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1 **Eyes in Staurozoa (Cnidaria): a review**

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23 **Abstract**

24 The presence of dark pigment spots associated with primary tentacles (or structures
25 derived from them, i.e., rhopaloids) in Staurozoa was recently overlooked in a study on the
26 evolution of cnidarian eyes (defined as a “region made of photoreceptor cells adjacent to pigment
27 cells”, irrespective of image formation, i.e., including all photoreceptive organs). Review of old
28 and recent literature on Staurozoa shows that dark pigment spots are present in virtually all
29 species of *Manania*, as well as some species of *Haliclystus*, *Stylocoronella*, and probably
30 *Calvadosia*. The known ultrastructure of ocelli seems to be compatible with light perception, but
31 no immediate response to changes in light intensity have been observed in the behavior of
32 staurozoans. Therefore, although further studies addressing photic behavior are required, we
33 discuss an earlier hypothesis that the dark spots in some stauromedusae may be related to
34 synchronous spawning, as well as the possible sensorial function of rhopaloids. Observations
35 summarized here suggest a possible ninth independent origin of eyes in Cnidaria, within a
36 lineage of benthic medusae. Alternatively, documented similarity across medusae of Cubozoa,
37 Scyphozoa, and Staurozoa – with eyes being topologically associated with primary tentacles in
38 each of these taxa – could indicate shared ancestry and a single origin of eyes in this clade
39 known as Acraspeda. Information on Staurozoa, one of the least studied groups within Cnidaria,
40 is often neglected in the literature, but correctly recognizing the characters of this class is crucial
41 for understanding cnidarian evolution.

42

43 **Introduction**

44 Staurozoa is a cnidarian class currently represented by 51 species classified in 11 genera
45 (Miranda et al., 2016a, 2018; Figure 1). They are all marine, benthic, and generally reported in

46 shallow temperate waters (Miranda et al., 2018). Staurozoa have a life cycle with two main
47 generations (i.e., metagenetic), known as the stauropolyp and the stauromedusa (Wietrzykowski,
48 1912; Kikinger & Salvini-Plawen, 1995; Miranda, Collins & Marques, 2010). However,
49 metamorphosis in Staurozoa is not so clearly defined as in other medusozoans (i.e., Cubozoa,
50 Hydrozoa, and Scyphozoa). In Staurozoa, metamorphosis is mainly observed in the apical region
51 (i.e., calyx) and the stauromedusa remains attached to the substrate by a basal peduncle
52 (Wietrzykowski, 1912; Kikinger & Salvini-Plawen, 1995; Miranda, Collins & Marques, 2010).
53 Therefore, the medusa stage has polypoid and medusoid characters (Miranda et al., 2016a).

54 During the metamorphosis of a stauropolyp into an adult stauromedusa, the eight primary
55 tentacles (four interradial and four perradial) can have four fates: 1) they disappear by resorption
56 (e.g., *Lucernaria*, *Craterolophus*, and some *Calvadosia*); 2) they metamorphose into adhesive
57 interradial and perradial rhopalioids (e.g., *Manania* and *Haliclystus*); 3) they remain as primary
58 tentacles but with a modified shape (e.g., some *Calvadosia*); 4) they change their shape (filiform
59 to capitate), migrate and cluster together with the secondary tentacles (e.g., *Stylocoronella*) (see
60 Miranda et al., 2016a). The secondary tentacles appear between two primary tentacles (one
61 perradial and one interradial), in adradial position, and progressively get united in clusters during
62 arm formation (Wietrzykowski, 1912) (Figure 2).

63 We recently hypothesized that rhopalioids are a synapomorphy of the family
64 Haliclystidae, which includes the genera *Manania*, *Haliclystus*, *Depastromorpha*, *Depastrum*,
65 and *Halimocyathus* (Miranda et al., 2016a; Figure 1). The structure likely has a role in temporary
66 substrate attachment, a hypothesis supported by stauromedusan behavior (Larson, 1988) and
67 histology (Miranda et al., 2016b). The genera *Lucernaria* and *Lipkea* are closely related, and we
68 hypothesized that *Stylocoronella* would fit this clade (Figure 1; see also Kikinger & Salvini-

69 Plawen, 1995) based on the morphology (e.g., presence of interradial longitudinal muscles in
70 peduncle and absence of primary tentacles or rhopaloids in stauromedusa) (Miranda et al.,
71 2016a). *Craterolophus* and *Calvadosia* belong to the suborder Amyostaurida as they are the only
72 two genera without interradial longitudinal muscles in peduncle (Miranda et al., 2016a).

73 In a recent article, Picciani et al. (2018) proposed that eyes (defined as a “region made of
74 photoreceptor cells adjacent to pigment cells”, irrespective of image formation, i.e., including all
75 photoreceptive organs) originated at least eight times in Cnidaria, even in the absence of a central
76 nervous system. Their study was mainly based on a review of the literature on the presence of
77 eyes for adult medusae (their table S1) in light of an extensive ribosomal and mitochondrial-
78 based molecular phylogeny for Cnidaria. Their analyses covered all of the cnidarian classes
79 (other than the unusual parasitic class Myxozoa), including Staurozoa (i.e., stalked jellyfishes,
80 Figure 2), which was characterized as lacking eyes (Picciani et al., 2018).

81 Picciani et al. (2018) considered eyes absent in Staurozoa based on a simple statement by
82 Mayer (1910 p. 520: “eyes [...] are absent in Stauromedusae”). Nevertheless, Picciani et al.
83 (2018) correctly included an exception for the genus *Stylocoronella*. Polyps of *Stylocoronella*
84 *riedli* Salvini-Plawen 1966 and *Stylocoronella variabilis* Salvini-Plawen 1987 possess dark
85 pigment spots on the oral side of the calyx, at the inner bases of the tentacles (Salvini-Plawen,
86 1966, 1987; Blumer et al., 1995; Kikinger & Salvini-Plawen, 1995). Based on histological
87 studies, these pigment spots were interpreted as being ocelli (Salvini-Plawen, 1966). Although
88 the polyps of *S. riedli* show no distinct reaction to light stimuli, ultrastructural results
89 corroborated the hypothesis that these structures are light-sensitive organs (Blumer et al., 1995).
90 The ocelli are composed of seven to nine monociliary sensory cells, that lie next to the tentacular
91 mesoglea, and one to four pigment cells (Blumer et al., 1995). The monociliary sensory cells of

92 *S. riedli* show all the features characteristic for photoreceptive cells, including the intraciliary
93 structure, demonstrating that these cilia are immobile (see Blumer et al., 1995). The pigment
94 cells enclose the photoreceptive cilia and have irregularly shaped pigment granules enveloped by
95 a membrane (Blumer et al., 1995). The dark pigment visible in living animals is associated with
96 these membranes (Blumer et al., 1995). In addition, the ocelli in *Stylocoronella* have
97 characteristics (e.g., arrangement of microtubules of photoreceptive cilia and membranous
98 elements of pigment granules) that were hypothesized as unique for *Stylocoronella* and unknown
99 within other metazoans (Blumer et al., 1995; Martin, 2002). The dark pigment spots are, in a
100 somewhat modified arrangement, retained in the medusa stage (Blumer et al., 1995; Kikinger &
101 Salvini-Plawen, 1995). The stauromedusa stage of *S. riedli* has “numerous tiny pigment spots in
102 the basal area of the capitate tentacles and four larger perradial ones at the subumbrellar margin”,
103 which were also presumed to be ocelli (Kikinger & Salvini-Plawen, 1995).

104 However, there are no molecular sequences for *Stylocoronella* species, so the presence of
105 these ocelli was disregarded in the analyses of eye origins within Cnidaria (Picciani et al., 2018).
106 On the other hand, Picciani et al. (2018) included two species of *Manania*: *Manania gwilliami*
107 Larson & Fautin 1989 and *Manania uchidai* (Naumov 1961) (their Table S1) in their phylogeny,
108 and both possess dark pigment spots (=eyes, *sensu* Picciani et al., 2018) associated with
109 rhopaloids (= anchors) in the stauromedusa stage (Naumov, 1961; Larson & Fautin, 1989).
110 Therefore, Picciani et al. (2018) neglected literature on Staurozoa and our aim is to review it and
111 discuss the evolutionary implications of correctly coding this character in Staurozoa.

112

113 **Survey methodology**

114 For recent papers reviewing the global diversity and natural history of stalked jellyfishes
115 (Miranda et al., 2018), and their systematics (Miranda et al., 2016a), we compiled and reviewed
116 literature containing every original description (51), occurrence, and morphological description
117 of staurozoan species to our knowledge (see Miranda et al., 2016a, 2018, and respective online
118 resources). For this study, we updated our list for any new additions to the literature and searched
119 for any detail that could speak to the possibility of eye-like structures using the terms “black”,
120 “dark”, “pigment”, “eye”, “spot” in English, French, and German in the accumulated literature
121 on Staurozoa.

122

123 **Results**

124 **The genus *Manania* Clark 1863 (family Haliclystidae)**

125 Currently, the genus *Manania* comprises seven valid species. Virtually all of them have
126 evidence of dark pigment spots in adult stauromedusae.

127

128 *Manania auricula* (Fabricius 1780) and *Manania hexaradiata* (Broch 1907)

129 There is not a clear mention of dark pigment spots associated with rhopaloids for these
130 two species in the literature. However, see Figure 2 (C–E) showing a specimen identified as *M.*
131 *auricula* from Svalbard with dark pigment spots associated with rhopaloids. Additionally, there
132 are doubts regarding the validity of *M. hexaradiata*. Mayer (1910) considered *M. distincta* (see
133 below) closely allied to *M. hexaradiata*. Uchida (1929) proposed that *M. hexaradiata* seemed a
134 “young specimen of a medusa closely allied to, if not identical with” *M. distincta*.

135

136 *Manania distincta* (Kishinouye 1910)

137 The first unequivocal mention on the dark pigment spots in *Manania* in the literature was
138 provided by Kishinouye (1910) in his description of *M. distincta*: “the eight primary tentacles are
139 transformed into small, cylindrical bodies. They are erect, hollow inside, and not adhesive. They
140 are black at base and along the axial median line. They serve probably as a sensory organ
141 standing in relation to light”.

142

143 *Manania atlantica* (Berrill 1962)

144 Berrill (1962) mentioned that, at the base of each rhopaloid of *M. atlantica*, there is a
145 “small spherical ocellus, apparently only a pigment spot”.

146

147 *Manania uchidai* (Naumov 1961)

148 Different records in the literature of *M. distincta* are actually *M. uchidai* (Figure 2, A–B),
149 as proposed by Naumov (1961). Uchida & Hanaoka (1933) mentioned that at the axial base of
150 the rhopaloids of *M. uchidai* (as *M. distincta*) “there exists a blackly pigmented spot which
151 probably serves as an organ standing in relation to light” and that “on the axial side of these
152 tentacles [= rhopaloids] there is a sensory organ for light which is blackly pigmented and
153 composed of exceedingly narrow cells arranged in a row”. Hanaoka (1935) added that “on the
154 axial side of primary tentacles [=rhopaloids]” of *M. uchidai* (as *M. distincta*) “there is a sensory
155 organ for light, which is blackly pigmented”. Uchida (1929), also for *M. uchidai* misidentified as
156 *M. distincta*, highlighted that “in my specimen the pigment has probably faded away on account
157 of preservation, or has not yet appeared owing to its being young”. Later, Hirano (1986)
158 observed that “the axial base of each primary tentacle (= rhopaloids)” was “provided with a
159 black spot”.

160

161 *Manania gwilliami* Larson & Fautin 1989

162 Larson & Fautin (1989) described that each rhopaloid of *M. gwilliami* has a “small, dark
163 pigment spot near margin”.

164

165 *Manania handi* Larson & Fautin 1989

166 In this unpublished thesis, Gwilliam (1956) described the species *Manania prasinus*,
167 mentioning that “the adaxial side of the primary tentacles [= rhopaloids] bears a dark pigment
168 fleck”. Later, the species was formally described by Larson & Fautin (1989) as *M. handi*, with a
169 “dark spot on adaxial side of each primary tentacle [= rhopaloids] near margin”.

170 Recently, Westlake & Page (2017) showed that the pigment spot at the base of the
171 rhopaloids of *M. handi* is associated with a greatly increased “concentration of FMRFamide-IR
172 neuronal cell bodies”.

173

174 **The genus *Haliclystus* Clark 1863 (family Haliclystidae)**

175 *Haliclystus* is the more diverse genus within Staurozoa, with 13 valid species (Miranda et
176 al., 2018), but the presence of dark pigment spots in stauromedusae of this genus is more elusive.
177 Naumov (1961) mentioned that the rhopaloids of *Haliclystus* are sometimes supplied with a
178 pigmented eyespot. Gwilliam (1956), while describing *Haliclystus* species, observed that “there
179 are no conspicuous pigment stripes other than the dark pigmented band on the subumbrellar side
180 of the anchor [= rhopaloid] peduncle”.

181 More specifically, Clark (1878; see also his figures 27 and 32) observed, for *Haliclystus*
182 *auricula* Clark 1863, “dark patch of color so noticeable at the proximal bases of tentacular

183 groups, and which remind one of eye-spots” and dark spots associated with rhopalioids. Then,
184 Clark (1878) clearly inferred the existence of a nervous system in *H. auricula* based on the
185 presence of “eye-spots” found in rhopalioids: “We speak of these eye-spots because they occupy
186 a position at the proximal side of the base of the anchors homologous with that in which a more
187 highly developed and even well defined optical apparatus is to be found in other Acalephae. In
188 our Lucernarian it amounts to a mere accumulation of pigment, in unusual quantity, in a small
189 circle, among the interstices of the prismatic cells of a specially thickened wall [...]. The boss-
190 like protuberance of the wall at these spots, conjoined with the conspicuous coloring matter
191 imbedded in it down to half its depth, give it strong claims to some special functional status, or to
192 a typical representation of what finds its full development in other Acalephs. The accumulation
193 of pigment matter at any point concentrates light there rather than any other force capable of
194 being taken note of by a nervous centre. Neither odor nor sound would be affected by it, nor does
195 it seem possible that taste could be seated at a point so distant from the digestive system. That it
196 is after all a mere foreshadowing, or a mimetism, of a more efficient organ of vision becomes
197 strongly probable when we learn that these spots lose their distinctness, or disappear altogether,
198 by the time the animal measures one-half an inch across the umbrella. When the latter is about
199 one-fifth of an inch across [...] the spots have attained to their greatest definiteness, and from
200 that period onward they gradually become obliterated; not so much, though, by fading out as by
201 the increase of pigment all around them, until they lose their distinctness for want of contrast”.

202 Clark (1878) added that “they are then probably to be set down rudimentary oculiferous tentacles
203 situated within the line along which the anchors are disposed. Now in all Acalephae the eye, so
204 called, stands in close proximity to the margin of the umbrella”. In addition, Clark (1878)
205 mentioned that “we find it [pigment matter] holding exactly the same relation to the prismatic

206 cells [...], i.e., forming a dark casing or envelope about them, as the pigment does to the facets of
207 the eyes of Articulata” and concluded “we have all that can be brought forward in favor of their
208 functional characters as elements of an optical apparatus”. Therefore, it seems that the dark
209 pigment spots associated with rhopalioids in *Haliclystus* disappear during the development, but
210 their rhopalioids might still have knots of FMRFamide- immunoreactive neurons in adult
211 stauromedusae (Westlake & Page, 2017).

212 In addition, but with less precision, Ling (1939) mentioned that *Haliclystus inabai*
213 (Kishinouye 1893) has “anchors [= rhopalioids] brown with brown spot in center”.

214

215 **The genus *Stylocoronella* Salvini-Plawen 1966 (family Lucernariidae)**

216 Dark pigment spots in the two species of *Stylocoronella* have been examined in detail
217 (Salvini-Plawen, 1966, 1987; Blumer et al., 1995; Kikinger & Salvini-Plawen, 1995) in both
218 stauropoly and stauromedusa stages. Polyps of *S. riedli* and *S. variabilis* possess dark pigment
219 spots on the oral side of the calyx, at the inner bases of the tentacles (Salvini-Plawen, 1966,
220 1987; Blumer et al., 1995; Kikinger & Salvini-Plawen, 1995). The youngest polyp observed of *S.*
221 *riedli* had eight primary tentacles and already possessed eight pigment spots (Kikinger &
222 Salvini-Plawen, 1995). The stauropoly has up to 24 pigment-spot ocelli (at 24-tentacle stage),
223 composed of monociliated sensory cells and pigment cells (Blumer et al., 1995; see also Martin,
224 2002). The cilia associated with the sensory cells (photoreceptive cilia) have a unique axonemal
225 pattern, with a third central microtubule at a certain point (9x1+3 arrangement) and a balloon-
226 like swelling of the distal portion of the cilium, with scattered microtubules in this area (Blumer
227 et al., 1995; see also Martin, 2002). The stauromedusa stage of *S. riedli* has “numerous tiny
228 pigment spots in the basal area of the capitulate tentacles and four larger perradial ones at the

229 subumbrellar margin”, which were presumed to be ocelli (Kikinger & Salvini-Plawen, 1995).

230 The stauromedusa stage of *S. variabilis* does not have four perradial pigment spots (Kikinger &

231 Salvini-Plawen, 1995).

232

233 **The genus *Calvadosia* Clark 1863 (family Kishinouyeidae)**

234 The genus *Calvadosia* is the second most diverse in Staurozoa, with 11 valid species

235 (Miranda et al., 2018). However, we found few mentions in the literature that could indicate

236 (with imprecision) the presence of dark pigment spots in this genus. Kishinouye (1902) observed

237 that the primary tentacles of *Calvadosia nagatensis* (Oka 1897) are absent and “in place of them

238 we see a dark pigment for each”. Ling (1937) described that a “semi-triangular purplish area is

239 seen in each of the eight marginal notches in close contact with the primary tentacles” for

240 *Calvadosia cruciformis* and a “semi-triangular purplish streak present at bottom of every

241 marginal notch” for *Calvadosia tsingtaoensis*, that faded away gradually after specimens are

242 preserved.

243

244 **Conclusions**

245 Based on our review, all dark pigment spots in stauromedusae are associated with the

246 primary tentacles, the region where they used to be, e.g., *Stylocoronella* and *Calvadosia*, or with

247 the rhopaloids that are derived from primary tentacles (e.g., *Manania* and *Haliclystus*). Because

248 the rhopalia of medusae of Cubozoa and Scyphozoa are also derived via metamorphosis of

249 primary tentacles of cubopolyps and scyphopolyps, respectively, rhopaloids and rhopalia are

250 hypothetically homologous (Thiel, 1966). However, whereas the rhopalia is clearly a sensory

251 structure (reviewed in Katsuki & Greenspan, 2013), an adhesive rather than sensorial function

252 thought to be associated with the benthic habit of staurozoans is often attributed to the
253 rhopaloids of stauromedusae (Larson, 1988; Miranda et al., 2016b, 2018).

254 Dark pigment spots associated with the eight rhopaloids have been observed in virtually
255 all species of *Manania* and in at least some species of *Haliclystus*. We found no records of dark
256 pigment spots for the other genera with rhopaloids, *Depaстроморpha*, *Depastrum*, and
257 *Halimocyathus* (Miranda et al., 2016a), in the literature, but this information could be
258 overlooked, since the dark spots disappear after preservation (Uchida, 1929) and their presence
259 can vary during development (Clark, 1878).

260 Unlike the pigment spots in polyps of *Stylocoronella* (Blumer et al., 1995), the
261 ultrastructure of dark pigment spots in stauromedusae of *Manania* and *Haliclystus* have never
262 been analyzed, potentially raising doubt about a sensorial function. However, recent evidence
263 (Westlake & Page, 2017) supports the idea of photo reception by these organs in both genera.
264 Westlake & Page (2017) analyzed the neuromuscular morphology of two stauromedusae,
265 *Manania handi* (Larson & Fautin 1989) and *Haliclystus "sanjuanensis"* (*nomen nudum*), using
266 whole mount immunohistochemistry with antibodies against FMRFamide and α -tubulin to label
267 neurons. Comparative observations on cnidarians indicate that photoreceptive organs are
268 consistently associated with the expression of these markers (see Westlake & Page, 2017),
269 although peptides of the RFamide family also occur in species or life stages of cnidarians that do
270 not have ocelli or known structures of photoreception (Plickert & Schneider, 2004).
271 Interestingly, the "transformed primary tentacles" of *M. handi* had a greatly increased
272 "concentration of FMRFamide-immunoreactive neurons at their base" associated with the dark
273 pigment spots, which they concluded to be "consistent with their homology with rhopalia" of
274 cubomedusae and scyphomedusae (Westlake & Page, 2017). Besides, a similar, but less

275 pronounced, knot of FMRFamide- immunoreactive neurons is present at the base of the
276 rhopalioids of *H. "sanjuanensis"*, although the species lacks a pigment spot in this area
277 (although, as we pointed out, there are changes during development in *Haliclystus*). This result
278 might be associated with the observed expression of opsin genes (the functional visual pigments
279 in vertebrate and most invertebrate photoreceptors; Martin, 2002) in species of *Haliclystus*
280 (Picciani et al., 2018), but raises interesting questions about the expression of medusozoan opsin
281 genes reported by Picciani et al. (2018) in staurozoan species whose adult stauromedusae
282 apparently lack both rhopalioids and dark pigment spots, such as *Lucernaria quadricornis*
283 Müller 1776 and *Craterolophus convolvulus* (Johnston 1835). For example, What is the diversity
284 of opsin genes across Staurozoa, and how have these genes evolved? Where and when are
285 different opsin genes expressed across staurozoan life cycles and body regions? Are there
286 mechanisms of extraocular photosensitivity in Staurozoa (see Martin, 2002)?

287 Although the dark pigment spots in *Manania* species were often associated with
288 hypotheses of light perception, Westlake & Page (2017) mentioned that *M. handi* showed no
289 immediate response to changes in light intensity, and hypothesized that "light sensitive neurons
290 in staurozoans may detect light to trigger synchronous spawning, rather than to modify
291 immediate behavior" (Westlake & Page, 2017). However, they highlighted that many
292 stauromedusae spawn in response to light (see Otto, 1976; Otto, 1978; Miranda et al., 2018)
293 despite having no obvious pigment spots (e.g., species of *Haliclystus*), which could be
294 interpreted as a counterargument for this hypothesis (see examples of extraocular
295 photosensitivity associated with cnidarian spawning in Martin, 2002). Besides, polyps of
296 *Stylocoronella* also have ocelli, show no distinct reaction to light stimuli (Blumer et al., 1995)
297 and do not spawn (as gonads are only fully developed in the stauromedusa stage; Kikinger &

298 Salvini-Plawen, 1995). Therefore, although the ultrastructure of ocelli are compatible with light
299 perception, at least in polyps of *S. riedli* (Blumer et al., 1995), the complete understanding of
300 their function is a challenge and further studies that specifically address photosensitive behavior
301 are necessary.

302 The presence of dark pigment spots (or ocelli) in Staurozoa suggests that rhopalioids in
303 stauromedusae can have both adhesive (see Miranda et al., 2016b) and sensorial functions. The
304 reasons related to the wide occurrence of these structures in the stauromedusa stage of species of
305 *Manania* needs further investigation. The homology of dark pigment spots in Staurozoa is
306 questionable, since the genera *Manania*, *Haliclystus*, *Stylocoronella*, and (possibly?) *Calvadosia*
307 do not form a monophyletic group (Miranda et al., 2016a; Figure 1). In this context,
308 ultrastructural studies on the dark pigment spots of different genera, especially *Manania*, and at
309 different stages (i.e., polyp and medusa) should also be encouraged as it could provide relevant
310 data on the evolution of this character in Staurozoa. However, in all these genera, the dark
311 pigment spots of stauromedusae were associated with primary tentacles, their region
312 (perradial/interradial) or structures derived from them. Therefore, based on our review and on the
313 phylogenetic topology obtained by Picciani et al. (2018), in which Staurozoa formed a sister
314 group relationship to the remaining medusozoans (Figure 3A), the presence of these structures in
315 Staurozoa could indicate at least a ninth independent origin of eyes in Cnidaria.

316 Alternatively, the observations reviewed here highlighting eyes associated with structures
317 derived from primary tentacles (rhopalioids/rhopalia) might indicate shared ancestry of eyes of
318 medusae across Cubozoa, Scyphozoa, and Staurozoa, which were recently shown to form the
319 clade Acraspeda based on phylogenomic data (Kayal et al., 2018; Figure 3B). The Acraspeda
320 hypothesis linking Staurozoa with Cubozoa and Scyphozoa goes back to the late 1880's

321 (Haeckel, 1880; Claus, 1883) and was also suggested by a cladistic analysis based on
322 morphology and life history characteristics (Marques & Collins, 2004). This explicit
323 phylogenetic analysis supported the assertion of Thiel (1966) that rhopalia and rhopalioids, both
324 derived from primary polyp tentacles, are shared across Acraspeda due to common ancestry.
325 That these apparently homologous structures are also always the position of eyes across
326 Acraspeda raises the possibility that light sensitivity has specific components that are shared
327 across the group due to common ancestry. On the other hand, the strength of the Picciani et al.
328 (2018) analysis was that it looked at species level observations of eyes, and while eyes are
329 ubiquitous across Cubozoa, documented eyes are less than universal in both staurozoans and
330 scyphozoans, supporting the idea that eyes may have evolved independently several times within
331 Acraspeda. We support that eyes in hydromedusae have an independent origin from those in the
332 medusae of Acraspeda, as sense organs in the former are not associated with metamorphosis of
333 primary polyp tentacles (Thiel, 1966; Salvini-Plawen, 1987; Marques & Collins, 2004; Picciani
334 et al., 2018). In short, correctly coding this character in Staurozoa, a lineage of benthic medusae,
335 has profound consequences for understanding the evolution of eyes and nervous systems in
336 Cnidaria and studies disregarding the presence of staurozoan ocelli should be reassessed.

337

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343

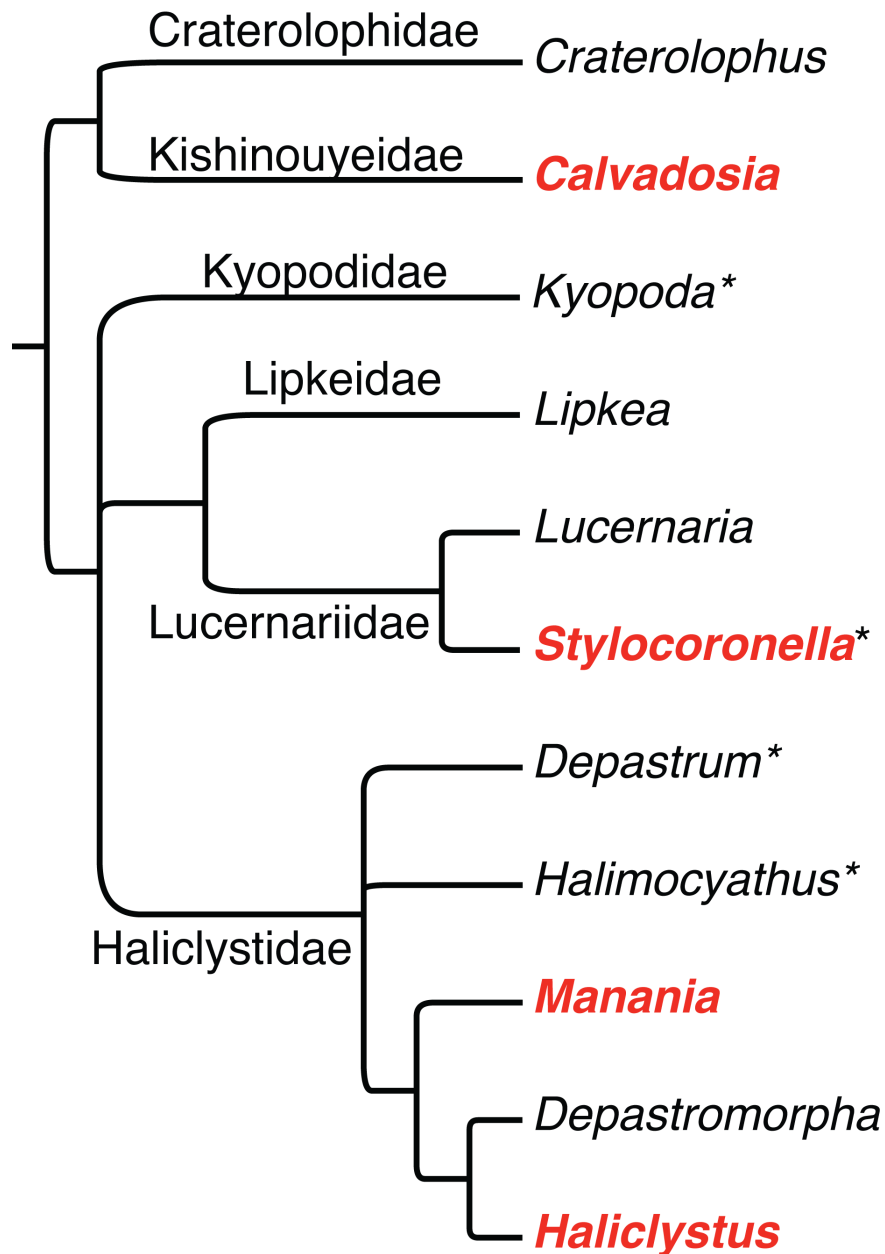
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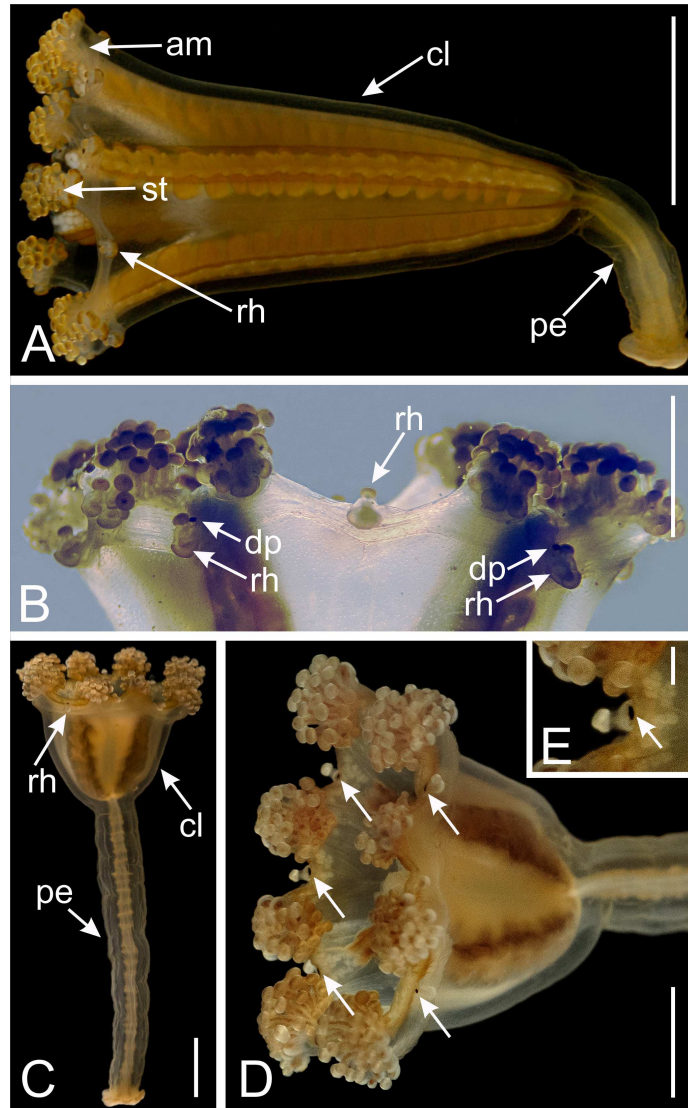
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436 **Figures**

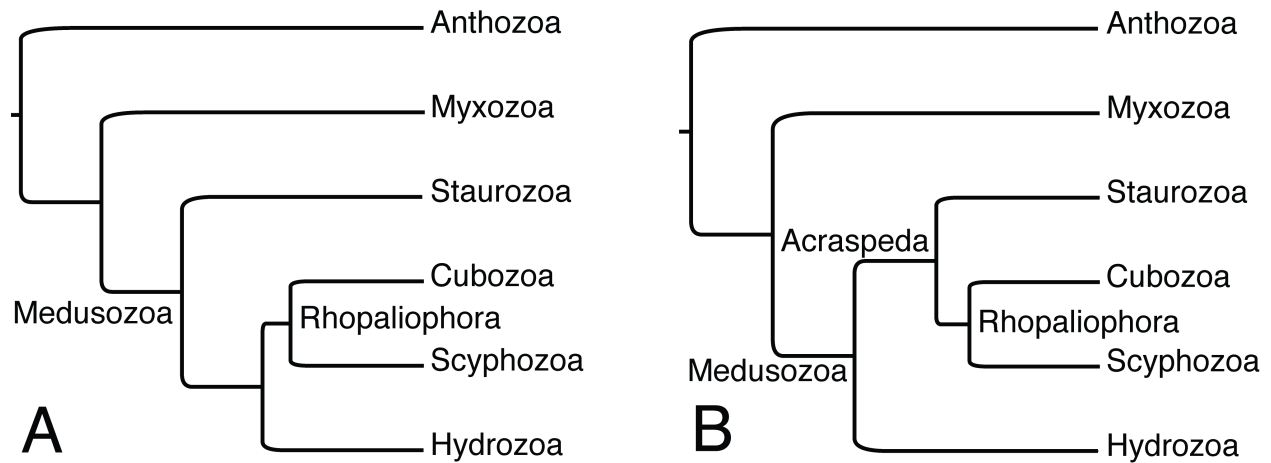
437

438 Figure 1: Phylogenetic hypothesis of relationships among staurozoan genera and families, based
 439 on Miranda et al. (2016a). Asterisks after generic names indicate that the genus has not yet been
 440 sampled for molecular data, and thus their respective positions in the phylogeny are based solely
 441 on inferences based on morphology. Bolded genera in red have been documented to have
 442 pigment spots, observations that are reviewed herein.



443

444 Figure 2: Dark pigment spots associated with rhopalioids in species of *Manania*. (A) General
 445 view of *Manania uchidai*. (B) View of calyx margin of *M. uchidai*, with rhopalioids and dark
 446 pigment spots associated with rhopalioids. (C) General view of *Manania auricula*. (D) View of
 447 calyx of *M. auricula* with arrows pointing to dark pigment spots associated with rhopalioids. (E)
 448 Arrow pointing to dark pigment spot associated with rhopalioid in *M. auricula*. Abbreviations:
 449 am, arm; cl, calyx; dp, dark pigment spot; pe, peduncle; rh, rhopalioid; st, secondary tentacles.
 450 Scale bar: A, C, D=0.5 cm; B=0.25 cm; E=0.1 cm. Photo credits: A–B, A.G.C.; C–E, courtesy of
 451 Maciej Mańko.



452

453 Figure 3: Alternative phylogenetic hypotheses for the placement of Staurozoa within Medusozoa.

454 (A) Based on analyses of ribosomal and mitochondrial markers of Picciani et al. (2018). (B)

455 Based on phylogenomic analyses of Kayal et al. (2018). Myxozoa has been added to (A) for

456 comparability, but this group has generally not been treated in analyses of ribosomal and

457 mitochondrial markers data due to highly accelerated rates of evolution.