# Gross and microscopic anatomy of the orbital glands of *Malaclemys* and other emydine turtles

F. B. M. Cowan
*Department of Zoology, University of Toronto, Toronto, Ontario*
Received March 24, 1969

Cowan, F. B. M. 1969. Gross and microscopic anatomy of the orbital glands of *Malaclemys* and other emydine turtles. Can. J. Zool. 47: 723–729.

A gross anatomical and light microscopic study of the orbital regions of several stenohaline and one euryhaline emydine turtles has shown that each has two orbital glands, the Harderian and the lachrymal. The Harderian is apparently serous and is identical in all species. The lachrymal gland of all the stenohaline species is a mixed seromucous gland. In the euryhaline *Malaclemys*, the lachrymal gland differs in being much larger and possessing a structure which resembles to a degree other known salt glands. The lachrymal glands from *Malaclemys* housed in salt and in fresh water differ in size and histochemistry. Whether these differences are related to salt secretion, corneal lubrication, or some other function is not known.

## Introduction

There are several detailed accounts of the anatomy of the orbital glands in the Squamata (Schwartz-Karsten 1937; Smith and Bellairs 1944), but similar glands in turtles have received little attention. The available reports on the latter deal almost exclusively with members of the marine Cheloniidae (Abel and Ellis 1966). The anatomy of the orbital glands in emydines has been virtually ignored.

This might represent a significant omission as one emydine, the euryhaline *Malaclemys*, is reported to have an orbital gland capable of hypertonic secretion (Schmidt-Nielsen and Fänge 1958; Dunson and Taub 1967), and to be able to survive in hyperosmotic media by the use of extrarenal salt secretion. Both reports record the collection of hypertonic secretion from the orbital region, but details of methodology are not given. Nor was the origin of the secretion known, although the assumption appears to be that the secretion originates as it does in the Cheloniidae, from the lachrymal gland. Direct cannulation of the duct of the gland has not been possible as it is with the larger turtles, and more indirect methods had to be used with *Malaclemys* (Dr. A. M. Taub, personal communication). Under these conditions, without a thorough knowledge of the anatomy of the orbit, the origin of the salt secretion and the reliability of the methods used remain uncertain.

In my attempt to confirm the presence of orbital hypertonic secretion in *Malaclemys*, the first step was to determine the glands that are actually present and to describe their anatomy fully. As more than one gland was found to be present, the next step was to identify the source of secretion.

## Material and Methods

The following species were examined: *Malaclemys terrapin*, *Pseudemys scripta elegans*, *Graptemys geographica*, *Graptemys barbouri*, and *Chrysemys picta marginata*. The *Malaclemys* were obtained through a local fish market from Chesapeake Bay, *Graptemys barbouri* came from Commercial Aquarium Suppliers, *Pseudemys* were acquired in a local pet shop, and I collected the rest in the St. Lawrence River in the vicinity of Kingston, Ontario.

The freshwater specimens were housed in dechlorinated tap water to which was added 1 gram each of sodium chloride and calcium chloride per liter of water. The *Malaclemys* were divided into three groups, one being kept as above and the others kept in 50% and 100% artificial seawater made from Instant Ocean (Aquarium Systems Inc., Ohio). The animals were fed shrimp every 3 days. Lettuce and fish scraps were occasionally included.

The animals were killed by decapitation, and the eyeball, eyelids, and associated structures were dissected out under fixative. This method was checked for completeness by serial sectioning of smaller heads. Baker's formol calcium, Lillie's acetic acid – alcohol – formalin, Bouin's, and glutaraldehyde (2% in 0.067 *M* phosphate buffer) were used as fixatives. Fixation was at 4 °C and for Bouin's and formalin containing fixatives lasted for 12 hours. Glutaraldehyde fixation lasted 2 hours and the fixation of whole heads 48 hours. Decalcification was done at room temperature with ethylenediaminetetraacetate and usually required 7 days for a head 1 cm in diameter.

The following stains were used: hematoxylin and eosin, periodic acid – Schiff (PAS) method with and without prior diastase digestion (McManus 1948), alcian blue at pH 3.0 and 0.5 (Lev and Spicer 1964), alcian blue in increasing concentrations of magnesium chloride (Scott and Dorling 1965), and toluidine blue at pH 4.5

and 2.5. Methylation and demethylation procedures before staining were also used with some of the above methods (Lillie 1965). Lillie's azure B – eosin method was carried out, and rhodamine B and Sudan III staining of cryostat sections was done in an attempt to identify simple lipids.

## Observations

All specimens examined possess a Harderian gland and a lachrymal gland (see discussion for basis of nomenclature). In addition, unicellular and simple tubular glands are found on the conjunctival surface of the upper and lower eyelids and on the deep aspect of the nictitating membrane.

In all specimens the Harderian gland is similar in both gross and microscopic anatomy. This gland lies mainly along the medial surface of the eyeball (Figs. 1A and 1B) where it is overlain by the insertions of the anterior rectus and superior oblique muscles. An anterior projection of the gland lies against the anterior bony wall of the orbit. Posteriorly the gland extends to the level of the optic nerve (Fig. 1A). The gland is invested in a thin connective tissue capsule which contains considerable pigment and thus gives the gland a mottled grey appearance.

The major axis of the gland is anteroposterior, but a portion of the gland leaves the main body along its ventral surface and curves ventrally and laterally around the ventral surface of the eyeball (Figs. 1A and 1B). This portion of the Harderian gland contains the ducts of the gland.

The ducts of the Harderian gland open into the conjunctival space along the medial surface of the nictitating membrane through openings which are approximately 50 μ in diameter (Fig. 3, arrows). These openings are arranged in a horizontal row near the base of the nictitating membrane (Fig. 1C). The initial segment of each duct is composed of columnar epithelium which may be either stratified or pseudostratified. The cells lining the ducts appear to be mucus-secreting as they are PAS-positive after diastase digestion and stain with alcian blue at pH 3.0 (Fig. 4). They stain metachromatically with toluidine blue. As the ducts follow the ventral curvature of the eyeball, the epithelium becomes simple columnar, retaining the staining characteristics just described. The lumen gradually widens and the ducts may bifurcate once or twice (Fig. 3). The transition from duct to tubules is abrupt, both in the structure of the epithelium and in the width of the lumen.

All the tubules in the gland have identical simple columnar, granular epithelium lining a wide irregular lumen (Fig. 5). Further branching of the tubules does not occur, although each tubule has large irregular outpocketings along its length (Fig. 2). The tubular epithelium is very regular, and each cell is filled with large

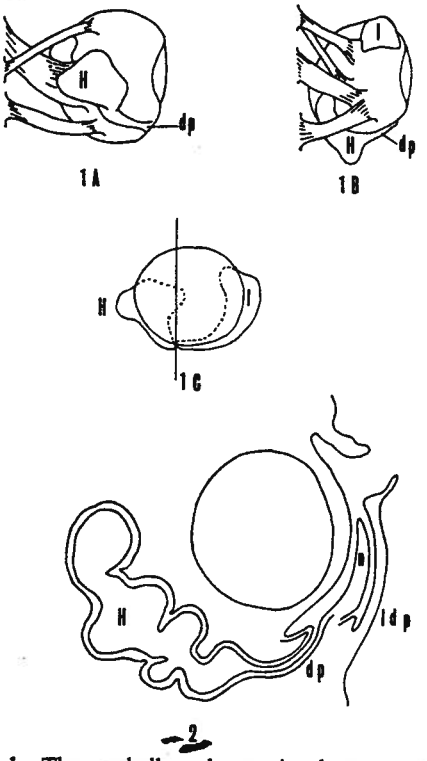


FIG. 1. The eyeball and associated structures of *Pseudemys scripta*. A. Anterior view showing the related muscles and the Harderian gland (H) and its duct or neck portion (*dp*) leading to the conjunctival space. B. Dorsal view showing the dorsal aspect of the Harderian gland (H) anteriorly and of the lachrymal gland (*I*) and associated muscles. C. Lateral view showing anteroposterior relationships of the Harderian and lachrymal glands. The relationships shown are common to all the species studied, but in *Malaclemys* the more posterior portion of the lachrymal gland is much enlarged.

FIG. 2. Diagram of a section through the most posterior duct of the Harderian gland and a segment of the most anterior duct of the lachrymal gland (for the level of the section see Fig. 1C). The duct (*dp*) of the Harderian gland leads from the medial surface of the nictitating membrane (*n*) and proceeds medially where it abruptly widens to form an unbranched tubule with a wide lumen. The most anterior duct of the lachrymal gland is also seen opening through the lateral aspect of the nictitating membrane (*ldp*) and proceeding posteriorly out of the plane of the section.

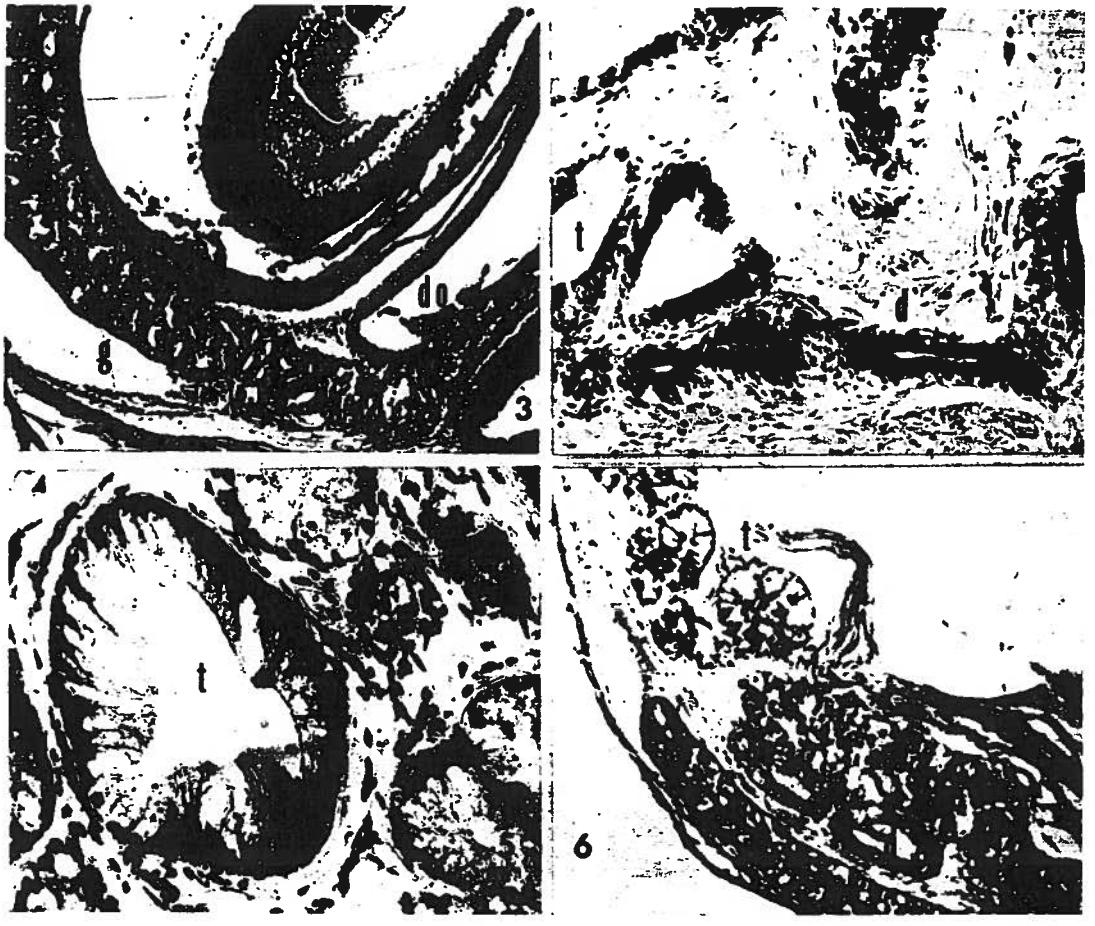


FIG. 3. Harderian gland of *Graptemys*, showing the main body of the gland (*g*) connecting through a neck region (*dp*) to the duct openings (*do*) on the medial surface of the nictitating membrane (*n*). Hematoxylin and eosin. 75 ×.

FIG. 4. Harderian gland of *Pseudemys*, showing dense staining with alcian blue of the ducts (*d*) and weak staining of the tubules (*t*). Alcian blue pH 3.0. 112 ×.

FIG. 5. Harderian gland of *Malaclemys* showing tubules (*t*) with large irregular lumen surrounded by tall columnar cells. Note the dense staining of the cytoplasm at the basal end of the cells. Toluidine blue 0.5% at pH 4.5. 750 ×.

FIG. 6. Lachrymal gland of *Graptemys*, showing ducts (*d*) and tubules (*t*) loosely aggregated in connective tissue. PAS – hematoxylin. 150 ×.

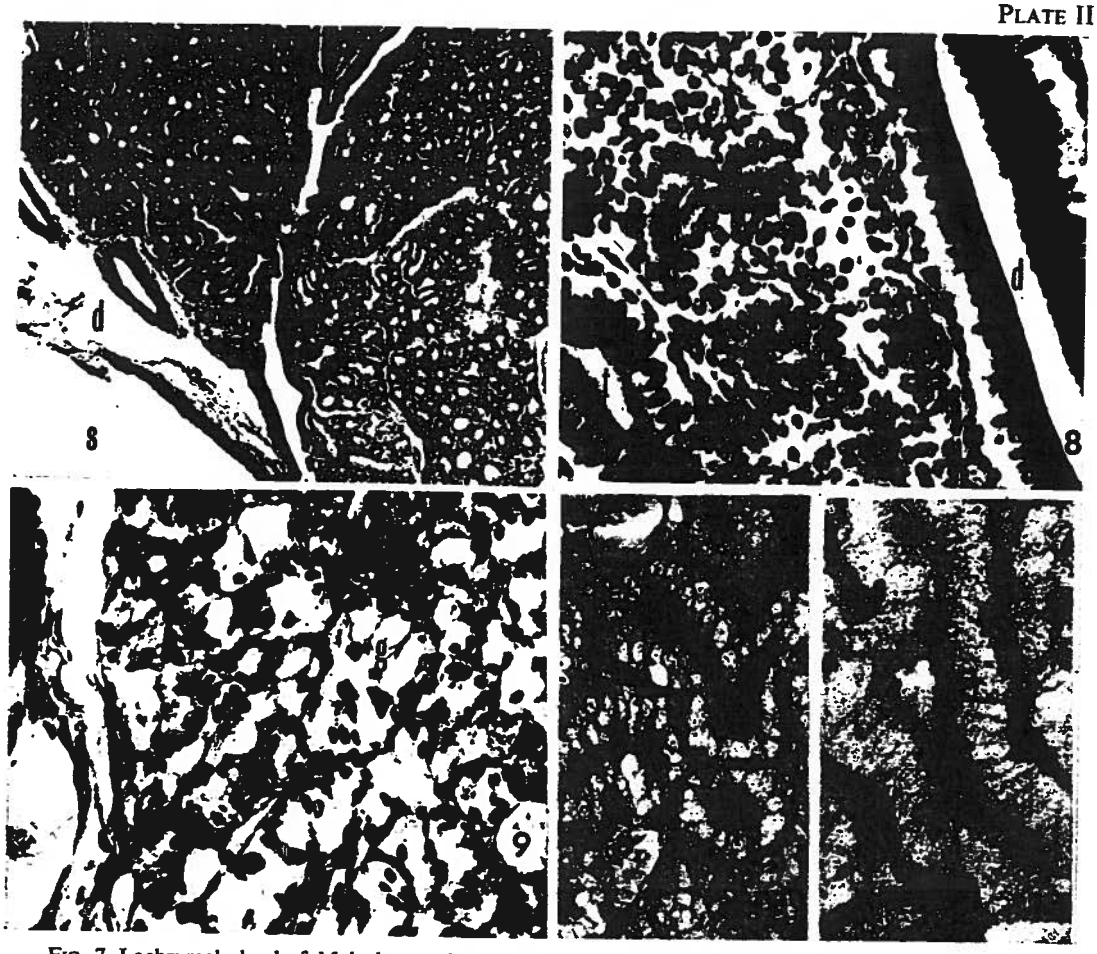


FIG. 7. Lachrymal gland of *Malaclemys*, showing ducts (*d*) emptying into conjunctival sac (*s*). Each duct drains one secretory lobule. Hematoxylin and eosin. 75 ×.

FIG. 8. Lachrymal gland of *Malaclemys*, showing dense metachromatic staining of the duct (*d*) and moderate staining of the tubules (*t*). Toluidine blue at pH 4.5. 370 ×.

FIG. 9. Lachrymal gland of *Pseudemys*, showing tubules with very slender, granular PAS-positive cells (*g*) wedged between larger PAS-negative cells. PAS – hematoxylin. 370 ×.

FIGS. 10, 11. Lachrymal glands of *Malaclemys* kept in 100% seawater (Fig. 10) and in fresh water (Fig. 11). Alcian blue at pH 0.5. 370 ×.

granules that do not stain with any of the methods used. The basal portion of the cell contains, in addition to a nucleus flattened against the basal membrane, a large quantity of a material which stains metachromatically with toluidine blue at pH 4.5 but not at pH 2.5 (Fig. 5). This material is particularly noticeable in tissue fixed in Lillie's acetic acid – alcohol – formalin.

The above morphology is found in the Harderian glands from fresh and brackish water species, and in the latter there are no differences in size or structure of the glands from animals kept in seawater or fresh water. This is in contrast to the differences in glandular size and histology of the other major orbital gland found in emydines, the lachrymal gland. This gland is probably the homologue of the gland described as salt-secreting in *Chelonia* and *Caretta* (see Discussion).

All specimens examined possess this second orbital gland. The gross anatomical relationships to other structures appear similar in all emydines, although the gland is much larger in *Malaclemys* than in the stenohaline species. The gland begins ventral to the lower lid. The most anterior portion lies just posterior to the last duct of the Harderian gland (Fig. 1C). In the freshwater species the gland is a small curved plate which runs posteriorly and somewhat medially in the connective tissue of the lower lid. Occasionally the gland extends posteriorly so that it leaves the connective tissue of the lid and curves dorsally along the posterior surface of the eyeball. In the freshwater species this posterior portion is invariably very small. In *Malaclemys*, while the origin of the gland is similar, the posterior extension is much enlarged, forming a globular mass against the posterior surface of the eyeball. This mass may be as large as the eyeball itself.

In all cases the ducts of the lachrymal gland open in a horizontal row along the lateral surface of the nictitating membrane or the medial surface of the lower lid, through pores of about the same size as those of the Harderian gland, around 50 μ. The ducts arising from these openings run ventrally and laterally towards the base of the lid. Here they branch repeatedly and run medially and posteriorly (Fig. 6). There are no interlobular ducts; each duct supplies one lobule and may branch within that lobule. In some cases, especially in the smaller freshwater emydines, the different lobules are only loosely aggregated and do not form a compact glandular unit. Usually in larger specimens the lobules, particularly the more posterior, do become aggregated and invested with a common connective tissue capsule. This is most complete and obvious in *Malaclemys* (Fig. 7).

The stratified epithelium of the ducts is PAS-positive after diastase digestion (Fig. 6). Some of the PAS staining is, however, removed by diastase, the resistant staining being found apically in the most superficial cell layer. This apical pattern of staining is also produced with alcian blue at pH 3.0, but is almost abolished at pH 0.5. Toluidine blue staining, which is metachromatic, is somewhat reduced by methylation and demethylation, but this is hard to quantitate.

In these features the lachrymal glands from all the emydines are similar. The tubule epithelium, however, distinguishes the lachrymal of *Malaclemys* from that of the freshwater species. In *Malaclemys* the major cell of the quite uniform epithelium is tall columnar and each cell is separated from its neighbors by a prominent intercellular space. This space is most obvious basally, and the apical ends of the cells seem closely apposed. The tubule cells stain with the PAS method; some of this stain is removed by diastase, leaving a strong apical staining. Toluidine blue at pH 4.5 and 2.5 stains the apex strongly and metachromatically (Fig. 8). The apices of the tubule cells stain strongly with alcian blue at pH 0.5, as well as at pH 3.0. A similar staining pattern is seen with alcian blue with 0.8 *M* magnesium chloride. With this technique the only other structure staining is the connective tissue and basement membrane matrix. The lipid stains used gave no indication of the presence of lipid.

The epithelium of the tubules of *Malaclemys* is not entirely uniform. Interspersed between the principal cells are occasional goblet-like cells that do not stain with any of the methods used. A third type of cell is found between the bases of the columnar cells. This cell typically has a large nucleus surrounded by a thin rim of cytoplasm which stains weakly with toluidine blue. These basal cells are being further investigated.

In *Pseudemys*, *Graptemys*, and *Chrysemys* the arrangement of ducts and lobules is basically

similar to that seen in *Malaclemys*. The tubules, however, are much shorter and appear more coiled. The tubule lumen is very small, and the diffuse nature of the apical membrane suggests the presence of microvilli. The epithelium is composed of two distinct cell types which occur in a ratio of roughly three to one. The more common type is a wide columnar cell without obvious granulation. It stains faintly with the PAS method, but not by any of the other methods used. The other type of cell is columnar but very thin, appearing wedged between the large pale cells. These cells are obviously granulated and the granules stain strongly with the PAS method after diastase (Fig. 9), with toluidine blue at pH 4.5, and with alcian blue at pH 3.0; they do not stain with alcian blue at low pH or with high concentrations of magnesium chloride.

Some animals were kept in media of different salinity to see if there were any obvious differences in the glands which might correlate with salinity and also to establish the tolerance to salinity of the various species used. *Malaclemys* can survive in apparent good health and with good appetite in seawater and fresh water. They have been kept in either of these media for periods of 14 months without supplemental drinking water. Weight drops off slightly during the winter months, as does the appetite, but in spring both rapidly return to the levels of the previous fall. *Pseudemys*, *Chrysemys*, and *Graptemys* were unable to survive in salinity equivalent to 50% seawater. The results of an attempt to acclimatize *Pseudemys* and *Chrysemys* slowly by slowly increasing salt concentration in the media are shown in Table I.

In gross morphology and histology there are no discernible differences in the glands of freshwater emydines kept in fresh or saline media. In *Malaclemys* however, two striking differences are seen. The alcian blue staining at low pH or with high magnesium chloride is much greater in epithelia from the lachrymal glands of animals kept in seawater. This staining of the tubule epithelium was virtually absent in animals kept in fresh water, but was very strong in the animals kept in seawater (Figs. 10 and 11). The second difference is in the wet weight of glandular tissue; in the *Malaclemys* kept in seawater it is significantly greater than in animals which were kept in fresh water (Table II).

TABLE I

Mortality studies of *Chrysemys* and *Pseudemys*

| Date | Experimental: Salt concn., g/l | Experimental: Animals dying in period | Controls: Salt concn., g/l | Controls: Animals dying in period |
|---|---|---|---|---|
| *Chrysemys* | | | | |
| June 18 | 5.0 | 0 | 1 | 0 |
| June 28 | 7.0 | 0 | 1 | 0 |
| July 5 | 10.0 | 0 | 1 | 0 |
| July 11 | 14.0 | 3 | 1 | 0 |
| July 18 | 16.0 | 2 | 1 | 0 |
| July 27 | 18.0 | 1 | 1 | 0* |
| *Pseudemys* | | | | |
| April 5 | Trace | 0 | 1 | 0 |
| April 8 | 4.5 | 0 | 1 | 0 |
| April 14 | 7.0 | 0 | 1 | 0 |
| April 19 | 9.0 | 0 | 1 | 1 |
| April 23 | 11.0 | 0 | 1 | 0 |
| April 29 | 13.0 | 0 | 1 | 0 |
| May 2 | 15.0 | 0 | 1 | 0 |
| May 7 | 17.0 | 0 | 1 | 0 |
| May 11 | 19.0 | 2 | 1 | 0 |
| May 14 | 19.0 | 4 | 1 | 0* |

* All controls survived for 1 more month.

TABLE II

Lachrymal gland weight of *Malaclemys* in fresh and salt water, expressed as wet weight percentage of body weight

| Animal | Sex | Body weight, g | Gland weight, g | Gland weight/100 g body weight |
|---|---|---|---|---|
| Fresh water | | | | |
| PTM 27 | F | 854 | 0.204 | 0.020 |
| PTM 28 | F | 756 | 0.143 | 0.018 |
| PTM 26 | M | 728 | 0.090 | 0.012 |
| PTM 47 | M | 616 | 0.100 | 0.016 |
| PTM 53 | F | 1204 | 0.156 | 0.013 |
| PTM 54 | M | 1036 | 0.161 | 0.015 |
| | | | Av. | 0.015 |
| Seawater | | | | |
| PTM 29 | M | 812 | 0.227 | 0.026 |
| PTM 31 | M | 672 | 0.141 | 0.020 |
| PTM 32 | F | 912 | 0.188 | 0.020 |
| PTM 33 | M | 896 | 0.225 | 0.025 |
| PTM 34 | F | 1064 | 0.220 | 0.020 |
| PTM 35 | M | 532 | 0.119 | 0.022 |
| PTM 38 | M | 728 | 0.177 | 0.023 |
| PTM 39 | M | 728 | 0.170 | 0.022 |
| PTM 46 | M | 470 | 0.114 | 0.026 |
| PTM 48 | F | 672 | 0.148 | 0.020 |
| PTM 51 | M | 350 | 0.069 | 0.019 |
| PTM 52 | M | 315 | 0.075 | 0.024 |
| | | | Av. | 0.022 |

## Discussion

Thus emydines possess two orbital glands. The more anterior of the two by virtue of its

anatomical position, the site of its duct openings through the deep surface of the nictitating membrane, and its histology, appears to be homologous with the Harderian gland of other reptiles (Peters 1892; Smith and Bellairs 1947). This term has been applied, in most reptiles, to the gland which opens through the deep aspect of the nictitating membrane near the anterior corner of the eye, the same position as the gland in the emydines considered here. Bojanus (1819–1821) called the gland of similar location in *Emys* the anterior lachrymal gland, while Peters (1892) in describing *Testudo* and *Chelonia* used the term the Harderian gland. That nomenclature seems now to be generally accepted.

The Harderian gland appears to be similar in the stenohaline and euryhaline emydines studied here. Its staining characteristics suggest that it is a serous gland with mucus-secreting ducts. It resembles to a degree the Harderian gland of lizards, which is also composed of serous and mucous portions; however, in that case each portion opens into the conjunctival sac through separate ducts (Schwartz-Karsten 1937). In the emydines the serous portion appears to drain through a mucous duct system, much as in the venom glands of snakes (Rosenberg 1967). In some lizards and snakes the Harderian gland drains directly into a lachrymal duct from the subbrillar space or conjunctival sac. The opening of the duct in the roof of the mouth near Jacobson's organ led to the suggestion that the secretion may be involved in chemoreception (Bellairs and Boyd 1950). This cannot be the function of the gland in emydines, where no lachrymal duct is found; the secretion is therefore lost by evaporation or, in aquatic environments, by diffusion from the conjunctival sac. In any case its large size in reptiles from a variety of environments and its apparently serous secretory nature suggest that the function of the Harderian gland is not primarily related to osmoregulation.

The more posterior of the two glands described here appears to be in a similar location to the gland which Bojanus (1819–1821) termed the external lachrymal gland. Here, also, the nomenclature of Peters (1892) has been followed, and the gland is known simply as the lachrymal gland. Peters also noted that the lachrymal gland of the marine Cheloniidae differs in size and structure from that of other chelonians studied. It was this gland which was studied by Schmidt-Nielsen and Fänge (1958) and described by Abel and Ellis (1966). Others, however, have referred to the gland responsible for hypertonic secretion in turtles as the Harderian (Dunson and Taub 1967) and the point needs further clarification.

The present study has shown that the lachrymal gland of the euryhaline *Malaclemys* differs markedly from the homologous gland in freshwater species. In the latter the lachrymal gland is very small and is apparently a mixed seromucous gland. The serous cells have been difficult to characterize by histochemical methods. The mucous cells apparently contain vicinal hydroxyl groups as shown by the PAS method. The alcian blue staining at higher pH and in low concentrations of magnesium chloride indicate that the polyanion indicated by toluidine blue metachromasia contains carboxyl but not sulfate groups.

In *Malaclemys* the tubular structure and staining characteristics are quite different. There is one predominant cell type, a tall columnar cell, separated from its neighbors by lateral intercellular spaces. The cells contain glycogen in histochemically demonstrable amounts. There are also very small amounts of a material which may be ribonucleic acid, being metachromatic at pH 4.5 but not at 2.5. The principal staining feature of the lachrymal gland of *Malaclemys* indicates the presence of a sulfated polyanion in the tubule epithelium. This is shown by strong staining with alcian blue at low pH or with 0.8 *M* magnesium chloride. The loss of toluidine blue staining after methylation–demethylation supports the idea that sulfate groups are present. Such a material is present only in the glands from *Malaclemys* kept in seawater; it is much reduced or absent in *Malaclemys* kept in fresh water, and a similar type of material was not seen in the lachrymal glands from the stenohaline species.

The function of this material is not known. If the lachrymal gland is serving an important role in osmoregulation, the polyanion might be related to transport. Such a substance has been reported in a variety of epithelia thought to be involved in active transport, for example the lachrymal gland of *Chelonia* (Abel and Ellis 1966), the rectal gland of elasmobranchs (Chan

and Phillips 1967), and the kidney of representatives of all vertebrate classes (Helmy and Hack 1967). Unfortunately no more than a correlation exists between the presence of this material and the supposed general function of the tissue. Few experimental studies have been done to investigate the suggested function of a sulfated polyanion, acting as an ion exchange resin (Abel and Ellis 1966). However, an increase in polyanion has been shown to occur in kidney tissue in states of dehydration and after the injection of antidiuretic hormone (Van Hegan 1967). In the present study I show only that a sulfated polyanion is present in large amounts in lachrymal glands from *Malaclemys* housed in hyperosmotic media. Similar material is absent in the glands from animals kept in distilled or fresh water. This indicates an alteration in the function of the gland. However, it is premature to infer a relationship between the sulfated material and salt transport.

In *Malaclemys* it must first be established that the gland's primary role is osmoregulation. Caution seems necessary in interpreting the results of Schmidt-Nielsen and Fänge (1958) and of Dunson and Taub (1967). These authors report the collection of hyperosmotic secretion from the orbital region of *Malaclemys*, but their methods were not described. In one case the collection was made by inserting a micropipette into the conjunctival sac and collecting the accumulated secretion (Dr. A. M. Taub, personal communication). Such a method could collect secretion from more than one gland. Furthermore, because of the very small multiple ducts, secretion rate would be slow and the secretion disseminated over a relatively large area. This could result in high evaporation losses and an overestimation of tonicity.

Another possible function of the gland is the maintenance of the corneal or associated epithelium. Such a role may be the primary function of the gland in emydines, as it is in many animals, and salt secretion may represent a secondary and obligatory function. In salivary glands isotonic salt secretion occurs along with the primary secretion of enzymes (Young and Schogel 1966). Much of the salt secreted by the acini is reabsorbed during the passage through the duct system, leaving a final hypotonic secretion. In hyperosmotic media, a hypotonic or even an isotonic secretion of salt would further aggravate the osmoregulatory stress. This could be combatted by shutting down glandular secretion in part or entirely, as in the renal glomeruli of reptiles subject to dehydration (Dantzler and Schmidt-Nielsen 1966). Slowing down secretion to prevent salt loss would result in accumulation of secretion, possibly explaining the increase of histochemically demonstrable sulfate. This, however, would not explain the increase in wet weight of the gland from animals kept in seawater.

In any case this paper provides a starting point, giving a detailed account of the glands present in the orbital region of the emydines. If hypertonic secretion is occurring, this study indicates that the lachrymal gland is the most likely source. The size of the gland and its histology suggest that in *Malaclemys* the lachrymal gland fulfills a function related to the salinity of the environment. Experiments are in progress in which improved methods of collection of secretion are being attempted, taking into account the anatomy of the duct system. Ultrastructural studies of the gland from animals in a variety of salinities are also being carried out.

## Acknowledgments

The author thanks Dr. V. E. Engelbert, Dr. T. S. Parsons, and Dr. D. G. Butler of the Department of Zoology, University of Toronto, for their guidance during this work. The work was supported by a National Research Council Grant A-838 to Dr. Engelbert, a National Research Council Grant A-1724 to Dr. Parsons, and an Ontario Graduate Fellowship to the author.

ABEL, J. H. and ELLIS, R. A. 1966. Histochemical and electron microscopic observations on the salt secreting lachrymal glands of marine turtles. Amer. J. Anat. **118**: 337–358.

BELLAIRS, A. D'A. and BOYD, J. D. 1950. The lachrymal apparatus in lizards and snakes. II. Proc. Zool. Soc. (London), **112**: 269–310.

BOJANUS, L. H. 1819–1821. Anatomie testudinis Europaeae. Vilnae.

CHAN, D. K. O. and PHILLIPS, J. G. 1967. The anatomy, histology and histochemistry of the rectal gland in the lipshark *Hemiscyllium plagiosum*. J. Anat. **101**: 137–157.

DANTZLER, W. H. and SCHMIDT-NIELSEN, B. 1966. Excretion in fresh water turtles (*Pseudemys scripta*) and desert tortoises (*Gopherus agassizii*). Amer. J. Physiol. **210**: 198–210.

DUNSON, W. A. and TAUB, A. M. 1967. Extrarenal salt excretion in sea snakes (*Laticauda*). Amer. J. Physiol. **213**: 975–982.

HELMY, F. M. and HACK, M. H. 1967. Comparative histochemical studies of the vertebrate kidney. Comp. Biochem. Physiol. **20**: 55–63.

LEV, R. and SPICER, S. S. 1964. Specific staining of sulphate groups with alcian blue at low pH. J. Histochem. Cytochem. **12**: 309–317.

LILLIE, R. D. 1965. Histopathologic technique and practical histochemistry. McGraw-Hill Book Co., New York.

MCMANUS, J. F. A. 1948. Histological and histochemical uses of periodic acid. Stain Technol. **23**: 99–108.

PETERS, A. 1892. Beitrag zur Kenntriss der Harder'schendrüse. Arch. Mikrosk. Anat. **36**: 192–203.

ROSENBERG, H. I. 1967. Histology, histochemistry and emptying mechanism of venom glands of some elapid snakes. J. Morphol. **123**: 133–156.

SCHMIDT-NIELSEN, K. and FÄNGE, R. 1958. Salt glands in marine reptiles. Nature, **182**: 783–785.

SCHWARTZ-KARSTEN, A. 1937. Über die Orbitaldrüsen von *Lacerta agilis, Lacerta muralis, Ophisops elegans, Tarentola mauretanica* und *Tropidonotus natrix*. Morph. Jahrb. **80**: 248–279.

SCOTT, J. E. and DORLING, J. 1965. Differential staining of acid glycosaminoglycans by alcian blue in salt solutions. Histochemie, **5**: 221–223.

SMITH, M. and BELLAIRS, A. D'A. 1947. Head glands of snakes. J. Linn. Soc. London Zool. **41**: 353–368.

VAN HEGAN, H. 1967. A method for the characterization of acid mucopolysaccharides in histological sections of renal medulla from animals in varying states of hydration. J. Physiol. **192**: 16P.

YOUNG, J. A. and SCHOGEL, E. 1966. Micropuncture investigation of sodium and potassium excretion in rat submaxillary saliva. Pflügers Arch. Gesamte Physiol. Menschen Tiere, **291**: 85–98.