be fatal, we also show that long-term tumor regression can occur even for turtles with advanced FP. The endemic Hawaiian green turtle stock was severely depleted by overexploitation prior to protection under the US Endangered Species Act in 1978. This stock has increased significantly ever since, despite exposure to a major chronic disease epidemic that is currently declining.

Key words: *Chelonia mydas*, fibropapillomatosis, green sea turtle, marine epizootic.

Chronic and acute diseases are a major concern for the health of human populations and are subject to extensive research (Anderson and May, 1991). While our knowledge of acute disease impact on free-ranging marine wildlife is increasing (Harkonen et al., 2006), this is not the case for chronic diseases; this is mainly due to a paucity of long-term information on disease prevalence or incidence (Harvell et al., 1999; Lloyd-Smith et al., 2005). The green sea turtle (*Chelonia mydas*) is one of the long-lived late-maturing vertebrates that comprise the charismatic marine megafauna (Chaloupka et al., 2008a). Many green turtle populations have been depleted by exploitation, leading to concern that the species might be globally endangered (Chaloupka et al., 2008a). An emerging worldwide threat to green turtles is fibropapillomatosis (Herbst, 1994), which is a pandemic disease associated with the presence of herpesviruses (Greenblatt et al., 2005).

Fibropapillomatosis (FP) is a neoplastic disease involving tumors in multiple cutaneous sites and connective tissue tumors in the viscera (Herbst, 1994; Fig. 1A). Fibropapillomatosis prevalence has apparently increased over the past 2–3 decades in green turtle populations in Australia, Indonesia, and the US (Herbst, 1994; Chaloupka and Balazs, 2005; Foley et al., 2005; Greenblatt et al., 2005). It is believed that FP might impair recovery of depleted populations (Herbst, 1994; Ene et al., 2005), especially the green turtle stock endemic to Hawaii (Balazs and Chaloupka, 2004; Chaloupka and Balazs, 2005). Despite a global distribution and high prevalence in some populations (Herbst, 1994), there has been no long-term assessment of FP for any marine turtle population (Chaloupka and Balazs, 2005). Long-term assessments of the prevalence of major chronic diseases like FP in marine vertebrates are critically needed to improve our understanding of marine disease epizootiology (Harvell et al., 1999).

We reviewed FP disease prevalence data for a green turtle population that has been monitored each year since 1982

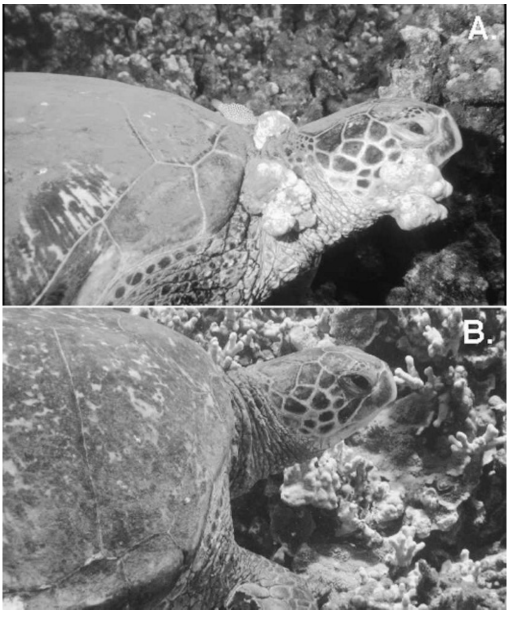

FIGURE 1. (A) Multitagged Hawaiian green turtle with advanced fibropapillomatosis in 1993. (B) Same turtle, tumor-free in 2004.

at Palaau (Molokai, Hawaii). This population has the highest recorded FP prevalence in the Hawaiian Archipelago (Balazs and Chaloupka, 2004; Chaloupka and Balazs, 2005), where the disease is endemic (Herbst, 1994). Annual disease monitoring was based on a capture-mark-recapture program, where each turtle was marked with metal flipper and/or passive integrated transponder tags (Balazs and Chaloupka, 2004; Chaloupka and Balazs, 2005). Each turtle was also evaluated at each annual sampling occasion for FP and assigned a severity score ranging from 0 (not affected) to 3 (severely affected) based on number, size, and location of tumors. This scoring system correlates well with a range of pathologic, hematologic, and physiologic parameters reflecting deteriorating immunocompetence with increasing tumor affliction (Work and Balazs, 1999; Work et al., 2001, 2003). We then estimated apparent FP prevalence as the proportion of green turtles at each annual sampling occasion with FP (Work and Balazs, 1999) based on 2,375 sampling records over a 26-yr sampling period (1982–2007).

A generalized smoothing spline regression (Gu, 2002) was fitted to the estimated apparent prevalence data to derive an epidemic curve. This robust nonparametric approach uses the data to determine the underlying linear or nonlinear trend without assuming any specific functional form or any particular error structure (for details, see Gu, 2002). No detection bias correction for these annual prevalence estimates was needed since there was no size class– or disease-specific difference in recapture probabilities for this sampled population. Briefly, we also used a multistate capture-mark-recapture model (Jennelle et al., 2007) to analyze 1,792 individually tagged turtles sampled over a 25-yr period. Size classes consisted of small and large immature green turtles at the Palaau study site. Each turtle was assigned at each encounter to a particular FP disease state (disease-free, diseased with FP scores > 0), where the transition probabilities among states, conditional on apparent survival, are analogous to probabilities of new infection and recovery from infection. One factor of relevance is that the estimated recapture probabilities were time-varying but independent of either disease state or size class, suggesting no sampling bias or behavioral differences for diseased turtles from this sampled population.

While FP is the most significant cause of stranding and mortality in green turtles in Hawaiian waters (Chaloupka et al., 2008b), not all diseased green turtles die, and our observations suggest that many green turtles with FP in Hawaiian waters can recover (Fig. 1B). Annual size class–specific disease recovery probabilities from our multistate capture-mark-recapture model were estimated at ~0.13–0.18 per annum. Meanwhile, the 26-yr epidemic curve for the Palaau (Molokai, Hawaii) green turtle population shows that apparent prevalence increased rapidly following the late 1980s outbreak, peaked during the

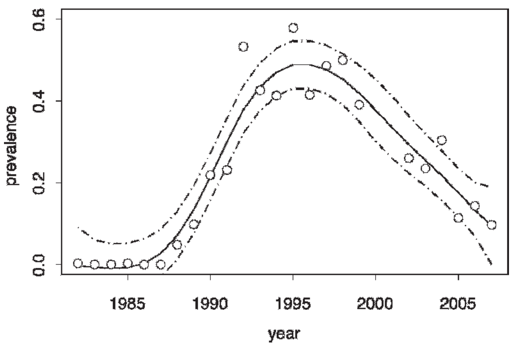

FIGURE 2. Epidemic curve with three phases: 1) rapid increasing phase (1988–1991), 2) peak phase (1992–1998), 3) slow decline phase from 1999 onward. Solid curve=smoothing spline fit, dashed curves=95% Bayesian confidence intervals, dots =apparent prevalence estimates.

mid-1990s, and then has declined steadily (Fig. 2). This curve reflects a chronic disease that persists for decades with the current prevalence in 2007 still around 9.4% (Fig. 2). The infection rate function derived from the multistate capture-mark-recapture model reflects the epidemic curve estimated for this population based on the prevalence data (Fig. 2).

Fibropapillomatosis is a major cause of stranding in Hawaiian green turtles (Chaloupka et al., 2008b) and is associated with an alphaherpesvirus, but the role of this virus in disease causation remains unknown (Quackenbush et al., 2001; Lackovich et al., 1999; Greenblatt et al., 2005). Interestingly, FP in Hawaiian green turtles was known long before the Palaau outbreak but was rare (Herbst, 1994). The endemic Hawaiian green turtle population is a genetically isolated metapopulation (Dutton et al., 2008). It was subject to extensive exploitation prior to complete protection in 1978 under the US Endangered Species Act, but it has since increased significantly (Balazs and Chaloupka, 2004; Chaloupka and Balazs, 2007). This ongoing stock recovery (Chaloupka and Balazs, 2007) has occurred in the presence of an epidemic disease (Chaloupka and Balazs, 2005), which has declined in recent years for the main Hawaiian FP enzootic focus (Fig. 2). Fibropapillomatosis severity has also declined at Palaau (Chaloupka and Balazs, 2005), and there is no evidence that FP significantly affects somatic growth or behavior (Chaloupka and Balazs, 2005) or diet (Seaborn et al., 2005) of Hawaiian green turtles nor the recovery of this once-severely depleted stock (Chaloupka and Balazs, 2007). However, this major chronic disease is apparently not evident in green turtles until they recruit from the open ocean to neritic or coastal developmental habitats (Ene et al., 2005), suggesting that the cause of the disease lies within the nearshore foraging habitats.

Because we do not know the cause of FP, the reasons why this disease was absent before the 1950s, peaked in the late 1990s, and has declined since are purely speculative. Two plausible explanations would include the development of herd immunity (Lloyd-Smith et al., 2005) to an infectious tumorigenic agent (if herpesvirus is contributing to disease) and/or removal of a tumor-inducing environmental insult in the nearshore foraging habitats around the island of Molokai (Herbst and Klein, 1995). Fibropapillomatosis has a wide distribution throughout the main Hawaiian Islands, except for the western coast of the island of Hawaii, where the disease is rare or absent (Work et al., 2004) in spite of apparently susceptible animals being present in those areas. This strongly suggests that an environmental cofactor is involved, but identification of the role of such cofactors would require tracking the virus in marine turtle populations. Unfortunately, robust serologic tests to assess exposure to the FP-associated herpesvirus remain elusive. While molecular virology has been helpful to demonstrate an association between FP and herpesvirus (Quackenbush et al., 1998; Lackovich et al., 1999), significant progress on understanding the role of this virus in the causation of FP will not be made until the virus can be grown and manipulated in an in vitro setting.

In contrast to Hawaii, where prevalence of the disease is declining, prevalence of FP in Florida appears to be more stable (Foley et al., 2005). The presence of FP in Florida has been known since the 1930s (Smith and Coates, 1938), so perhaps in that region, herd immunity is playing less of a role compared to environmental cofactors or nature of causative agents. There is evidence that in Florida, the FP-associated herpesvirus is different than that found in Hawaiian waters (Ene et al., 2005), and this may partly explain the differences between regions. Clearly, more research on the role of FP-associated turtle herpesvirus in actual causation of disease is needed if progress is to be made on disentangling the importance of environmental versus infectious factors in the epidemiology of FP. Unlike more acute viral diseases, the epidemic curve of FP in Hawaiian green turtles is more akin to that of chronic diseases such as cancer, which have durations of many years (Weiss, 1982). Meanwhile, the FP epidemic decline at Palaau (Molokai, Hawaii) is encouraging news for other marine turtle populations afflicted more recently with this chronic and often fatal disease.

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