

mitotic figures in circulating blood. Different types of Leucocytes were not founded in all the observed smears. Eosinophils, Basophils, Lynphocytes, Monocytes and Azupholis were higher in hatchlings coming from hatchery, on the other hand, Thrombocytes were more numerous in hatchlings incubated in boxes.

ESTIMATING AGE IN HAWAIIAN GREEN SEA TURTLES BY SKELETOCHRONOLOGY

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Green sea turtles, *Chelonia mydas*, in Hawaiian coastal waters range from 35 cm SCL juveniles to 106 cm SCL adults (Balazs *et al.*, 1987). An earlier skeletochronological analysis (Zug and Balazs, 1985) of a small sample suggested that in Hawaii, large juveniles to adults might be as old as 43-81 years. A larger sample salvaged since 1991 to examine the effects and onset of fibropapillomas also permits us to use skeletochronology to estimate the ages of these turtles. Skeletochronology is based on the reptilian aspect of skeletal growth by appositional deposition of new bone on old. This periosteal growth is cyclic, producing discrete bony layers in response to environmental and/or physiological cycles. The number of layers serves as an age index, and we assume that each layer represents one year of growth, *i.e.*, the number of layers equals the estimated age of the turtle in years.

For skeletochronological analysis, we use 0.6-0.8 mm cross-sections of the humerus from the middle of the shaft. Although analogous to dendrochronology in determining the number of years of growth, bone is a dynamic tissue. Resorption and remodeling in the core of the humerus destroys the earlier periosteal growth layers, and a protocol must be developed to estimate the number of lost layers. We use the correction-factor protocol (Parham and Zug, 1998) which estimates the number of lost layers by determining the average growth vector of increasing humerus diameter in the youngest/smallest turtle in the sample. This vector via a regression equation provides an estimate of the lost/resorbed layers. The total number of layers (also the estimated age in years) is the sum of the number of estimated lost layers and the number of remaining periosteal layers.

Age estimates range from 4.8 to 34.6 yr (Figure 1). The smallest turtle in the sample has the lowest age estimate and the two largest individuals the oldest estimates. Are the age estimates strictly the function of size and the mechanics of the age-estimation protocol? Because the correction-factor protocol and, for that matter, any other technique rely on the observed (remaining) growth layer and resorption-core diameters, a component of the estimates is likely always protocol-linked, because the diameter of the humerus enlarges

with increasing age and size. This link of age and size is a physiological and evolutionary component of the growth pattern of every animal. Why is there such a difference between these and the 1985 estimates? The 1985 estimates derive from an average growth width protocol that was noted then to provide overestimates in large turtles with large resorption cores in the humerus.

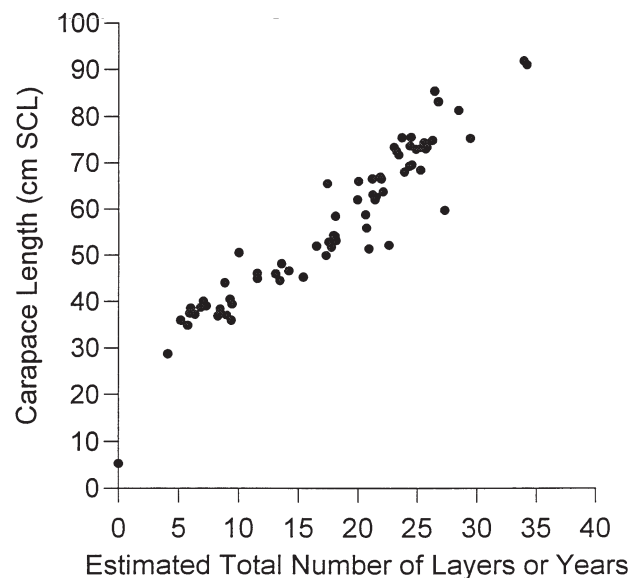


Figure 1. The association of age estimates and size in the Hawaiian green sea turtle sample. The 29 cm turtle was captured in the pelagic habitat in a commercial highseas driftnet fishery.

Linear regression of the age estimates yields a positive and strong association of age (X) and size (Y) [$Y = 22.8 + 1.91 X$, $r^2 = 0.90$]. Because of the strong linearity of these data, exponential growth models do not fit these data. Examining the data for local growth trends with a smoothing function, *e.g.*, LOWESS, suggests different growth rates at different life stages, as in an Australian population of *C. mydas* (Limpus and Chaloupka 1997).

Skeletochronology yields age estimates, not actual ages, of sea turtles, because resorption destroys the earlier

layers. Another limitation of the skeletochronological data is that age estimates are not equally extractable from the different age classes. Skeletochronology relies on a pattern of distinct layering within the bony elements. Such patterns are most evident in the smaller turtle, and their occurrence becomes less frequent in the larger individuals. Age can be estimated for 89% of the 30-69 cm SCL turtles, 72% for 70-79 cm, 18% for 80-89 cm, and 29% for >89 cm turtles. Another aspect of bone layering in Hawaiian *C. mydas* is the predominance of “annuli” to “lines of arrested growth.” This observation suggests continuous growth in most individuals in this population, although retaining a cycle of rapid versus slow growth, and thus producing layers and permitting skeletochronological analysis.

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