2012

Morphology of Rathke's Glands in the Snapping Turtle, *Chelydra serpentina*, with Comments on the Presence of Multilaminar Lamellar Bodies in Turtles

S. E. Trauth

Arkansas State University, strauth@astate.edu

Follow this and additional works at: <https://scholarworks.uark.edu/jaas>

Part of the [Zoology Commons](#)

Recommended Citation

Trauth, S. E. (2012) "Morphology of Rathke's Glands in the Snapping Turtle, *Chelydra serpentina*, with Comments on the Presence of Multilaminar Lamellar Bodies in Turtles," *Journal of the Arkansas Academy of Science*: Vol. 66 , Article 28.

DOI: <https://doi.org/10.54119/jaas.2012.6621>

Available at: <https://scholarworks.uark.edu/jaas/vol66/iss1/28>

This article is available for use under the Creative Commons license: Attribution-NoDerivatives 4.0 International (CC BY-ND 4.0). Users are able to read, download, copy, print, distribute, search, link to the full texts of these articles, or use them for any other lawful purpose, without asking prior permission from the publisher or the author.

This Article is brought to you for free and open access by ScholarWorks@UARK. It has been accepted for inclusion in Journal of the Arkansas Academy of Science by an authorized editor of ScholarWorks@UARK. For more information, please contact scholar@uark.edu.

Morphology of Rathke's Glands in the Snapping Turtle, *Chelydra serpentina*, with Comments on the Presence of Multilaminar Lamellar Bodies in Turtles

S. E. Trauth

Department of Biological Sciences, Arkansas State University, State University, AR 72467-0599

Correspondence: strauth@astate.edu

Abstract

I examined the histology and ultrastructure of Rathke's glands in hatchling, juvenile, and adult snapping turtles (*Chelydra serpentina*). This species possesses four pairs of Rathke's glands (i.e., one axillary and three inframarginals) that are embedded beneath marginal bones and are named primarily according to the anatomical location of their orifices. These integumentary glands are similar anatomically and ultrastructurally to one another. Each gland is comprised of a single, highly vascularized, secretory lobule, which is surrounded by a thick tunic of asymmetrically arranged, striated muscle. Two types of large secretory vacuoles characterize most of the holocrine cells produced by a relatively thin secretory epithelium. My results support previous studies that suggest that the chief secretory material of the mostly larger, dark-staining Type 1 secretory vacuole is a glycoprotein complex. The mostly translucent Type 2 secretory vacuole may contain osmiophilic granules that exhibit variously sized lamellar bodies, whose multilaminar structural design is reminiscent of an epidermal lipid delivery system in many vertebrates. The functional role of Rathke's gland secretions in *Chelydra serpentina* and in other turtles remains unknown.

Introduction

Rathke's glands occur in members of 13 of the 14 living chelonian families (Waagen 1972, Ehrenfeld and Ehrenfeld 1973, Solomon 1984, Plummer and Trauth 2009). These large exocrine integumentary glands, also known as musk or scent glands, number from one to five pairs and exude a sometimes-malodorous secretion through external epidermal pores. They are named based upon the general location of their orifices (axillary and inguinal) and/or the proximity of the orifices to scutes (e.g., inframarginal). Rathke's glands consist of one or more lobules encased within a striated muscle tunic, and the secretory epithelium liberates

mostly ovoid-to-spherical holocrine cells (Ehrenfeld and Ehrenfeld 1973, Solomon 1984, Plummer and Trauth 2009). The secretions released by these cells are primarily proteins and, to a lesser extent, lipids, as well as various acids (Seifert et al. 1994, Weldon et al. 2008). Lamellar bodies may also be present within the secretory vacuoles of these cells (Plummer and Trauth 2009). The function of Rathke's gland secretions as well as their comparative anatomy among chelonians remains unknown despite our increasing knowledge about their morphology and glandular chemistry (Weldon et al. 2008). Few detailed investigations have focused on the histology and/or ultrastructure of Rathke's glands in chelonians (Zangerl 1941, Ehrenfeld and Ehrenfeld 1973, Solomon 1984, Plummer and Trauth 2009, Trauth and Plummer 2012).

Lamellar bodies (also referred to as multilaminar bodies, multivesicular bodies, lamellar granules, and lipid bodies, to name a few) are intracellular tubulovesicular organelles composed of concentric phospholipid bilayers. These osmiophilic structures typically occur in epithelial cells (e.g., type II alveolar cells, corneocytes, and mesothelial cells) in humans (Schmitz and Müller 1991, Fartasch 2004, Kennish and Reidenberg 2005, Spener et al. 2006, Sato and Ghazizadeh 2009, Vanhecke et al. 2010) and in Rathke's glands of turtles (Ehrenfeld and Ehrenfeld 1973, Maltoltsy and Bednarz 1975, Alibardi and Toni 2006, Plummer and Trauth 2009, and Trauth and Plummer 2012). Lamellar bodies serve as lipid storage and secretion sites, and the current functional characterization of lamellar bodies is mostly restricted to descriptive studies using electron microscopy (Spener et al. 2006). Plummer and Trauth (2009) and Trauth and Plummer (2012) reported on the presence of lamellar bodies in softshell turtles and hatchling box turtles, respectively, using transmission electron microscopy; other than a study by Mahmoud and Alkindi (2008), which showed the ultrastructure of a lipoidal body within the corpus luteum of the snapping turtle, no additional ultrastructural studies have depicted lamellar bodies in turtles.

Morphology of Rathke's Glands in the Snapping Turtle, *Chelydra serpentina*

Snapping turtles (*Chelydra serpentina*) often produce a musky odor (Ernst and Lovich 2009) from four pairs of Rathke's glands (Zangerl 1941, Waagen 1972). My objectives in the present study were to investigate the morphology and ultrastructure of Rathke's glands in the snapping turtle and report on the comparative ultrastructure of multilaminar lamellar bodies found within secretory vacuoles of the holocrine cells of Rathke's glands among box, mud, musk, and softshell turtles.

Materials and Methods

I dissected Rathke's glands from hatchling, juvenile, and adult snapping turtles collected from northeastern Arkansas. The glands were prepared for light microscopy (LM) and scanning and transmission electron microscopy (SEM and TEM, respectively) in the Electron Microscopy Facility at Arkansas State University. Rathke's glands of other turtles (i.e., Mississippi mud turtle, *Kinosternon subrubrum hippocrepis*, the eastern musk turtle, and *Sternotherus odoratus*) previously necropsied and prepared as mentioned below were also investigated to determine the presence of lamellar bodies in Rathke's glands. Voucher specimens were deposited in the Arkansas State University herpetological collection (ASUMZ). Turtles were sexed, measured (i.e., carapace lengths, CL, in mm), sacrificed with an intra-pleuroperitoneal injection of sodium pentobarbital, macro-photographed, and dissected to reveal glands.

Rathke's glands were extracted from beneath the lateral edges of carapaces using a Dremel Multi-Max™ oscillating tool (Fig. 1). The glands were then fixed in either 10% neutral buffered formalin, NBF (for SEM) for 48 h or in a 2% glutaraldehyde (GTA) solution buffered with 0.1 M sodium cacodylate at a pH of 7.2 (for plastic sectioning) for 2 h. For post fixation, I used 1% osmium tetroxide, buffered as above, for 2 h.

For LM of plastic-embedded tissues, I dehydrated tissues in a graded series of increasing ethanol solutions (50-100%), placed in a 50/50% acetone/plastic mixture for overnight infiltration, and then embedded tissue in Mollenhauer's Epon-Araldite #2 (Dawes 1988). For thick sectioning (approximately 1 µm in thickness) and staining, I used glass knives on an LKB Ultratome (Type 4801A) with Ladd® multiple stain (LMS), respectively. For photomicroscopy, I utilized a Nikon Eclipse 600 epi-fluorescent light microscope with a Nikon DXM 1200C digital camera (Nikon Instruments Inc, Melville, NY). I also used a Konica Minolta Maxxum 7D digital single lens reflex

camera fitted with a ProMaster AF to photograph turtle shells and glandular tissues.

For SEM, I dehydrated tissues in a graded series of increasing ethanol solutions (50-100%), followed by several fluid exchanges in 100% ethanol. An Autosam-dri-815 critical point drier (Tousimis Research Corporation, Rockville, MD) was used (31°C, 1072 psi, ventilation rate ~100 psi/min) to remove excess ethanol. Tissue samples were then mounted on 25.4 mm aluminum pin stub specimen mounts and coated with gold using a Cressington 108 sputter coater (Cressington Scientific Instruments Ltd, Watford, UK). Tissues were analyzed both qualitatively and quantitatively with a Vega TS 5136XM digital scanning electron microscope (Tescan USA Inc., Cranberry Township, PA) at 19.5 kV.

Plastic-embedded samples prepared for LM were also utilized for TEM. Tissue blocks were sectioned with a diamond knife. Sections were picked up with 150 - 200 mesh copper grids, stained with uranyl

Figure 1. Rathke's glands in a juvenile male *Chelydra serpentina* (ASUMZ 30973; CL = 155 mm). A. Dissection of glands using dorsal and lateral incisions into carapace and marginals. B. Arrow points to the orifices of the axillary gland (far left) and three inframarginal glands along bridge. C. Arrow points to the axillary gland and inframarginal glands (left to right) revealed by reflecting lateral incisions as shown in A; metric scale provided in A - C.

acetate (3% aqueous) and lead citrate for 40 min each. Grids were examined with a JEOL 100 CX-II transmission electron microscope (JEOL USA, Inc., St. Louis, MO) at 60 kV (55 μ A). Positive digital images were generated by scanning developed TEM negatives using an Epson Perfection 4990 scanner (Epson America, Inc., Long Beach, CA).

For the descriptive terminology of Rathke's glands, I followed Ehrenfeld and Ehrenfeld (1973), Solomon (1984), Plummer and Trauth (2009), and Trauth and Plummer (2012). For describing the ultrastructure of lamellar bodies, I followed a previous investigation on hatchling three-toed box turtles (Trauth and Plummer 2012).

Results

Gross Morphology

From a dorsal perspective, the four pair of Rathke's glands lie beneath the posterolateral edge of pleural scute 1 and extends to the middle of costal scute 3 (Fig. 1A and C). The orifices of these epidermal glands are mostly inconspicuous; however, the three inframarginal orifices are embedded in the posterolateral surfaces of three bridge scutes, whereas the orifice of the axillary gland appears along the interface between abdominal skin and the 4th marginal scute (Fig. 1B). Internally, the glands are situated in a linear series within slight depressions along the interior peripheral bones (Ernst and Lovich 2009). Their gland dimensions are variable, but fall between 8 - 12 mm in length and 3 - 5 mm in width.

Light Microscopy

The internal anatomy of axillary glands in *Chelydra serpentina* is comprised of a single lobule, which possesses a secretory epithelium that rests upon a thin basement membrane (Fig. 2B; Fig. 4C). A thin-to-moderately thick layer of dense connective tissue is contiguous with the basal lamina (Figs. 3A and 5A). In general, the secretory epithelium is comprised of a single basal cell layer, which produces roughly oblong-to-spheroid shaped holocrine cells (Figs. 2 and 3). These epithelial cells proliferate outward into an expansive lumen (Fig. 2). The exposed wall of each gland is made of a relatively thick muscular tunic (Figs. 2A and 4A). At some point following their release from the secretory epithelium, the secretory cells lose their structural integrity and degenerate, dumping their cellular contents into the glandular

Figure 2. Light micrographs of axillary gland and 1st inframarginal gland in a hatchling and juvenile male *Chelydra serpentina* (A, ASUMZ 31248, CL = 63 mm; B, ASUMZ 31358, CL = 150 mm), respectively. Ladd® multiple stain. A. Section showing gland lumen (Lu) filled with holocrine cells containing smaller, dark-staining, Type 1 secretory vacuoles and larger, lipid droplets (clear spheres) characteristic of Type 2 secretory vacuoles. The secretory vacuoles are interspersed within secretory cell/cellular debris matrix. Sm, striated muscle. B. Section revealing the boundary among the striated muscle, dense connective tissue, and secretory epithelium. Dct, dense connective tissue; Sv-1, Type 1 secretory vacuole; Sv-2, Type 2 secretory vacuole.

lumen. This mostly flocculent cellular debris becomes the material that is eventually passed into a duct leading to the exterior.

Secretory cells are also characterized by the presence of two different types of secretory vacuoles: Type 1 and Type 2 (Figs. 2 - 5). Type 1 secretory vacuoles are generally slightly larger than those of Type 2, appear as singular mostly spherical masses, and range from 20 - 30 μ m at their greatest diameter. They may occur in clusters as small dark-staining spherical or oval masses. Their matrix is not removed during tissue preparation. When stained with LMS, Type 1 secretory vacuoles appear bright red to purple in color, a positive indication of the presence of carbohydrate substances.

Morphology of Rathke's Glands in the Snapping Turtle, *Chelydra serpentina*

Figure 3. Light micrographs of axillary gland and 1st inframarginal gland in a hatchling and juvenile male *Chelydra serpentina* (A, ASUMZ 31248, CL = 63 mm; B, ASUMZ 31358, CL = 150 mm), respectively. Ladd® multiple stain. A. Section showing secretory epithelium with numerous holocrine cells containing smaller, dark-staining, Type 1 secretory vacuoles and larger, lipid droplets (clear spheres) characteristic of Type 2 secretory vacuoles. Some Type 2 secretory vacuoles contain osmiophilic lamellar bodies. B. Individual holocrine cells exhibiting secretory vacuoles. Nu, nucleus; No, nucleolus. See Figs. 1 and 2 for additional abbreviations.

In contrast, Type 2 secretory vacuoles are mostly oblong when fully distended, range from 15 – 25 µm in diameter, and appear mostly devoid of material. Type 2 secretory vacuoles generally contain lipoidal material that is normally referred to as lipid droplets. Soluble lipids of these lipid droplets are normally removed from these vacuoles during histological preparation; however, osmiophilic, membrane-bound, lipoidal structures (Figs. 5 and 6) can also found within Type 2 secretory vacuoles.

Scanning Electron Microscopy

When transversely sectioned, Rathke's glands in *Chelydra serpentina* appear to be asymmetrical with respect to the width of the muscular tunic (Figs. 2A and 4A). The unattached (free) surface of each gland is dominated by a thick layer of striated muscle, whereas the attached surface is comprised primarily of a broad layer of dense connective tissue. This layer is continuous with the innermost layer of dense connective tissue that encapsulates the glandular lumen. As individual holocrine cells proliferate away from the secretory epithelium, they fail to maintain their plasma membrane integrity. Holocrine cell apoptosis is characterized by the appearance of holes within the plasma membrane (Fig. 4D). Upon liberation, the glandular constituents of cells as well as the cellular debris create a honeycomb configuration within the lumen (Fig. 4B and C).

Transmission Electron Microscopy

Ultrastructural features of secretory epithelial cells in Rathke's glands of *Chelydra serpentina* are shown in Fig. 5. Primary cytological components of a typical developing secretory cell (Fig. 5A) include a single or small cluster of Type 1 secretory vacuoles situated apically and in a polar-opposite position to a single, large Type 2 secretory vacuole (see also Fig. 3). Circular-to-oblong osmiophilic granules were observed within Type 2 secretory vacuoles (Fig. 5B). These granules produce concentric whorls of multilaminar, phospholipid bilayers (Fig. 5C) and are individually referred to as lamellar bodies. Lamellar bodies enlarge as consecutive bilayers are added to the membrane complex. Lamellar bodies vary greatly in size and complexity. A uniform chromatin fills the circular-shaped secretory cell nucleus (Fig. 5A) of secretory cells. Upon cell death and fragmentation, the cytoplasm of the secretory cells becomes highly vacuolated due to destructive lysosomal activity. Numerous lamellar bodies of varying sizes, shapes and stages of development can be seen inside an osmiophilic granule in Fig. 5C.

Discussion

Most morphological features of Rathke's glands in the snapping turtle resemble those previously described in other non-marine turtles. For example, the thin secretory epithelium of each gland is normally incased within a thick tunic of striated muscle. One notable

difference found in the snapping turtle was an asymmetry with respect to the thickness of the muscle layer; i.e., the attached surface of these glands rests on a dense layer of connective tissue that lacks an associated muscular envelope in this region. I have observed a similar condition in Rathke's glands of the alligator snapping turtle, *Macrochelys temminckii* (unpublished data). Only one secretory lobule was observed in the snapping turtle (a feature also observed

in hatchling box turtles, *Terrapene carolina triunguis* and *Terrapene ornata luteola* [Trauth and Plummer 2012]), whereas two lobules are characteristic of softshell turtles (Plummer and Trauth 2009) and the Mississippi mud turtle, *Kinosternon subrubrum hippocreps* (Webb 2010). Another feature of Rathke's glands shared by the snapping turtle and other turtles is the presence of holocrine cells that exhibit Type 1 and Type 2 secretory vacuoles (Ehrehfeld and Ehrenfeld

Figure 4. Scanning electron micrographs of Rathke's glands in an adult male *Chelydra serpentina* (ASUMZ 31239, CL = 302 mm). A. Transverse section of axillary gland filled with holocrine cells and cellular materials. B. Magnification of A revealing honeycomb appearance of holocrine cells within lumen. C. Magnification of B revealing the thin secretory epithelium. D. Axillary gland showing the release of holocrine cells. Arrow points to a hole in the plasma membrane of a cell. Abbreviations are the same as in previous figures, unless otherwise stated.

Morphology of Rathke's Glands in the Snapping Turtle, *Chelydra serpentina*

Figure 5. Transmission electron micrographs of holocrine cells in Rathke's glands of male *Chelydra serpentina* (A-B, ASUMZ 31358; C, ASUMZ 31248, CL = 63 mm). A. Secretory epithelium of an axillary gland shows developing Type 1 secretory vacuole. The cell is positioned atop a basal cell (Bc), which rests upon the basal lamina (Bl). B. Type 1 and Type 2 secretory vacuoles of a development cell. An osmiophilic granule can be seen inside the Type 2 secretory vacuole. Early formation of lamellar bodies has begun within this granule. C. Cluster of lamellar bodies developing within an osmiophilic granule of a Type 2 secretory vacuole.

1973). Two recent studies examined the ultrastructure of these secretory vacuoles (Plummer and Trauth 2009, Trauth and Plummer 2012) and have revealed the presence of lamellar bodies in Type 2 secretory vacuoles.

The secretory material within Type 1 secretory vacuoles is common to all turtles, including marine species. The secretory material also appears mostly homogenous and has been identified as a carbohydrate-protein substance using a periodic acid-Schiff reaction

(Ehrenfeld and Ehrenfeld 1973, Plummer and Trauth 2009, Trauth and Plummer 2012). Type 2 secretory vacuoles are basically lipid droplets, and although much of this lipid material is removed during histological preparation, osmiophilic granules often persist in some vacuoles. These granules appear to undergo a progressive development, which culminates in the production of numerous multilaminar lamellar bodies. By definition, lipid droplets are storage compartments for neutral lipids, whereas lamellar

bodies are organelles for the storage and secretion of polar lipids (Spener et al. 2006).

I compared the lamellar bodies of the snapping turtle of the family Chelydridae with members of three other families of turtles (Trionychidae—midland smooth softshell, *Apalone mutica mutica*; Emydidae—three-toed box turtle, *Terrapene carolina triunguis*; Kinosternidae—eastern musk turtle, *Sternotherus odoratus* and the Mississippi mud turtle, *Kinosternon subrubrum hippocrepsis*) and found common features among them (Fig. 6). All lamellar bodies among these species exhibit phospholipid bilayers, but the size and overall complexity of these bodies differ among these species. Although I was able to examine only a limited

number of specimens for each species, lamellar bodies of the Mississippi mud turtle were most similar to those of the snapping turtle (Figs. 5C and 6D). The lamellar bodies found in *A. mutica* (Fig. 6A, from Plummer and Trauth 2009) were much smaller than those observed in any of the other species. Hatchling *T. carolina* (adults box turtles lack Rathke's glands) exhibited lamellar bodies that were not only larger than those found within the softshell species, but they often exhibited tightly compressed bilayers along with circular bodies not seen in the other turtle species (Fig. 6B).

The bilayers within the lamellar bodies in *Sternotherus odoratus* (Fig. 6C) were more

Figure 6. Transmission electron micrographs of lamellar bodies of Rathke's glands in selected turtle species. A. Adult male *Apalone mutica* (ASUMZ 30748; CL = 175 mm). B. Hatchling *Terrapene carolina triunguis* (ASUMZ 31242; CL = 29 mm). C. Adult female *Sternotherus odoratus* (ASUMZ 31139; CL = 110 mm). D. Adult female *Kinosternon subrubrum hippocrepsis* (ASUMZ 31195; CL = 98 mm). See text for explanation of micrographs.

Morphology of Rathke's Glands in the Snapping Turtle, *Chelydra serpentina*

pronounced and less well organized compared to the other species. The lamellar bodies in this species also exhibit a highly vacuolated, desquamated appearance.

The formation of lamellar bodies within osmiophilic granules of the snapping turtle and the Mississippi mud turtle (Figs. 5C and 6D) yields a variety of bilayered configurations. Most of these entities develop as haphazard arrangements of bilayers within a single developing body and best described as longitudinal, circular, or whorl layering.

In conclusion, resolving the function of Rathke's glands will require additional histological and ultrastructural investigations and offers a unique opportunity for microscopists. Knowledge about the development of lamellar bodies and their ultimate disposition should enhance our understanding of role of these epidermal derivatives.

Acknowledgments

Collection of turtles was authorized by a scientific collecting permit from the Arkansas Game and Fish Commission. I thank Mike Plummer, Matthew Connior, and Phillip Jordan for their assistance in the collection of specimens. This work was conducted under established protocols set by the IACUC at Arkansas State University.

Literature Cited

- Alibardi L** and **M Toni**. 2006. Skin structure and cornification proteins in the soft-shelled turtle *Trionyx spiniferus*. *Zoology (Jena)* 109:182-195.
- Dawes CJ**. 1988. Introduction to biological electron microscopy: theory and techniques. Ladd Research Industries, Inc., Burlington (VT). 315 p.
- Ehrenfeld JG** and **DW Ehrenfeld**. 1973. Externally secreting glands of freshwater and sea turtles. *Copeia* 1973:305-314.
- Ernst CH** and **JE Lovich**. 2009. Turtles of the United States and Canada. 2nd ed. Johns Hopkins University Press, Baltimore, MD. 827 p.
- Fartasch M**. 2004. The epidermal lamellar body: a fascinating secretory organelle. *Journal of Investigative Dermatology* 122, xi-xii;doi:10.1111/j.0022-202X.2004.22541.x.540.
- Kennish L** and **B Reidenberg**. 2005. A review of the effect of occlusive dressings on lamellar bodies in the stratum corneum and relevance to transdermal absorption. *Dermatology Online Journal* 11:7.
- Mahmoud IY** and **AYA Alkindi**. 2008. Reproductive physiology of the snapping turtle. In Steyermark AC, MS Finkler and RJ Brooks, editors. *Biology of the snapping turtle (Chelydra serpentina)*. Johns Hopkins University Press, Baltimore, MD. p. 59-70.
- Matoltsy AG** and **JA Bednarz**. 1975. Lamellar bodies of the turtle epidermis. *Journal of Ultrastructural Research* 53:28-132.
- Plummer MV** and **SE Trauth**. 2009. The structure of Rathke's glands in the softshell turtles *Apalone mutica* and *A. spinifera*. *Herpetological Conservation and Biology* 4:207-220.
- Sato S** and **M Ghazizadeh**. 2009. Lamellar body formation and dark multivesicular body pockets in fetal and postnatal normal rat alveolar type II cells: ultrastructural study. *Experimental Lung Research* 35:748-755.
- Schmitz G** and **G Müller**. 1991. Structure and function of lamellar bodies, lipid-protein complexes involved in storage and secretion of cellular lipids. *Journal of Lipid Research* 32:1539-1570.
- Seifert WE, SW Gotte, TL Leto and PJ Weldon**. 1994. Lipids and proteins in the Rathke's gland secretions of the North American mud turtle (*Kinosternon subrubrum*). *Comparative Biochemistry and Physiology* 109B:459-463.
- Solomon SE**. 1984. The characterization and distribution of cells lining the axillary gland of the adult green turtle (*Chelonia mydas* L.). *Journal of Anatomy* 138:267-279.
- Spener F, SE Kohlwein and G Schmitz**. 2006. Lipid droplets and lamellar bodies – from innocent bystanders to prime targets of lipid research for combating human diseases. *European Journal of Lipid Science and Technology* 108:541-543.
- Trauth SE and MV Plummer**. 2012. Comparative histology, histochemistry, and ultrastructure of Rathke's glands in hatchlings of two species of North American box turtles (*Terrapene carolina* and *T. ornata*). *Chelonian Conservation and Biology* 11 (*in press*).
- Vanhecke D, G Herrmann, W Graber, T Hillmann-Marti, C Mühlfeld, D Studer and M Ochs**. 2010. Lamellar body ultrastructure revisited: high-pressure freezing and cryo-electron microscopy of vitreous sections. *Histochemistry and Cell Biology* 134:319-326.

- Waagen GN.** 1972. Musk glands in recent turtles. MS Thesis, University of Utah, Salt Lake City, Utah, USA. 64 p.
- Webb S.** 2010. The histology and histochemistry of Rathke's glands in the Mississippi mud turtle, *Kinosternon subrubrum hippocrepis*. MS Thesis, Arkansas State University, Jonesboro. 37 p.
- Weldon PJ, B Flachsbarth and S Schulz.** 2008. Natural products from the integument of nonavian reptiles. *Natural Products Reports* 25:738–756.
- Zangerl R.** 1941. A series of lateral organs found in embryos of the snapping turtle (*Chelydra serpentina*). *Papers of the Michigan Academy of Science Letters* 26:339–341.