DOI:
10.1111/jfb.14302

Document Version
Peer reviewed version

Holocephalan (Chondrichthyes) dental plates with hypermineralized dentine as a substitute for missing teeth through developmental plasticity

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Abstract

All extant holocephalans (Chimaeroidei) have lost the ability to make individual teeth, as tooth germs are neither part of the embryonic development of the dental plates, nor of their continuous growth. Instead, a hypermineralized dentine with a unique mineral, whitlockin, is specifically distributed within a dentine framework into structures that give the dental plates their distinctive, species-specific morphology. Control of the regulation of this distribution must be cellular, with a dental epithelium initiating the first outer dentine, and via contact with ectomesenchymal tissue as the only embryonic cell type that can make dentine.

Chimaeroids have three pairs of dental plates within their mouth, two in the upper jaw and one in the lower. In the genera *Chimaera, Hydrolagus* and *Harriotta*, the morphology and distribution of this whitlockin within each dental plate differs both between different plates in the same species and between species. Whitlockin structures include ovoids, rods and tritoral pads, with substantial developmental changes between these. For example, rods appear before the ovoids, and result from a change in the surrounding trabecular dentine. In *Harriotta*, ovoids form separately from the tritoral pads, but also contribute to tritor development, while in *Chimaera* and *Hydrolagus*, tritoral pads develop from rods that later are perforated to accommodate the vasculature. Nevertheless, the position of these structures, secreted by the specialized odontoblasts (whitloblasts), appears highly regulated in all three species. These distinct morphologies are established at the aboral margin of the dental plate, with proposed involvement of the outer dentine. We observe that this outer layer forms into serially added lingual ridges, occurring on the anterior plate only. We propose that positional, structural specificity must be contained within the ectomesenchymal populations, as stem cells below the dental epithelium, and a coincidental occurrence of each lingual, serial ridge with the whitlockin structures that contribute to the wear resistant oral surface.

Keywords Chimaeroidei, dentine, dentition, Holocephali, development, whitlockin

1 INTRODUCTION

Among chondrichthyans, the extant holocephalan dentition is specialized and unique, with three pairs of continuously growing tooth plates. Compared to other chondrichthyans (sharks and rays), these extant holocephalans have lost the ability to make individual teeth (tooth agenesis), with tooth germs not being observed during embryonic development. However, teeth are present in fossil, stem holocephalans such as *Helodus* and *Cladoselache* (e.g., Stahl, 1999; Coates *et al*., 2018; Frey *et al*., 2019), and are arranged in tooth files or families, comparable to the shark and ray dentitions (Dean, 1894; Moy-Thomas, 1936; Johanson *et al*., in press). The phylogenetic position of these stem taxa supports a loss of teeth in extant holocephalans.

Tooth plates in these extant groups are entirely composed of three types of dentine, one of which is hypermineralized, forming into distinct patterns among taxa, despite the lack of teeth in development and growth. The focus of our paper is to describe the histogenesis and renewal of dentine in upper jaw dental plates of *Chimaera* and *Hydrolagus* (Family Chimaeridae), following on from a recent description in *Harriotta* (Family Rhinochimeridae;

Smith *et al*., 2019). The arrangement of the dentine types, each of a different hardness, accounts for the morphology of the worn oral surface and is specific to each taxon, once the juvenile morphology has transformed into that of the adult (Smith *et al*., 2019). Different rates of wear during feeding create raised areas and fossae that combine to make a highly adapted and functional dental surface (e.g., Figure 1c, d). All the tissues are renewed by aboral growth, possibly with epigenetic input to the late timing of extra mineralization (Smith *et al*., 2019), potentially controlled by the odontoblasts within the trabecular dentine.

Previously, we described a new type of extra hard, hypermineralized dentine (renamed whitlockin, from the generic term pleromin) in the dental plates of *Harriotta* of both adult and juvenile forms (Smith *et al.*, 2019). This whitlockin contains the magnesium-rich mineral whitlockite ((ß-Ca_3 $Mg_X(PO4)_2$)) that, during dentine mineralization towards the oral surface of the tooth plate, replaces the more typical hydroxyapatite, and becomes more compact. This hypermineralized tissue was described as tritoral dentine (reviewed in Didier, 1995; Stahl, 1999) forming the distinctive patterns of specifically ordered ovoids (beads), rods, and tritoral pads, that after wear of the surrounding softer trabecular dentine, become exposed at the oral surface as projections, representing the morphological pattern of the wear-resistant whitlockin. It was proposed that a pre-pattern of spaces in the trabecular dentine, at the growing aboral surface, preceded formation of the ovoids, rods and tritoral pads, with secretion of whitlockin into these spaces by specialized odontoblasts (whitloblasts). In establishment of this pattern, whitlockin was confined within these spaces, where a single layer of the specialized whitloblasts generate massive numbers of ramifying tubules that formed this highly mineralized dentine (Smith et al., 2019: figs. 4, 6). However, details of the formation of this dentine in development were confined to the lower dental plates; the role of the dental epithelium surrounding the plates in forming any tissues that shaped the structure of the dental plate, or the positioning and timing of the whitlockin within the dental plate, is completely unknown.

Here we focus on tissues of the upper dental plates, from a variety of chimaeroid taxa including different growth stages of *Chimera monstrosa* Linnaeus, 1758 and *Hydrolagus mirabilis* Collett, 1904, as well as a juvenile *Chimaera* sp. and a specimen of *Harriotta.* The study primarily used µCT-scanning and relative density differentiation (3µm–12µm voxels) to evaluate the morphology and distribution of whitlockin relative to trabecular dentine, and how these mineralized elements changed through growth. The tritoral tissues of all three plates of each side of the upper and lower jaws have specific intra-topographic patterns to the arrangement of the whitlockin. In terms of potential mechanisms for patterning this tissue distribution, we have observed uniquely iterative serial ridges of the outer dentine layer, only present on the epithelially covered lingual surface of the anterior upper dental plates. All trabecular dentine forms from odontoblasts at the aboral surface of the plate, within the outer dentine (forming the ridges), these developmental events are located in the cartilage furrow, nominally the location where the dental epithelium also occurs (Smith *et al.*, 2019). We also observed a co-incidence of new ridges with that of whitlockin formation and hypothesize a developmental model in which the ridges on the outer dentine layer are relevant to the pattern of rods and ovoids, whose formation is regulated at the aboral surface, the putative location of odontogenic stem cells of the pulp cavity associated with the dental plate (Smith *et al*. 2019).

2 MATERIALS AND METHODS

All specimens from UK waters were obtained from scientific trawls by Marine Scotland (*Chimaera monstrosa*, *Hydrolagus mirabilis*, *Harriotta raleighana* Goode & Bean, 1895), with all individuals arriving on deck already dead as is the case with deep water chimaeroids (Finlay Burns, Marine Scotland, *pers. comm*.). The specimen of *Chimaera* sp. is from a commercial trawl off Taiwan but further details are lacking. *Callorhinchus milii* Bory de Saint-Vincent, 1823 was obtained from Dr Catherine Boisvert (Monash University, Victoria, Australia, Monash Animal Services (MAS) ethics permit MAS/ARMI/2010/01), with animals euthanized via an overdose of tricaine in seawater (see Johanson *et al*., 2015 for further details). No surgical procedures, nor experiments, were performed.

3 RESULTS

3.1 *Chimaera monstrosa* and *Chimaera* sp.

In *Chimaera*, the close fit of the anterior and posterior upper dental plates is apparent (Figures 1c, d, 2a, 3a, c, a.dpl, p.dpl, 2b, white arrows; Supplementary Information File 1), such that when the density of whitlockin is segmented out (Figures 1d, arrow, 2c, 3c), the highly mineralized rods and ovoids of the anterior plate are in close alignment with those of the posterior, presumed to result from tight morphological constraints. One feature on the lingual surface of the anterior plate in all growth stages is the iterative, sequentially developed ridges of the outer dentine layer. In the youngest individual examined (14cm long; Figure 1c, rdg), 3–4 widely spaced ridges are present, with small oval dentine units at the midline (*) of each dental plate. Notably these are not hypermineralized tissues as are the rods found in these plates (Figure 1d, ap.rd, pp.rd) but just elevations of the outer dentine layer. Also at this growth stage, the hypermineralized rods are shorter, and do not appear to extend through the dental plate.

At the next growth stage (Figure 2), there has been substantial wear on both anterior and posterior dental plates, as indicated by the thickness of the sclerotic dentine (*sensu* Smith *et al.*, 2019; Figure 2b, g, h, sod), forming in response to wear. The hypermineralized rods of the anterior plate are longer, and extend through more of the anterior plate, while on the posterior dental plate, more rods are present when compared to the 14cm individual (Figure 2c). The number of ridges on the lingual surface of the plate has increased as well, to seven, with the most oral ridge showing wear (Figure 2a, b). These are absent from the comparable surface on the posterior dental plate (Figure 2a). The posterior end of these ridges is expanded and bulbous, particularly the most aboral ridges (Figure 2b, d, f, white asterisk), the histology of the lower jaw plates of *Harriotta* showing that this outer dentine layer does become even more mineralized (Smith *et al.*, 2019). Just below these ridges, the aboral margin of the plate is composed of an open network of trabecular dentine (Figure 2f, h, tb). In section, the ridges surround trabecular dentine (Figure 2g, h), with coincidence of early whitlockin formation of the rod and the ridge itself (Figure 2g, opposing white arrows). Sclerotic dentine fills the ridges in the same manner as it fills the spaces within the trabecular dentine closer to the oral surface (Figure 2g, h). Additionally, the ridges themselves appear to be set off from the rest of the plate, with a shallow furrow developing posteriorly (Figure 2f, arrow 1) and an elongate oral-aboral ridge anteriorly (Figure 2f, arrow 2).

The next two growth stages are represented by a subadult *C. monstrosa* and a juvenile *Chimaera* sp. (Figure 3), and in the upper dental plate of the younger of these individuals (Figure 3a–e), the hypermineralized dentine forms a series of ovoids in both the anterior and posterior dental plates (Figure 3b, ov). Along with this is a small rod located posteriorly and parasymphysially (Figure 3b). The posterior dental plate includes multiple series of ovoids, along with a small number of rods, at the anterior end. As well, there are two elongate rods, lingual and medial to the ovoids (Figure 3a, b). There are 6–7 ridges on the lingual surface of the anterior plate, although the bulbous ends are not as apparent (Figure 3c, e), and a shallow furrow and elongate ridge mark the boundaries of this region within the plate (Figure 3e, arrows 1 and 2). In section (Figure 3d), there again appears to be some spatial relationship between the series of rods and the lingual ridges, including those with developing rods that are less mineralized (grey (d) in Figure 3d; see also Figure 8). In the older individual (Figure 3f–i), the number of lingual ridges stays more or less constant. The anterior dental plate includes both a series of ovoids and of rods, but in the posterior dental plate, the rods are being modified to produce the tritoral pads, with spaces for the vasculature appearing aborally within the whitlockin tissue (Figure 3g).

3.2 *Hydrolagus mirabilis*

Anterior and posterior dental plates of a young juvenile and adult of *Hydrolagus* (Figures 4, 5) are comparable to those of *Chimaera*, with an ovoid series of whitlockin in the anterior plate and a parasymphyseal rod in the adult (Figure 5b), and a small number of extra rods in the ovoid series in the juvenile (Figure 4b). With respect to the posterior plate, in the earlier growth stage, both an ovoid series and rods are present along the labial margin of the plate, with two rods present lingually and medially. As well, a tritoral pad is developing posterior to these two rods, appearing to form via the incorporation of individual ovoids (Figure 4b, black arrowheads on left side of image, Supplementary Information files 1–3). In the adult, broad, well–developed tritoral pads are present in this position, and with respect to the more medial rods, the more posterior has become intensely vascularized to resemble the more labial pad. However, the more anterior rod shows only a few openings that represent incorporation of blood vessels within the mineralized dentine. Along the labial margin, both series of ovoids and rods are present (Figure 5b).

In both growth stages, trabecular dentine below the oral surface has been infilled with sclerotic dentine (Figures 4d, e, 5d, e), and becomes deeply worn, exposing the ovoids and tritoral pads that resist deep wear (Figures 4c, d, f, 5c, d, f). Serial ridges are present on the lingual surface of the anterior dental plate, with five thick ridges in the juvenile, comparable in morphology to those in *Chimaera* (Figure 4). However, in the adult, the ridges are more numerous but less distinct (Figure 5). Bulbous expansions at the end of these ridges are absent in both these growth stages (Figures 4c, f, 5c, f). As in *Chimaera*, there may be correspondence between the ridges and the forming ovoids (Figures 4e, 5e, double arrow). As well, trabecular dentine forms in advance of these ridges forming in both growth stages (Figures 4f, 5f) but always inside a shell of outer dentine. The posterior furrow and oral-aboral ridge are present in both growth stages although the furrow appears to be deeper in the adults (Figures 4f, 5f, arrows 1 and 2).

3.3 *Harriotta raleighana*

In this juvenile specimen (Figure 6; Smith *et al.*, 2019: fig.17a), with respect to whitlockin, the anterior dental plate has both rods and ovoids, while in the posterior plate, tritoral pads and ovoids are present (Figure 6C). In older individuals, these rods are replaced within the dental plate by ovoids, and the tritoral pads are better developed (Smith *et al.*, 2019: figs. 1D, 17). On the specimens examined, there is no indication of posterolingual rods on the posterior dental plate (Smith *et al.*, 2019: fig. 17), and instead, in the youngest individual available (Figure 6C), two short series of ovoids are present. However, these do not transform into tritoral pads (Smith *et al.*, 2019: fig. 17). One important difference between the Chimaeridae and *Harriotta* is the absence of lingual ridges on the anterior dental plates of *Harriotta* (Figure 6a, b, e). However, while these are not conspicuous on the lingual surface of the outer dentine, the distinct region of the plate associated with the ridges in the other taxa described here can be recognized and defined by the furrow and oral-aboral ridge (Figure 6e, arrows 1, 2).

3.4 Anterior versus posterior dental plates

With respect to the whitlockin in the anterior dental plate, only rods or series of ovoids are formed, with rods appearing dominant in early growth stages (e.g., Figure 2c), and ovoids later, implying a developmental change in the shape of the space, but not their location, within the trabecular dentine. Rods are retained within the anterior dental plate, normally in a parasymphysial position. By comparison, the posterior dental plate is more variable and includes broad tritoral pads, which are blocks of highly vascularized whitlockin (e.g., Figure 5b, 6a), more characteristic of adult dentitions. In earlier growth stages the position of the tritors is occupied by whitlockin rods (Figures 3b, g, 4b) that subsequently become vascularized (Figures 3g, 5b). This characterizes the more lingual, midline tritors, but in the case of labial tritors, evidence from the earlier growth stage of *Hydrolagus* (Figure 4b; Supplementary Information) indicates that the tritor is forming from the incorporation of ovoids into the tritor. Thus, it seems that the tritors develop differently in the posterior dental plates, by intrusion into whitlockin rods by blood vessels lingually, and by addition of material to surround preexisting blood vessels, labially. These dental plates show more morphological and developmental diversity than previously appreciated.

3.5 Synthesis of interpretation

In all of the extant holocephalan species studied here the hypermineralised tissue, whitlockin, whether ovoids, rods or tritoral pads, occurs in different arrangements within the dental plate, all highly regulated. As part of this regulation (Smith *et al*., 2019), spaces for the whitlockin are preformed in the trabecular dentine comprising much of the dental plate, at the aboral surface, with whitlockin deposited within the spaces by specialized odontoblasts; our new observations implicate superficial dentine ridges, also newly forming lingually at the aboral surface of the anterior dental plate, in this patterning of the rods and ovoids. These ridges are less mineralized than the whitlockin, with locations that seem to concur with the position of the newly forming rods and ovoids (Figures 2–4); a superimposed colour image (Figure 8) summarizes our interpretation of the process by which dental epithelium could induce the canonical ectomesenchymal tissue to make the whitlockin type of dentine in the correct arrangement for each species, discussed further below. Along with this, a clear ontogenetic pattern is present, with the whitlockin rods dominating the lateral parts of the plates of the younger individuals; positionally these rods are replaced by ovoid series in older individuals. As well, rods appear to be located medially in younger individuals, to be replaced in these positions by the tritoral pads.

4 Discussion

Previously Smith *et al*. (2019) showed the detailed arrangement of the specialized hypermineralized dentine forming in set spaces within the trabecular dentine, and that its mineral composition incorporated a novel mineral form, called whitlockin, in preference to the more generic term pleromin (Kemp, 1984; Ørvig, 1985; Didier *et al*., 1994; Stahl, 1999). Only the lower dentition of *Harriotta* (representing the Rhinochimaeridae) was studied but it was concluded that this whitlockin determined the specific shape of the oral surface, as it was more resistant to wear than the supporting dentine, and was regulated by renewal at the aboral surface by an unknown process.

Here we have demonstrated the different morphologies of the oral surface in the upper four plates, compared to the two lower jaw plates, sculpted by wear resistant dentine (Figure 7a–c). These differences between the plates, as well as different morphologies among the taxa, are determined by intraspecific modulation of each dental plate. These surface morphologies are dependent on the properties and arrangement of the tissues, as modified in occlusion. For example, in *Hydrolagus* upper dental plates, a marginal antero-labial ridge with hard, translucent whitlockin ovoids, set in less hard sclerotic osteodentine, surrounds a deep, extensive fossa of the less mineralized sclerotic osteodentine (double-headed arrow, Figure 7a, see also μCT scans Figure 5a–f). Notably, the occlusion of the four upper plates can be appreciated in lateral and frontal views (Figure 1a, b), where the close alignment of the anterior with the posterior plate is shown, also in occlusal views (Figures 2b, 3e, f, 4a, f). They act as one unit of the upper jaw to meet with the anterior edges of the two lower jaw plates. Also, the close alignment of the hypermineralized rods and ovoids between anterior and posterior plates adds to this functional link, as a feature suggesting common morphological control to form a morphology against which the lower plates bite (Figures 1d, 2c, 3g, 4b, 5b, 6c, lower jaw ovoid stack, 7b).

We discuss the likely control of this patterning by a dental epithelium surrounding the plate margins, of the continuous growth plates, modeled on new observations of distinctive ridged, lingual surfaces on the anterior upper dental plate (Figures 1a, 2a–h, 3a–l, 4a–f, 5a–f, 7a, 8b). During dental plate development, an outer shell forms, composed of dentine formed by odontoblasts creating two layers, surrounding the whitlockin and trabecular dentine. The outer layer forms by apposition to the inner, demonstrated by the odontoblast tubules directed inwards from cells located on the outermost surface of the dental plate (Smith *et al*., 2019: figs. 3A, 4B), while the inner layer forms from odontoblasts associated with the trabecular dentine. The outer layer, in particular, is heavily mineralized and hence translucent, revealing the structures beneath (Figure 7b, double arrow; Smith *et al.*, 2019: figs 1A, 2A, 3A, 7C). We have assumed these odontoblasts are ectomesenchymal cells that deposit the dentine layer from beneath a layer of epithelium, but currently have no supporting soft tissue histology (see Figure 7e). Nevertheless, if this is accepted as a cellular model, then control of growth at the aboral surface can be by epithelial-mesenchymal interactions that regulate the timing and positioning of new tissues.

Observations from the anterior dental plates of chimaeroids (*Hydrolagus*, Figure 7a, c, with plates *in situ*) provide a model for proposed epithelial control for the serial production of ridges on the lingual surface of the plate but also dentine produced by ectomesenchymal derivatives (see below). Notably, these ridges are not heavily mineralized (low density, missing when whitlockin structures are segmented out, e.g., Figure 2b), and form aborally in advance of the trabecular dentine spaces where rods, ovoids and tritors will develop (Figures 2g, 4e). The ridges coincide with these developing whitlockin elements on the labial surface, contributing to the antero-labial ridge (Figures 2g, 3d, 4e, 5e, 8b, nos. 0–4), as well, sclerotic dentine forms within the ridges, spreading through the plate to form the fossa of the wear surface (Figures 2g, 7d).

We also compared the distribution of the hypermineralized dentine in embryos of *Callorhinchus milii* (red, Figure 7e, ple; Kemp 1984: figs. 3B, 5) and that of the adult (Figure 7f, false colored red), with the whitlockin of rhinochimaeroids and chimaerids, forming as separate ovoids within trabecular dentine (Figure 7d, false colored red, rings denoting capsular dentine). In *Callorhinchus* (Callorhinchidae, sister group to Rhinochimaeridae + Chimaeridae) the whitlockin comprises continuous antero-posterior ridges, surrounded by trabecular dentine, continuous growth of each is aboral to form low contoured wear surfaces (Figure 7f). Control of the specific arrangement of this whitlockin must be generated in the embryo by pattern regulation of the cell layers, dental epithelium and ectomesenchyme, as described in the analysis of embryos of *Callorhinchus* (Kemp, 1984). The ectomesenchyme is canonically accepted to be necessary to produce dentine, also is derived from cranial neural crest, as demonstrated in teeth of an osteichthyan fish (Kundrat *et al.*, 2008).

The generic model based on *Callorhinchus milii* shows both an enclosing layer of epithelium of the blastema for the entire dental plate including a 'diffuse mesenchyme' at the formative, aboral side, with a 'dense 'mesenchyme' at the postero-lingual, aboral surface (ide, md, Figure 7e; from Kemp, 1984: fig. 5B). We propose a similar tissue arrangement in *Chimaera* and *Hydrolagus*, creating the serial pattern of ridges on the anterior dental plate (Figure 8b, nos. 0–4). In the youngest specimen of *Chimaera*, one symphysial and two marginal rods are present, whereas the older juvenile has four marginal plus a symphysial rod (Figures 1c, d, 2a–c), coinciding with serial addition to the lingual ridges (Figure 2g). This suggests an increase in the number of rods forming the anterior margin with regulated growth, as apparent in the growth series of *Chimera monstrosa* (Figure 8a, nos. 0–2, 8b, nos. 0–4).

Also, there appears to be a correlation of lingual ridges with new formation of ovoids (replacing the rods) in the oldest *Chimaera* dental plate (arrows, Figures 2g, 3d, 8d) with regulation by the epithelium in close contact with this surface. A coloured schematic superimposed on one half on the anterior plate (Figure 8b, nos. 0–4) presents an explanation of the relationships of ridges to internal plate structure. Although the lingual ridges appear to show some positional relationship to the developing rods and ovoids of the antero-labial margin of the plate, a distinct region forms at the symphysial margin (Figure 8a, b, rod no. 0), being separated by an oral-aboral ridge, as well, the posterior part of the plate is defined by a distinct furrow (Figures 4f, 5f, arrows 1 and 2).

It is important to note that although there is a potential developmental relationship between the lingual ridges and both the developing trabecular dentine and hypermineralized whitlockin (lime green, orange, Figure 8b), the ridges have formed in advance of the mineralized ovoids, as well as the patterned spaces within the trabecular dentine (Figure 4d–f). And, the ridges are absent from the posterior dental plate in the three taxa examined here, despite possessing substantially organized whitlockin, while the ridges appear to become less distinct with growth although whitlockin deposition is ongoing (Figure 5d, e). Moreover, these ridges are absent from *Harriotta*, whose anterior upper dental plate is also characterized by a series of patterned rods and ovoids, while the lingual face where these ridges occur in *Chimaera* and *Hydrolagus* can also be identified in *Harriotta* (Figure 6e, arrows 1 and 2).

5 Conclusions

Individual teeth are present in stem group holocephalans such as *Helodus* (e.g., Stahl, 1999), arranged in tooth families comparable to other chondrichthyans such as sharks and rays. The dentition of *Helodus* also presents evidence for tooth fusion (Moy-Thomas, 1936; Patterson, 1964; Stahl, 1999; Johanson et al. in press), making it an ideal intermediate in the evolutionary transition from stem- to crown group holocephalans, with complete tooth fusion in the latter (e.g., Ørvig, 1957). Previously, researchers have attempted to identify evidence for this tooth fusion in extant taxa, such as the recognition of discrete and stacked oral and aboral territories in *Callorhinchus*, said to be equivalent to the separate teeth of shark and ray tooth families (Didier *et al*., 1994). Separate growth phases were also described in *Harriotta* (Smith et al. 2019: fig. 7A, C), although this was not thought to represent the development of individual teeth. Developmentally, the tooth plates in crown group holocephalans have been demonstrated to be initiated in a similar manner to sharks and rays (e.g., Martin *et al.* 2016; Rasch *et al*. 2016), with an infolding of epithelial tissue and corresponding aggregation of mesenchymal cells (Didier *et al.*, 1994). The tooth plate formed from this single primordium (Schauinsland, 1903; Kemp, 1984; Didier, 1995), but separate tooth germs were not observed as part of the embryonic development of the dental plates, nor of their continuous growth. Despite this loss, significant patterning of dental elements, related to highly functional dentitions, occurs in all extant taxa. How this patterning is regulated is unknown, but may be correlated with development of outer and inner dentine tissue, controlled by a dental epithelium and dentally committed ectomesenchyme.

Acknowledgements

We would like to thank Marine Scotland for holocephalan specimens used in this study, as well as all those who generously contributed money for project costs through the crowdfunding site *Experiment*. Catherine Boisvert and Peter Currie (Australian Regenerative Medicine Institute, Monash University, Melbourne, Australia) are thanked for providing a specimen of *Callorhinchus milii*.

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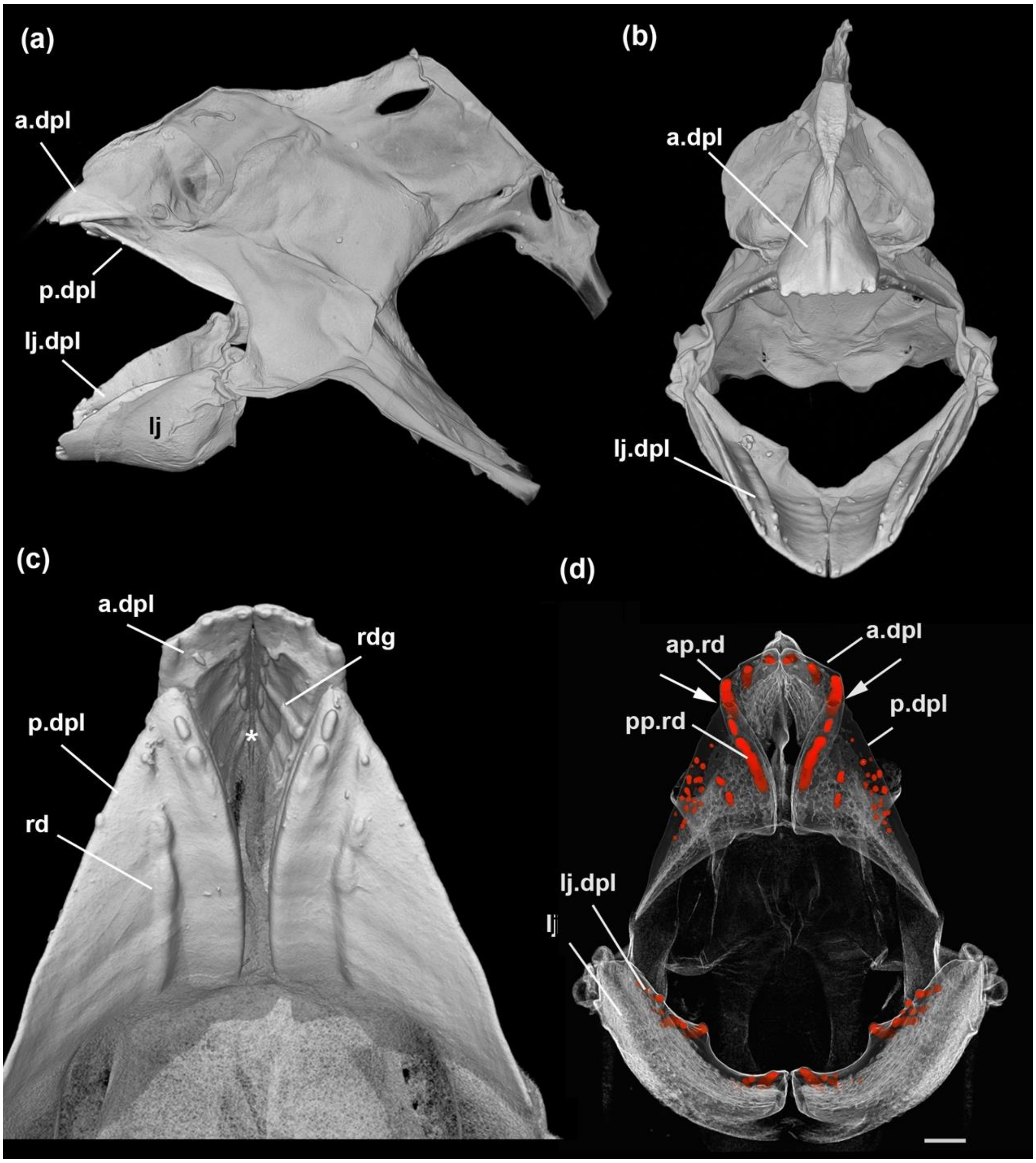

FIGURE 1 *Chimaera monstrosa* Linnaeus, 1758, 14 cm juvenile, µCT-scans. Skull, jaws and dentition in (a) lateral view, (b) anterior view, (c) upper dentition including anterior and posterior dental plates in oral view, (d) upper and lower dentitions in antero-oral view, rendered (Drishti) to show mineralized tissue of the dental plates (false colored red). a.dpl, anterior dental plate; ap.rd, rods of hypermineralized dentine (whitlockin) in the anterior dental plate, lj, lower jaw; lj.dpl, lower jaw dental plate; p.dpl, posterior dental plate; pp.rd, rods of hypermineralized dentine (whitlockin) in the

posterior dental plate, rdg, ridges on lingual surface of the anterior dental plate. White arrows in (d) indicate position of close contact between the anterior and posterior dental plates, scale bar=1mm. White asterisk in (c) indicates small oval dentine units at the midline of each dental plate.

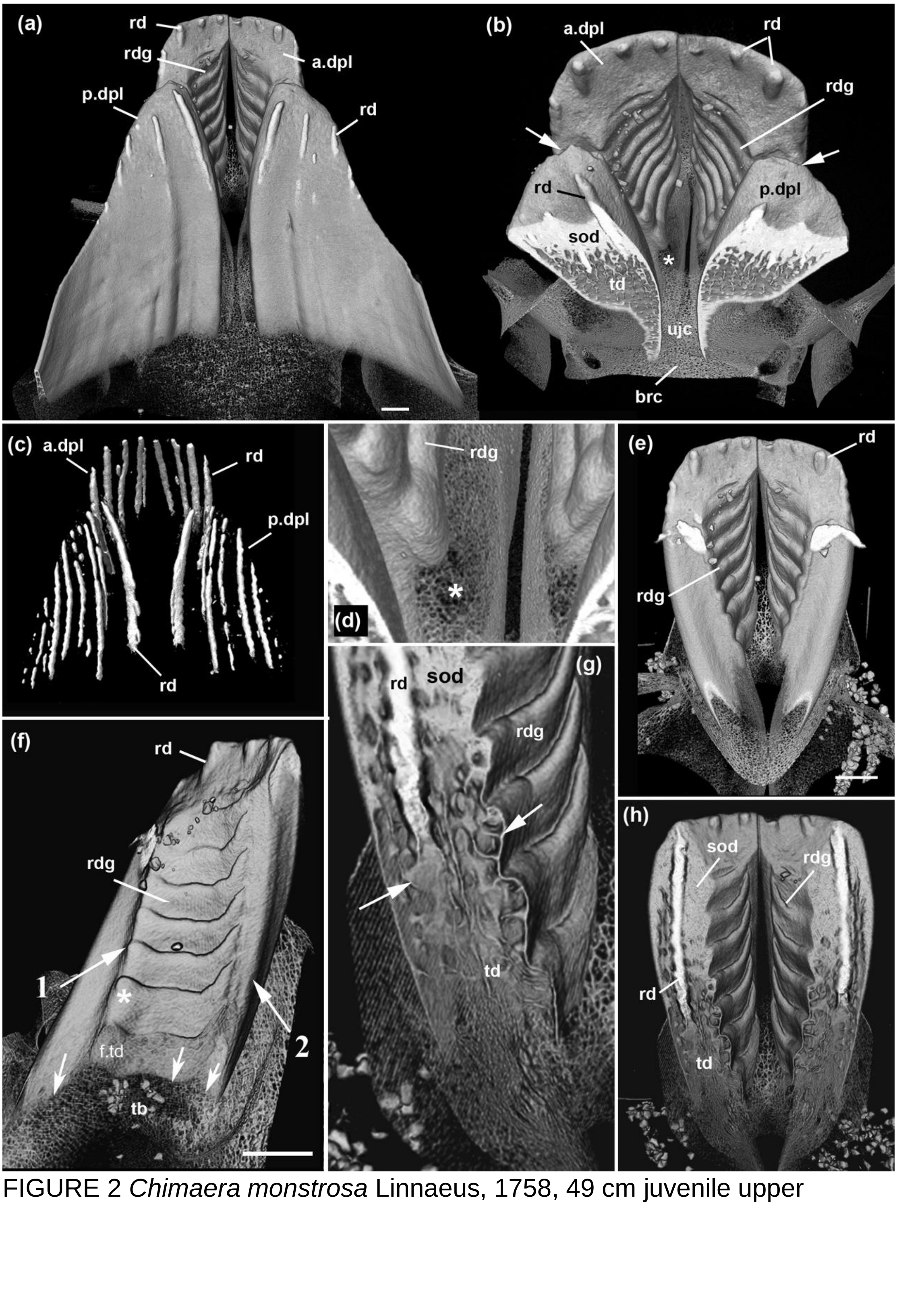

FIGURE 2 *Chimaera monstrosa* Linnaeus, 1758, 49 cm juvenile upper

dentition, µCT-scans. In (a) oral view, including anterior dental plate with ridges on the lingual surface, (b) upper dentition in oral view, with posterior dental plate cut away to show internal structure, and ridges on the anterior dental plate, asterisk indicates region shown in (d), white arrows indicate close correspondence between anterior and posterior dental plates, (c) upper dental plates rendered (Avizo) showing rods of hypermineralized dentine (whitlockin) in both, (d) closeup of aboral anterior dental plate, showing newest, bulbous postero-lingual end of the ridge, asterisk indicates newly developing trabecular dentine below the most aboral ridge, (e) anterior dental plate, with posterior plate almost completely cut away to show lingual ridges relative to cartilage of the upper jaw, (f) symphyseal view of anterior dental plate, with opposing plate almost completely cut away to show the ridges, asterisk indicates bulbous aboral new ridge tissue, also three separate growth regions (white arrows indicate direction of growth), anterior ridge, or column, and posterior furrow on each side of the ridged zone (arrows labelled 1 and 2), (g, h) anterior dental plate, virtual section through developing rod, framework trabecular dentine, ridges in section with forming sclerotic dentine inside, (g) arrows indicate correspondence between the ridge and developing rod. Abbreviations as in Figure 1, also brc, cartilage of the braincase; f.td, forming trabecular dentine; rd, whitlockin rods; sod, sclerotic osteodentine, ujc, upper jaw cartilage. Scale bars, (a)=1.5cm, (e, f)= 1mm.

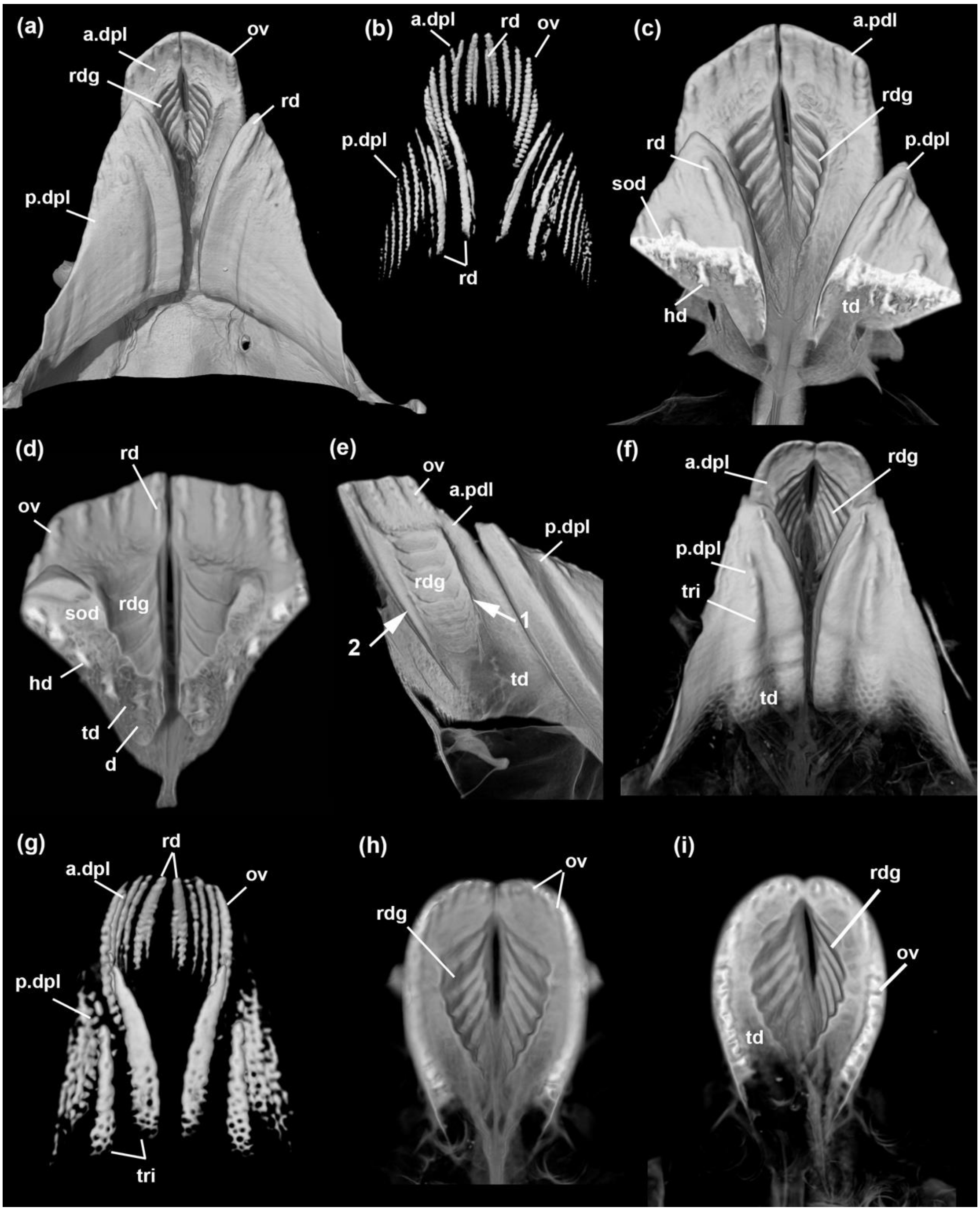

FIGURE 3 *Chimaera* spp., upper dentition, µCT-scans, (a-e) subadult *Chimaera monstrosa* Linnaeus, 1758, (f-i) juvenile (20cm) *Chimaera* sp. (unidentified), Taiwan. (a) *C. monstrosa*, (f) *Chimaera* sp., upper dentition in oral view, including anterior dental plate with ridges on the lingual surface of the dental plate, (b) *C. monstrosa*, (g) *Chimaera* sp., upper dentition rendered (Avizo) showing rods and ovoids of hypermineralized dentine (whitlockin) in the both dental plates, and tritoral pads in the posterior plate (g), (c) *C.*

monstrosa, upper dentition in oral view, with posterior dental plate cut away to show internal structure, including ridges on the anterior dental plate, (d) *C. monstrosa*, (i) *Chimaera* sp., anterior dental plate, cut away to show section through developing ovoids (hypermineralized whitlockin), surrounding trabecular dentine, sclerotic (hypermineralized) trabecular dentine and the ridges, in (d) the less mineralized ovoids can be seen at the aboral surface, (e) *C. monstrosa*, anterior and posterior dental plates, with opposing plates cut away to show symphyseal face of the anterior plate and the ridges, anterior ridge, or column, and posterior furrow on each side of the ridged zone (arrows labelled 1 and 2, as in Figure 2f), (h) *Chimaera* sp., anterior plate in oral view, with posterior dental plate cut away to show ridges on the anterior dental plate. Abbreviations as in previous Figures, also d, less mineralized dentine; tri, tritoral pad.

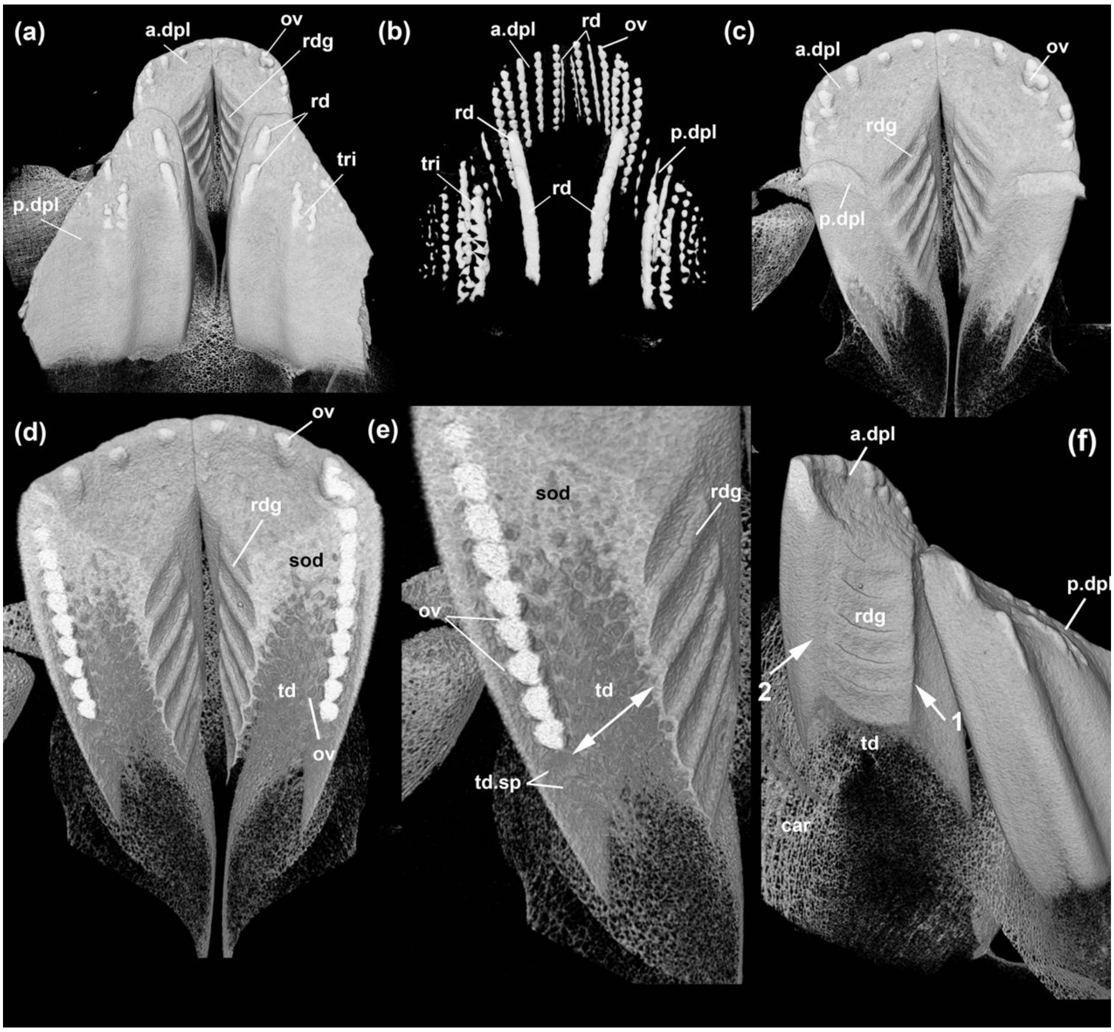

FIGURE 4 *Hydrolagus mirabilis* Collet, 1904, µCT-scans. (a) upper dentition in oral view, including anterior dental plate with distinct ridges on the lingual surface of the dental plate, (b) upper dentition rendered (Avizo) showing ovoids and rods of hypermineralized dentine (whitlockin) in both dental plates, black arrowheads on developing tritoral pad on the left side of the image showing ovoids being added to form the pad, (c) upper dentition in oral view, with posterior dental plate cut away to show ridges on the anterior dental plate, (d, e) anterior dental plate, cut away to show section through developing ovoids (hypermineralized), surrounding trabecular dentine, sclerotic (hypermineralized) trabecular dentine and the ridges, (f) anterior and posterior dental plates in symphyseal view, with opposing plates cut away to

show symphyseal face of the anterior plate and the ridges, numbered arrows different regions of the plate (as in Figures 2f, 3e). Abbreviations as in previous Figures.

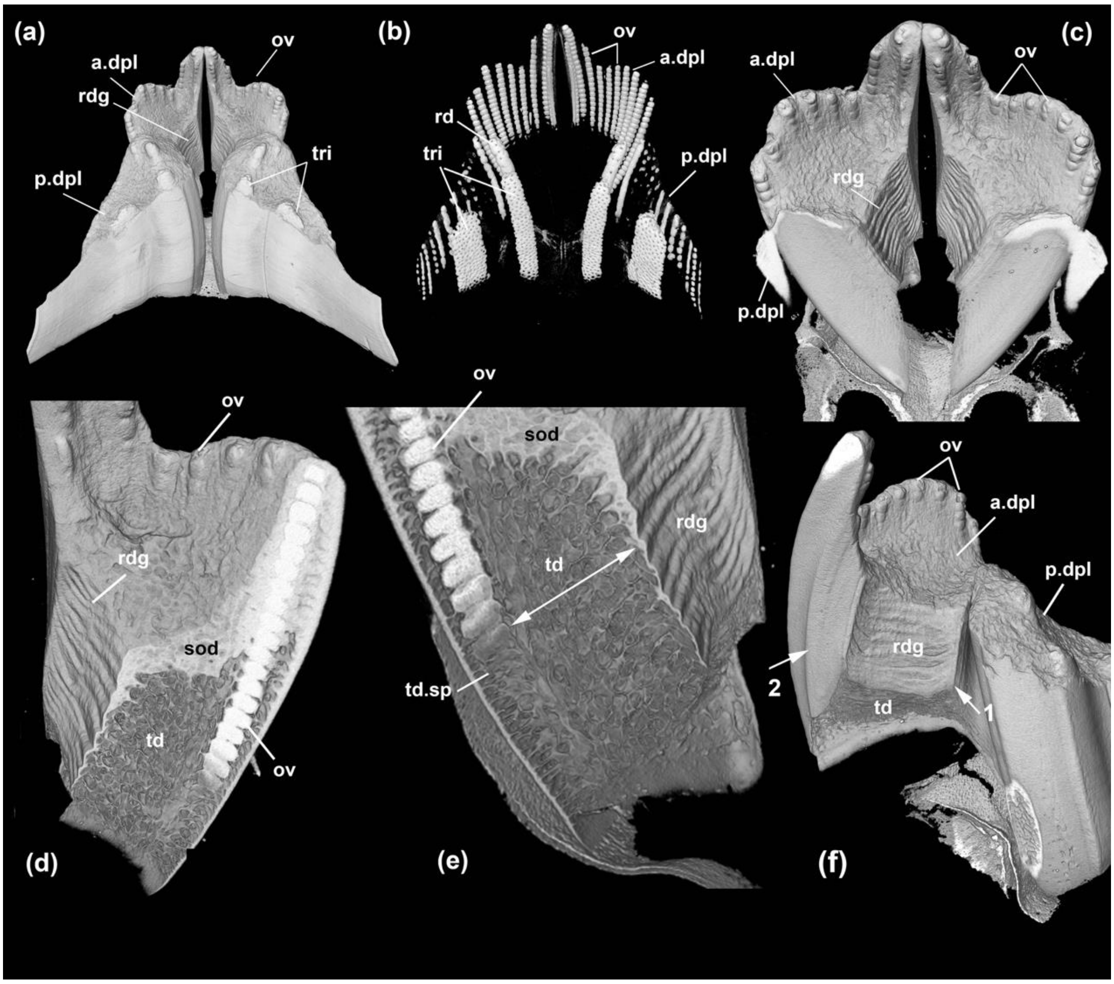

FIGURE 5 *Hydrolagus mirabilis* Collet, 1904, µCT-scans, (a) upper dentition in oral view, including anterior dental plate with ridges on the lingual surface of the dental plate, (b) upper dentition rendered (Avizo) showing ovoids and rods of hypermineralized dentine in the both dental plates, and tritoral pads on the posterior plate, (c) in oral view, with posterior dental plate cut away to show posterior face of the anterior dental plate and lingual ridges, (d, e) anterior dental plate, cut away to show section through developing ovoids (hypermineralized), surrounding trabecular dentine, sclerotic

(hypermineralized) trabecular dentine and the ridges, double-headed arrow (e) indicating correspondence of developing ovoids and lingual ridges, (f) anterior and posterior dental plates in symphyseal view, with opposing plates cut away to show symphyseal face of the anterior plate and the ridges, numbered arrows mark different regions of the plate as in Figures 2f, 3e, 4f. Abbreviations as in previous Figures.

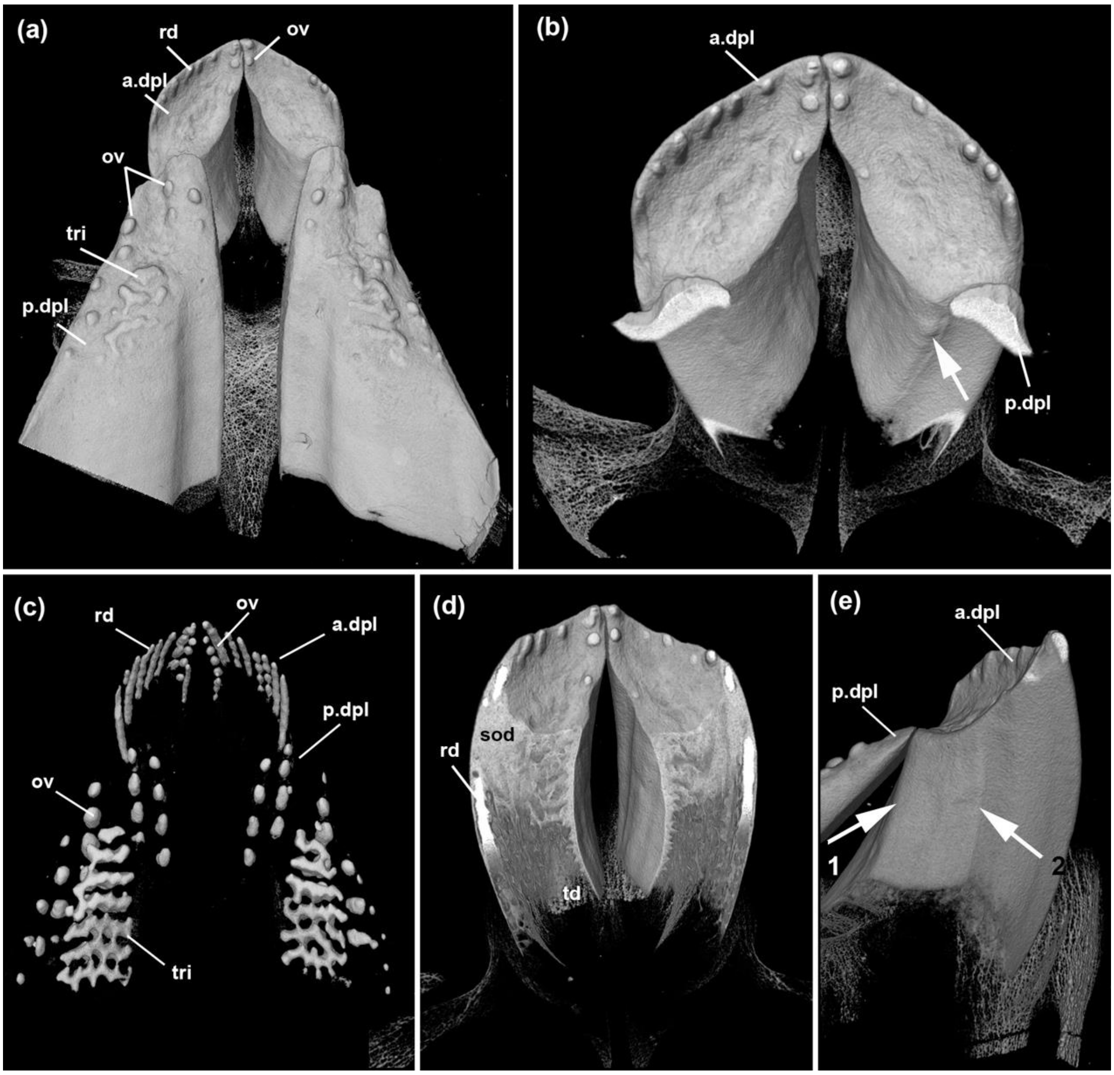

FIGURE 6 *Harriotta raleaghana* µCT-scans, (a-e) upper dentition in oral view, (a) anterior and posterior dental plate showing close fit between them and tritoral pad on posterior plate, (b) anterior dental plate with posterior dental plate cut away to show posterior and lingual faces of the anterior plate,

lacking prominent ridges on the lingual surface of the dental plate, but with some bulbous expansion that was associated with ridges in *Chimaera* (Figure 2), indicated by white arrow, (c) four upper dental plates rendered (Avizo) showing ovoids and rods (whitlockin) as extensive below the worn surface, and developing tritoral pads on the posterior plate, ovoids in symphyseal row with antero-labial set of rods in anterior plate, (d) anterior dental plate, virtual section through developing rod (whitlockin), surrounding trabecular dentine, sclerotic dentine near the wear surface, trabecular dentine only at forming aboral surface, (e) symphyseal surface with opposing left plates cut away, showing anterior dental plate (arrow 2 marks antero-symphyseal portion) and posterior in close alignment at oral surface (arrow 1). Abbreviations as in previous Figures.

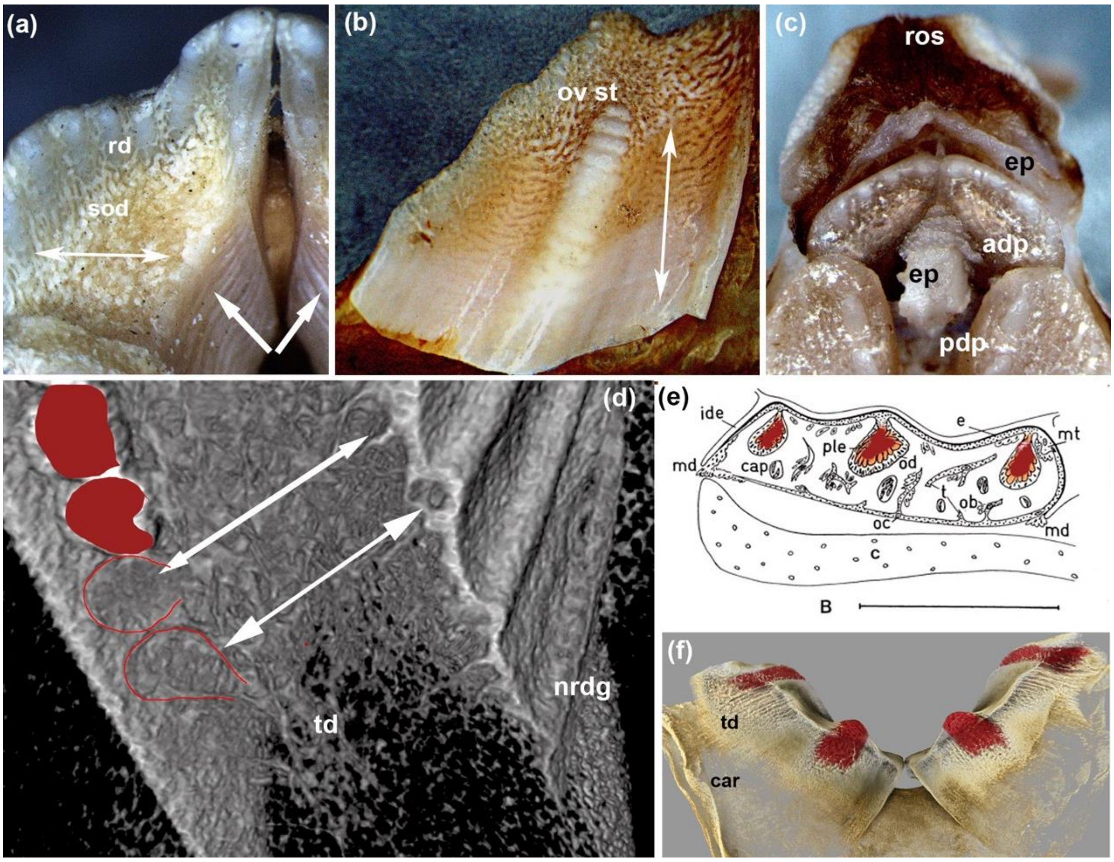

FIGURE 7 *Hydrolagus mirabilis*, (a-c) photomicrographs in incident light of

upper and lower dental plates, (a) anterior upper plate shows ridges on lingual surface (arrows), extent of worn tissue fossa (double arrow), translucent grey rods, (b) lower dental plate tissue, transparency of most mineralized tissues as in outer dentine (white double arrow) surrounding trabecular dentine, ovoid stack not fully mineralized, (c) upper dentition *in situ* with soft tissue, plates lined by epithelium, rostral snout present, (d) close up of virtual section through anterior upper plate as in Figure 4d, e, colored false red to show whitlockin as mineralized ovoids, and red rings the pre-formed capsular spaces in the trabecular dentine, newest ridge forming aborally, coincidence of ridges with new whitlockin forming (double arrows), (e) *Callorhinchus milii* Bory de Saint-Vincent, 1823, schematic drawing of section through lower jaw embryonic plate (Kemp 1984), false coloured red indicates whitlockin formation (ple = pleromin) under an epithelium, with modified trabecular dentine (mt), condensed mesenchyme (md), cartilage (c), scale bar=1mm, (f) *Callorhinchus milii*, adult lower jaw, µCT rendered with whitlockin segmented, false coloured red (Drishti), showing low ridges on the surface that form deep into the trabecular dentine, above a cartilage furrow. Abbreviations as in previous Figures, also car, cartilage furrow, ep, epithelium; nrdg, newest ridge; ov st, ovoid stack; ros, rostral snout.

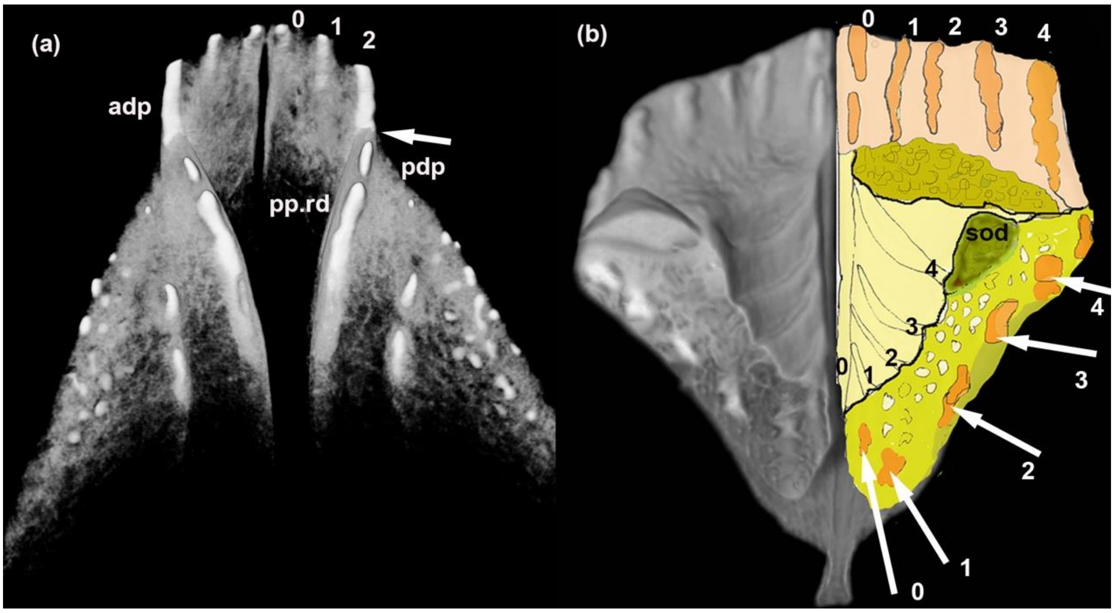

FIGURE 8 *Chimaera monstrosa*, µCT-scans, sub-adult upper dentitions, (a) from Figure 1, (b) from Figure 3a-e, with the rod series explained as whitlockin formed to an timed order at the aboral surface in the trabecular dentine, determined by a dental epithelium, where on the lingual side, ridges of the outer dentine layer form (Figure 8b, also Figures 1a, 2b, 3 d-e). (a) partially segmented upper dentition (Avizo) showing three rods of the anterior plate, no. 2 aligns with the rods of the posterior plate), no. 0 forms in the symphyseal segment of the plate, 1, 2 along the labial ridge, (b) anterior dental plate with five rods and a coloured overlay to explain the co-incidence of ridges with rods (0-4), tissues seen in virtual section relative to the surface anatomy of the oral and lingual surfaces, whitlockin, orange; lime green, trabecular dentine; olive green, sclerotic osteodentine dentine; moss green, wear surface of the sclerotic dentine, arrowed in figure 7(a). Abbreviations as in previous figures.

Supplementary information

Hydrolagus mirabilis upper dental plates. Supplementary Information File 1, movie of upper dental plates, Supplementary Information Files 2, 3 rotatable .stl files of mineralized whitlockite components within the posterior dental plates. Available on request from the Zerina Johanson (NHM, z.johanson@nhm.ac.uk).