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

4

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25

26 **Abstract**

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

36 Chimaeroids have three pairs of dental plates within their mouth, two in
37 the upper jaw and one in the lower. In the genera *Chimaera*, *Hydrolagus* and
38 *Harriotta*, the morphology and distribution of this whitlockin within each dental
39 plate differs both between different plates in the same species and between
40 species. Whitlockin structures include ovoids, rods and tritoral pads, with
41 substantial developmental changes between these. For example, rods appear
42 before the ovoids, and result from a change in the surrounding trabecular
43 dentine. In *Harriotta*, ovoids form separately from the tritoral pads, but also
44 contribute to tritor development, while in *Chimaera* and *Hydrolagus*, tritoral
45 pads develop from rods that later are perforated to accommodate the
46 vasculature. Nevertheless, the position of these structures, secreted by the
47 specialized odontoblasts (whitloblasts), appears highly regulated in all three
48 species. These distinct morphologies are established at the aboral margin of
49 the dental plate, with proposed involvement of the outer dentine. We observe
50 that this outer layer forms into serially added lingual ridges, occurring on the

51 anterior plate only. We propose that positional, structural specificity must be
52 contained within the ectomesenchymal populations, as stem cells below the
53 dental epithelium, and a coincidental occurrence of each lingual, serial ridge
54 with the whitlockin structures that contribute to the wear resistant oral surface.

55

56 **Keywords Chimaeroidei, dentine, dentition, Holocephali, development,**
57 **whitlockin**

58

59 **1 INTRODUCTION**

60 Among chondrichthyans, the extant holocephalan dentition is specialized and
61 unique, with three pairs of continuously growing tooth plates. Compared to
62 other chondrichthyans (sharks and rays), these extant holocephalans have
63 lost the ability to make individual teeth (tooth agenesis), with tooth germs not
64 being observed during embryonic development. However, teeth are present in
65 fossil, stem holocephalans such as *Helodus* and *Cladoselache* (e.g., Stahl,
66 1999; Coates *et al.*, 2018; Frey *et al.*, 2019), and are arranged in tooth files or
67 families, comparable to the shark and ray dentitions (Dean, 1894; Moy-
68 Thomas, 1936; Johanson *et al.*, in press). The phylogenetic position of these
69 stem taxa supports a loss of teeth in extant holocephalans.

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

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

85 Previously, we described a new type of extra hard, hypermineralized
86 dentine (renamed whitlockin, from the generic term pleromin) in the dental
87 plates of *Harriotta* of both adult and juvenile forms (Smith *et al.*, 2019). This
88 whitlockin contains the magnesium-rich mineral whitlockite ($(\beta\text{-Ca}_3$
89 $\text{Mg}_x(\text{PO}_4)_2$) that, during dentine mineralization towards the oral surface of
90 the tooth plate, replaces the more typical hydroxyapatite, and becomes more
91 compact. This hypermineralized tissue was described as tritoral dentine
92 (reviewed in Didier, 1995; Stahl, 1999) forming the distinctive patterns of
93 specifically ordered ovoids (beads), rods, and tritoral pads, that after wear of
94 the surrounding softer trabecular dentine, become exposed at the oral surface
95 as projections, representing the morphological pattern of the wear-resistant
96 whitlockin. It was proposed that a pre-pattern of spaces in the trabecular
97 dentine, at the growing aboral surface, preceded formation of the ovoids, rods
98 and tritoral pads, with secretion of whitlockin into these spaces by specialized
99 odontoblasts (whitloblasts). In establishment of this pattern, whitlockin was
100 confined within these spaces, where a single layer of the specialized

101 whitoblasts generate massive numbers of ramifying tubules that formed this
102 highly mineralized dentine (Smith et al., 2019: figs. 4, 6). However, details of
103 the formation of this dentine in development were confined to the lower dental
104 plates; the role of the dental epithelium surrounding the plates in forming any
105 tissues that shaped the structure of the dental plate, or the positioning and
106 timing of the whitlockin within the dental plate, is completely unknown.

107 Here we focus on tissues of the upper dental plates, from a variety of
108 chimaeroid taxa including different growth stages of *Chimera monstrosa*
109 Linnaeus, 1758 and *Hydrolagus mirabilis* Collett, 1904, as well as a juvenile
110 *Chimaera* sp. and a specimen of *Harriotta*. The study primarily used μ CT-
111 scanning and relative density differentiation ($3\mu\text{m}$ – $12\mu\text{m}$ voxels) to evaluate
112 the morphology and distribution of whitlockin relative to trabecular dentine,
113 and how these mineralized elements changed through growth. The tritoral
114 tissues of all three plates of each side of the upper and lower jaws have
115 specific intra-topographic patterns to the arrangement of the whitlockin. In
116 terms of potential mechanisms for patterning this tissue distribution, we have
117 observed uniquely iterative serial ridges of the outer dentine layer, only
118 present on the epithelially covered lingual surface of the anterior upper dental
119 plates. All trabecular dentine forms from odontoblasts at the aboral surface of
120 the plate, within the outer dentine (forming the ridges), these developmental
121 events are located in the cartilage furrow, nominally the location where the
122 dental epithelium also occurs (Smith *et al.*, 2019). We also observed a co-
123 incidence of new ridges with that of whitlockin formation and hypothesize a
124 developmental model in which the ridges on the outer dentine layer are
125 relevant to the pattern of rods and ovoids, whose formation is regulated at the

126 aboral surface, the putative location of odontogenic stem cells of the pulp
127 cavity associated with the dental plate (Smith *et al.* 2019).

128

129 **2 MATERIALS AND METHODS**

130 All specimens from UK waters were obtained from scientific trawls by Marine
131 Scotland (*Chimaera monstrosa*, *Hydrolagus mirabilis*, *Harriotta raleighana*
132 Goode & Bean, 1895), with all individuals arriving on deck already dead as is
133 the case with deep water chimaeroids (Finlay Burns, Marine Scotland, *pers.*
134 *comm.*). The specimen of *Chimaera* sp. is from a commercial trawl off Taiwan
135 but further details are lacking. *Callorhinchus milii* Bory de Saint-Vincent,
136 1823 was obtained from Dr Catherine Boisvert (Monash University, Victoria,
137 Australia, Monash Animal Services (MAS) ethics permit MAS/ARMI/2010/01),
138 with animals euthanized via an overdose of tricaine in seawater (see
139 Johanson *et al.*, 2015 for further details). No surgical procedures, nor
140 experiments, were performed.

141

142 **3 RESULTS**

143 **3.1 *Chimaera monstrosa* and *Chimaera* sp.**

144 In *Chimaera*, the close fit of the anterior and posterior upper dental plates is
145 apparent (Figures 1c, d, 2a, 3a, c, a.dpl, p.dpl, 2b, white arrows;
146 Supplementary Information File 1), such that when the density of whitlockin is
147 segmented out (Figures 1d, arrow, 2c, 3c), the highly mineralized rods and
148 ovoids of the anterior plate are in close alignment with those of the posterior,
149 presumed to result from tight morphological constraints. One feature on the
150 lingual surface of the anterior plate in all growth stages is the iterative,

151 sequentially developed ridges of the outer dentine layer. In the youngest
152 individual examined (14cm long; Figure 1c, rdg), 3–4 widely spaced ridges are
153 present, with small oval dentine units at the midline (*) of each dental plate.
154 Notably these are not hypermineralized tissues as are the rods found in these
155 plates (Figure 1d, ap.rd, pp.rd) but just elevations of the outer dentine layer.
156 Also at this growth stage, the hypermineralized rods are shorter, and do not
157 appear to extend through the dental plate.

158 At the next growth stage (Figure 2), there has been substantial wear on
159 both anterior and posterior dental plates, as indicated by the thickness of the
160 sclerotic dentine (*sensu* Smith *et al.*, 2019; Figure 2b, g, h, sod), forming in
161 response to wear. The hypermineralized rods of the anterior plate are longer,
162 and extend through more of the anterior plate, while on the posterior dental
163 plate, more rods are present when compared to the 14cm individual (Figure
164 2c). The number of ridges on the lingual surface of the plate has increased as
165 well, to seven, with the most oral ridge showing wear (Figure 2a, b). These
166 are absent from the comparable surface on the posterior dental plate (Figure
167 2a). The posterior end of these ridges is expanded and bulbous, particularly
168 the most aboral ridges (Figure 2b, d, f, white asterisk), the histology of the
169 lower jaw plates of *Harriotta* showing that this outer dentine layer does
170 become even more mineralized (Smith *et al.*, 2019). Just below these ridges,
171 the aboral margin of the plate is composed of an open network of trabecular
172 dentine (Figure 2f, h, tb). In section, the ridges surround trabecular dentine
173 (Figure 2g, h), with coincidence of early whitlockin formation of the rod and
174 the ridge itself (Figure 2g, opposing white arrows). Sclerotic dentine fills the
175 ridges in the same manner as it fills the spaces within the trabecular dentine

176 closer to the oral surface (Figure 2g, h). Additionally, the ridges themselves
177 appear to be set off from the rest of the plate, with a shallow furrow
178 developing posteriorly (Figure 2f, arrow 1) and an elongate oral-aboral ridge
179 anteriorly (Figure 2f, arrow 2).

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

199

200 **3.2 *Hydrolagus mirabilis***

201 Anterior and posterior dental plates of a young juvenile and adult of
202 *Hydrolagus* (Figures 4, 5) are comparable to those of *Chimaera*, with an ovoid
203 series of whitlockin in the anterior plate and a parasymphyseal rod in the adult
204 (Figure 5b), and a small number of extra rods in the ovoid series in the
205 juvenile (Figure 4b). With respect to the posterior plate, in the earlier growth
206 stage, both an ovoid series and rods are present along the labial margin of the
207 plate, with two rods present lingually and medially. As well, a tritoral pad is
208 developing posterior to these two rods, appearing to form via the incorporation
209 of individual ovoids (Figure 4b, black arrowheads on left side of image,
210 Supplementary Information files 1–3). In the adult, broad, well–developed
211 tritoral pads are present in this position, and with respect to the more medial
212 rods, the more posterior has become intensely vascularized to resemble the
213 more labial pad. However, the more anterior rod shows only a few openings
214 that represent incorporation of blood vessels within the mineralized dentine.
215 Along the labial margin, both series of ovoids and rods are present (Figure
216 5b).

217 In both growth stages, trabecular dentine below the oral surface has
218 been infilled with sclerotic dentine (Figures 4d, e, 5d, e), and becomes deeply
219 worn, exposing the ovoids and tritoral pads that resist deep wear (Figures 4c,
220 d, f, 5c, d, f). Serial ridges are present on the lingual surface of the anterior
221 dental plate, with five thick ridges in the juvenile, comparable in morphology to
222 those in *Chimaera* (Figure 4). However, in the adult, the ridges are more
223 numerous but less distinct (Figure 5). Bulbous expansions at the end of these
224 ridges are absent in both these growth stages (Figures 4c, f, 5c, f). As in
225 *Chimaera*, there may be correspondence between the ridges and the forming

226 ovoids (Figures 4e, 5e, double arrow). As well, trabecular dentine forms in
227 advance of these ridges forming in both growth stages (Figures 4f, 5f) but
228 always inside a shell of outer dentine. The posterior furrow and oral-aboral
229 ridge are present in both growth stages although the furrow appears to be
230 deeper in the adults (Figures 4f, 5f, arrows 1 and 2).

231

232 **3.3 *Harriotta raleighana***

233 In this juvenile specimen (Figure 6; Smith *et al.*, 2019: fig.17a), with respect to
234 whitlockin, the anterior dental plate has both rods and ovoids, while in the
235 posterior plate, tritoral pads and ovoids are present (Figure 6C). In older
236 individuals, these rods are replaced within the dental plate by ovoids, and the
237 tritoral pads are better developed (Smith *et al.*, 2019: figs. 1D, 17). On the
238 specimens examined, there is no indication of posterolingual rods on the
239 posterior dental plate (Smith *et al.*, 2019: fig. 17), and instead, in the youngest
240 individual available (Figure 6C), two short series of ovoids are present.

241 However, these do not transform into tritoral pads (Smith *et al.*, 2019: fig. 17).

242 One important difference between the Chimaeridae and *Harriotta* is the
243 absence of lingual ridges on the anterior dental plates of *Harriotta* (Figure 6a,
244 b, e). However, while these are not conspicuous on the lingual surface of the
245 outer dentine, the distinct region of the plate associated with the ridges in the
246 other taxa described here can be recognized and defined by the furrow and
247 oral-aboral ridge (Figure 6e, arrows 1, 2).

248

249 **3.4 Anterior versus posterior dental plates**

250 With respect to the whitlockin in the anterior dental plate, only rods or series
251 of ovoids are formed, with rods appearing dominant in early growth stages
252 (e.g., Figure 2c), and ovoids later, implying a developmental change in the
253 shape of the space, but not their location, within the trabecular dentine. Rods
254 are retained within the anterior dental plate, normally in a parasymphysial
255 position. By comparison, the posterior dental plate is more variable and
256 includes broad tritoral pads, which are blocks of highly vascularized whitlockin
257 (e.g., Figure 5b, 6a), more characteristic of adult dentitions. In earlier growth
258 stages the position of the tritors is occupied by whitlockin rods (Figures 3b, g,
259 4b) that subsequently become vascularized (Figures 3g, 5b). This
260 characterizes the more lingual, midline tritors, but in the case of labial tritors,
261 evidence from the earlier growth stage of *Hydrolagus* (Figure 4b;
262 Supplementary Information) indicates that the tritor is forming from the
263 incorporation of ovoids into the tritor. Thus, it seems that the tritors develop
264 differently in the posterior dental plates, by intrusion into whitlockin rods by
265 blood vessels lingually, and by addition of material to surround preexisting
266 blood vessels, labially. These dental plates show more morphological and
267 developmental diversity than previously appreciated.

268

269 **3.5 Synthesis of interpretation**

270 In all of the extant holocephalan species studied here the hypermineralised
271 tissue, whitlockin, whether ovoids, rods or tritoral pads, occurs in different
272 arrangements within the dental plate, all highly regulated. As part of this
273 regulation (Smith *et al.*, 2019), spaces for the whitlockin are preformed in the
274 trabecular dentine comprising much of the dental plate, at the aboral surface,

275 with whitlockin deposited within the spaces by specialized odontoblasts; our
276 new observations implicate superficial dentine ridges, also newly forming
277 lingually at the aboral surface of the anterior dental plate, in this patterning of
278 the rods and ovoids. These ridges are less mineralized than the whitlockin,
279 with locations that seem to concur with the position of the newly forming rods
280 and ovoids (Figures 2–4); a superimposed colour image (Figure 8)
281 summarizes our interpretation of the process by which dental epithelium could
282 induce the canonical ectomesenchymal tissue to make the whitlockin type of
283 dentine in the correct arrangement for each species, discussed further below.
284 Along with this, a clear ontogenetic pattern is present, with the whitlockin rods
285 dominating the lateral parts of the plates of the younger individuals;
286 positionally these rods are replaced by ovoid series in older individuals. As
287 well, rods appear to be located medially in younger individuals, to be replaced
288 in these positions by the tritoral pads.

289

290 **4 Discussion**

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

300 Here we have demonstrated the different morphologies of the oral
301 surface in the upper four plates, compared to the two lower jaw plates,
302 sculpted by wear resistant dentine (Figure 7a–c). These differences between
303 the plates, as well as different morphologies among the taxa, are determined
304 by intraspecific modulation of each dental plate. These surface morphologies
305 are dependent on the properties and arrangement of the tissues, as modified
306 in occlusion. For example, in *Hydrolagus* upper dental plates, a marginal
307 antero-labial ridge with hard, translucent whitlockin ovoids, set in less hard
308 sclerotic osteodentine, surrounds a deep, extensive fossa of the less
309 mineralized sclerotic osteodentine (double-headed arrow, Figure 7a, see also
310 μ CT scans Figure 5a–f). Notably, the occlusion of the four upper plates can
311 be appreciated in lateral and frontal views (Figure 1a, b), where the close
312 alignment of the anterior with the posterior plate is shown, also in occlusal
313 views (Figures 2b, 3e, f, 4a, f). They act as one unit of the upper jaw to meet
314 with the anterior edges of the two lower jaw plates. Also, the close alignment
315 of the hypermineralized rods and ovoids between anterior and posterior plates
316 adds to this functional link, as a feature suggesting common morphological
317 control to form a morphology against which the lower plates bite (Figures 1d,
318 2c, 3g, 4b, 5b, 6c, lower jaw ovoid stack, 7b).

319 We discuss the likely control of this patterning by a dental epithelium
320 surrounding the plate margins, of the continuous growth plates, modeled on
321 new observations of distinctive ridged, lingual surfaces on the anterior upper
322 dental plate (Figures 1a, 2a–h, 3a–l, 4a–f, 5a–f, 7a, 8b). During dental plate
323 development, an outer shell forms, composed of dentine formed by
324 odontoblasts creating two layers, surrounding the whitlockin and trabecular

325 dentine. The outer layer forms by apposition to the inner, demonstrated by the
326 odontoblast tubules directed inwards from cells located on the outermost
327 surface of the dental plate (Smith *et al.*, 2019: figs. 3A, 4B), while the inner
328 layer forms from odontoblasts associated with the trabecular dentine. The
329 outer layer, in particular, is heavily mineralized and hence translucent,
330 revealing the structures beneath (Figure 7b, double arrow; Smith *et al.*, 2019:
331 figs 1A, 2A, 3A, 7C). We have assumed these odontoblasts are
332 ectomesenchymal cells that deposit the dentine layer from beneath a layer of
333 epithelium, but currently have no supporting soft tissue histology (see Figure
334 7e). Nevertheless, if this is accepted as a cellular model, then control of
335 growth at the aboral surface can be by epithelial-mesenchymal interactions
336 that regulate the timing and positioning of new tissues.

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

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

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

374 the number of rods forming the anterior margin with regulated growth, as
375 apparent in the growth series of *Chimera monstrosa* (Figure 8a, nos. 0–2, 8b,
376 nos. 0–4).

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

388 It is important to note that although there is a potential developmental
389 relationship between the lingual ridges and both the developing trabecular
390 dentine and hypermineralized whitlockin (lime green, orange, Figure 8b), the
391 ridges have formed in advance of the mineralized ovoids, as well as the
392 patterned spaces within the trabecular dentine (Figure 4d–f). And, the ridges
393 are absent from the posterior dental plate in the three taxa examined here,
394 despite possessing substantially organized whitlockin, while the ridges appear
395 to become less distinct with growth although whitlockin deposition is ongoing
396 (Figure 5d, e). Moreover, these ridges are absent from *Harriotta*, whose
397 anterior upper dental plate is also characterized by a series of patterned rods

398 and ovoids, while the lingual face where these ridges occur in *Chimaera* and
399 *Hydrolagus* can also be identified in *Harriotta* (Figure 6e, arrows 1 and 2).

400

401 **5 Conclusions**

402 Individual teeth are present in stem group holocephalans such as *Helodus*
403 (e.g., Stahl, 1999), arranged in tooth families comparable to other
404 chondrichthyans such as sharks and rays. The dentition of *Helodus* also
405 presents evidence for tooth fusion (Moy-Thomas, 1936; Patterson, 1964;
406 Stahl, 1999; Johanson et al. in press), making it an ideal intermediate in the
407 evolutionary transition from stem- to crown group holocephalans, with
408 complete tooth fusion in the latter (e.g., Ørvig, 1957). Previously, researchers
409 have attempted to identify evidence for this tooth fusion in extant taxa, such
410 as the recognition of discrete and stacked oral and aboral territories in
411 *Callorhinchus*, said to be equivalent to the separate teeth of shark and ray
412 tooth families (Didier *et al.*, 1994). Separate growth phases were also
413 described in *Harriotta* (Smith et al. 2019: fig. 7A, C), although this was not
414 thought to represent the development of individual teeth. Developmentally, the
415 tooth plates in crown group holocephalans have been demonstrated to be
416 initiated in a similar manner to sharks and rays (e.g., Martin *et al.* 2016; Rasch
417 *et al.* 2016), with an infolding of epithelial tissue and corresponding
418 aggregation of mesenchymal cells (Didier *et al.*, 1994). The tooth plate formed
419 from this single primordium (Schauinsland, 1903; Kemp, 1984; Didier, 1995),
420 but separate tooth germs were not observed as part of the embryonic
421 development of the dental plates, nor of their continuous growth. Despite this
422 loss, significant patterning of dental elements, related to highly functional

423 dentitions, occurs in all extant taxa. How this patterning is regulated is
424 unknown, but may be correlated with development of outer and inner dentine
425 tissue, controlled by a dental epithelium and dentally committed
426 ectomesenchyme.

427

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434 *mili*.

435

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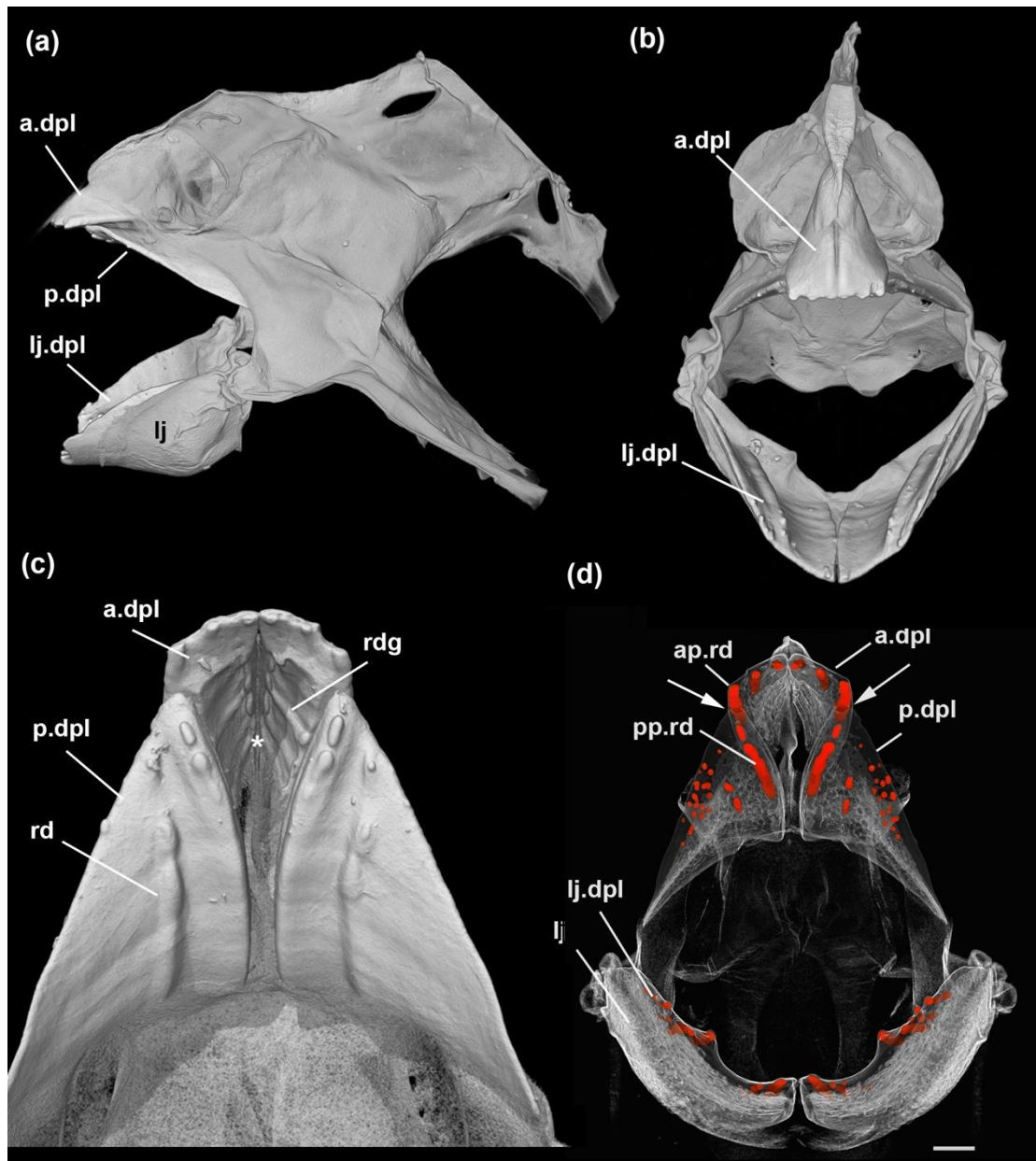
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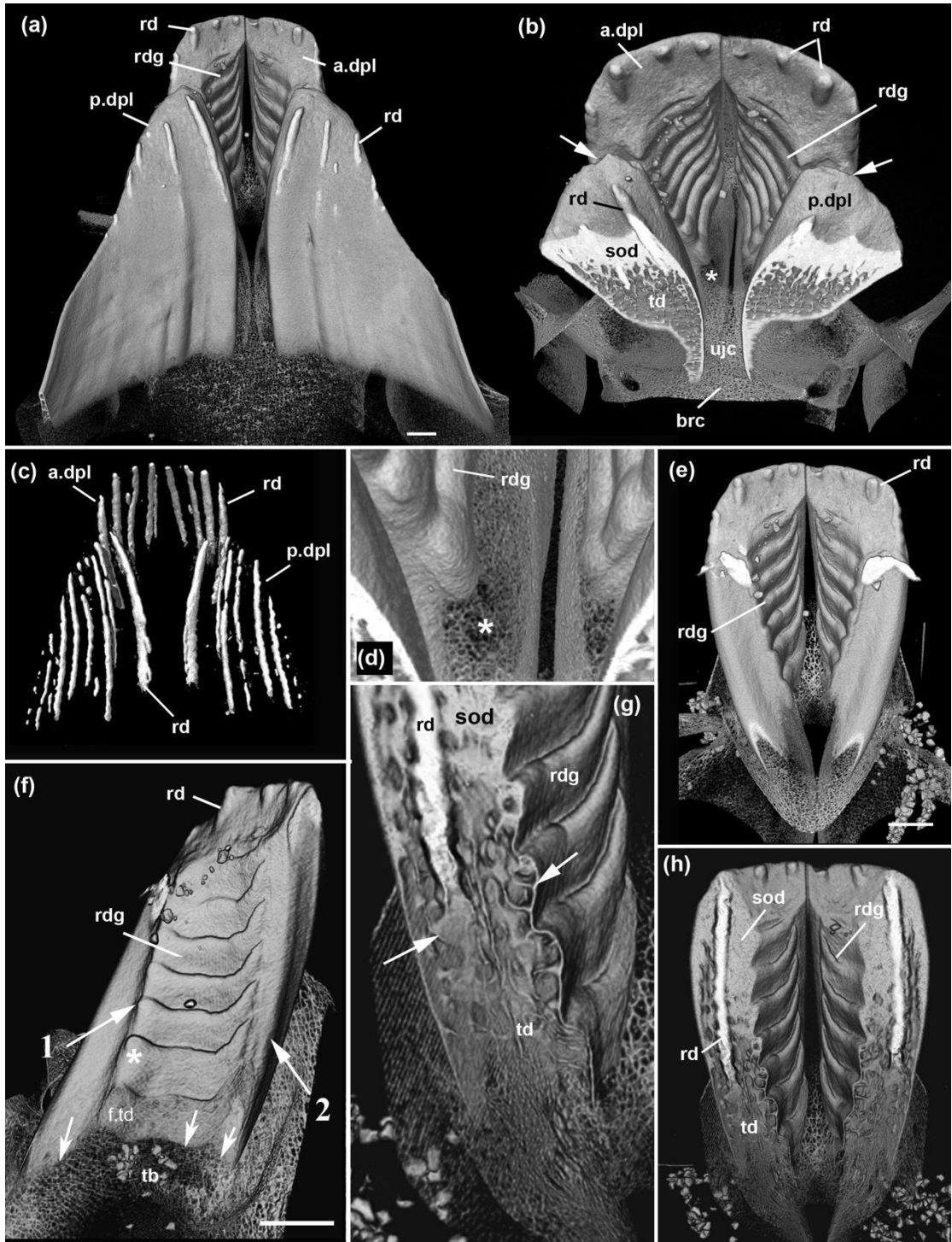


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FIGURE 1 *Chimaera monstrosa* Linnaeus, 1758, 14 cm juvenile, μ CT-scans.

518 Skull, jaws and dentition in (a) lateral view, (b) anterior view, (c) upper
519 dentition including anterior and posterior dental plates in oral view, (d) upper
520 and lower dentitions in antero-oral view, rendered (Drishti) to show
521 mineralized tissue of the dental plates (false colored red). a.dpl, anterior
522 dental plate; ap.rd, rods of hypermineralized dentine (whitlockin) in the
523 anterior dental plate, lj, lower jaw; lj.dpl, lower jaw dental plate; p.dpl, posterior
524 dental plate; pp.rd, rods of hypermineralized dentine (whitlockin) in the

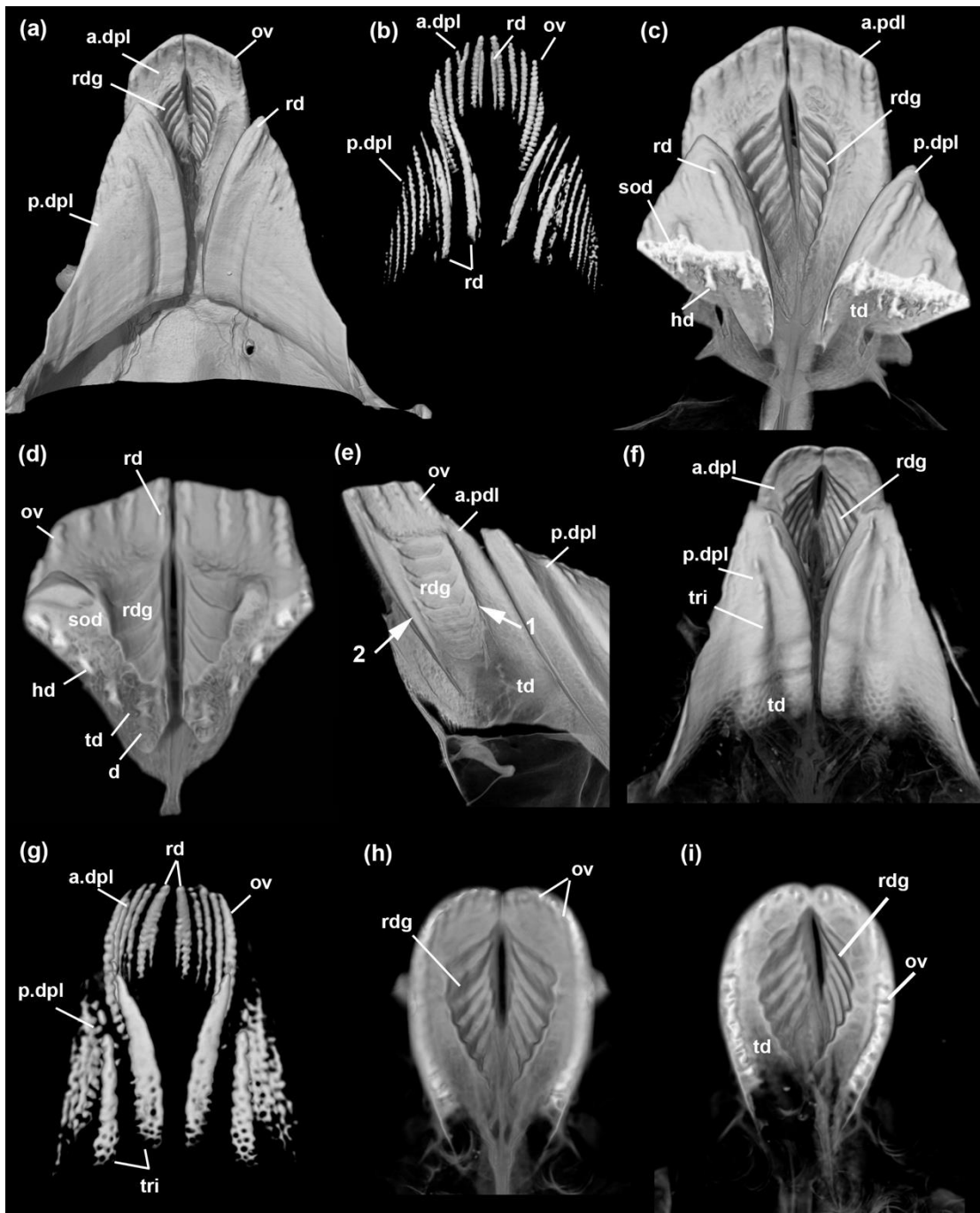
525 posterior dental plate, rdg, ridges on lingual surface of the anterior dental
 526 plate. White arrows in (d) indicate position of close contact between the
 527 anterior and posterior dental plates, scale bar=1mm. White asterisk in (c)
 528 indicates small oval dentine units at the midline of each dental plate.
 529



530
 531 **FIGURE 2** *Chimaera monstrosa* Linnaeus, 1758, 49 cm juvenile upper

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

553



554
555

FIGURE 3 *Chimaera* spp., upper dentition, μ CT-scans, (a-e) subadult

556

Chimaera monstrosa Linnaeus, 1758, (f-i) juvenile (20cm) *Chimaera* sp.

557

(unidentified), Taiwan. (a) *C. monstrosa*, (f) *Chimaera* sp., upper dentition in

558

oral view, including anterior dental plate with ridges on the lingual surface of

559

the dental plate, (b) *C. monstrosa*, (g) *Chimaera* sp., upper dentition rendered

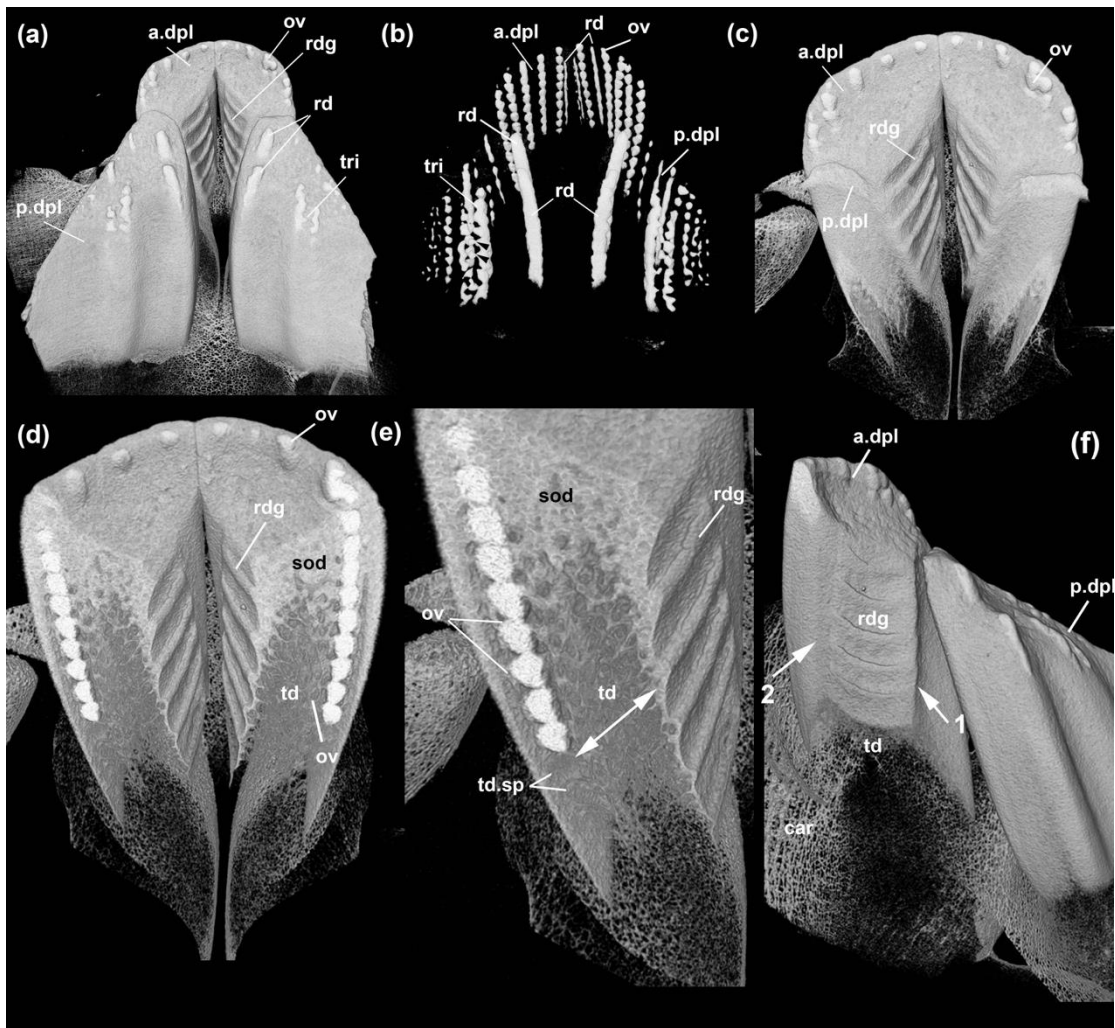
560

(Avizo) showing rods and ovoids of hypermineralized dentine (whitlockin) in

561

the both dental plates, and tritoral pads in the posterior plate (g), (c) *C.*

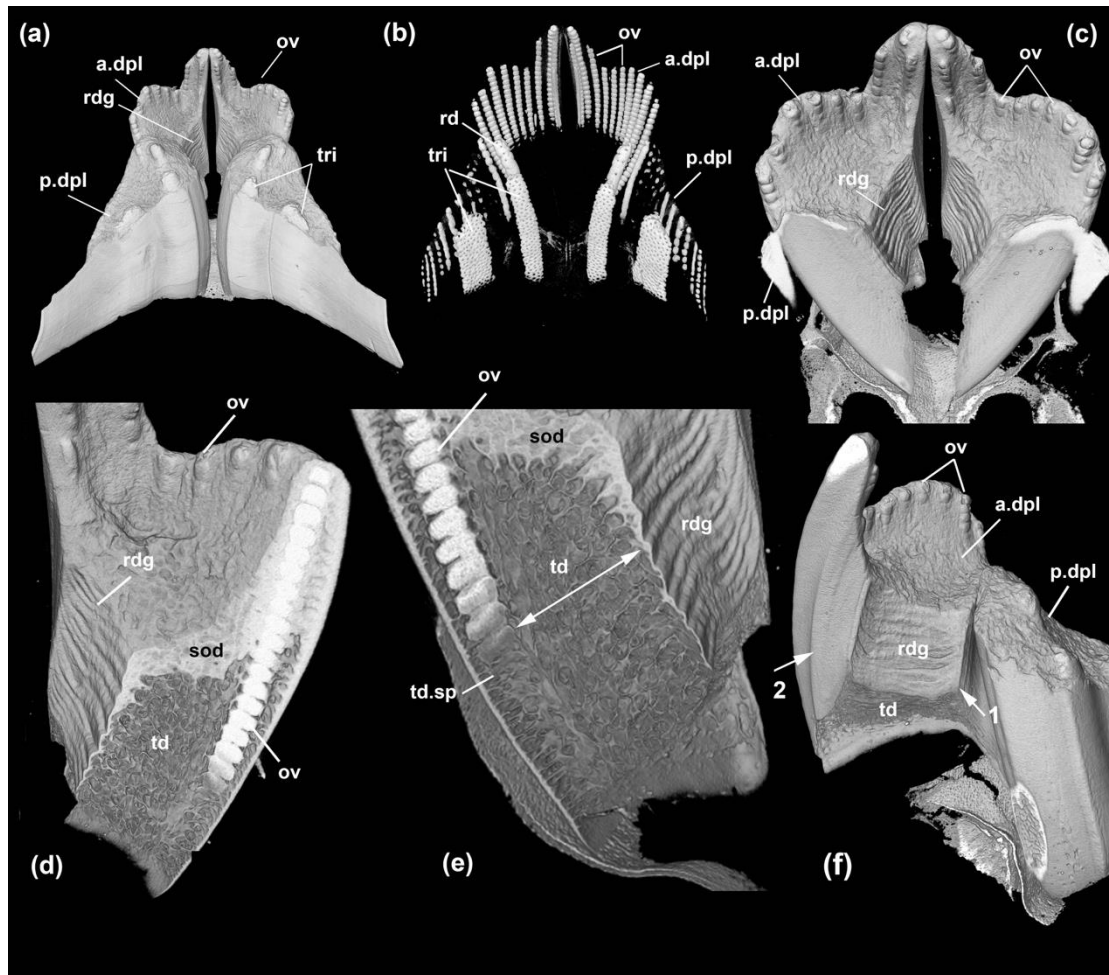
562 *monstrosa*, upper dentition in oral view, with posterior dental plate cut away to
563 show internal structure, including ridges on the anterior dental plate, (d) *C.*
564 *monstrosa*, (i) *Chimaera* sp., anterior dental plate, cut away to show section
565 through developing ovoids (hypermineralized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



575

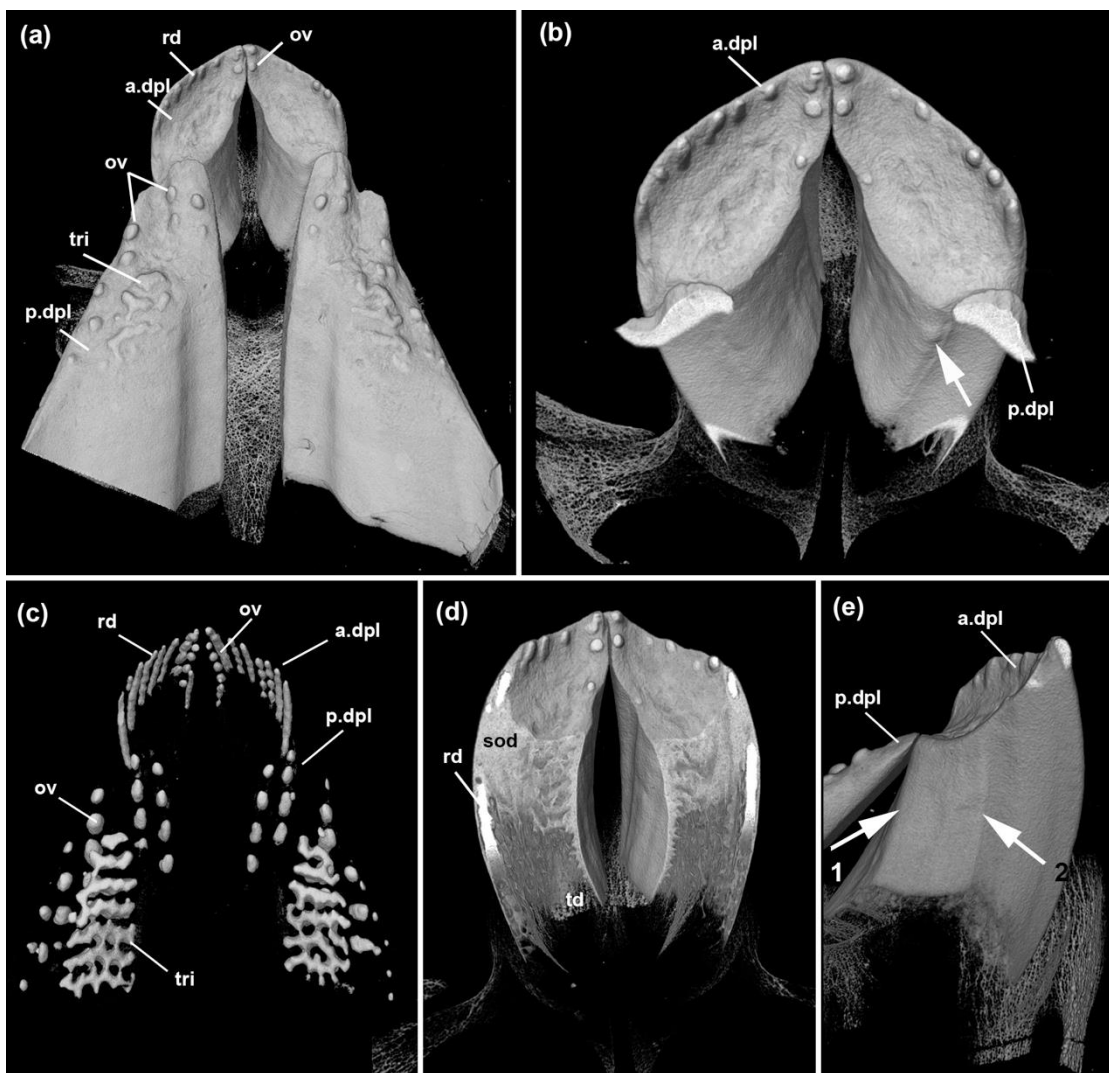
576 FIGURE 4 *Hydrolagus mirabilis* Collet, 1904, μ CT-scans. (a) upper dentition
 577 in oral view, including anterior dental plate with distinct ridges on the lingual
 578 surface of the dental plate, (b) upper dentition rendered (Avizo) showing
 579 ovoids and rods of hypermineralized dentine (whitlockin)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

587 show symphyseal face of the anterior plate and the ridges, numbered arrows
 588 different regions of the plate (as in Figures 2f, 3e). Abbreviations as in
 589 previous Figures.
 590



591
 592 **FIGURE 5** *Hydrolagus mirabilis* Collet, 1904, μ CT-scans, (a) upper dentition
 593 in oral view, including anterior dental plate with ridges on the lingual surface of
 594 the dental plate, (b) upper dentition rendered (Avizo) showing ovoids and rods
 595 of hypermineralized dentine in the both dental plates, and tritoral pads on the
 596 posterior plate, (c) in oral view, with posterior dental plate cut away to show
 597 posterior face of the anterior dental plate and lingual ridges, (d, e) anterior
 598 dental plate, cut away to show section through developing ovoids
 599 (hypermineralized), surrounding trabecular dentine, sclerotic

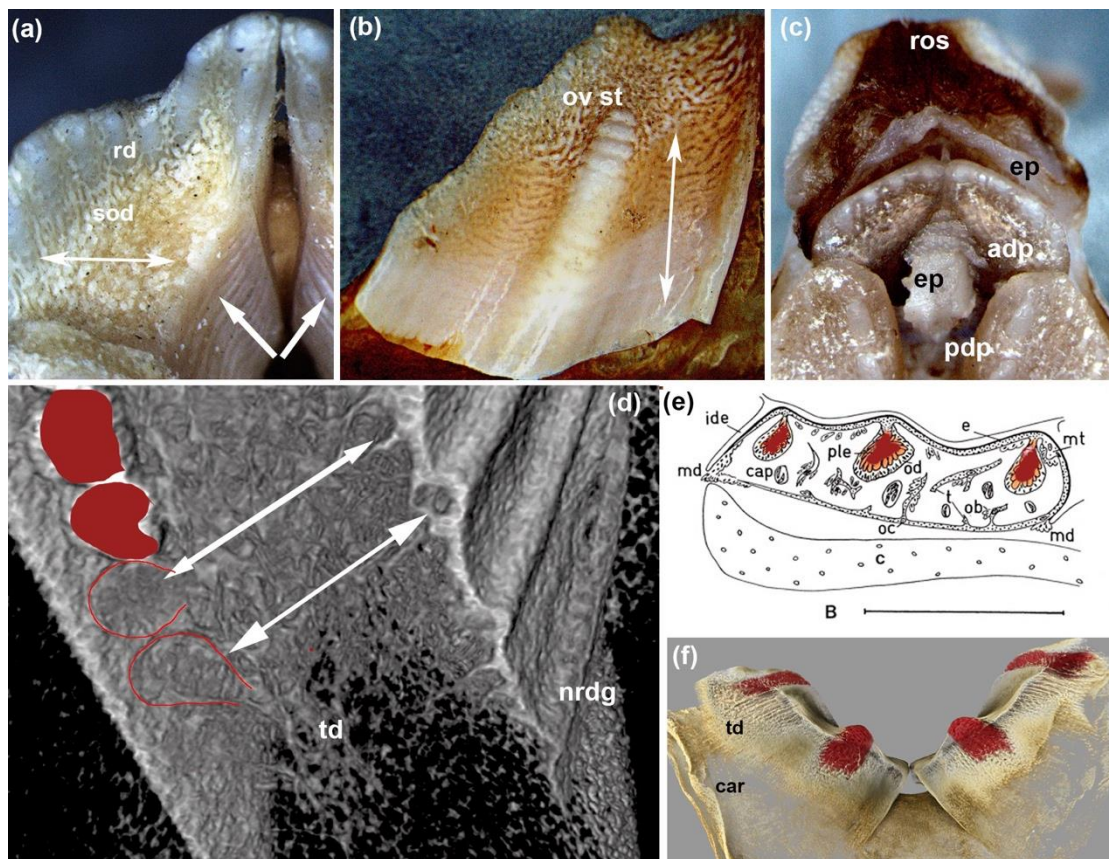
600 (hypermineralized) trabecular dentine and the ridges, double-headed arrow
 601 (e) indicating correspondence of developing ovoids and lingual ridges, (f)
 602 anterior and posterior dental plates in symphyseal view, with opposing plates
 603 cut away to show symphyseal face of the anterior plate and the ridges,
 604 numbered arrows mark different regions of the plate as in Figures 2f, 3e, 4f.
 605 Abbreviations as in previous Figures.
 606



607
 608 FIGURE 6 *Harriotta raleighana* μ CT-scans, (a-e) upper dentition in oral view,
 609 (a) anterior and posterior dental plate showing close fit between them and
 610 tritoral pad on posterior plate, (b) anterior dental plate with posterior dental
 611 plate cut away to show posterior and lingual faces of the anterior plate,

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

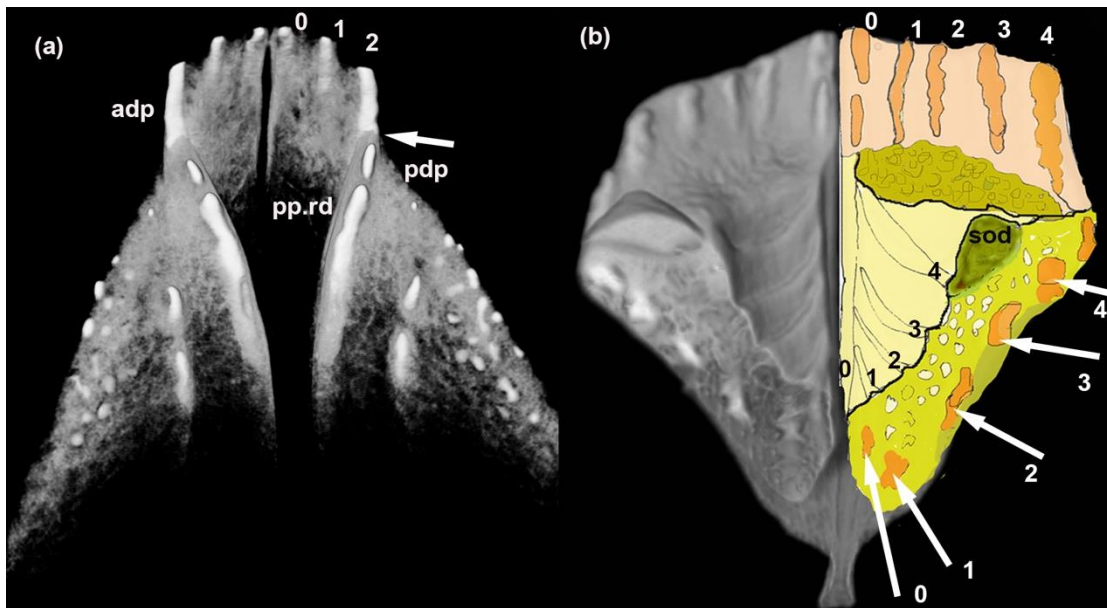
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FIGURE 7 *Hydrolagus mirabilis*, (a-c) photomicrographs in incident light of

627 upper and lower dental plates, (a) anterior upper plate shows ridges on lingual
628 surface (arrows), extent of worn tissue fossa (double arrow), translucent grey
629 rods, (b) lower dental plate tissue, transparency of most mineralized tissues
630 as in outer dentine (white double arrow) surrounding trabecular dentine, ovoid
631 stack not fully mineralized, (c) upper dentition *in situ* with soft tissue, plates
632 lined by epithelium, rostral snout present, (d) close up of virtual section
633 through anterior upper plate as in Figure 4d, e, colored false red to show
634 whitlockin as mineralized ovoids, and red rings the pre-formed capsular
635 spaces in the trabecular dentine, newest ridge forming aborally, coincidence
636 of ridges with new whitlockin forming (double arrows), (e) *Callorhinchus milii*
637 [Bory de Saint-Vincent](#), 1823, schematic drawing of section through lower jaw
638 embryonic plate (Kemp 1984), false coloured red indicates whitlockin
639 formation (ple = pleromin) under an epithelium, with modified trabecular
640 dentine (mt), condensed mesenchyme (md), cartilage (c), scale bar=1mm, (f)
641 *Callorhinchus milii*, adult lower jaw, μ CT rendered with whitlockin segmented,
642 false coloured red (Drishti), showing low ridges on the surface that form deep
643 into the trabecular dentine, above a cartilage furrow. Abbreviations as in
644 previous Figures, also car, cartilage furrow, ep, epithelium; nrdg, newest
645 ridge; ov st, ovoid stack; ros, rostral snout.
646



647

648

FIGURE 8 *Chimaera monstrosa*, μ CT-scans, sub-adult upper dentitions, (a)

649

from Figure 1, (b) from Figure 3a-e, with the rod series explained as whitlockin

650

formed to an timed order at the aboral surface in the trabecular dentine,

651

determined by a dental epithelium, where on the lingual side, ridges of the

652

outer dentine layer form (Figure 8b, also Figures 1a, 2b, 3 d-e). (a) partially

653

segmented upper dentition (Avizo) showing three rods of the anterior plate,

654

no. 2 aligns with the rods of the posterior plate), no. 0 forms in the

655

symphyseal segment of the plate, 1, 2 along the labial ridge, (b) anterior

656

dental plate with five rods and a coloured overlay to explain the co-incidence

657

of ridges with rods (0-4), tissues seen in virtual section relative to the surface

658

anatomy of the oral and lingual surfaces, whitlockin, orange; lime green,

659

trabecular dentine; olive green, sclerotic osteodentine dentine; moss green,

660

wear surface of the sclerotic dentine, arrowed in figure 7(a). Abbreviations as

661

in previous figures.

662

663

Supplementary information

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

669

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