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The Thyroid Gland in Bottlenose Dolphins (*Tursiops truncatus*) from the Texas Coast of the Gulf of Mexico: Normal Structure and Pathological Changes

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Summary

Fresh thyroid glands ($n = 60$) from Atlantic bottlenose dolphins that died after stranding along the Texas coast between 1991 and 2005 were examined. Organ weight ranged from 11 g in a neonate (length 109 cm) to 58 g in a large (249 cm) male. More typical weights were 25–45 g (mean = 30.6 g). Glands tended to be larger in pregnant and lactating females (mean 37.4 g; $n = 5$) than in non-pregnant animals of comparable size. In infancy, the gland tended to be compact, relatively homogeneous, and sometimes partly lobular, but with advancing age it became more lobular, the lobules being defined by fibrous bands. In one 8-year-old female (233 cm), and in a large male (295 cm) aged > 25 years the gland was represented by a cluster of lobules. Lobulation was not necessarily accompanied by increased weight, distinguishing it from hyperplasia. With age, variation in follicle size and colloid density tended to increase. Two animals (3%) had adenomas and five (8%) had discrete hyperplastic nodules, not to be confused with lobulation. Five (8%) had macroscopically identifiable colloid-filled cysts (1–4 mm in diameter). Nine animals (15%) had squamous cysts (4–15 mm) containing creamy white fluid. Other abnormalities included patchy or diffuse interstitial fibrosis (six cases, 10%) amyloidosis (two cases), thyroiditis (one case) and vasculitis (one case). No malignant neoplasms were found. Cells presumed to be C cells (light cells, parafollicular cells) were identified immunohistochemically with synaptophysin antibody.

Keywords: amyloidosis; arteritis; cetacean; dolphin; nodular hyperplasia; thyroid gland; *Tursiops truncatus*

Introduction

There is little reliable published information on the adaptive endocrine organs (thyroid, adrenal and pituitary glands) of dolphins and other cetaceans. The scant older (pre-1920) literature on the cetacean thyroid was reviewed by Arvy (1971). Cowan (1966a, b) described the thyroid glands of North Atlantic pilot whales (*Globicephala melaena* [*melas*]) and Harrison (1969) recognized colloid depletion in the cetacean thyroid. St Aubin and Geraci (1989) reported seasonal variation in the histology of the thyroid of the white whale (*Delphinapterus leucas*). Because of the important adaptive role of the thyroid gland, a number of authors studied its weight in relation to body weight (Cowan, 1966b; Harrison, 1969; Harrison and Young, 1970; Ridgway and Patton, 1971). Colloid depletion and fibrosis in harbour porpoises (*Phocoena phocoena*) was described by Schumacher *et al.* (1993).

The purpose of the present study was to describe the gross and histological features of the thyroid gland in bottlenose dolphins (*Tursiops truncatus*) from the western Gulf of Mexico.

Materials and Methods

Animals and Thyroid Specimens

Thyroid glands were recovered from carcasses of Atlantic bottlenose dolphins collected by the Texas

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Marine Mammal Stranding Network (TMMSN), under a Letter of Authorization from the National Marine Fisheries Service (NMFS), from the Texas coast between 1991 and 2005. Most animals, except for a few that died during rehabilitation, were beach-retrieved and considered to be "well preserved". The usual interval between death and collection (6–24 h) resulted, however, in a degree of autolysis. Sixty animals (13 immature males, 20 mature males, 13 immature females, and 14 mature females, maturity referring to sexual maturity as determined by examination of the gonads) were found to be suitable for inclusion in the study.

Standard total body length was measured in all 60 animals according to the procedure specified by the NMFS, from the tip of the rostrum to the notch in the flukes. Total body weights were determined in 53 animals with a Western DF-2000S Digital Weight Indicator scale (Western Scale Co., Port Coquitlam, BC, Canada). The weights of thyroids in the unfixed state were determined in 49 animals, with a Sartorius model 4800P electronic platform scale (Data Weighing Systems, Elk Grove, IL, USA) until 2001, and with a Mettler-Toledo AB54-S (Mettler-Toledo International, Columbus, OH, USA) thereafter. The ages of 40 of the animals were determined either from tooth eruption or by counting dentinal growth layer groups (GLGs; Hohn *et al.*, 1989); one GLG being taken to be equivalent to 1 year of age. For animals dying in the first year of life a fraction of a year was assigned, ranging from 0.1 year (neonates) to 0.9 (animals dying at the end of their first year).

Histology

Thyroid glands were carefully dissected away from the trachea, weighed, serially sliced at 3-mm intervals, and promptly fixed in 10% neutral-buffered formalin at ambient temperature. After routine processing, sections (5 μm) were cut and stained with either haematoxylin and eosin (HE) or haematoxylin phloxine and safranin (HPS; a trichrome stain).

Immunohistochemistry (IHC)

An effort was made to confirm the identity of presumed C cells (light cells, parafollicular cells) by calcitonin IHC, but because no calcitonin-specific antibody was found to react with dolphin C cells (Cowan and Gatalica, 2002), a monoclonal antibody (Boehringer Mannheim Biochemicals, Indianapolis, Indiana, USA), for the less specific neuroendocrine marker synaptophysin was used, diluted 1 in 40. This antibody is known to react with dolphin tissue. The methods used were those recommended by Naish (1989).

Statistical Analysis

The results were analysed (means, standard deviation [SD], and analysis of variance [ANOVA]) with MINITAB (Minitab Inc., State College, Pennsylvania, 1996).

Results

The 60 animals included in the study are listed in Table 1 in order of body length, each animal having been allocated a TMMSN field number indicating the coastal region by letters, and the sequence number within the region by digits. Table 1 contains details of age, sex and reproductive status, month of collection, body length and weight, thyroid weight, and the indices for thyroid weight (g)/body length (cm) and for thyroid weight (g)/body weight (kg).

The thyroid gland was located in the upper anterior mediastinum, in close association with the trachea, just above the level of the aortic arch. This location reflected the greatly foreshortened neck in the dolphin. Four different gross configurations were noted, as follows: two lobes joined by an isthmus (45% of animals); two separate lobes, usually of equal size, one on each side of the trachea, with no connecting isthmus (28%); a shield-like, single mass, roughly diamond-shaped, placed anteriorly (ventrally) on the trachea (20%); an irregular, multilobular grape cluster-like mass, with adjacent but separate lobules (7%). Lobulation varied, many animals showing some degree of lobulation within other configurations. The gland, which was invariably enveloped in a thin fibrous capsule, was particularly compact in younger animals, but even young animals sometimes showed a degree of lobulation. Fig. 1a shows the thyroid of a large (238 cm) female animal with the commonest configuration (see above) and also pronounced lobulation. Fig. 1b illustrates an unusual thyroid configuration, shown by a large (295 cm) male, in which the thyroid gland was closely associated with the thymus and accompanied by small lymph nodes.

Thyroid weight ($n = 49$) ranged from 11 g in a 109-cm-long neonate to 58 g in a large (249 cm) male. More typical weights ranged from 25 to 45 g (mean, 30.6 g, SD ± 9.84 g). Mean weight for 12 immature males was 25.25 g ± 7.58 g; and for eight immature females 25.87 g ± 3.27 g. Mean weights for 17 mature males was 33.88 g ± 10.79 and for 14 mature females (including pregnant and lactating animals) 34.36 g ± 8.89 g. Glands tended to be larger in pregnant or lactating females ($n = 5$, mean 37.4 g ± 5.22 g) than in non-pregnant females in the same size range ($n = 7$, mean 33.75 g ± 10.99 g).

Both body length and thyroid weight were recorded for 49 animals, for which the mean thyroid weight/

Table 1
Sex, reproductive status, and age and weight parameters of 60 bottlenose dolphins

Field no.	*Sex*	*Age (years)*	*BL (cm)*	*BLI*	*BW (kg)*	*BWI*	*Month of collection*	*Thyroid weight (g)*
GA 947	MI	0·1	109	0·1	16·4	0·67	4	11
GA 769	FI	0·1	115	—	—	—	3	—
PA 638	FI	0·9	120	0·22	56	0·46	11	26
CC 187	MI	5	120	0·17	56	0·36	6	20
SP 190	FI	0·4	138	—	44	—	8	—
GA 668	FI	0·6	145	0·17	46	0·59	10	27
PA 572	MI	0·6	150	0·15	41	0·54	8	22
PA 355	FI	—	175	0·13	51	0·43	1	22
GA 1000	FI	2·5	178	0·15	56	0·48	9	27
PA 614	FI	2	181	0·15	69	0·39	9	27
PA 375	MI	0·6	187	0·11	53	0·4	8	21
GA 1130	MI	5	189	—	79	—	2	—
PO 372	MI	—	189	0·15	78	0·37	4	29
CC 186	MI	2	190	0·1	88	0·22	3	19
PO 249	FI	—	190	0·12	62	0·35	9	22
CC 162	FI	2·5	196	0·12	162	0·15	3	24
PA 381	MI	4·5	199	0·15	117	0·25	10	29
LA 038	MI	2·5	200	0·1	87	0·24	8	21
GA 407	F	—	206	—	—	—	4	—
GA 705	MI	3·5	210	0·18	97	0·39	6	38
LA 040	MI	5·5	215	0·13	103	0·26	9	27
GA 535	FI	9	216	—	118	—	8	—
GA 484	F	6	219	0·15	—	—	2	32
PO 353	FI	—	222	—	—	—	9	—
GA 1119	M	—	224	0·15	137	0·25	10	34
GA 476	MI	6·5	226	0·14	112	0·29	12	32
GA 1122	M	—	226	0·12	55	0·49	1	27
PA 387	F	19	230	0·11	145	0·17	12	25
PA 236	F	12	230	0·1	107	0·22	2	24
PO 432	F	27	232	—	112	—	7	—
GA 1247	M	—	232	0·08	56	0·34	8	19
SP 189	FL	8	233	0·17	151	0·26	8	40
GA 1248	M	—	236	0·14	145	0·23	11	34
SP 153	F	16	237	0·11	169	0·16	2	27
GA 710	M	24	238	0·17	186	0·22	1	40
GA 1339	FP	38	238	0·12	145	0·2	10	29
GA 1148	M	—	240	0·11	160	0·17	3	27
GA 775	FL	13	240	0·15	153	0·24	3	37
PI 128	M	—	242	—	159	—	11	—
CC 110	F	15	242	0·08	139	0·14	2	19
PA 361	FP	20	243	0·16	165	0·23	2	38
GA 675	FL	—	245	0·18	218	0·2	1	43
CC 208	F	—	245	0·17	210	0·19	9	41
GA 1239	F	—	245	0·13	175	0·19	4	33
GA 1292	M	31	245	0·13	210	0·16	8	34
GA 699	F	11	245	0·21	141	0·36	4	51
SP 321	M	—	249	0·23	—	—	12	58
GA 1212	M	—	255	0·11	187	0·14	2	27
GA 740	M	13	255	0·21	159	0·33	1	53
GA 664	F	19	255	0·16	130	0·32	8	42
LA 042	M	—	256	—	218	—	2	—
GA 803	M	17	256	—	—	—	4	—
PA 397	M	14	265	—	225	—	3	—
PA 608	M	—	266	0·14	160	0·23	10	37
GA 1244	M	27	269	0·1	159	0·17	5	27
PA 292	M	13	271	0·09	—	—	4	25
PO 256	M	—	272	0·08	258	0·09	1	23
GA 1027	M	14	277	0·14	211	0·19	2	40
PO 331	M	—	294	0·16	226	0·02	3	46
PA 748	M	> 25	295	0·08	229	0·11	10	25

M, male; F, female; I, immature; P, pregnant; L, lactating; BLI, body length index; BWI, body weight index.

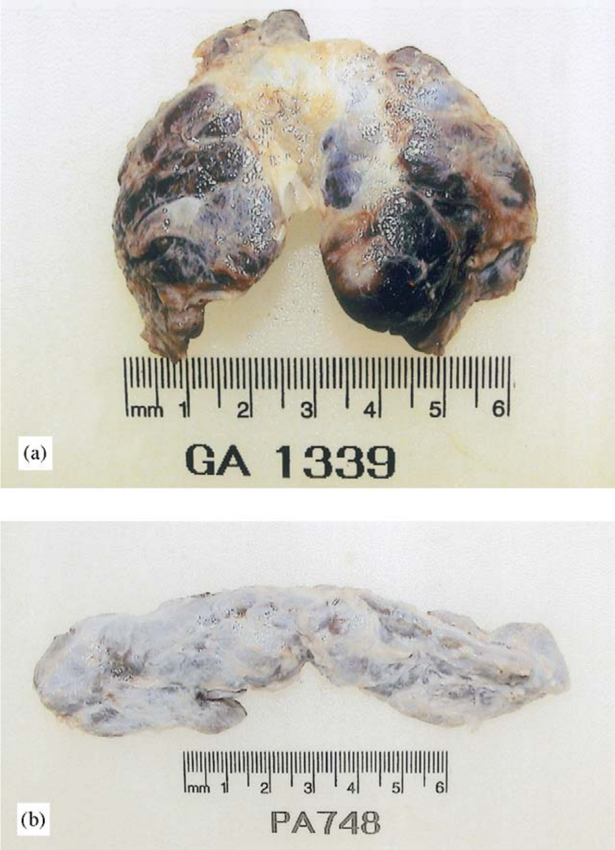

Fig. 1a,b. Gross appearance of representative thyroids. (a) Dolphin GA 1339, displaying the typical lobes plus isthmus, with lobularity. (b) Dolphin PA 748, with an elongate strap-like configuration of lobules within a more dense capsule.

body length ratio or index (BLI) was 0.14 ± 0.04. Both body weight and thyroid weight were recorded for 46 animals, for which the mean thyroid weight/body weight index (BWI) was 0.28 ± 0.14.

To determine the effect of increasing body size on the BWI and BLI, the indices (with standard deviations) of the 10 smallest and 10 largest animals were compared. The mean BWI values were 0.47 ± 0.10 and 0.20 ± 0.11, respectively, and the mean BLI values were 0.15 ± 0.03 and 0.14 ± 0.05, respectively.

In infancy, the gland was usually compact and relatively homogeneous, but with advancing age it tended to become lobular, with sub-dividing fibrous bands (Fig. 2). In one mature but not aged female (field number SP 189, GLG 8, 233 cm length) the gland was represented by a cluster of lobules, but lobulation was not accompanied by increased weight.

Histology and IHC

The histological appearance was variable, with a mixture of small and medium sized follicles and occasionally large (diameter $\geqslant$ 1 mm) follicles in the same gland. In young animals, areas predominantly occupied by small follicles were scarcely recognizable as thyroid tissue, the colloid-free follicles appearing as cell clusters rather than follicles. A distinctive feature in some glands was the folding or tufting of follicular epithelium and apparent detachment of epithelium. With increasing age, histological variability of the thyroid increased, with the occurrence of large and irregular follicles. Occasionally follicles appeared to coalesce, forming small colloid cysts.

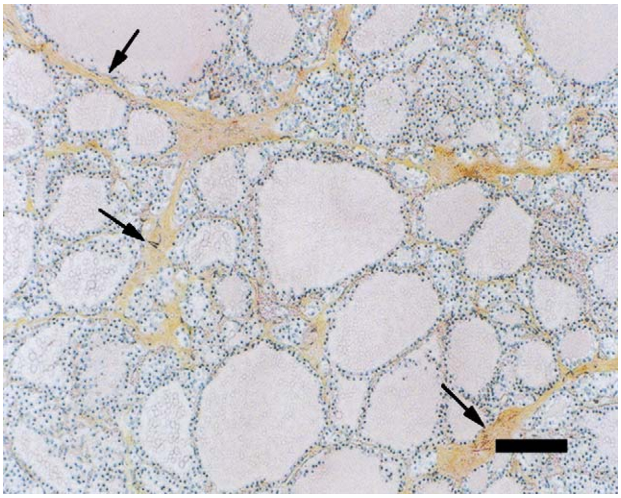

Fig. 2. Developing lobular septation. Fibrous bands staining orange (arrows) eventually join to define lobules. HPS. Bar 0.1 mm.

C cells (light cells, parafollicular cells) were identified by IHC with the synaptophysin antibody marker, which reacted strongly with cells consistent with C cells by size and location. (Fig. 3a,b) These cells were not evenly distributed throughout the gland, being heavily concentrated in some areas and almost absent from others.

With age, variability of follicle size and colloid density tended to increase. Six animals (10%) had discrete nodules diagnosable either as follicular adenoma (two) (Fig. 4) or as hyperplastic nodules (four), a separate condition from lobulation. Four (7%) had grossly identifiable colloid-filled cysts (size range 1–4 mm) (Fig. 5). Nine animals (16%) had squamous cysts (Fig. 6) (size range 4–15 mm) containing creamy white fluid material, easily recognizable on gross examination. In all the cysts the squamous lining was incomplete, or represented only by a small patch of epithelium. "Slit spaces" (cholesterol clefts) were prominent in the cyst material. In one instance, the cyst lining was squamous in part but in other areas consisted of pseudo-stratified ciliated columnar (respiratory) epithelium (Fig. 7). Several glands with squamous cysts also had separate small islands of squamous epithelium, without cyst formation. Other abnormalities included patchy or diffuse

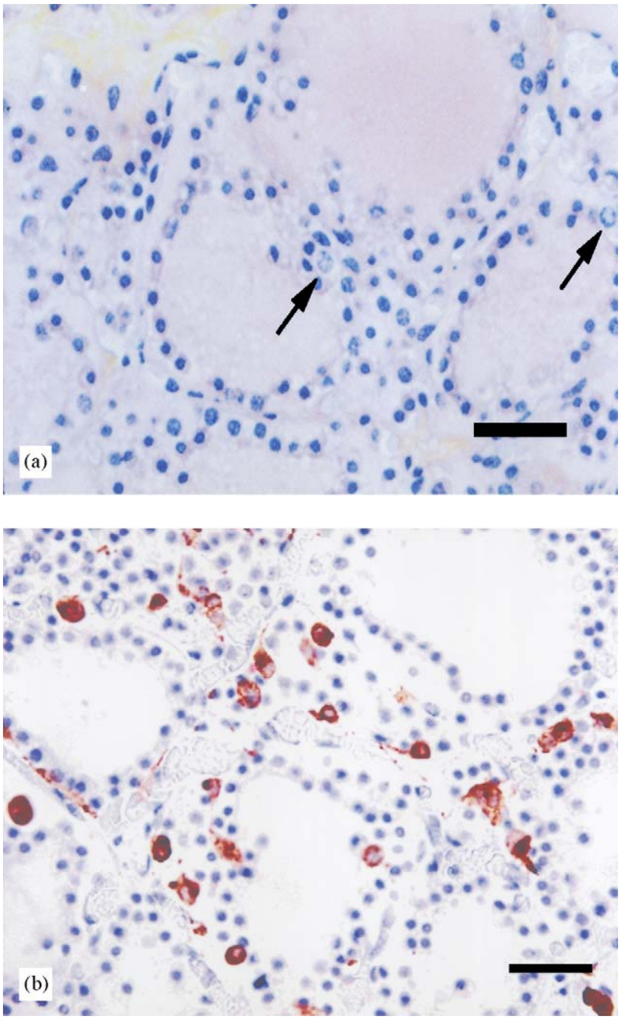

Fig. 3a,b. (a) Thyroid from an immature animal (PA 572, 6 month old male), displaying clear or C cells (arrows). HPS. Bar, 0.05 mm. (b) Adjacent section from the same gland, labelled with an antibody against synaptophysin. C cells are brown. IHC. Bar, 0.05 mm.

interstitial fibrosis (five cases, 8%) (Fig. 8), amyloidosis (two cases, 3%) (Fig. 9) thyroiditis (one case) (Fig. 10) and vasculitis (one case) (Fig. 11). No malignant neoplasms were found.

Weights of all thyroids were sorted according to the month of the year in which the animal stranded. Since as many as 9 animals stranded in August, but only one in July over the study years, rather than analyze gland weights by month, they were clustered into seasons: December–February, March–May, June–August, September–November. The mean weights and SD for the seasons were, respectively: 33.12 g ± 11.31, $n = 16$; 29.90 g ± 11.47, $n = 11$; 28.56 g ± 9.70, $n = 9$; 29.61 g ± 5.65, $n = 13$. Comparisons with the mean weight over the entire year (30.79 g ± 9.84 g) indicate little seasonal variation over the entire year (ANOVA, $P = 0.65$).

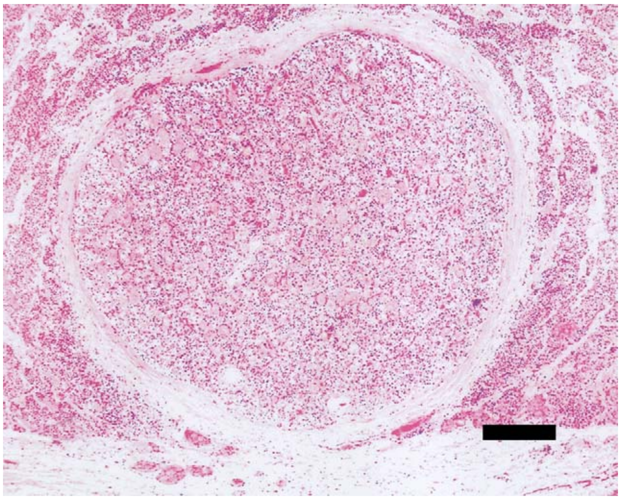

Fig. 4. Encapsulated nodule within a gland, diagnosed as a follicular adenoma. Dolphin GA 675. HPS. Bar, 0.1 mm.

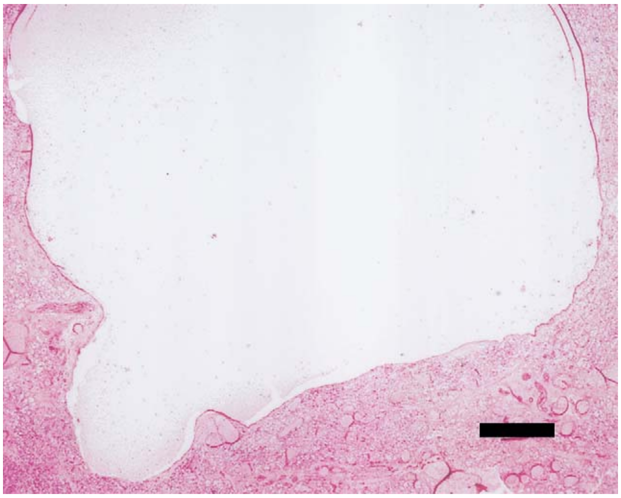

Fig. 5. A colloid cyst, the content of which is homogeneous and pale-staining. Dolphin PA 361. HPS. Bar, 0.1 mm.

Discussion

The relative size of the thyroid gland of marine mammals has been of interest for some time, as it has been suggested that adaptation to maintain body temperature in a cold or relatively cold aquatic environment includes modulation of metabolic rate and physical activity (Slijper, 1958, 1979). The finding that some marine mammals have relatively large thyroids and a high thyroid weight to body weight ratio (Slijper 1958; Harrison, 1969) is seen as supporting this suggestion.

Slijper (1958) explored the relation between organ weight and body weight in the common porpoise (harbour porpoise; *Phocoena phocoena*), expecting to find that organ weights would lag behind as body weight

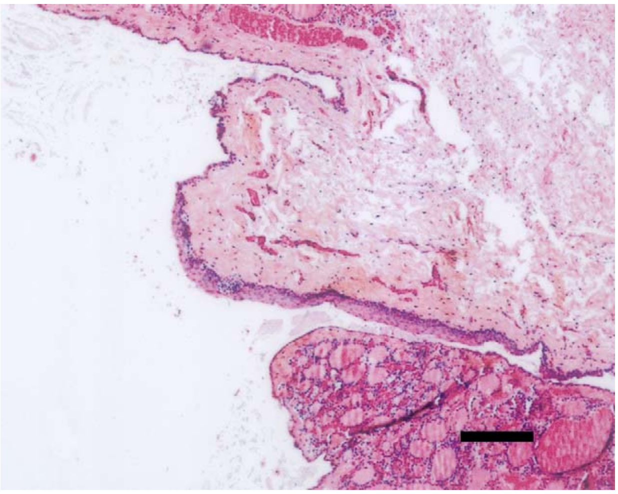

Fig. 6. Portion of a cyst lined in part by squamous epithelium. The material in these cysts tends to be pasty and white. Dolphin GA 1027. HPS. Bar, 0.1 mm.

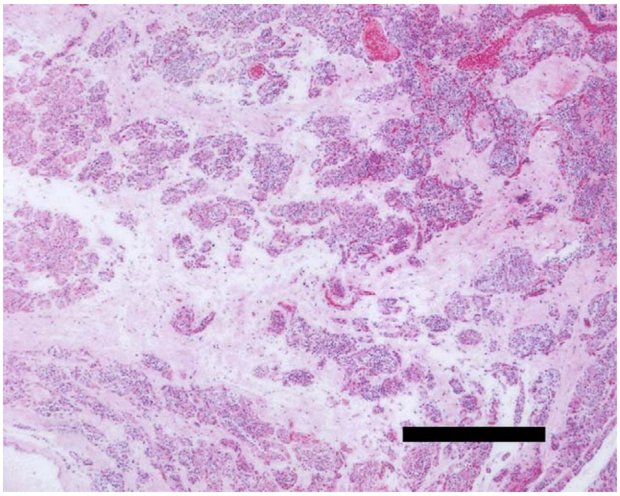

Fig. 8. Gland with diffuse fibrosis, not to be confused with the common lobular septation. Dolphin GA 675. HPS. Bar, 0.1 mm.

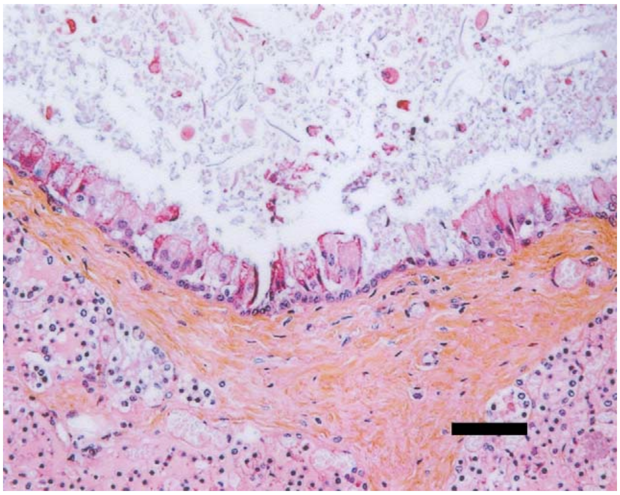

Fig. 7. Pseudostratified columnar epithelium forming part of the lining of a combined cyst. Dolphin GA 1130. HPS. Bar, 0.1 mm.

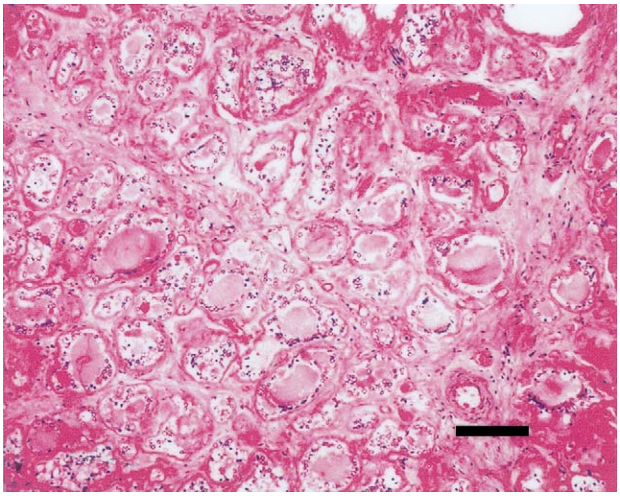

Fig. 9. Diffuse amyloidosis. Amyloid is deposited in the interstitium and around follicles. Dolphin PA 361. HPS. Bar, 0.075 mm.

increased, but finding instead that the relative weight of the thyroid and visceral organs increased as body weight increased. This was attributed to the relative decrease in weight of the blubber layer as the animal increased in size, as a result of decreasing surface area in relation to mass. The conclusion from the present study that the thyroid lags behind as the animal enlarges but retains a relatively constant proportional body length throughout life does not accord with Slijper's conclusion. However, the species and climates differ. The harbour porpoise is a petite animal by comparison with the brawny bottlenose dolphin.

The thyroid weight/body length ratio was preferable to the thyroid weight/body weight ratio as an indicator of the relationship between animal size and gland size, because many of the animals were abnormally thin or even emaciated when stranded. Differences in BLI and BWI between small and large animals may be explained in part by disproportionate weight loss in the larger animals, but also by the fact that the small animals are normally slimmer and less brawny than the adults. Length is constant, regardless of what happens to weight. Thus, as a proportion of body weight, the thyroid weight lags behind as the animal enlarges, but its relation to body length remains about the same throughout life.

In general, the bottlenose dolphin thyroid and its abnormalities are histologically similar to those of man and other terrestrial animals, making diagnosis

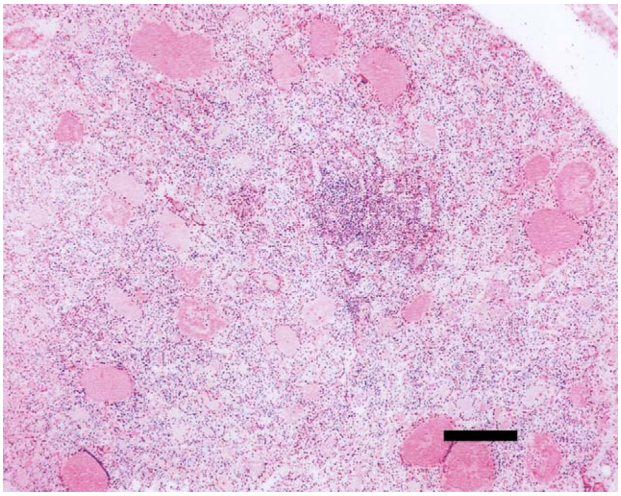

Fig. 10. Chronic thyroiditis. Diffuse infiltration by lymphocytes, with aggregates, and destruction of follicles. Dolphin PA 381. HPS. Bar, 0.1 mm.

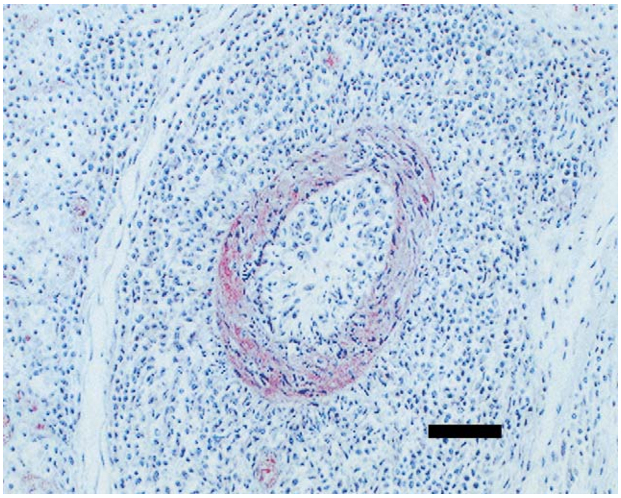

Fig. 11. Small artery cuffed by a mixed inflammatory infiltrate, with polymorphs in the vessel wall and marked intimal fibrosis. Dolphin GA 668. HPS. Bar, 0.075 mm.

relatively simple. In man, intraluminal colloid is pale-staining and homogeneous, with scalloped borders in follicles with active secretory function, and more dense and intensely eosinophilic in inactive follicles. In old age, the colloid tends to lose its homogeneous character and become broken up and granular or lumpy (LiVolsi, 1992). This also appears to be the case in the bottlenose dolphin, colloid tending to become more irregular as age advances.

The 55 thyroid glands of Newfoundland pilot whales (*G. melaena* = *melas*) reported by Cowan (1966a) displayed marked variability in size and histology, glands of similar weight being dissimilar histologically. The glands of younger animals tended to have small regular follicles with thin weakly staining colloid and large epithelial (follicular) cells, while older animals showed more variability of follicle size and colloid density. A prominent finding was large numbers of intra-follicular, variably basophilic, lamellar corpora amylacea. A number of glands contained areas of epithelium deemed to be hyperplastic. Four glands were considered to be diseased, being up to 10 times the weight of glands from animals of similar size. These abnormal glands were nodular, with many macroscopic follicles containing dense colloid, as in simple macrofollicular goitre (colloid goitre) of man. Two glands contained granulomata, consistent with a foreign body reaction to dense colloid. This suggests that disparities in weight may be disease-related. Only the beluga whales described by Mikaelian *et al.* (2003) displayed as much histological variability. It may be relevant that both species inhabit cold northern waters.

Measured thyroid weight/body weight ratios in healthy dolphins are consistently higher than those of terrestrial mammals (Harrison, 1969). Various comparisons between the mass of the cetacean thyroid gland and body mass have been made; gland-to-body weight ratios are said to be particularly high in aquatic mammals and in terrestrial mammals that live in the Arctic (Crile and Quiring, 1940). As a generality, the gland weight-to-body weight ratio for the horse, a terrestrial mammal of about the size of a white whale (*D. leucas*), is 0.015%, while in the white whale it is 0.05% (Crile and Quiring, 1940).

Ambient water temperatures in the western Gulf of Mexico range from about 12 to 14 °C in January, and from about 27 to 30 °C in August. The Newfoundland summer water temperature is similar to the Gulf of Mexico winter temperature. The BLI for Newfoundland pilot whales was 0.142 (Cowan, 1966b) while in Gulf of Mexico bottlenose dolphins in the present study it was 0.14, i.e., remarkably similar.

Harrison (1969) recognized colloid depletion in sick, captive and stranded adult dolphins, with follicles about 40 μm in diameter. In these animals some follicles had only remnants of colloid. Harrison and Young (1970) described the histology of the thyroid gland of 12 common (Pacific) dolphins, *Delphinus delphis bairdi* of various ages. The gland was described as made of small often irregular colloid-containing, basement membrane-bound follicles with a diameter of 35–125 μm, lined by cuboidal cells about 10–20 μm in height. "Light cells" were also recognized. No abnormalities were found. Harrison (1969) reported that the thyroid of newborn *T. truncatus* and *Delphinus* sp. was irregularly subdivided by relatively thick connective tissue septa, the interstices being filled by groups of differentiating follicle cells. Some small follicles of 30–100 μm diameter were present in the periphery of the gland. Epithelial cells were cuboidal and 6–10 μm in height, and the colloid was lightly stained. Suckling *Lagenorrhynchus obliquidens* had follicles in all stages of development, with the average follicle diameter ranging from 0.2 to 0.35 mm. The thyroid of suckling *Stenella* sp. was similar, but follicles were occasionally as large as 0.2 mm in diameter. The thyroid was similar in young *Globicephala scammoni* and *G. melaena = melas*; and also in *Phocoena*, *Delphinus*, *Tursiops, Stenella* and *Lagenorhynchus* spp. taken at sea, in which follicle diameters were about 0.2 mm. The cuboidal follicle cells were of similar appearance in all species cited by Harrison (1969).

Young and Harrison (1969, 1977) investigated the ultrastructure of "light cells" (parafollicular cells, clear cells, C cells) in the dolphin thyroid, and found small (up to 0.15 μm) dense, membrane-bounded granules, singly or in pairs on the follicular basement membrane, sometimes between the follicular cells, and sometimes in the intercellular space. They associated clear cells with the secretion of calcitonin. In the present study, positivity for synaptophysin was taken to indicate C cells.

It appears that local environmental influences other than temperature may have a marked effect on the dolphin thyroid. Garner *et al.* (2003) described diffuse hyperplastic goitre associated with perinatal mortality in 11 captive-born bottlenose dolphins from four different collections in the United States and Canada. Histological changes included reduced follicular diameter, markedly reduced or absent luminal colloid, hypertrophy of follicular epithelium, and follicular dysplasia. The glands were scarcely recognizable as thyroid. The aetiology was obscure but thought to be related to captivity (Garner *et al.*, 2003). The thyroids of the affected animals were quite unlike those of very young stranded animals in the present study, which invariably showed follicle formation.

Mikaelian *et al.* (2003) found adenomatous hyperplasia in the thyroids of nine of 16 belugas (*D. leucas*) from the estuary of the St Lawrence River, Quebec, collected from stranded animals between 1996 and 1998, and in six of 14 belugas from Hudson's Bay collected during subsistence hunting in 1995. The proportion of the glands affected tended to increase with age. The authors suggested environmental contaminants as a cause of the lesions. Seasonal changes in the thyroids of far-northern beluga whales were noted by St Aubin and Geraci (1989). The lesions of the belugas reported by Mikaelian *et al.* (2003) from the St Lawrence estuary are rarely seen in cetaceans and may, as suggested by the authors, have resulted from endocrine disruption by environmental contaminants.

Even in human pathology, in which large numbers of thyroid lesions have been studied over many years, it may be difficult to distinguish between nodular hyperplasia, follicular adenoma (Derwahl and Studer, 2002) and follicular carcinoma. Nodules found as isolated lesions are interpreted as adenomas, while those occurring in a diffusely nodular gland are interpreted as nodular hyperplasia. Derwahl and Studer (2002) suggest that adenoma and nodular or adenomatous hyperplasia represent opposite ends of a single spectrum, rather than discrete entities. Papillary lesions in man are taken to be carcinoma (Rosai, 1996). No such lesions were found in the dolphins reported here.

In man, the most common squamous cysts of the thyroid gland are thyroglossal duct cysts which, although developmental in origin, may be found at any age (Rosai, 1996). They are typically mid-line in the neck, an important diagnostic feature. Squamous cysts are evidently rare in animals, receiving no mention in a standard textbook on the pathology of domestic animals (Jubb *et al.*, 1993). All the dolphin cysts reported here were found lateral to the midline, usually well within the lateral masses of thyroid tissue, and are therefore best interpreted as resulting from squamous metaplasia of follicular epithelium, with subsequent cyst formation. This interpretation is supported by the finding of islands of squamous epithelium in several glands. The combined squamous/respiratory mucosal cyst suggests developmental origin, and if found in the midline would readily be diagnosed as a thyroglossal duct cyst. It is concluded that the mixed epithelial lining indicates a thyroglossal duct cyst.

The fibrosis found in the present study was not severe and no cause was identified. Whether it should be regarded as pathological or normal is not clear. Das *et al.* (2006) suggested toxic endocrine disruption as a cause of fibrosis of the thyroid in the harbour porpoise.

Amyloidosis is well recognized in the population of bottlenose dolphins in the present study, occurring in about 15–19% (Cowan, 1995). It occurs mainly in the kidneys, but also in vessels of the spleen, lung and heart, and around acini in the palatal gland, as well as in the thyroid. Its fibrillar structure on electron microscopy, staining with Congo red, and dichroism, abolished by pre-treatment with permanganate solution, all indicate that the amyloid is of the AA variety (Wright *et al.*, 1977).

The aetiology of the cases of thyroiditis and vasculitis in the present study remains obscure. The vasculitis was chronic, and in an active phase to judge by the nature of the inflammatory infiltrate. It affected several but not all small muscular arteries in the thyroid, but no other organ. This type of lesion may be autoimmune in origin (Rosai, 1996).

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References

Arvy, L. (1971). Endocrine glands and hormonal secretion in cetaceans. *Investigations in Cetacea (Berne)*, **3**, 229–300.

Cowan, D. F. (1966a). Pathology of the pilot whale. *Archives of Pathology*, **82**, 178–189.

Cowan, D. F. (1966b). Observations on the pilot whale *Globicephala melaena*: organ weight and growth. *Anatomical Record*, **155**, 623–628.

Cowan, D. F. (1995). Amyloidosis in the bottlenose dolphin, *Tursiops truncatus*. *Veterinary Pathology*, **32**, 311–314.

Cowan, D. F. and Gatalica, Z. (2002). Immunohistochemistry in Cetaceans. In: *Molecular and Cell Biology of Marine Mammals*, C.J. Pfeiffer, Ed., Kreiger Publishing Co., Malabar, FL, pp. 280–288.

Crile, G. C. and Quiring, D. P. (1940). A comparison of the energy releasing organs of the white whale and the thoroughbred horse. *Growth*, **4**, 291–298.

Das, K., Vossen, A., Tolley, K., Vikingsson, G., Thron, K., Muller, G., Baumgartner, W. and Siebert, U. (2006). Interfollicular fibrosis in the thyroid of the harbour porpoise: an endocrine disruption? *Archives of Environmental Contamination and Toxicology*, in press.

Derwahl, M. and Studer, H. (2002). Hyperplasia versus adenoma in endocrine tissues: are they different? *Trends in Endocrinology and Metabolism*, **13**, 23–28.

Garner, M. M., Schwetz, S., Ramer, J. C., Rasmussen, J. M., Petrini, K., Cowan, D. F., Raymond, J. T., Bossart, G. D. and Levine, G. A. (2003). Congenital diffuse hyperplastic goiter associated with perinatal mortality in 11 captive born bottlenose dolphins (*Tursiops truncatus*). *Journal of Zoo and Wildlife Medicine*, **33**, 350–355.

Harrison, R. J. (1969). Endocrine organs: hypophysis, thyroid and adrenal. In: *The Biology of Marine Mammals*, H.T. Andersen, Ed., Academic Press, New York, pp. 365–372.

Harrison, R. J. and Young, B. A. (1970). The thyroid gland of the common (Pacific) dolphin, *Delphinus delphis bairdi*. *Journal of Anatomy*, **106**, 243–254.

Hohn, A. A., Scott, M. D., Welles, R. S., Sweeney, J. S. and Irvine, A. B. (1989). Growth layers in teeth from known age free-ranging bottlenose dolphins. *Marine Mammal Science*, **5**, 315–342.

Jubb, K. V. F., Kennedy, P. C. and Palmer, N. C. (Eds) (1993). *Pathology of Domestic Animals*, Vol. 3, 4th Edit., Academic Press, London.

LiVolsi, V. A. (1992). Thyroid. In: *Histology for Pathologists*, S.S. Sternberg, Ed., Raven Press, New York, pp. 301–310.

Mikaelian, I., Labelle, P., Kopal, M., De Guise, S. and Martineau, D. (2003). Adenomatous hyperplasia of the thyroid gland in beluga whales (*Delphinapterus leucas*) from the St. Lawrence estuary and Hudson Bay, Quebec, Canada. *Veterinary Pathology*, **40**, 698–703.

Naish, S. J. (1989). *Immunochemical staining methods*, DAKO Corporation, Carpinteria, California, USA.

Ridgway, S. H. and Patton, G. S. (1971). Dolphin thyroid: some anatomical and physiological findings. *Zeitschrift für Vergleichenden Physiologie*, **71**, 129–141.

Rosai, J. (1996). *Thyroid gland. Surgical Pathology*, Mosby, St Louis, MO pp. 493–567.

Schumacher, U., Zahler, S., Horny, H. -P., Heidemann, G., Skirnisson, K. and Welsch, U. (1993). Histological investigations on the thyroid glands of marine mammals (*Phoca vitulina*, *Phocoena phocoena*) and the possible implications of marine pollution. *Journal of Wildlife Diseases*, **29**, 103–108.

Slijper, E. J. (1958). Organ weights and symmetry problems in porpoises and seals. *Archives Neerlandaises de Zoologie*, **13**, 97–113.

Slijper, E. J. (1979). *Whales*, 2nd Edit., English Edition, Cornell University Press, Ithaca, NY.

St Aubin, D. J. and Geraci, J. R. (1989). Seasonal variation in thyroid morphology and secretion in the white whale, *Delphinapterus leucas*. *Canadian Journal of Zoology*, **67**, 263–267.

Wright, J. R., Calkins, E. and Humphrey, R. L. (1977). Potassium permanganate reaction in amyloidosis. A histologic method to assist in differentiating forms of this disease. *Laboratory Investigation*, **36**, 274–281.

Young, B. A. and Harrison, R. J. (1969). Ultrastructure of light cells in the dolphin thyroid. *Zeitschrift für Zellforschung und Mikroskopische Anatomie*, **96**, 222–228.

Young, B. A. and Harrison, R. J. (1977). The ultrastructure of the thyroid and pars distalis of the dolphin. *Aquatic Mammals*, **5**, 1–8.

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