Veterinary Ophthalmology (2008) **11**, 2, 99–110

Ocular morphology of the Leatherback sea turtle (*Dermochelys coriacea*)

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This work was sponsored in part by an unrestricted sabbatical grant from Research to Prevent Blindness, NY, United States of America and by Prevent Blindness Foundation, Brisbane, Australia, through Viertels Vision. Further funding was received from the US National Marine Fisheries Service (Pacific Islands Fisheries Science Center) and the State of Hawaii's Fisheries Disaster Relief Program.

Abstract

Objective The Leatherback sea turtle is the largest extant reptile and the sole member of the family Dermochelyidae. Here, the eye of this critically endangered marine turtle was investigated to determine the anatomy, optics, and optical sensitivity.

Animals studied Three Leatherback sea turtles, *Dermochelys coriacea*.

Results The eye is small in proportion to body size of the adult compared to other vertebrates, with prominence of the retractor bulbi and pyramidalis muscles. The nictitans shows extensive folding of the bulbar conjunctiva as an apparent mechanism to increase the surface area for mucus secretion. The intraocular anatomy is consistent with an eye adapted to aquatic vision with minimal curvature of the cornea, a near-spherical lens, deep ciliary cleft and highly vascularized ciliary body. The optical sensitivity, a measure of the sensitivity to light of a given optical system, is higher than in other marine turtles studied but lower than those found in teleost fish that share a habitat with the Leatherback sea turtle.

Conclusions The Leatherback sea turtle shows ocular features that are characteristic of Chelonians with similarities to aquatic mammals. The calculated optical sensitivity suggests that compared to pelagic fishes, for instance, the Leatherback sea turtle eye is not particularly well adapted for vision in dim light even though this species is known to venture into deep, dark waters, and might feed at night.

Key Words: accommodation, anatomy, *Dermochelys*, eye, Leatherback sea turtle, optical sensitivity

INTRODUCTION

The Leatherback sea turtle (*Dermochelys coriacea*) is thought to represent an early divergence of the marine turtle (*Chelonioidea*) lineage[1] and is characterized by a number of fundamental differences in morphology, physiology, and behavior compared to other marine turtles.

With weights of up to 900 kg, Leatherback sea turtles are the largest of extant turtles.[2] They are the most pelagic of the marine turtles, undertake large cross-oceanic migrations,[3,4] and feed mainly on gelatinous prey such as jellyfish and salps. While juvenile animals are found in the tropics of the world's oceans, adults can expand their feeding range into temperate waters and are capable of thermoregulation.[5] The Leatherback sea turtle holds the record as the deepest diving marine turtle and is one of the deepest diving air-breathing vertebrates, with an estimated diving depth of greater than 1000 m.[6]

Leatherback sea turtles are now critically endangered worldwide. The Pacific population is on the verge of extinction with a 23–33% annual mortality rate of female Leatherbacks.[7] The global population decline is largely attributed to incidental capture by gill-net and longline fisheries,[7] with a likely worldwide catch by pelagic longlines of 50 000 Leatherbacks in the year 2000.[8] In this study we aim to investigate the morphology of the eye in the adult animal, as there is a long-standing need to understand how the Leatherback may use its vision. The clear-water, pelagic habitat suggests that vision is likely to be an important sense for these animals for finding food and mates and avoiding predation.

As all marine turtles, Leatherback sea turtles spend the majority of their lives in the ocean, only interrupted by short periods on land when mature females lay their eggs, and hatchling turtles emerge from these nests and immediately enter the water. These are activities for which sea turtles appear to use their visual sense,[9] hence it is likely that their eyes have adapted to vision in both air and water.

Refraction is strongly affected by moving from air to water, and there are conflicting reports on the accommodative

status of sea turtle eyes in both media.[10,11] A closer investigation of the accommodative structures of the Leatherback sea turtle eye is attempted here and compared to those structures in aquatic mammals.

Upon entering the aquatic phase of their lives, Leatherback sea turtles undertake regular dives varying between several minutes and over 1 h in length.[6,12] While this species has been observed feeding on the surface,[13] many of the dives are presumed to reach prey organisms at depths throughout the water column.[6,14,15] As downwelling light is reduced with increasing depth in the water, the question arises of how well this species is adapted to vision in dim light. This anatomical study also investigates the optics and anatomy of the retina to allow an estimate of light sensitivity.

MATERIALS AND METHODS

Specimens were collected from three adult Leatherback sea turtles found deceased off Prince Edward Island (turtle 1) and Nova Scotia (turtles 2 and 3). Both the eyes were dissected from each turtle and fixed in either 70% ethanol (turtles 1 and 2) or 10% formalin (turtle 3). The eyes were transported to the University of Queensland for investigation under approval of the convention on International Trade in Endangered Species (CITES). Due to the critically endangered status of the species and difficult access to samples, fresh Leatherback turtle eyes were not available for investigation.

Eyes were assigned for gross morphology, photoreceptor modeling, histopathology, or frozen for measurement of internal ocular dimensions, based on the assessment of eye quality. The optical axis for each eye was determined as each eye was identified as left or right, and by identifying the extraocular muscles using previous anatomical reports[16–18] and by location of the third eyelids. Radiography and special staining for scleral ossicle morphology were performed on two globes. Both globes were bleached in 3% hydrogen peroxide in 1% potassium hydroxide, followed by staining with alizarin red and alcian blue–acetic acid solution. Two eyes were opened in a parasagittal direction for collection of intraocular tissue samples. One of these eyes was demineralized with 10% EDTA prior to sectioning. Histopathology was performed on a series of 5 μm sections of nictitans, anterior globe, and posterior globe, and stained with H&E, PAS, or Gamori's trichrome. Two globes were frozen in dry ice followed by sectioning in an anterior–posterior direction, with one oriented vertically and the other horizontally through the center of the cornea, to obtain internal ocular dimensions.

The dimensions of the photoreceptors were assessed using floating preparations. Small areas of the retina were removed from five different regions (dorsal, ventral, medial, lateral, and central). The tissue was gently teased apart in a drop of 0.1 M phosphate buffer (pH 7.2) on a microscope slide using a hypodermic needle. The resulting dissociated photoreceptors were photographed using a spot RT color camera (Diagnostic Instruments Inc., Sterling Heights, MI) through a light microscope (Zeiss Axioscope, Carl Zeiss, Jena, Germany) and the width and length of the photoreceptor outer segments measured on printed copies of the photographs.

As a measure of sensitivity of the eye to light, the optical sensitivity of the Leatherback sea turtle eye was calculated.[19,20] In brief the optical sensitivity (S[μm^2 steradian (sr)]) considers the optical features of the eye (A aperture, the diameter of the circular pupil; d diameter of individual photoreceptors; f as a function of the focal length; and F is the fraction of incident light absorbed by each receptor) in the following expression:

$$S(\mu m^2\ sr) = (\pi/4)^2 \times A^2 \times (d/f)^2 \times F$$

The value for focal length used in this equation is that for the focal length in water. The focal length f is measured from the center of the lens to the point at which parallel rays of light are brought into focus[20] which was taken to be the retina in this case, making the assumption that the Leatherback sea turtle is emmetropic in water. F can be calculated for white light $F_{\text{white light}} = \kappa l/(2.3 + \kappa l)$,[21] approximating the turtle's light environment on the surface of the water; and for monochromatic light at the receptors' preferred wavelength, approximating the conditions encountered at depths where the downwelling light is nearly monochromatic $F_{max} = 1 - e^{-\kappa l}$.[19] The optical sensitivity calculated at white light conditions is approximately half that found at the preferred wavelength.[21] The value for the photoreceptor absorbance coefficient (κ) was estimated to be similar to bony fishes at 0.03[20,22] while l is the length of the photoreceptor outer segment. The lack of access to fresh tissue meant that measurements of optical dimensions and photoreceptors were undertaken in fixed tissue and shrinkage could not be estimated. It is possible that shrinkage affected the photoreceptors more than the cartilaginous globe, resulting in a conservative estimate of the optical sensitivity.

RESULTS

Gross globe

All six eyes were similar in shape and dimensions with a mild variation among the three turtles (Table 1). The globes were oval in shape from the anterior aspect (Fig. 1a,b) with the dorsoventral dimension longer than the mediolateral dimension. The anteroposterior dimension was less than the dorsoventral and mediolateral dimension, but slightly greater ventral to the cornea on two eyes that were completely cleaned of extraocular attachments. Corneal clarity and the degree of visualization of the anterior chamber were used as an indicator of intraocular quality and degree of decomposition prior to fixation. Quality was assessed from best to worse in the order turtle 2–turtle 1–turtle 3. No outer eyelids were present but all eyes had extraocular muscle attachments of variable amounts.

Nictitans

The nictitans was investigated in three eyes where it was still present in part or total after removal from the head. The nictitans

Table 1. Measurements of the general and ocular dimensions for the three Leatherback turtles in the study*

	Turtle 1		Turtle 2		Turtle 3	
	Mean	SD	Mean	SD	Mean	SD
Weight (kg)	276.0		336.8		359.1	
Sex	Male		Female		Male	
Curved carapace length (cm)	145.2		151.0		168.3	
Curved carapace width (cm)	98.0		100.8		114.9	
Dorsoventral globe (mm)	44.6	0.4	42.1	0.1	45.5	0.4
Mediolateral globe (mm)	40.0	0.6	38.1	0.5	44.2	1.5
Anterior–posterior globe (mm)	27.1	0.5	29.0	2.3	32.9	0.8
Dorsoventral corneal diameter (mm)	12.6	0.1	11.6	0.4	13.5	0.6
Mediolateral corneal diameter (mm)	11.7	0.1	9.8	0.6	12.5	0.7
Pupil diameter *A* (mm)†	–		4.3	0.2	–	
Lens anterior–posterior length (mm)	6.7	0.8	6.3	0.7	–	
Lens equatorial width (mm)	8.2	1.1	7.1	1.0	–	
Posterior cornea–anterior lens capsule (mm)	2.0		1.1			
Focal length (lens center-retina) *f* (mm)†	18.7		–		–	
Retinal thickness (μm)	–		241.0		–	
Cone OS length *l**†	24.4	4.7	–		–	
Cone OS width *d**†	5.3	1.0	–		–	
$S_{\text{white light}}$ (μm^2 sr)	0.23		–		–	
$S_{\lambda\text{max}}$ (μm^2 sr)	0.49		–		–	

*Measurements are given as mean and SD based on OU from each animal, apart from the dimensions of the cones (*) where $n = 53$ for the width and $n = 30$ for the length of the cone outer segment (OS).
†Measurements used to provide the data for calculating the optical sensitivity *S*.

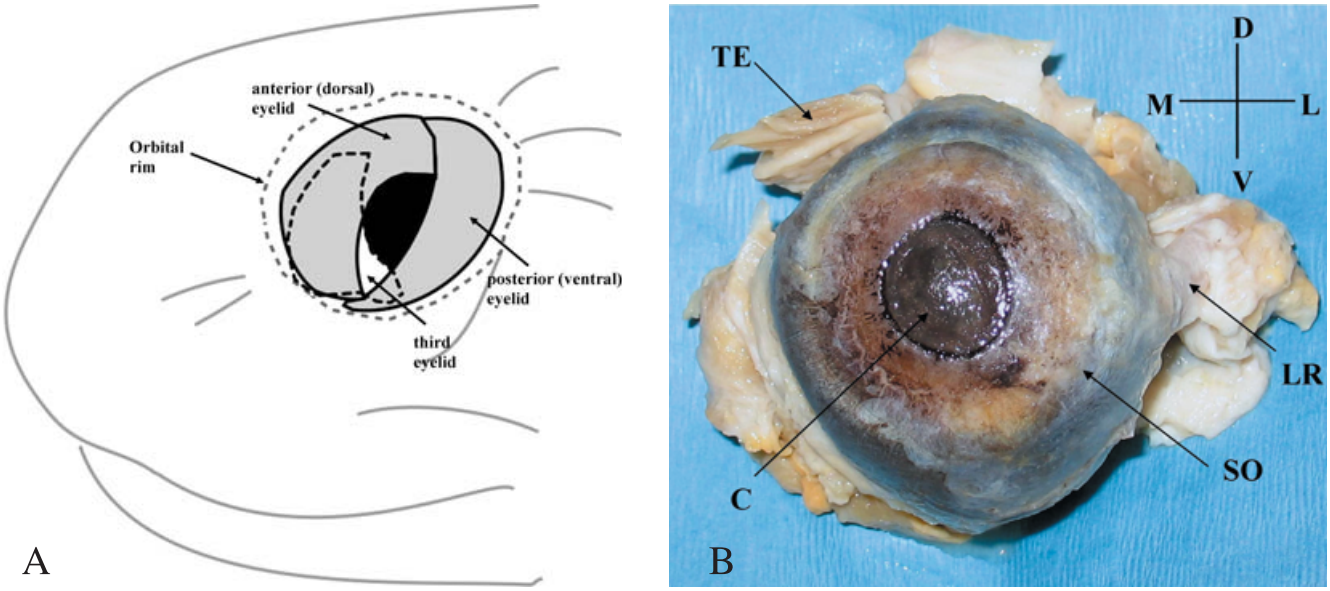

Figure 1. Leatherback eye (a) Diagram of left side of head to illustrate the position of the outer eyelids and third eyelids (modified from Wyneken J, The Anatomy of Sea Turtles, 2001). (b) Gross appearance of the left globe illustrating cornea (C), scleral ossicle rim (SO), remnant of third eyelid (TE), and lateral rectus (LR).

canted slightly ventromedially from a dorsoventral position and its bulbar conjunctiva was arranged as a series of deep pleats (Fig. 2a,b). There were over 25 pleats which traversed the length of the nictitans, and many shorter pleats that originated and inserted part way along a longer pleat, resulting in most pleats at the midsection. Each pleat had extensive folding of the mucosal surface, which was more evident at a microscopic level (Fig. 3a). PAS staining was positive for intracellular and surface glycoprotein consistent with mucus (Fig. 3b).

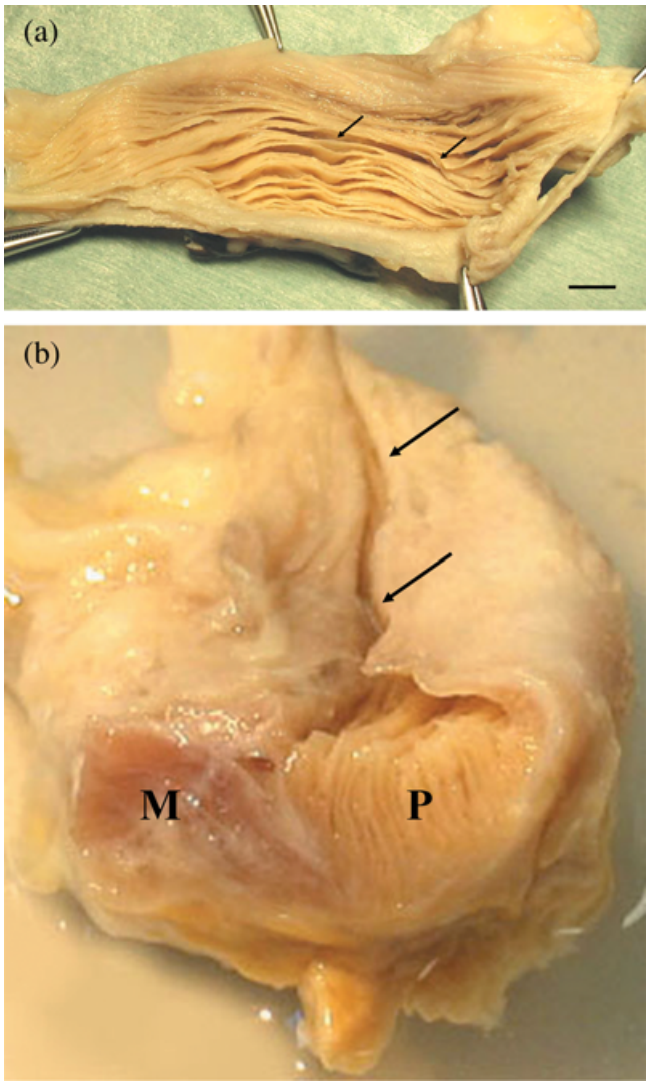

Figure 2. Dissected nictitans of OD. (a) Longitudinal view exposing conjunctival pleats of bulbar aspect (arrows). Bar = 5 mm.
(b) Cross-sectional view through mid-nictitans showing muscle at base of nictitans (M), orderly arrangement of conjunctival pleats (P) and leading edge of nictitans (arrows).

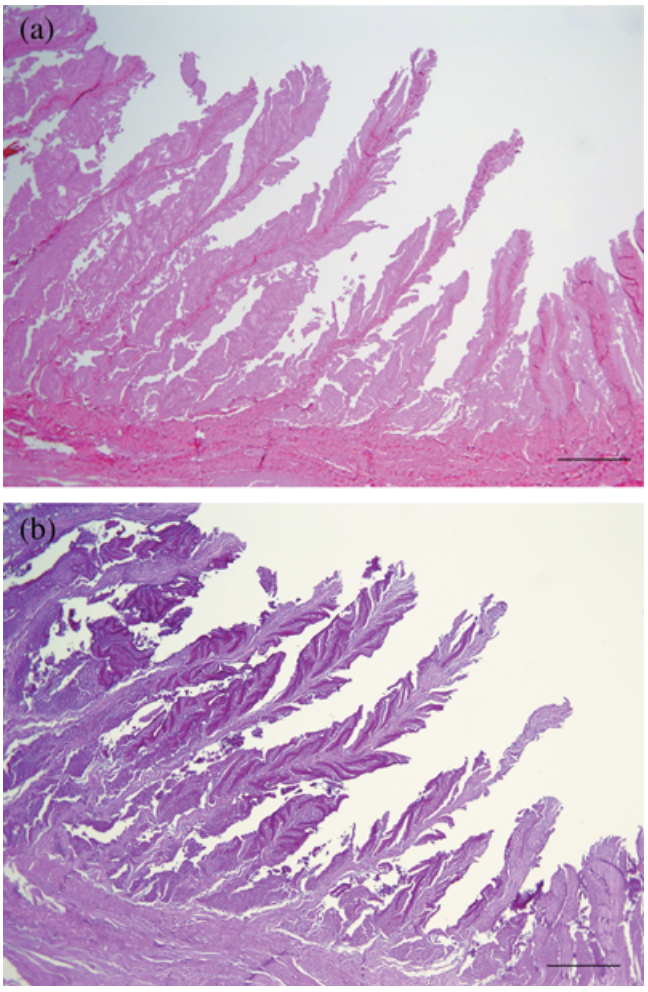

Figure 3. Histopathology of nictitans of OD. (a) Extensive folding of the mucosal surface exhibited on each pleat. H&E stain. (b) PAS staining illustrating intracellular and surface glycoprotein consistent with mucus. Bars = 500 μm.

Extraocular muscles

All globes had a dorsal, ventral, medial, and lateral rectus, dorsal and ventral oblique, with a prominent retractor bulbi and pyramidalis muscle. Muscle insertion sites for the rectus and oblique muscles were located between 8 and 14 mm posterior to the scleral ossicle rim; the lateral rectus insertion site being the most anterior and the ventral rectus the most posterior. All rectus and oblique muscles inserted into the collagenous framework of the sclera. Muscle tissue inserted along the length of the nictitans base, being thickest at the central region (Fig. 2a). This muscle merged with the aponeuroses of the dorsal, medial, and ventral rectus muscles as they traveled in a posterior direction. The retractor bulbi and pyramidalis muscles were the largest of all the extraocular muscles. The pyramidalis muscle was located on the dorsomedial aspect of the posterior globe and divided into two branches, with one muscle branch traveling to the ventrolateral anterior aspect and the other traveling to the dorsolateral anterior aspect. The optic nerve traveled between the muscle fibers of the retractor bulbi, with this muscle inserting on the dorsal, lateral, and ventral posterior globe.

Bulbar conjunctiva

The epithelium of the bulbar conjunctiva consisted of five to six layers of lightly pigmented, stratified squamous cells at the corneal limbus increasing to 15–16 cell layers towards the periphery of the scleral ossicles and rim of the cartilaginous cup (Fig. 4).

Sclera

The sclera consisted of a ring of overlapping ossified plates anteriorly and a cartilaginous cup posteriorly, embedded within a collagenous matrix. The scleral ossicles protruded anteriorly forming an angle of 35° to the cartilaginous cup. The individual

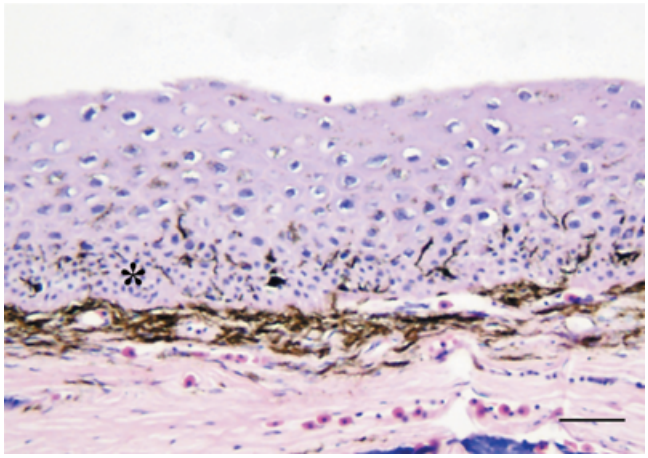

Figure 4. Bulbar conjunctiva external to scleral ossicles, consisting of a thick basal layer (*) with intermingling melanin pigmentation progressing into stratified squamous, lightly pigmented epithelium. H&E stain, bar = 50 μm.

ossicle plates were not clearly identifiable on radiography (Fig. 5a) or by further staining, but overlapping of bony plates was identified microscopically (Fig. 5b). The cartilaginous scleral cup measured between 12.0 and 12.4 mm at maximum thickness of three eyes. The posterior sclera was traversed by multiple vascular channels with a large collecting sinus at the ventral aspect.

Cornea

The cornea continued the curvature of the scleral ossicle plates and consisted of four layers: epithelium, stroma, Descemet's membrane, and endothelium (Fig. 6). The epithelium was absent over the majority of the cornea, due to cell loss prior to fixation. The stroma comprised 95% of the total corneal thickness. Descemet's membrane measured 4 μm in thickness, and stained PAS-positive. A single layered endothelium consisted of flattened cells, with the density at the most preserved region estimated at 2000 cells/mm^2 with 15–20 cell nuclei per ×40 microscopic field. Total corneal thickness as measured on histologic sections ranged from 650 μm at the limbus to 300 μm centrally.

Iris

A round pupil was visible through the cornea of OU of turtle 2 (Table 1). Histologically a large number of small vessels and melanocytes were present on the anterior surface of the iris (Fig. 7a). A large, thick-walled vessel was present at the periphery of the anterior iris stroma, consistent with a major arterial circle. A large number of small diameter vessels were present throughout the iris stroma, these vessels increased in size at the posterior iris base. A well-developed sphincter muscle extended 1.5 mm peripherally from the pupil margin, and thin dilator muscle fibers extended 2.9 mm from the inner third of the iris to the iris root, adjacent to the posterior iris epithelium (Fig. 7b). Tissue quality precluded further

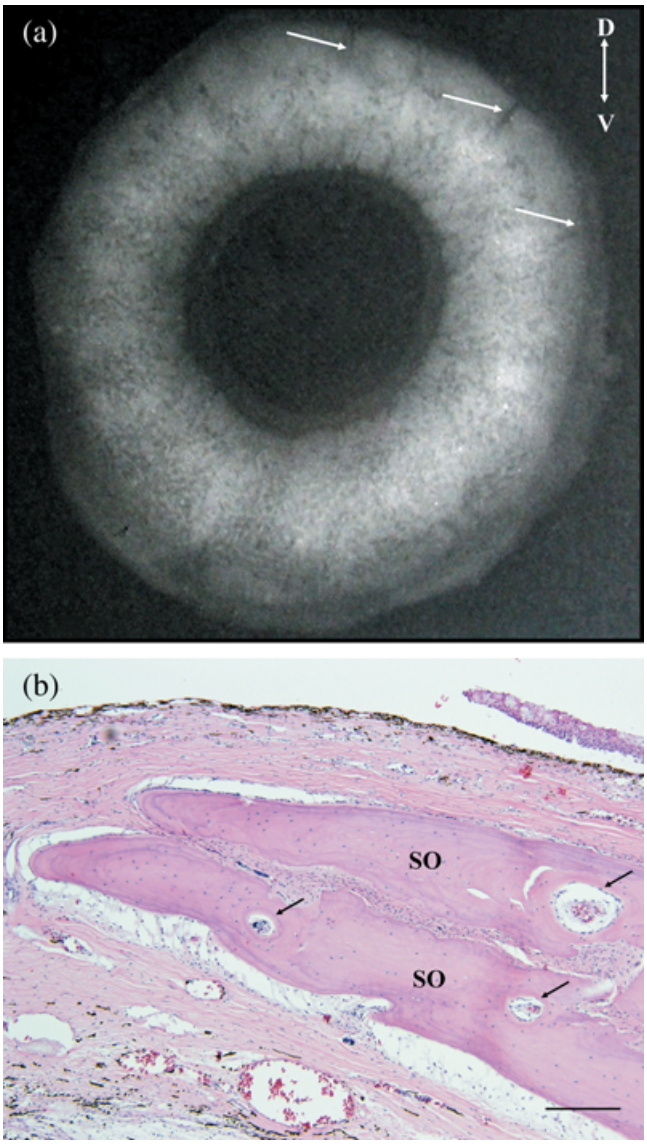

Figure 5. Sclera (a) Radiograph of scleral ossicles of OD showing almost complete ossification. Radiolucent lines are suggestive of the previous outline of individual plates (arrows). The ossicle ring is orientated with the dorsal aspect uppermost. (b) Complex overlapping and interdigitation of the scleral ossicle plates (SO). Vessels are evident within individual ossicles (arrows). H&E stain, bar = 200 μm.

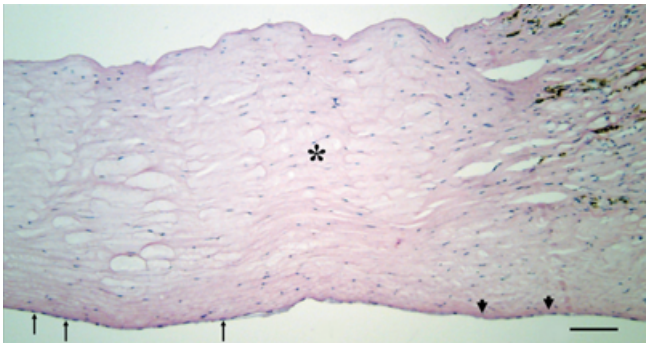

Figure 6. Peripheral cornea showing stroma (*), Descemet's membrane (arrowheads) and endothelium (arrows). Epithelium was lost prior to fixation. H&E stain, bar = 100 μm.

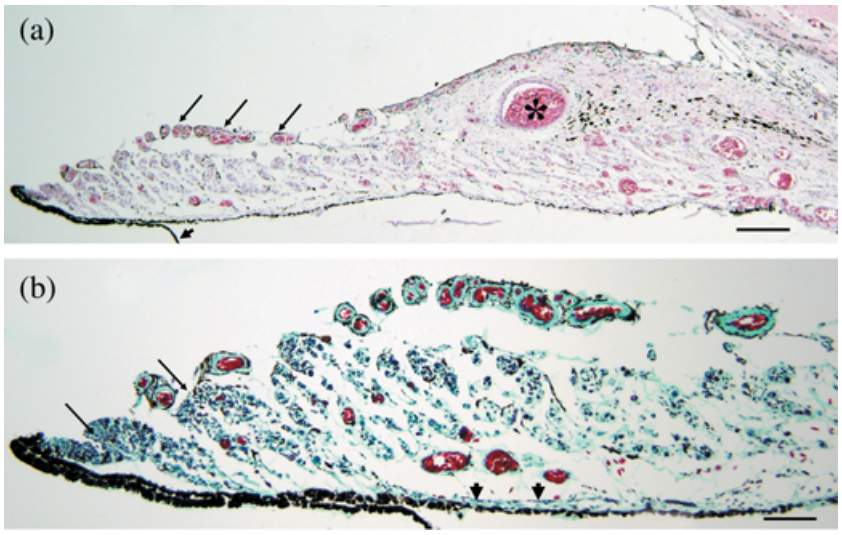

Figure 7. Iris (a) Large numbers of small diameter vessels (arrows) covered with a single layer of pigmented epithelium of the anterior iris. A large, thick-walled vessel at peripheral iris is consistent with a major arterial circle (*). Separation of the double-layered pigmented posterior iris epithelium has occurred as a fixation artefact (arrowhead). H&E stain, bar = 250 µm. (b) Gamori's trichrome stain illustrating iris sphincter muscle bundles adjacent to the pupil margin (arrows) and thin dilator muscle layer adjacent to the posterior iris epithelium (arrowheads). Bar = 100 µm.

determination of this muscle as smooth or striated type. A double-layered pigmented epithelium was present on the posterior iris surface. Melanin pigment was scattered within cells of the iris stroma.

Anterior chamber and iridocorneal angle

The anterior chamber depth measured 2 mm on frozen ($n = 1$) and 1.1 mm on histologic section ($n = 1$) (Table 1). The opening of the iridocorneal angle was traversed by fine, pigmented pectinate ligament strands (Fig. 8a). The ciliary cleft was long and narrow, contained a fine meshwork of pigmented uveal trabeculae with large spaces of Fontana, and extended 3.2 mm posterior to the primary pectinate ligament to the level of the mid-ciliary body. The cleft was lined by an inner and outer layer of connective tissue containing numerous vessels, with a large sinus-like vessel at the posterior aspect of the cleft evident in some sections (Fig. 8b). An angular aqueous plexus was present but less prominent than the ciliary body/suprachorioidal vessels, suggesting the uveoscleral route is significant for aqueous humor drainage.

Ciliary body

The ciliary body region was highly vascularized with large thin-walled, sinus-like vessels within the connective tissue stroma adjacent to the ciliary epithelium; which itself consisted of a single inner layer of nonpigmented epithelium overlying a single layer of pigmented epithelium (Fig. 9a). Ciliary muscle fibers were meridionally arranged, thin bundles with a maximum width of 120 µm, which extended from the posterior aspect of the ciliary cleft for 2.8 mm to the ora ciliaris retinae, just posterior to the rim of the scleral cartilage cup (Fig. 9b). Tissue quality precluded further determination of this muscle as smooth or striated type.

Lens

The lens was almost spherical measuring between 6 and 7 mm for anterior–posterior diameter and 6.2–7 mm in

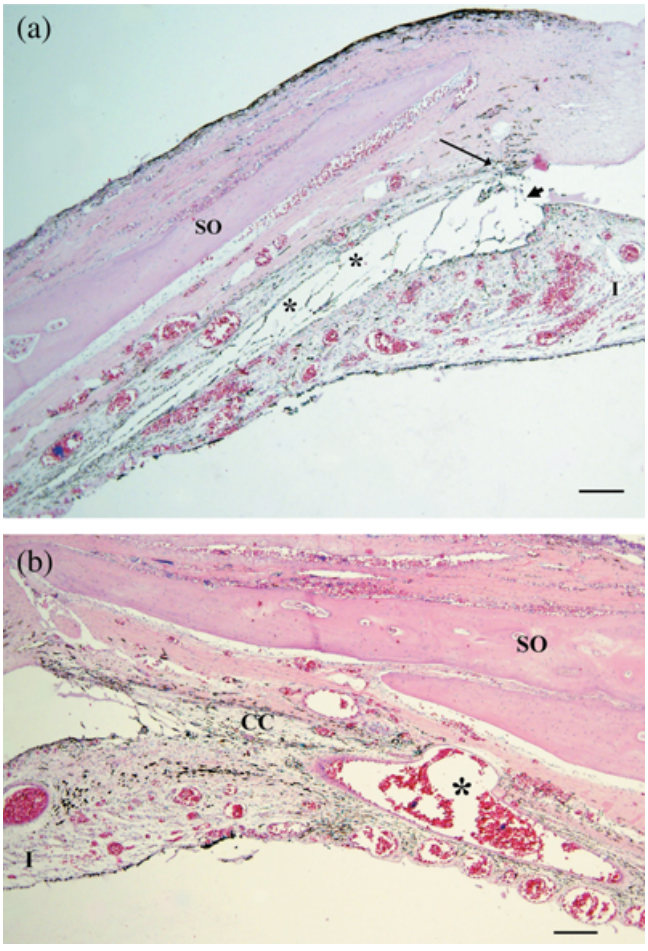

Figure 8. Iridocorneal angle illustrating (a) the primary pectinate ligament (arrowhead), large spaces of Fontana (*), opening to the angular aqueous plexus (arrow) and (b) a large sinus-like vessel (*) at the posterior extent of the ciliary cleft. I = iris, SO = scleral ossicle, CC = ciliary cleft. H&E stain, bar = 250 µm.

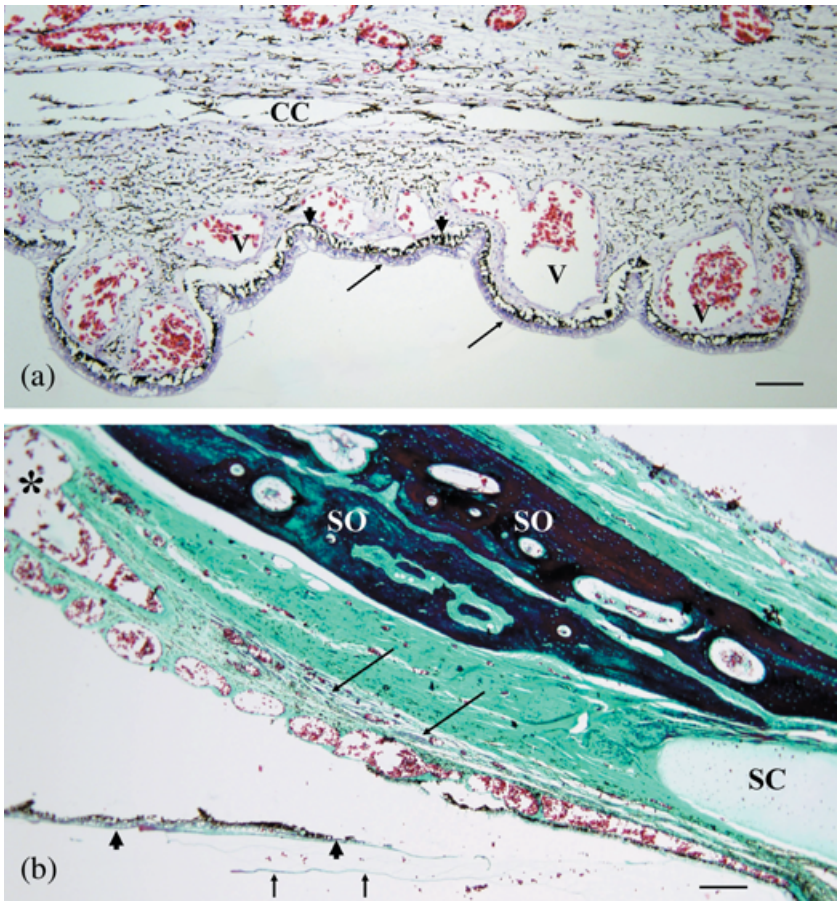

Figure 9. Ciliary body (a) Double layered epithelium, consisting of an inner nonpigmented (long arrows) and outer pigmented layer (arrowhead), overlying large thin-walled vessels (V) within the stroma. CC = ciliary cleft. H&E stain, bar = 100 µm. (b) Thin bundles of meridionally arranged ciliary muscle fibers (long arrows) and fine collagenous zonular fiber (short arrows) demonstrated with Gamori's trichrome stain. Separation of ciliary body epithelium due to fixation artefact (arrowheads). SO = scleral ossicle, SC = scleral cartilage. Bar = 250 µm.

equatorial diameter for three lenses taken from frozen ($n = 2$) (Fig. 10a) and histologic sections ($n = 1$) (Fig. 10b). A PAS-positive capsule measured 12 µm in thickness anteriorly, 5 µm at the equator and 3 µm posteriorly. A single layer of cuboidal epithelial cells was present underneath the anterior lens capsule, which became progressively elongated and columnar at the equator and site of insertion of the zonular fibers and may represent a vestigial remnant of an annular pad (Fig. 10c). A lens bow was evident at the equator.

Choroid

The choroid was extensively vascularized with large, thin-walled, sinus-like vessels interspersed with pigmented stroma (Fig. 11). There was a progressive reduction in vessel size from the outer to inner choroid, with an almost continuous layer of small vessels intimately associated with the basement membrane of the retinal pigment epithelium (RPE), consistent with a choriocapillaris. There was no identifiable tapetum.

Retina

The retina was avascular and layers conformed in principle to the standard vertebrate arrangement (Fig. 12) with a mean retinal thickness of 241 µm on histologic section (Table 1). The cells of the RPE were rectangular in cross-section at their nucleated base and arranged on a clearly defined basement membrane. The inner aspect of each RPE cell showed multiple, finely pigmented villous-like cytoplasmic extensions. Photoreceptor cells were orderly aligned with all nuclei at the same level within the retina vitread to a distinct outer limiting membrane. The floating preparation revealed the presence of rods and cones, as well as distinct cone types that varied in the length and width of the outer segments. The width and length of cone outer segments were measured in the floating preparation (Table 1) with dimensions varying slightly in the different areas measured, from outer segment width 4.8 µm (SD 0.8 µm) to 5.9 µm (SD 0.8 µm) and outer segment length 20.5 µm (SD 3.6 µm) to 27.0 µm (SD 3.2 µm).

Optic nerve

The optic nerve head was mildly elevated above the surface of the retina, with small vessels within the nerve head close to the vitreal surface (Fig. 13). Several vessels traveled within the substance of the optic nerve. The optic nerve head was situated just dorsal of the optical axis of the globe, with the optic nerve exiting through the sclera in a ventral direction at 45° to the retinal surface.

Optics and optical sensitivity

Internal dimensions of the right globes of turtles 1 and 2 showed a small lens and a long focal length (Table 1, Fig. 10a). The sectioned eye of turtle 2 revealed a fracture of the scleral ossicles altering the internal dimensions of the globe, hence

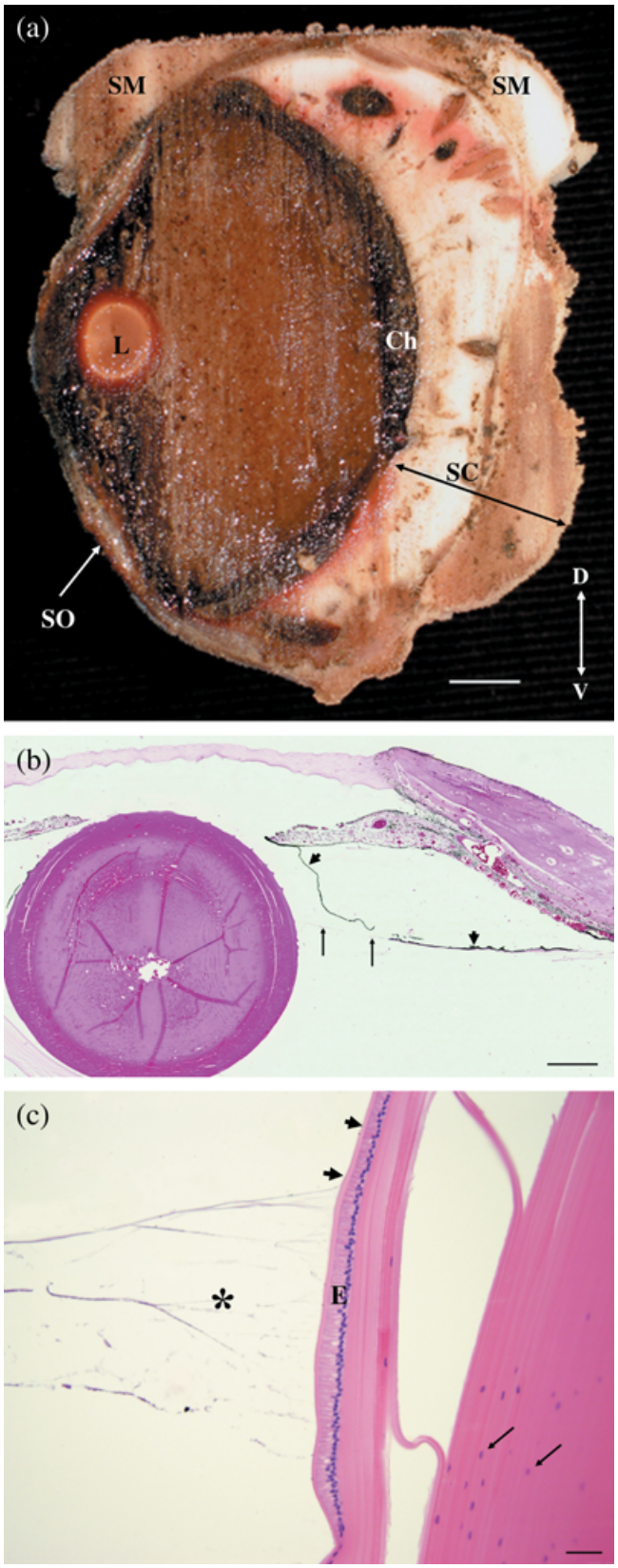

Figure 10. Lens (a) Cross-section of deep-frozen right globe illustrating small lens in relation to globe size. L = lens, SC = scleral cartilage, thickness indicated by double ended arrow, SO = scleral ossicle, Ch = choroid, SM = frozen supporting media. Bar = 5 mm. (b) Near-spherical lens demonstrated on histologic section with zonular process (arrows) attaching at lens equator. Separation of posterior iris epithelium and ciliary body epithelium (arrowheads) due to fixation artefact. H&E stain, bar = 750 µm. (c) High power view illustrating insertion site of a zonular process at the lens equator (*), lens capsule (arrowheads), single layered lens epithelium (E) and nuclei of lens bow (arrows). H&E stain, bar = 50 µm.

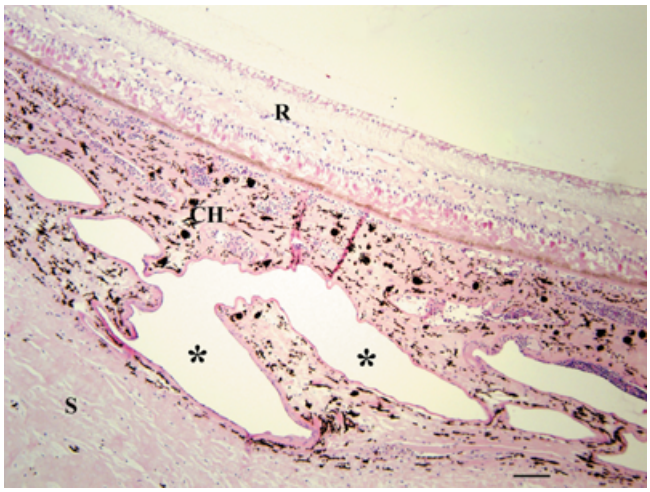

Figure 11. Large thin-walled, sinus-like vessels (*) demonstrated within the extensively vascularized choroid (CH). R = retina; S = sclera. H&E stain, bar = 100 µm.

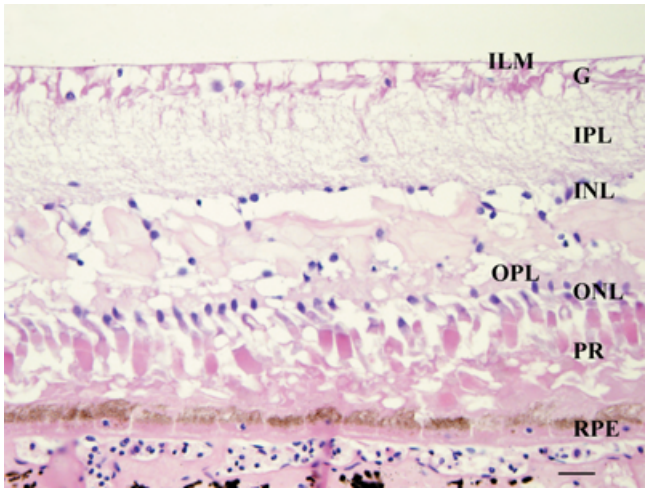

Figure 12. Microscopic section of retina. RPE = retinal pigment epithelium; PR = photoreceptor outer segments; ONL = outer nuclear layer; OPL = outer plexiform layer; INL = inner nuclear layer; IPL = inner plexiform layer; G = ganglion cell layer; ILM = inner limiting membrane. H&E stain, bar = 25 µm.

further estimates of the optical properties of the eye and the optical sensitivity could only be based on the eye of turtle 1. Given the relatively similar size of the eyes of turtles 1 and 2, the pupil dimensions of turtle 2 were used to calculate the aperture. Due to the difficulty in accessing live material, pupil measurements were only available for formalin preserved

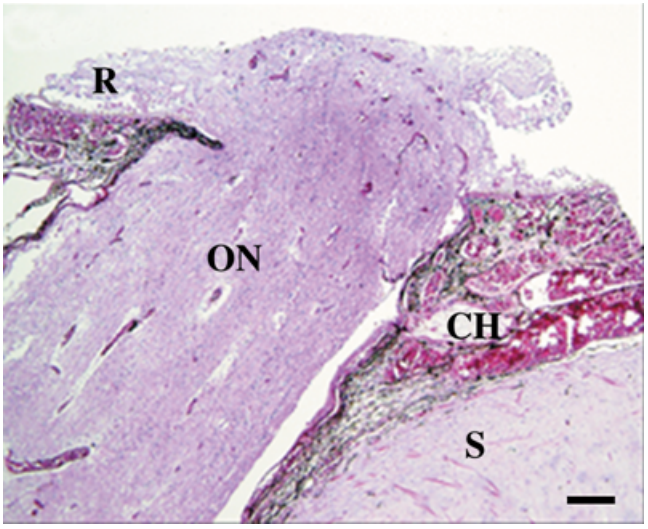

Figure 13. Optic nerve (ON) exiting through the sclera (S) at 45° to the retina (R), with extensive vascularization of the adjacent choroid (CH). H&E stain, bar = 250 μm.

tissue. In turtle 1, a focal length of 18.7 mm and pupil diameter of 4.3 mm revealed an *F*-number (focal length/aperture) of 4.3. The *F*-number would decrease to 2.3 if the pupil of the Leatherback dilated to expose the entire lens, a possibility given the iris musculature found in this study.

Apart from the optical dimensions of the eye, optical sensitivity considers light sensitivity at the level of the photoreceptors. Mean photoreceptor dimensions were used from all areas of the retina measured and optical sensitivity for light at the water surface (white light) was calculated at $S = 0.23\ \mu m^2$ sr and for monochromatic light at the preferred wavelength of the receptor (λ_{max}) $S = 0.49\ \mu m^2$ sr (Table 1). The latter value assumes that the Leatherback has a cone photoreceptor matched in its spectral sensitivity to the prevailing near-monochromatic wavelength at depth in the open ocean.

DISCUSSION

Ocular anatomy

There is little previously published information describing the ocular anatomy of marine turtles. The Leatherbacks described in this study have several characteristics in common with previous descriptions of Chelonians, that is, a small globe in comparison to the size of the animal, scleral ossicles, a thick-scleral cartilage cup, and an avascular retina,[23–25] with minor differences. The vertical diameter is consistently greater than the horizontal diameter in Leatherback eyes in contrast to equal vertical and horizontal diameters reported previously in Chelonians.[24] Scleral ossicle plates in Dermochelyidae have been reported as having irregular overlapping borders and ranging from 10 to 12 in number.[17] Ossification of scleral ossicles in this study prevented identification of individual plates by radiography or special staining. Interdigitation of ossicle plates was identified on histopathology sections.

The scleral cartilage of Leatherbacks has previously been reported as a centimeter in thickness,[17,23] which is similar to our findings. Thickened sclera can also be found in large sharks and aquatic mammals, especially notable in whales, and has been proposed as an adaptation to prevent deformation of the globe due to water pressure at depth[26] or to maintain ocular rigidity against the pull of large extraocular muscles,[24] which could apply to the Leatherback. As pressure is distributed evenly throughout the globe under water and the Leatherback cornea is relatively thin, as is the case in whales, then scleral thickness is unlikely to be related to high pressure as such.[24] A thickened sclera is required though in a nonspherical globe, to maintain peripheral rigidity and prevent deformation of the cornea at differential pressures.[24]

The plications of the nictitans appear unique to Leatherback sea turtles and have previously been described by Burne (1905).[27] These plications result in an enormous surface area for mucus secretion, and we propose that this large volume of mucus protects the cornea from the copious volumes of highly concentrated secretions from the salt glands that drain to the lateral canthus of the eye.[28,29] Although salt glands are present in other marine turtles, Leatherbacks are reported as secreting salt gland fluids of considerably higher osmotic pressure than the Loggerhead, Green and the Hawksbill turtle.[29,30] The large retractor bulbi and pyramidalis muscles are consistent with both active globe retraction and movement of the nictitans across the cornea. The plications of the nictitans may also assist spreading of the mucus across the cornea similar to the action of the plica marginalis of the avian nictitans.

The Leatherback has a blood and tissue oxygen store larger than that of the lung, and a blood oxygen-carrying capacity twice that of smaller, shallow-diving sea turtles,[31] which use their lungs as the primary oxygen store while diving.[32] The hematocrit, hemoglobin and myoglobin concentrations of Leatherbacks are among the highest recorded for reptiles, and approach levels found in diving mammals.[33] The Leatherback globe is highly vascularized throughout, with vessels being especially prominent within the choroidal layer as previously described in sea turtles.[24] The large sinusoidal vessels of the choroid allow a large quantity of blood to be stored within the globe as a mechanism of maintaining high oxygen concentrations immediately adjacent to the photoreceptors. This is especially useful during diving when the heart rate is significantly lower than that at the surface, having been recorded as low as 1.05 beats/min.[12] Vascularization of the iris and ciliary body is discussed below in respect to accommodation.

Accommodative mechanisms

In this study, the ciliary body musculature is found to be of minor significance although it has previously been reported as massive in sea turtles.[24] The lens is strongly curved and the cornea relatively flat as previously reported in sea turtles.[10,18,23–25,34] The zonular processes consist of fine strands

of collagenous tissue which support the lens in the anterior segment. The ciliary body musculature in Leatherbacks may not cause significant anterior or posterior displacement of these processes in accommodation, although it may play a role in tensioning the zonular processes and thereby supporting the lens against iris sphincter contraction or changes in aqueous humor formation. An intraocular transversalis muscle is not found in this study, with decomposition prior to fixation considered unlikely in view of the condition of the internal eye. A transversalis muscle that moves the lens nasally in binocular vision, and passes through the choroidal fissure to the ventral side of the lens and the connective tissue of the ciliary body, has been reported in the Order Testudines,[17] specifically the Loggerhead turtle.[34]

The presence of an iris dilator muscle and a prominent iris sphincter muscle suggests that the pupil in Leatherbacks is not immobile as previously reported.[23,24] Turtles, as a group, have been reported to squeeze the anterior lens with the iris sphincter muscle to produce an anterior lenticonus to supplement accommodation,[23,24] so this may also occur in Leatherbacks. A well-developed iris musculature acting on a spherical lens has also been postulated to focus light underwater in other aquatic species including water snakes, otters[35] and cetaceans.[36]

The iridocorneal angle of Leatherbacks, with its long and narrow ciliary cleft, poorly developed ciliary musculature and highly vascularized ciliary body and iris, appears to have several features in common with aquatic mammals. Little to no ciliary body musculature has been found in the West Indian manatee, the short-finned pilot whale[36] and the Antarctic Weddell seal,[37] and all three are reported to have increased ciliary body vasculature. Increased numbers and prominence of iridal and ciliary body blood vessels have also been found in other aquatic mammals such as the beluga, narwhal and fin whale.[36] Previously, the ciliary body musculature was thought to be the only means of accommodation in mammals, but the lack of ciliary body musculature is now considered as a likely adaptation to aquatic life.[38] The ability to increase aqueous outflow via both the intrascleral vessels and ciliary body vessels as identified in the manatee and pilot whale, and as appears to be the situation in Leatherbacks, has been surmised to facilitate changes in the IOP by allowing more control over the production of aqueous humor. This, in turn, may be a method of adapting to variations in osmotic conditions, outside pressure changes, and the need for accommodation.[36] Marine mammals may utilize aqueous humor dynamics to move the lens through changes in the depth of the anterior chamber for accommodation and/or reshaping of the ciliary body.[36] It is interesting that a sea turtle has evolved convergent ocular features to mammals in the same environment.

Are Leatherback sea turtles adapted to bright-light vision?

The Leatherback has a small eye relative to body size when compared to other reptiles and indeed vertebrates as a whole,[39] especially as the scleral wall thickness contributes significantly to axial length measurement. The size of the eye and the pupil in relation to the size of the globe and the lens, indicated that the eye of the Leatherback is relatively insensitive to light. In comparison, the blue marlin, a large teleost predator known to hunt to depths of 300 m which inhabits the same oceanic conditions as the Leatherback, has eyes that are highly adapted to dim light conditions.[40] For instance a blue marlin of similar body length to the Leatherback has a globe that is about 50% larger, a lens that is almost twice the diameter and a pupil that is four times larger in diameter (Fritsches, unpublished observations) than the Leatherback sea turtle measured here. Of particular interest is the small aperture (pupil) and lens of the Leatherback eye, resulting in an *F*-number from 4.3, when using the pupil diameter measured here as aperture, to 2.3 if the pupil was fully dilated. These *F*-numbers are similar to those found in Green turtles,[20] but much higher than those found in fishes such as the blue marlin (1.6, $n = 6$, Fritsches, unpublished observations) or other animals adapted to very dim light.[19,41]

As a further indicator for sensitivity to light we calculated optical sensitivity, a model that considers the optics of the eye and also the size of the photoreceptors. The optical sensitivity of the Leatherback was four times higher than that of the green turtle,[20] mainly because the outer segments of Leatherback cones were wider (5.3 μm compared to 2.9 μm in the Green turtle), allowing more photons to be absorbed per photoreceptor. However the optical sensitivity of the blue marlin based on cone photoreceptors was higher than the Leatherback turtle by a factor of five,[40] mainly due to the larger aperture, allowing more light to enter the eye and reach the retina. A possible maximal dilation of the Leatherback's pupil to fully expose the lens would result in a doubling of the optical sensitivity ($S = 0.98$ μm^2 sr and 0.46 μm^2 sr for monochromatic and white light, respectively). Whether the animal is capable of such extensive dilation of the pupil requires further investigation.

Given its relatively poor adaptations to dim light what is the predominant light environment for the Leatherback sea turtle? The exceptionally deep dives to 1000 m and beyond will lead the animal to an environment that completely lacks light from the surface, with the only source of light being the dim flashes of bioluminescence animals.[42] However, Leatherbacks appear to venture only very occasionally to these depths, spending only a few minutes there before ascending to the surface to breathe.[6] It is also unclear if the animals are actually feeding at these depths, with behavioral thermoregulation (cooling down) cited as a possible reason for these deep spike-dives.[6] Our findings suggest that visually guided feeding is unlikely at these depths, as also speculated by Levenson *et al.*[43]

Far more common are shallower dives below 150 m,[6,44] and adult Leatherbacks are also found close to shore in shallow water[14,15] and feed on the surface during the day.[13] The eyes of Leatherbacks would be well-suited for these bright light conditions. Photopic spectral sensitivities of Green and Loggerhead turtles indicate that they are also

well-suited for well-lit, shallow water marine environments.[43] Leatherback sea turtles are known to possess a visual streak along the nasotemporal retinal axis and an area temporalis of the dorsotemporal retina which corresponds to improved acuity and detection of prey of the visual field in front, ventral and lateral to the Leatherback's head and mouth.[45] Green and Loggerhead turtles also possess a visual streak,[45,46] and visual acuity thresholds measured in juvenile Loggerheads have been analogous to other benthic, shallow water species.[47]

At night the dives become more regular and shallow, leading to the suggestion that Leatherbacks may hunt their prey in the deep scattering layer,[6] which ascends at night. Our results suggest that given its visual optics the Leatherback sea turtle is not specifically adapted to visually feeding at night, however, our study does not consider possible neural mechanisms of improving the sensitivity to light.[48] Given the muscular and well developed iris it might also be possible that this species is capable of increasing its pupil diameter and hence aperture of its optics, improving its ability to see prey and avoid predators in dim light.

ACKNOWLEDGMENTS

The authors wish to thank the Nova Scotia Leatherback Turtle Working Group for the collection of the specimens, and Paul Addison for his expertise and technical assistance in preparation of the histological samples. We also thank Robert Donnell for assistance with preparation of the photomicrographs and Chris Pollitt for the use of his equipment.

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