

1 **Title Page**

2 Running Head: Salp colonial ontology

3 Title: A developmental ontology for the colonial architecture of salps

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26 **Abstract**

27 Colonial animals are composed of clonal individuals that remain physically connected and
28 physiologically integrated. Salps are urochordates with a dual life cycle including an
29 asexual solitary stage that buds sexual colonies composed of jet-propelling zooids that
30 efficiently swim together as a single unit by multi-jet propulsion. Colonies from different
31 species develop distinct architectures characterized by their zooid arrangement patterns,
32 but this diversity has received little attention. Thus, these architectures have never been
33 formally defined using a framework of variables and axes that would allow comparative
34 analyses. We set out to define a homology-oriented ontology of the salp colony
35 architecture morphospace and characterize the developmental pathways that build the
36 different architectures. We collected and photographed live specimens of adult and
37 developing colonies through offshore SCUBA diving. Since all salp colonies begin their
38 development as a transversal double chain, we characterized each adult colonial
39 architecture as a series of developmental transitions, such as rotations and translations
40 of zooids, relative to their orientation at this early shared stage. We found that all adult
41 architectures are either final or intermediate stages within three developmental pathways
42 towards either bipinnate, cluster, or helical forms. The architectural variation found among
43 salp colonies is the most diverse in any clade of clonal colonial animals. We hypothesize
44 that this remarkable breadth of geometries is a product of adaptive evolution to distinct
45 multijet colonial locomotory strategies. This framework will enable comparative studies
46 on the biomechanical implications, ecological functions, evolutionary history, and
47 engineering applications of the diversity of salp colony architectures.

48

49 **Introduction**

50 Salps (Chordata: Tunicata: Thaliacea: Salpida) are marine pelagic urochordates
51 that filter-feed on phytoplankton and bacteria. The salp life cycle (Fig. 1) consists of a
52 solitary stage (oozoid) that asexually buds colonies of the aggregate stage
53 (blastozooids) along a ventral projection (stolon). Aggregate blastozooids are
54 protogynous and can sexually reproduce, brooding embryonic solitary oozoids in a
55 placenta as females (Bone, 1998). While solitary oozoids move using single-jet
56 propulsion (such as solitary medusae), salp aggregate colonies move in an integrated,
57 coordinated manner through multi-jet propulsion.

58 Compared to other multijet colonies, e.g., siphonophores or pyrosomes, salps
59 present a much broader set of architectural configurations among free-swimming
60 colonial animals (Madin, 1990). Salp colony architectures vary across the 48 described
61 species of salps, and include transversal chains, oblique chains, linear chains, whorls,
62 clusters, and helical solenoids (Fig. 2). This diversity and complexity in the
63 arrangements of zooids across species represent a morphological phenotype above the
64 individual's morphology. A familiar analogy would be the variation in the quaternary
65 structure of proteins as an emergent property of lower-level structural changes. While
66 these colonial architectures look radically different from one another, all species have
67 one early developmental stage in common where the stolon of the solitary progenitor
68 segments into a double chain of paired chiral zooids arranged in a transversal double
69 chain (Bone, 1998).

70 As the young colony is released from the solitary, the zooids grow rapidly in size,
71 develop their anatomical features, and in most species, they shift their arrangement in

72 the colony into the different architectures we observe in adults. While these colony
73 architectures have been described qualitatively (Madin, 1990), they have received little
74 attention in the past three decades and lack a formal definition. The primary gaps of
75 knowledge include a breakdown of the traits that define the ontology of these
76 architectures, a quantitative framework to measure those traits, and a detailed
77 comparison of the developmental processes that give rise to the different architectures.
78 An ontology is defined as “a set of concepts and categories in a subject area or domain
79 that shows their properties and the relations between them” (Oxford Languages, 2023).
80 In biology, ontologies serve as conceptual frameworks to designate categories,
81 identities, and relationships of parts and variations in complex systems (Bard & Rhee
82 2004). In this context, we refer to an ontology of salp colony architectures as the
83 categorization of the forms, the description of their characteristics, the definition of their
84 ontogenetic relationships, and their relationships to geometric transformations.

85 Here we aim to leverage the shared earliest stage in their colonial development
86 to (1) define a set of homologous axes, variables, and planes of observation in all salp
87 colonies, (2) map the different architectures based on (1), and (3) define a hierarchical
88 classification of the distinct types and degrees of developmental translations and
89 rotations of the zooids. The ultimate goal is to enable comparative analyses of variation
90 in zooid arrangements between and within architectures. Using this framework,
91 comparative studies will be able to investigate the biomechanical implications,
92 ecological functions, evolutionary history, and engineering applications of the extant
93 architectonic diversity of salp colonies. Moreover, this work will shed light on the
94 broader design space of clonal coloniality among animals.

95

96 **Materials and Methods**

97 We observed, videographed, and collected live specimens of both adult salp
98 blastozooid colonies and developing colonies in the stolons of solitary salp oozoids.
99 These specimens were collected while SCUBA diving untethered from a small vessel off
100 the coast of Kailua-Kona (Hawai'i Big Island, 19°42'38.7" N 156°06'15.8" W), at an
101 offshore location with a bottom depth of over 2000m. Some dives were conducted
102 during the day, where we encountered most of the specimens of *Iasis cylindrica* (Cuvier,
103 1804), *Pegea* sp., *Cyclosalpa affinis* (Chamisso, 1819), and *Brookgia rostrata*
104 (Traustedt, 1893). The rest of the species included in this study were collected during
105 night dives when many salps perform diel vertical migration to shallower depths.

106 After the dive, salp specimens were anesthetized in 0.2% MS222 buffered with
107 sodium bicarbonate in seawater to facilitate photography. Developing stolons were
108 dissected from the anesthetized solitary oozoid before photographing. We
109 photographed anesthetized adult and developing blastozooid colonies in glass
110 crystallization dishes with a black background using a Canon 6D DSLR camera with a
111 35mm lens mounted on an inverted tripod used as a copy stand. Specimens were
112 photographed from different orientations relative to the constituent zooids' bilateral
113 symmetry (oral, aboral, dorsoventral, and lateral), with a ruler in the frame for scale
114 reference. We collected and photographed blastozooids across 22 salp species. In
115 addition, we supplemented our gaps in taxon sampling using underwater photos and
116 videos of live salps from previous expeditions and from online sources.

117 From these images, we examined the colonial arrangement from the earliest
118 stage of stolon development to adulthood. In some taxa, the temporal axis of
119 blastozoid development can be observed spatially in a continuous gradation of
120 blastozoid development (Fig. 3). This is the case in *Cyclosalpa* spp. (Fig. 3E-F),
121 *Brooksia* spp., *Soestia* spp., and *Helicosalpa* spp. (Fig. 3G). In other taxa, the temporal
122 axis of blastozoid development can also be observed spatially in discretely segmented
123 cohort blocks with synchronous development within each block, such as in *Salpa* spp.,
124 *Ritteriella* spp. (Fig. 3D), and *Thalia* spp. Other taxa, however, produce only a single
125 cohort block with synchronous development, such as in the case of *I. cylindrica* (Fig.
126 3C), *Thetys vagina* Tilesius, 1802 (Fig. 3B), *Pegea* spp. (Fig. 3A), and *Traustedtia*
127 *multotentaculata* (Quoy & Gaimard, 1834). We examined the development of the
128 blastozoid chain in these taxa by keeping the solitaries alive in seawater and observing
129 the developmental transitions overnight.

130

131 **Results**

132 **Defining the observation framework** - The arrangement and relative orientation of
133 blastozoids in different colony architectures present a 3-dimensional problem, where
134 the axes and angles of reference shift in ways that are challenging to compare from a
135 single viewpoint. Using the transversal double-chain architecture found in the earliest
136 developmental stage of every species (as well as in adult colonies of *Pegea* spp. and
137 *Traustedtia* spp.), in addition to the bilateral symmetry of salp blastozoids, we defined
138 three orthogonal axes and their corresponding normal planes (Fig. 4). These are: (1)
139 The dorsoventral axis of the colony is defined as the axis parallel to the dorsoventral

140 axis of the zooids in the transversal double chain, with a normal (perpendicular) plane of
141 observation corresponding to viewing the dorsal side of the zooids on either side of the
142 transversal double chain. (2) The oral-aboral axis is defined as the axis parallel to the
143 oral-aboral axis of the zooids in the transversal double chain, with a perpendicular plane
144 of observation corresponding to viewing the oral or aboral end of the zooids on either
145 the frontal or rear side of the transversal double chain. The zooid oral-aboral axis of
146 each zooid is defined as the line parallel to the endostyle. (3) The stolon axis is defined
147 as the axis of chain growth parallel to the stolon, perpendicular to a plane of observation
148 that corresponds with looking directly at either end of the transversal double chain, with
149 a lateral view of the zooids.

150 With these axes and planes delineated, we can then describe developmental
151 changes in zooid orientation characters relative to these three planes of observation.
152 Based on this universal observation framework, we then defined the following
153 characters: (A) the dorsoventral zooid-stolon angle is the angle formed between the
154 oral-aboral axis of the zooid and the elongation axis on the colony as viewed from the
155 dorsal side of the developing zooids, driving the formation of oblique, linear, and
156 bipinnate chains; (B) the lateral chiral angle, defined as the angle formed between the
157 oral-aboral axes of a pair of chiral zooids as viewed from the zooids' lateral orientation,
158 driving the formation of bipinnate chains; (C) zooid autorotation, defined as rolling of the
159 zooid around its own oral-aboral axis also driving the formation of bipinnate chains; (D)
160 serial stolon-normal angle, defined as the angle formed between a zooid's oral-aboral
161 axis and the oral-aboral axis of its lateral neighbor as viewed from one end of the
162 colony, driving the formation of a solenoid double helix chains; (E) peduncle length ratio

163 (the peduncle is an extension of the tunic that connects the zooids to their chiral pair or
164 to the stolon during development, present in most *Cyclosalpa* species), defined as the
165 ratio between the total oral-aboral length of the zooids relative to the longest axis of
166 their peduncle, driving the formation of whorls and clusters; and finally (F) neighbor
167 attachment, defined as direct contact between lateral neighbors, its loss drives the
168 formation of clusters with loosely-attached zooids.

169 We examined changes in these variables across the development of colonies in
170 different salp species and characterized a developmental ontology of salp colony
171 architecture by first describing the set of developmental transformations that give rise to
172 each architecture, then identifying which intermediate stages in the formation of more
173 derived architectures in some species are equivalent to the adult finalized architectures
174 in other species, to build a process-based hierarchical ontology of the architectures
175 within colonial developmental pathways.

176 **A developmental ontology of architectural transition pathways** - Some taxa, such
177 as *Pegea* spp. and *Traustedtia* spp., retain the transversal double-chain architecture
178 throughout the growth and development of the blastozoooids in the chain. This
179 architecture is characterized by a dorsoventral zooid-stolon angle of ~90°, with ventral
180 attachment to the chiral neighbor and lateral attachment to the lateral neighbors (Fig.
181 2A). These chains move parallel to the oral-aboral axis of their zooids, at an angle
182 orthogonal to the length of the chain. Often, we find colonies of *Pegea* species moving
183 in a coiled formation, where the transversal chain is curled up on the oral-aboral-normal
184 plane. Most other species do not retain this developmentally basal architecture, but
185 instead modify the orientation, rotation, and position of the zooids relative to each other

186 and the axis of the chain during development (Fig. 5). We observed that the
187 developmental series of all salp species (Fig. 3) with non-transversal adult architectures
188 fall under three distinct developmental transition pathways (Fig. 6).

189 First there is the pathway that leads to the architectures found in *Cyclosalpa* spp.
190 These blastozooid chains continue their development in a transversal arrangement
191 (dorsoventral zooid-stolon angle of 90°) but grow peduncles that separate the zooids
192 from the stolon attachment point and from their chiral ventral neighbor. Moreover,
193 discrete sets of budding salps bundle together, where the attachment points of their
194 peduncles remain attached to a central point and detached from other sets. These
195 radial aggregations (whorls) are formed by two chiral, bilaterally symmetrical,
196 semicircular sets of zooids (Ritter & Johnson 1911). In the first stage of the
197 transformation, the whorl architecture is formed. These zooids are packed together
198 tightly in a wheel shape due to the short peduncles (Fig. 2B). These are found in many
199 *Cyclosalpa* species such as *C. affinis*, *C. quadriluminis* Berner, 1955, and *C. bakeri*
200 Ritter, 1905. In *C. affinis*, these whorls remain attached to each other for a longer time
201 than in other species and adult whorls can often be found conjoined. A further stage in
202 this transformation is the cluster architecture, where the peduncles grow so long that
203 zooids are no longer attached laterally to each other (Fig. 2C) and can freely bob
204 around and end up arranged in hemispherical or spherical sets. These are found in
205 other *Cyclosalpa* species such as *C. sewelli* Metcalf, 1927, *C. pinnata* (Forskål, 1775),
206 and *C. polae* Sigl, 1912. These cluster bundles typically contain many more zooids than
207 those in whorls. It is unclear whether these cluster architectures ever go through a
208 distinct ‘whorl’ stage in their development. The developing colonies we observed do go

209 through a similar zooid-stolon allometry as their whorl-shaped counterparts, though the
210 serial neighbor zooid detachment (Fig. 6) may occur before the release of the radial
211 aggregations.

212 Second, there is the pathway leading to the helical double-chain architecture in
213 *Helicosalpa* spp. (Fig. 2D). These transversally budded double chains undergo stolon
214 torsion into a solenoid shape (Fig. 3G), and zooid become angled relative to their chiral
215 pair to accommodate this conformation. Finally, there is the pathway leading to
216 streamlined chains through the alignment of zooid orientations to the axis of the stolon
217 during development. These start with the partial dorsoventral rotation of zooids into the
218 oblique architecture (Fig. 3B), which is the final form for species like *Thetys vagina* and
219 *Thalia* spp. (Fig. 2E), with zooid-stolon angles of 50-60°. The movement direction of
220 these colonies is closely aligned (but not perfectly parallel) with the stolon axis. This
221 torsion goes a step further toward near-complete alignment (15-30°) of the oral-aboral
222 axis of the zooids to the axis of the stolon in linear architecture, found in taxa such as
223 *Ihlea*, *Iasis*, *Metcalfina*, and *Salpa*. In the species *Soestia zonaria* (Pallas, 1774), we
224 find the most extreme version of this architecture, with zooid-stolon angles close to 0°
225 (Fig. 2F). The final transformation stage in this pathway is the bipinnate architecture
226 found in *Brookzia* spp. and *Ritteriella* spp., where in addition to the linear dorsoventral
227 alignment of zooids to the stolon, there is a mirror-symmetry outward lateral flare of the
228 aboral ends of zooids in the plane normal to the oral-aboral-normal; and a rotation of
229 zooids where the ventral (and dorsal) sides of every zooid are all facing the same side
230 (Fig. 2G). This pathway has been challenging to observe empirically because *Ritteriella*
231 does not undergo transformation past the oblique stage in colonies retained by the

232 oozooid (Fig. 3D), and in *Brookzia* the transformation occurs at a very small scale in the
233 most proximal and underdeveloped end of the budding colony. It is possible that the
234 order of developmental transitions that lead to the bipinnate morphology differs from the
235 one hypothesized here (Fig. 6), though it always occurs during or after the process of
236 dorsoventral zooid stolon rotation that produces oblique and linear chains.

237 Each developmental transition is characterized by variation across specific
238 continuous morphological traits (Fig. 6). The transversal-to-whorl transformation is
239 mediated by an increase in the peduncle-to-zooid length ratio and a continuous
240 allometric shift in zooid-to-stolon size as the zooids grow and develop asynchronously
241 along the stolon length. The subsequent whorl-to-cluster transformation also relies on
242 further peduncle elongation but is marked by a loss of neighbor zooid attachment that
243 allows neighboring zooids to bob around freely. The transversal-to-helical pathway is
244 characterized by a continuous shift in the serial neighbor stolon-normal angle, where the
245 orientation of neighboring zooids breaks parallelism and starts to offset by a few
246 degrees like stairs in a spiral staircase. The transversal-to-oblique-to-linear pathway is
247 driven solely by changes in the dorsoventral zooid-stolon angle. Finally, the linear-to-
248 bipinnate transformation is characterized by an increased oral-aboral chiral angle and
249 zooid autorotation, where the oral-lateral facets of chiral zooids face each other, the
250 aboral ends turn outwards, and their ventral sides face the same side of the colony.

251

252 **Discussion**

253 Ontologies in biology are helpful conceptual tools to characterize, categorize, and
254 compare variation between and within species. We leveraged homologies in the

255 development of salp colonies across species to categorize and geometrically compare
256 the different architectures. From this developmental perspective, we were able to
257 establish an ontology for salp colony architecture by defining the developmental
258 transitions in the zooid arrangements that lead to the different architectural endpoints
259 and identifying which adult endpoints are homologous to intermediate stages in the
260 development of other endpoints. These ontological definitions and reference
261 frameworks are essential to measure and compare standing variation in colony
262 architecture and its emergent properties between salp species.

263 One of the most immediate emergent properties of salp colony architecture is the
264 potential implications for locomotion. Different salp colony architectures present different
265 relative orientations of the individual jets to each other and to the overall colony motion
266 axis. In addition, we hypothesize that different architectures differ in how the number of
267 zooids in the colony scales with cross-sectional area relative to motion. These
268 hydrodynamic properties can have further consequences on the locomotory efficiency of
269 different architectures. Swimming in linear salp chains is hypothesized to be more
270 economical due to the reduction of drag (Bone & Trueman 1983). A salp colony is
271 equipped with multiple propelling jets rather than one, which increases its propulsive
272 power. Drag experienced during swimming depends on the total area exposed to the
273 fluid as well as the frontal (motion-orthogonal) projected area (Alexander 1968). Skin
274 drag will increase with the number of zooids in the colony in a predictable manner that is
275 independent of their zooid arrangement. However, frontal drag is drastically reduced in
276 linear chains compared to the sum of each separate zooid (Mackie 1986). We
277 hypothesize that frontal drag will vary across architectures and therefore impact the

278 relative speed attained by each species. In addition to changing the way the frontal area
279 scales with the number of zooids, we hypothesize that architecture may also impact the
280 angles of the jets relative to the axis of colony motion. In siphonophores, the velum of
281 the nectophore is used to orient the jet to prioritize torque or thrust (Sutherland et al
282 2019). In salps, these orientations are usually fixed in a colony (Sutherland & Weihs
283 2017), but the angle of the exhalant jets relative to the swimming of the colony will
284 dictate the thrust-to-torque ratio, which will determine their propulsive efficiency.

285 Understanding the hydrodynamic advantages and implications of each colonial
286 architecture can be valuable beyond basic science since it may yield interesting
287 applications to bioinspired underwater vehicles. Pulsatile jet propulsion is increasingly
288 inspiring underwater vehicle engineering (Mohensi 2006, Yue et al. 2015). Multijet
289 systems comprised of collaboratively interactive propeller units could revolutionize the
290 field of underwater vehicles (Chao et al. 2017, Costello et al. 2015) with designs
291 inspired by gelatinous invertebrates such as salps (Marut 2014, Krummel 2019, Bi et al
292 2022). Some of these bio-inspired solutions are stimulating novel solutions in the field of
293 soft robotics (Renda et al. 2015, Krummel 2019), as deformable body shapes can
294 augment propulsive forces (Giorgio-Serchi & Weymouth 2017). Understanding the
295 biomechanical underpinnings of the diversity of salp colony architectures would reveal
296 nature's broadest design space for underwater multi-jet-propelled soft locomotors and
297 their inherent trade-offs.

298 Another potential contribution of this architectural ontology is the characterization
299 of colonial morphology from a comparative, evolutionary perspective. Salp colony
300 architectures are distributed across the phylogenetic diversity of salp species, but their

301 evolutionary history remains unknown. The two main obstacles to the reconstruction of
302 the evolutionary history of salp colony architecture have been (1) the lack of a homology
303 framework to compare and characterize variation, and (2) a phylogenetic tree that
304 resolves the position of every architecture in every lineage where it has evolved.
305 Govindarajan et al. (2011) reconstructed the first thaliacean molecular phylogeny using
306 18S sequences. While this phylogeny included many of the known salp species, it
307 cannot fully resolve the evolutionary history of salp colony architecture since the
308 position of *Pegea* and *Thalia* are poorly resolved and the position of *Helicosalpa* is
309 unknown. A phylogenetic comparative approach to the diversity of colonial architectures
310 will facilitate further research on its evolutionary, ecological, and biomechanical
311 underpinnings. If evolutionary shifts in the architecture of salp chains bring on changes
312 in their locomotory efficiency, it is possible that these shifts are related to different
313 selective pressures such as predatory pressure, habitat nutritional patchiness, or
314 vertical migration behavior.

315 The research directions outlined above would advance our understanding of salp
316 biology across their species diversity. Salps have attracted significant scientific interest
317 in the past decade since they are essential consumers in oceanic ecosystems that feed
318 on microbial plankton production and can grow explosively following phytoplankton
319 blooms (Henschke et al. 2016). Salp fecal pellets play an important role in the biological
320 carbon pump and are responsible for a large fraction of the biological carbon pump
321 (Decima et al. 2023), responsible for the trapping of gigatons of carbon fixed from the
322 atmosphere into the deep sea and are therefore a key mediator for atmospheric CO₂
323 concentration and global change (Buesseler et al. 2020). Many of the salp species that

324 contribute most to this process are vertical migrants that respire and deposit (by
325 defecation and predation) carbon during the day in the mesopelagic zone, after feeding
326 during the night near the surface (Steinberg et al 2023). Many of the vertically migrating
327 salp species (such as *Salpa* spp.) present a linear architecture (Madin et al. 1996) and
328 their migratory behavior varies with colonial development (Henschke et al. 2021). While
329 some of these linear, vertically migrating species have been extensively investigated,
330 the ecology and natural history of the broader diversity of salps remains understudied.
331 Characterizing the relationship between colonial architecture, locomotion, and migratory
332 behavior is key to understanding the ecological implications of shifting salp species
333 compositions and distributions with global change (Lavaniegos & Ohman 2003).

334 Finally, we believe the colonial ontology presented here expands our
335 understanding of the development and evolution of colonial animals in general. Colonial
336 animals (modular colonies, not including eusocial colonies) are composed of clonal
337 individuals produced by asexual reproduction that remain physically connected and
338 physiologically integrated (Harvell, 1991). Most animal colonies are arranged with their
339 zooids in parallel to each other forming 2D sheets with one pole, typically the oral end,
340 exposed to the external environment. Some of these topologically simple planar
341 colonies can form complex 3D shapes by folding this sheet. In benthic species, the
342 sheet is often folded around an endogenous skeleton or an object in the environment
343 (e.g., corals, ascidians, millepores). Bryozoans also tend to develop into sheets though
344 also into branching structures. In pelagic species, colonies are free-living and capable of
345 swimming around by the combined (and often coordinated) action of their zooids (Du
346 Clos et al. 2022). Therefore, their shapes are often directional, with a front and a rear

347 end defined by their colonial locomotion (Mackie, 1986). On one hand, pyrosomes
348 (Chordata: Tunicata) use the same 2D-sheet template as their benthic relatives, yet in
349 their case, the sheet grows folded forming a closed-ended tube where all the exhalant
350 flow from the inner side aboral ends of the zooids are canalized to a single jet stream.
351 On the other hand, siphonophores and doliolids typically form 1D colonies with sub-
352 specialized zooid types, where only one or few frontal locomotory zooids (nectophores
353 and nurse zooid, respectively) propel a linear colony with non-swimming zooids
354 dragging behind. Siphonophore (Cnidaria: Hydrozoa) colonies can be topologically
355 complex in benthic rhodaliids or in the pleustonic Portuguese Man-o-war, but most
356 planktonic free-swimming siphonophore colonies have their zooids arranged bi-serially
357 or mono-serially along a stem (Mackie et al., 1988). Among them, physonect
358 siphonophores bear multiple nectophores (swimming bodies) that propel the colony
359 through multijet propulsion (Sutherland et al. 2019) in a similar fashion to linear salp
360 colonies. Compared to siphonophores or pyrosomes, salps present a much broader set
361 of architectural configurations among free-swimming colonial animals (Madin, 1990),
362 thus expanding the boundaries of our known design space for both form and function of
363 coloniality in the pelagic realm.

364

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373

374 **Literature Cited**

375 Alexander, W., 1968, September. A discussion of governing decelerator performance
376 and design parameters in the supersonic flight regime. In *2nd Aerodynamic
377 Deceleration Systems Conference* (p. 938).

378 Bard, J. B., & Rhee, S. Y., 2004. Ontologies in biology: design, applications and future
379 challenges. *nature reviews genetics*, 5(3), 213-222.

380 Bi, X., Tang, H., & Zhu, Q., 2022. Feasibility of hydrodynamically activated valves for
381 salp-like propulsion. *Physics of Fluids*, 34(10), 101903.

382 Bone, Q., 1998. *The biology of pelagic tunicates*.

383 Bone, Q. and Trueman, E.R., 1983. Jet propulsion in salps (Tunicata: Thaliacea).
384 *Journal of Zoology*, 201(4), pp.481-506.

385 Buesseler, K. O., Boyd, P. W., Black, E. E., & Siegel, D. A., 2020. Metrics that matter for
386 assessing the ocean biological carbon pump. *Proceedings of the National
387 Academy of Sciences*, 117(18), 9679-9687.

388 Chao, S., Guan, G., & Hong, G. S., 2017, September. Design of a finless torpedo
389 shaped micro AUV with high maneuverability. In *OCEANS 2017-Anchorage* (pp.
390 1-6). IEEE.

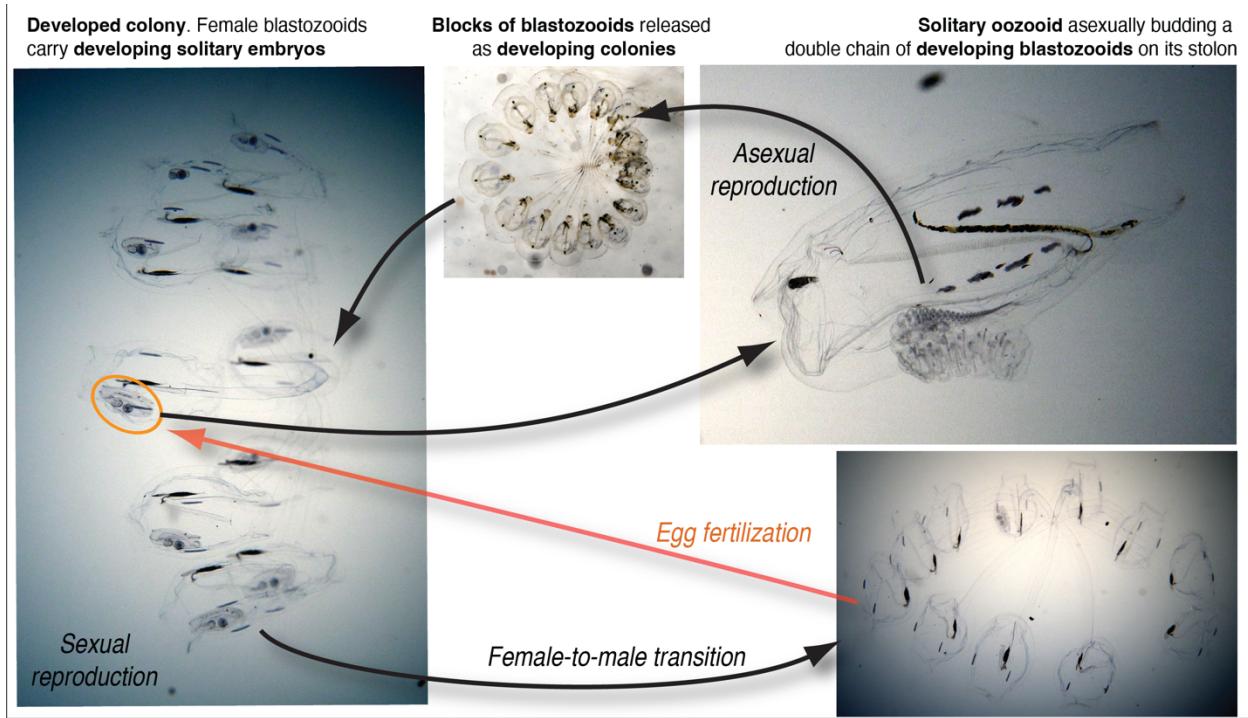
391 Colin, S. P., Gemmell, B. J., Costello, J. H., & Sutherland, K. R. (2022). In situ high-
392 speed brightfield imaging for studies of aquatic organisms. *Protocolio*.

- 393 Costello, J. H., Colin, S. P., Gemmell, B. J., Dabiri, J. O., & Sutherland, K. R., 2015.
- 394 Multi-jet propulsion organized by clonal development in a colonial siphonophore.
- 395 *Nature communications*, 6(1), 8158.
- 396 Décima, M., Stukel, M. R., Nodder, S. D., Gutiérrez-Rodríguez, A., Selph, K. E., Dos
397 Santos, A. L., ... & Pinkerton, M., 2023. Salp blooms drive strong increases in
398 passive carbon export in the Southern Ocean. *Nature communications*, 14(1),
399 425.
- 400 Du Clos, K. T., Gemmell, B. J., Colin, S. P., Costello, J. H., Dabiri, J. O., & Sutherland,
401 K. R., 2022. Distributed propulsion enables fast and efficient swimming modes in
402 physonect siphonophores. *Proceedings of the National Academy of Sciences*,
403 119(49), e2202494119.
- 404 Giorgio-Serchi, F., & Weymouth, G. D., 2017. Underwater soft robotics, the benefit of
405 body-shape variations in aquatic propulsion. In *Soft Robotics: Trends,*
406 *Applications and Challenges: Proceedings of the Soft Robotics Week, April 25-*
407 *30, 2016, Livorno, Italy* (pp. 37-46). Springer International Publishing.
- 408 Govindarajan, A.F., Bucklin, A. and Madin, L.P., 2011. A molecular phylogeny of the
409 Thaliacea. *Journal of Plankton Research*, 33(6), pp.843-853.
- 410 Harvell, C.D., 1991. Coloniality and inducible polymorphism. *The American Naturalist*,
411 138(1), pp.1-14.
- 412 Henschke, N., Everett, J. D., Richardson, A. J., & Suthers, I. M., 2016. Rethinking the
413 role of salps in the ocean. *Trends in Ecology & Evolution*, 31(9), 720-733.

- 414 Henschke, N., Cherel, Y., Cotté, C., Espinasse, B., Hunt, B. P., & Pakhomov, E. A.,
415 2021. Size and stage specific patterns in *Salpa thompsoni* vertical migration.
416 *Journal of Marine Systems*, 222, 103587.
- 417 Krummel, G. M., 2019. *Locomotion and Control of Cnidarian-Inspired Robots* (Doctoral
418 dissertation, Virginia Tech).
- 419 Lavaniegos, B. E., & Ohman, M. D., 2003. Long-term changes in pelagic tunicates of
420 the California Current. *Deep Sea Research Part II: Topical Studies in*
421 *Oceanography*, 50(14-16), 2473-2498.
- 422 Mackie, G.O., 1986. From aggregates to integrates: physiological aspects of modularity
423 in colonial animals. *Philosophical Transactions of the Royal Society of London.*
424 *B, Biological Sciences*, 313(1159), pp.175-196.
- 425 Mackie, G.O., Pugh, P.R. and Purcell, J.E., 1988. Siphonophore biology. In *Advances in*
426 *Marine biology* (Vol. 24, pp. 97-262). Academic Press.
- 427 Madin, L.P., 1990. Aspects of jet propulsion in salps. *Canadian Journal of Zoology*,
428 68(4), pp.765-777.
- 429 Madin, L. P., Kremer, P., & Hacker, S., 1996. Distribution and vertical migration of salps
430 (Tunicata, Thaliacea) near Bermuda. *Journal of Plankton Research*, 18(5), 747-
431 755.
- 432 Sutherland, K. R., Gemmell, B. J., Colin, S. P., & Costello, J. H., 2019. Propulsive
433 design principles in a multi-jet siphonophore. *Journal of Experimental Biology*,
434 222(6), jeb198242.
- 435 Marut, K. J., 2014. *Underwater Robotic Propulsors Inspired by Jetting Jellyfish* (Doctoral
436 dissertation, Virginia Tech).

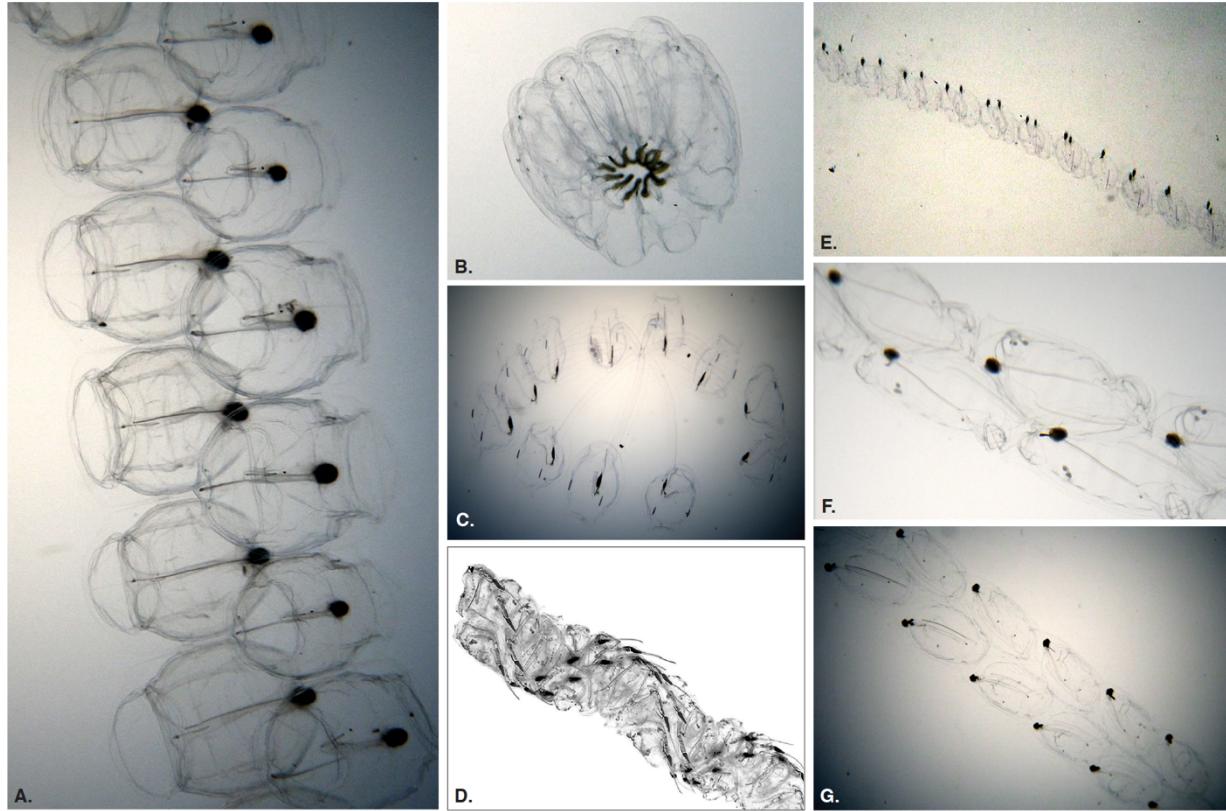
- 437 Mohensi, K., 2006. Pulsatile vortex generators for low-speed maneuvering of small
438 underwater vehicles. *Ocean Eng.* 33, 2209–2223.
- 439 Oxford Languages, 2023. Oxford Languages and Google - English. Oup. com.
440 <https://languages.oup.com/google-dictionary-en/>
- 441 Renda, F., Serchi, F. G., Boyer, F., & Laschi, C., 2015. Structural dynamics of a pulsed-
442 jet propulsion system for underwater soft robots. *International Journal of
443 Advanced Robotic Systems*, 12(6), 68.
- 444 Ritter, W. E., & Johnson, M. E., 1911. The growth and differentiation of the chain of
445 *Cyclosalpa affinis* Chamisso. *Journal of Morphology*, 22(2), 395-453.
- 446 Steinberg, D. K., Stamieszkin, K., Maas, A. E., Durkin, C. A., Passow, U., Estapa, M. L.,
447 ... & Siegel, D. A., 2023. The Outsized Role of Salps in Carbon Export in the
448 Subarctic Northeast Pacific Ocean. *Global Biogeochemical Cycles*, 37(1),
449 e2022GB007523.
- 450 Sutherland, K. R., & Weihs, D., 2017. Hydrodynamic advantages of swimming by salp
451 chains. *Journal of The Royal Society Interface*, 14(133), 20170298.
- 452 Sutherland, K. R., Gemmell, B. J., Colin, S. P., & Costello, J. H., 2019. Maneuvering
453 performance in the colonial siphonophore, *Nanomia bijuga*. *Biomimetics*, 4(3),
454 62.
- 455 Yue, C. et al., 2015. Mechantronic system and experiments of a spherical underwater
456 robot: SUR-II. *J. Intell. Robot Syst.* doi:10.1007/s10846-015-0177-3.
- 457
- 458 **Tables**
- 459

460 **Figure Legends**



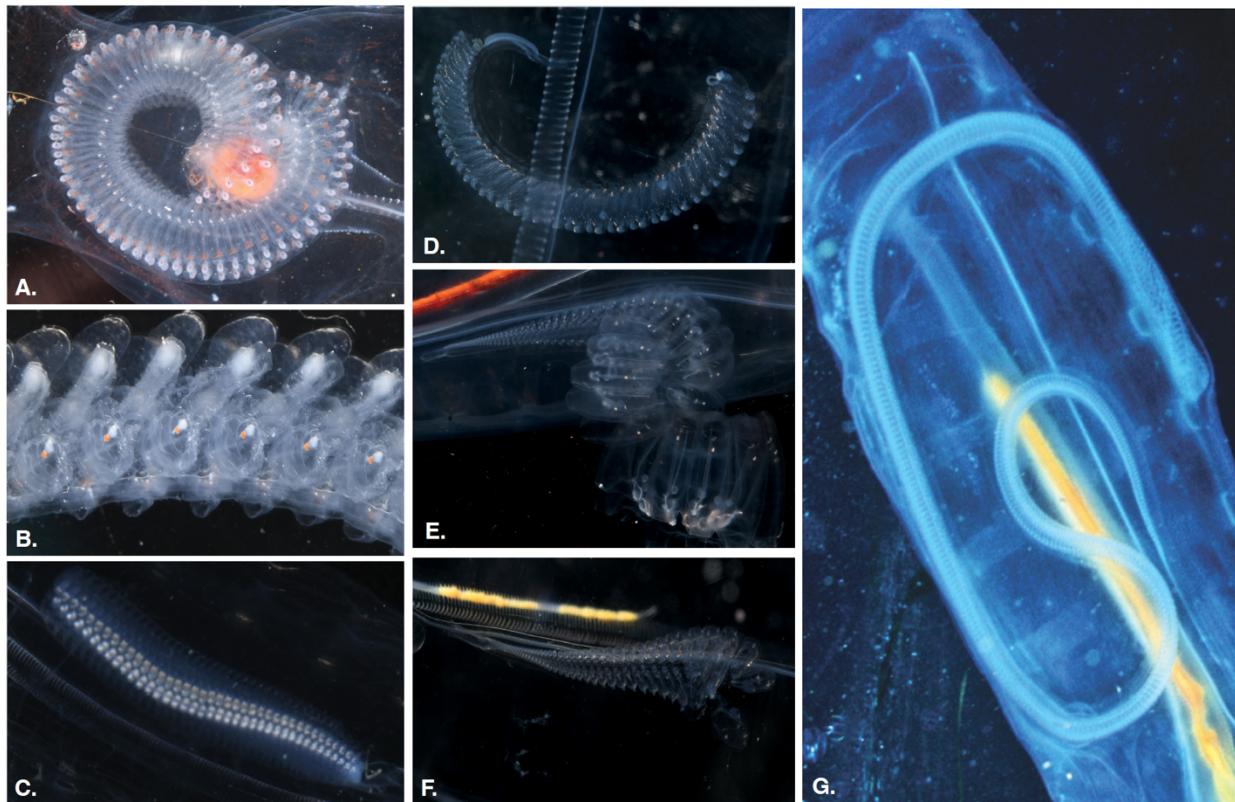
461

462 Figure 1. Salp life cycle using the species *Cyclosalpa sewelli* as an example. Frame
463 captures from brightfield in situ videos by Brad Gemmell following method from Colin et
464 al. 2022.



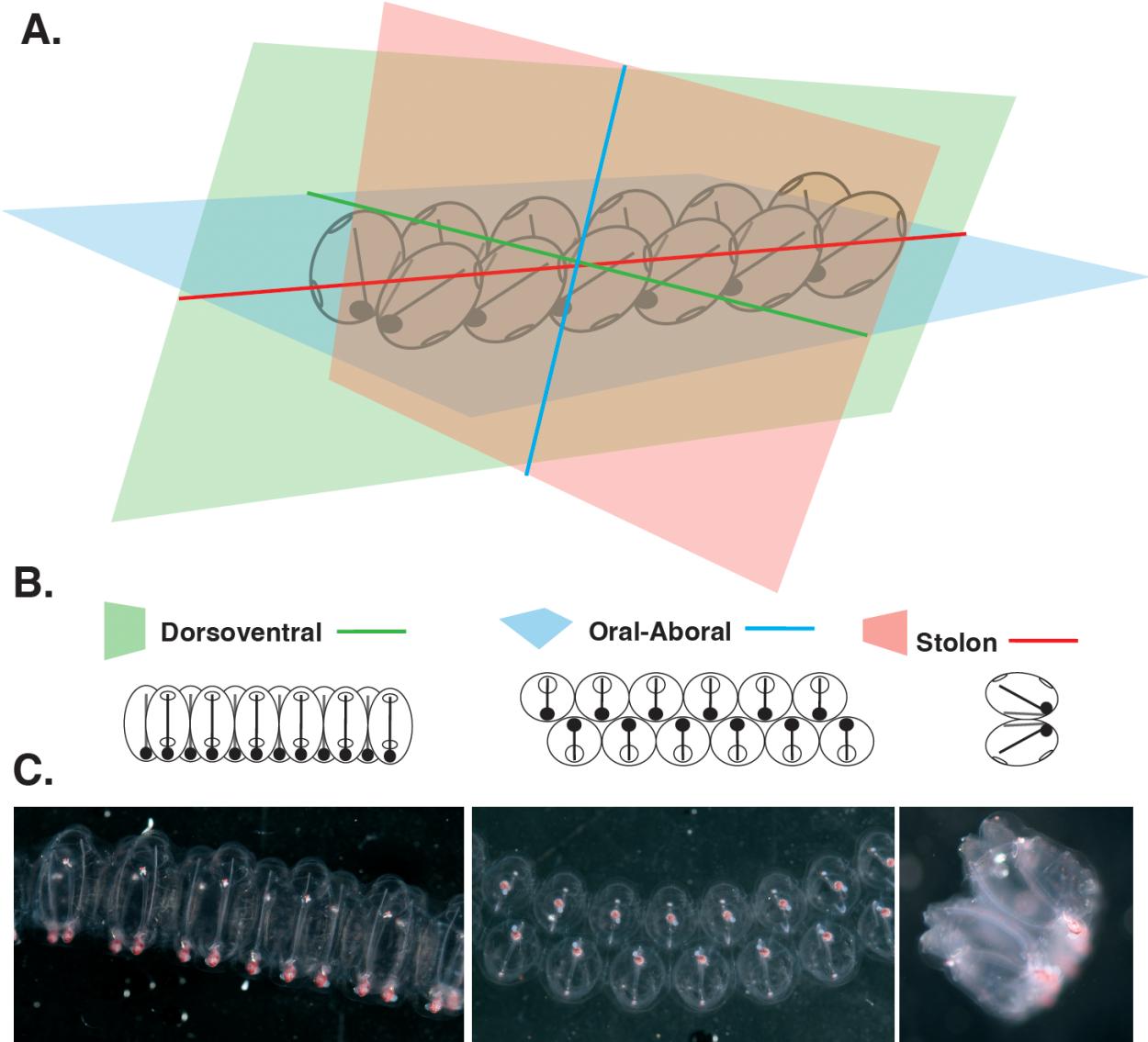
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466 Figure 2. Adult salp colonies representing every distinct colonial architecture observed
 467 across all salp species. (A) transversal chain (*Pegea* sp.), (B) whorl (*Cyclosalpa affinis*),
 468 (C) cluster (*Cyclosalpa sewelli*), (D) helical chain (*Helicosalpa virgula* (Vogt, 1854),
 469 photograph by Nils Aukan), (E) oblique chain (*Thalia longicauda* (Quoy & Gaimard,
 470 1824)), (F) bipinnate chain (*Ritteriella amboinensis* (Apstein, 1904)). Frame captures A,
 471 B, C, E, F, G from brightfield in situ videos by Brad Gemmell.



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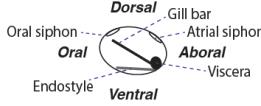
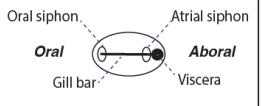
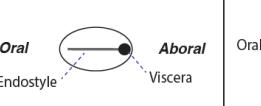
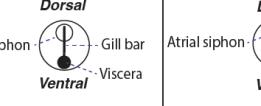
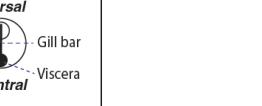
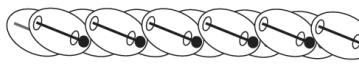
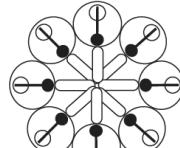
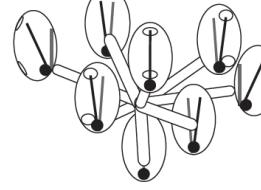
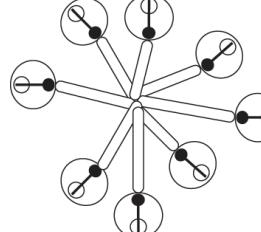
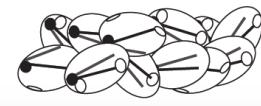
473 Figure 3. Developing blastozooid colonies produced by the budding stolons of solitary
 474 oozoids across different salp architectures. (A) transversal chain buds (*Pegea* sp.), (B)
 475 oblique chain buds (*Thetys vagina*), (C) linear chain buds (*Iasis cylindrica*), (D)
 476 bipinnate chain buds (*R. amboinensis*), (E) whorl buds (*C. affinis*), (F) cluster buds
 477 (*Cyclosalpa polae*), (G) helical chain buds (*H. virgula*, photograph by David Wrobel).



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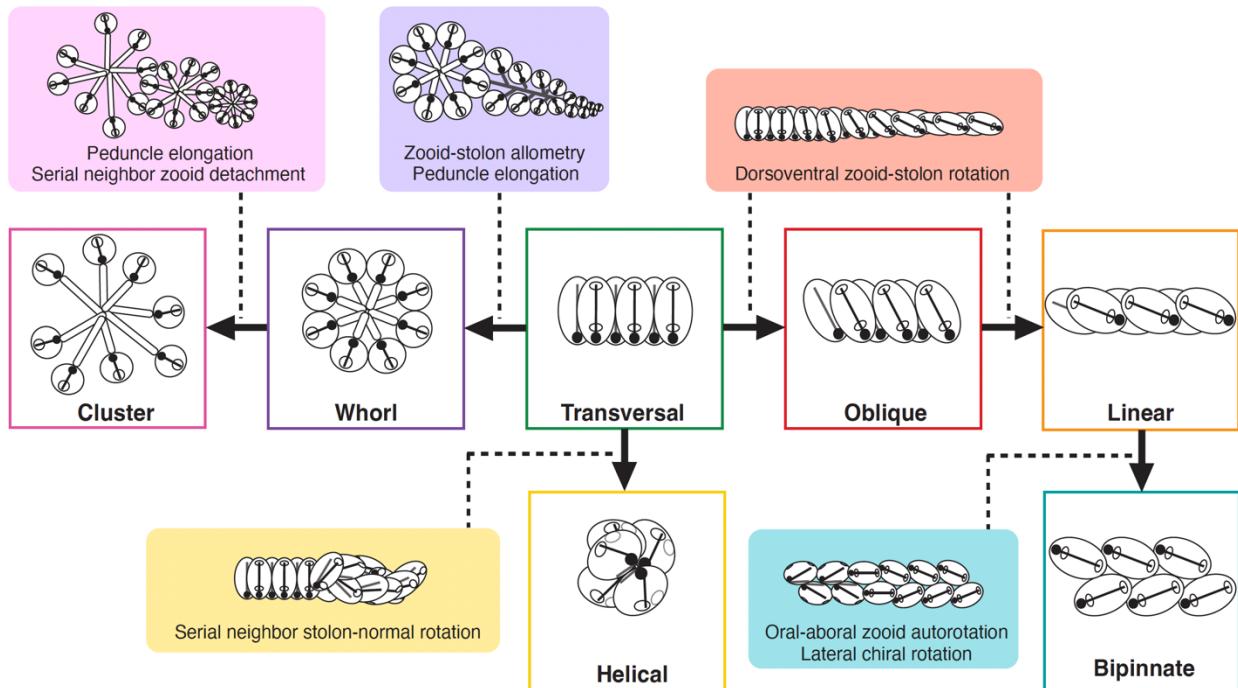
479 Figure 4. (A) Definition of the homologous universal axes and planes of observation
 480 relative to the orientation of the colony in the early transversal chain stage. (B)
 481 Diagrams representing the cross-sectional views of a transversal chain from each of
 482 these three planes of observation. (C) Photographs of a transversal chain of early-
 483 developing *Pegea* sp. blastozoids taken from each of the abovementioned
 484 orientations.

485

Zoid planes of observations				
Lateral	Dorsal	Ventral	Oral	Aboral
				
Colony architecture	Ontogenetically-homologous planes of colony observation			Stolon
Transversal	 Zoid-Stolon angle: 90°			
Oblique	 Zoid-Stolon angle: 40-70°			
Linear	 Zoid-Stolon angle: 0-30°			
Bipinnate	 Zoid-Stolon angle: 0-30°			
Whorl	 Zoid-Stolon angle: 90°			
Cluster	 Zoid-Stolon angle: 90°			
Helical				

486

487 Figure 5. Sketches of individual zooids and adult colonies representative of every
 488 architecture as viewed from each plane of observation.



489

490 Figure 6. Developmental transition pathways and mechanisms leading to the different
 491 adult blastozoooid colony architectures. The transversal architecture is found in the
 492 earliest developmental stage of every species as well as in the adult stage of some
 493 species.

494

495 **Ethical Care Considerations**

496 Our specimen collection and protocol were compliant with all local regulations. Since no
 497 vertebrates or cephalopods were involved, we did not need oversight from an animal
 498 care board.

499

500 **Data Accessibility**

501 All photographs will be made available in a Dryad repository.