SUPPLEMENTARY FIGURES

Cell differentiation and morphogenesis in the colony-forming choanoflagellate *Salpingoeca rosetta*

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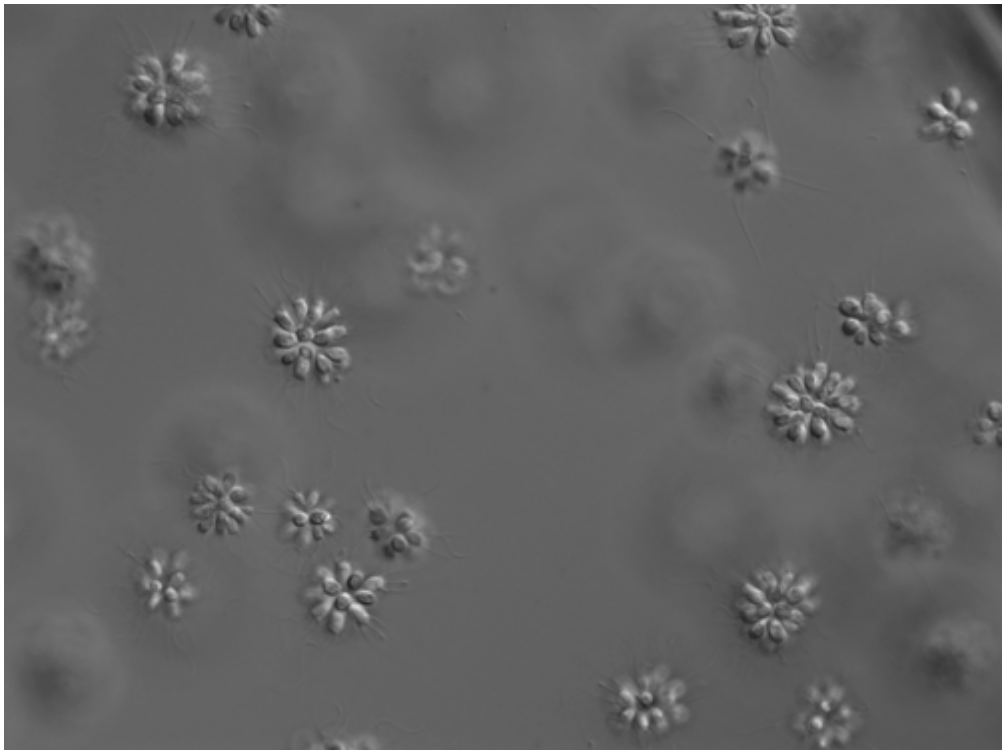


Figure S1: *S. rosetta* rosette colonies dominate in a monoxenic culture grown exclusively with *A. machipongonensis*

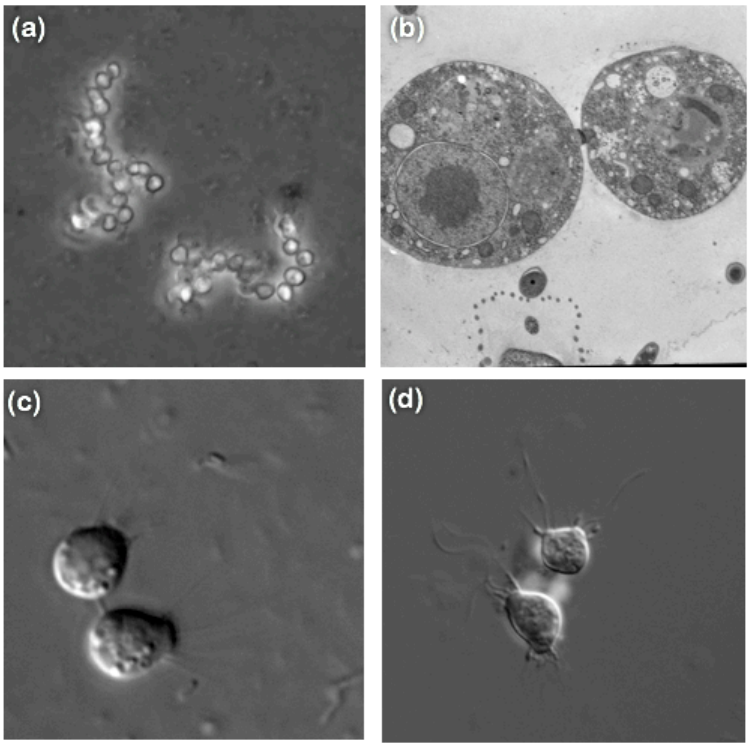


Figure S2: *S. rosetta* chain colony morphology, showing ECM and intercellular bridges **(a)** Longer chain colonies (~20 cells) are often branched **(b)** TEM of intercellular bridge attaching adjacent cells in chain colony **(c)** Intercellular bridges are especially visible between pairs of cells. **(d)** Chain colonies in rapidly growing culture often contain large cells with multiple flagella and collars (up to 4 per cell)

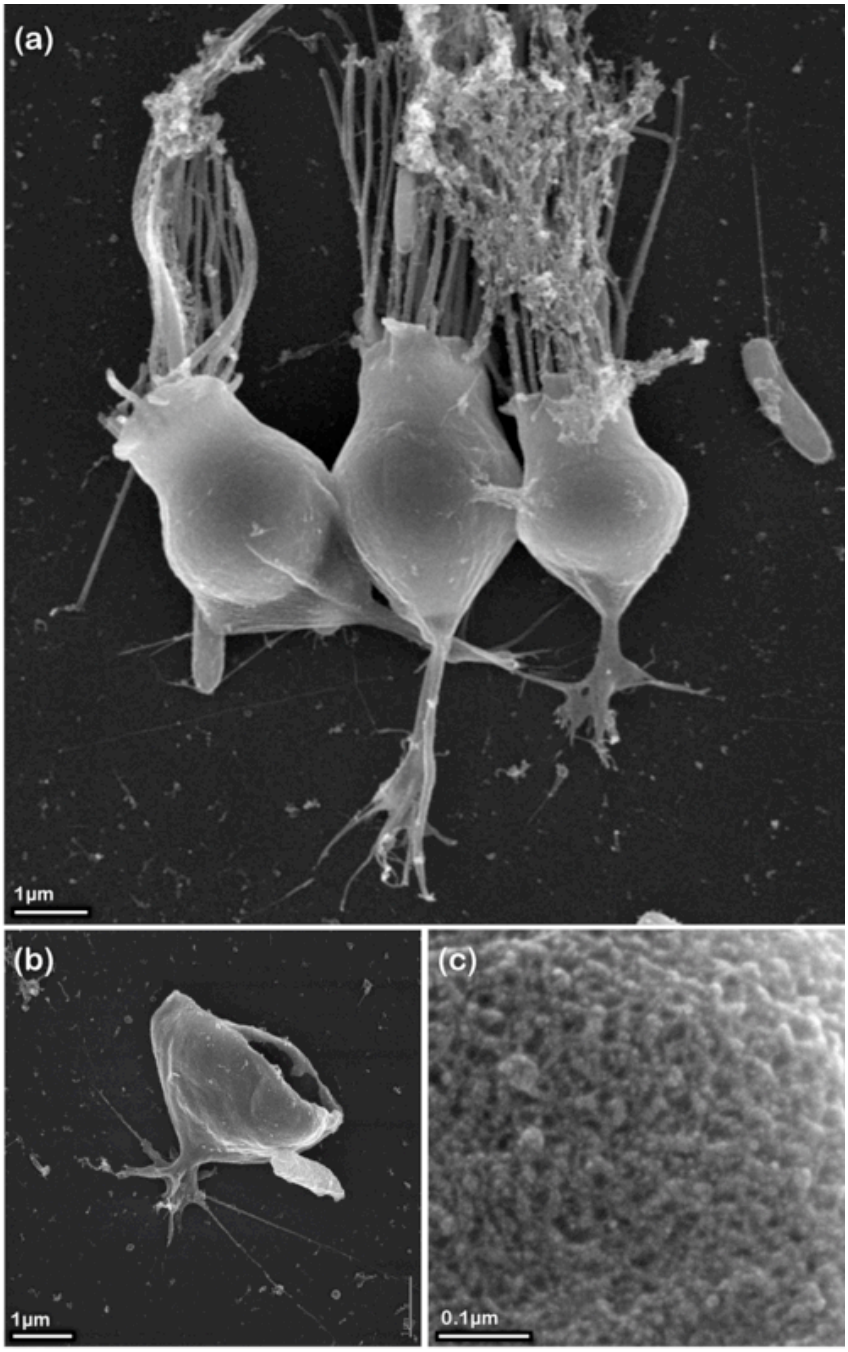


Figure S3: Theca morphology. **(a)** SEM of typical thecate cells. Stalks are tubes continuous with the thecal cup, extending down to splay out at the base and attach to the substrate. (note: the two cells on the right are not connected; the junction-like extension between them is debris) **(b)** Length of stalk varies and the thecal cup is capable of rooting directly onto substrate **(c)** High magnification view shows tightly woven texture of theca.

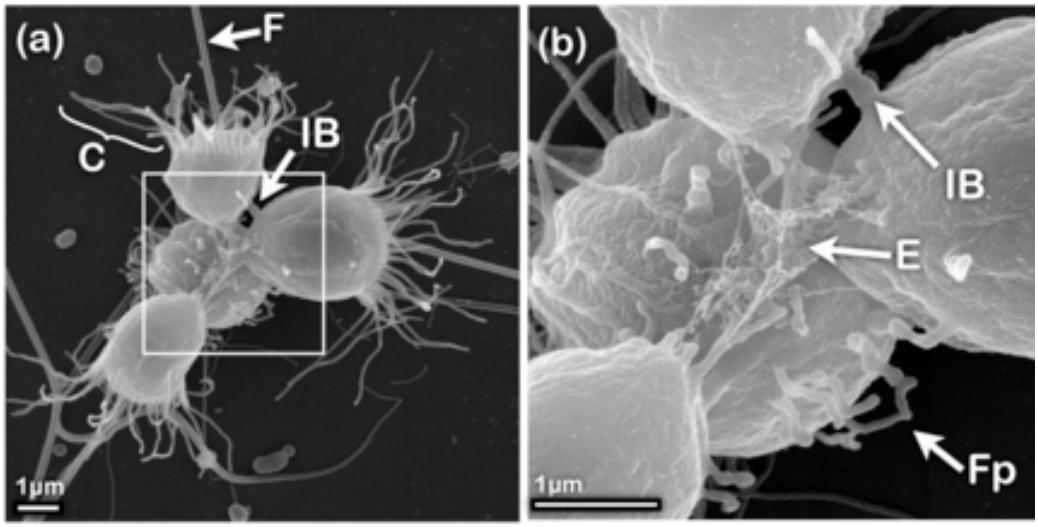
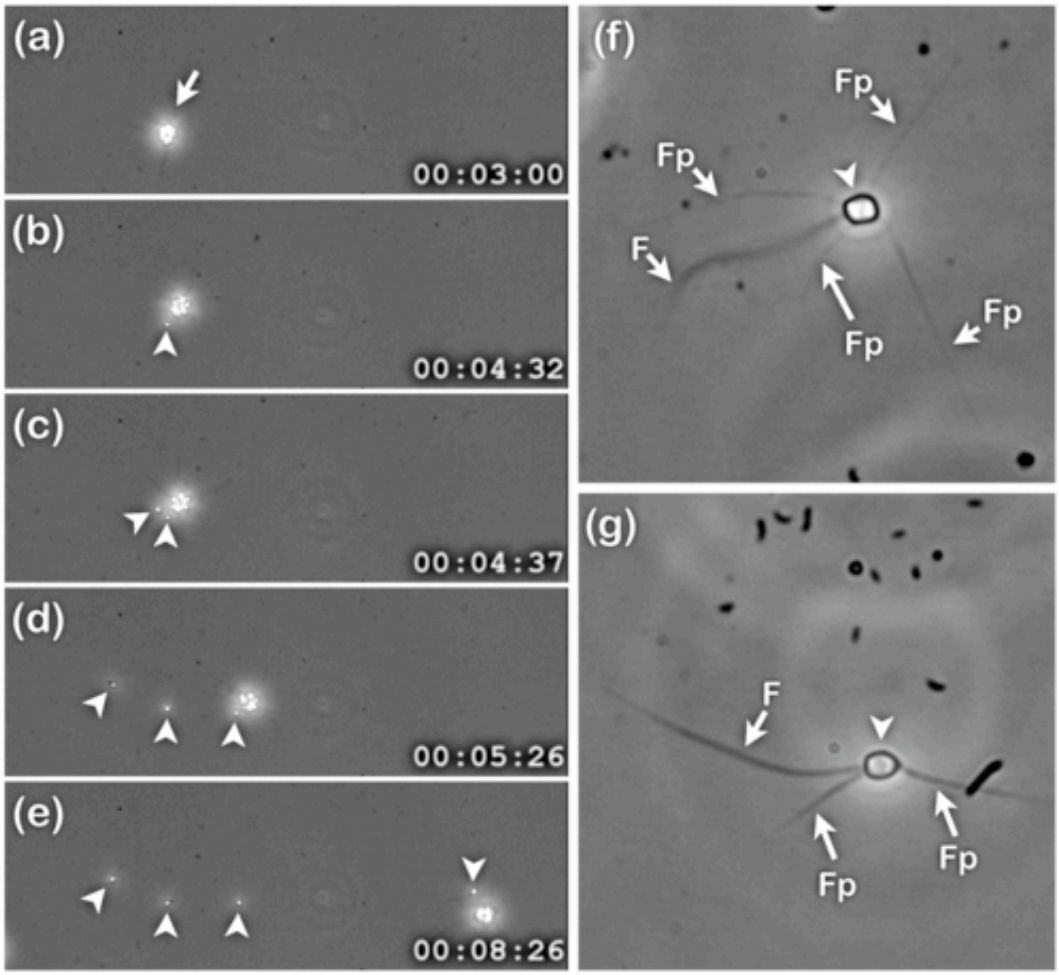


Figure S4: *S. rosetta* rosette colony core revealed using room temperature glutaraldehyde fixation, which removes most of the obscuring ECM. **(a)** Center of the colony shows intercellular bridge and filopodia extending into small amount of residual ECM at the colony core. **(b)** Inset from (a)**.**



S5: Minute cells (a-e) Timecourse showing rosette colony (arrow) ejecting minute cells (arrowheads), which adhere to the coverslip. See Supplemental Movie 5. (f,g) Minute cells attach to coverslip via long filopodia (Fp). Beating flagellum (F) is visible but cells appear to lack collars.

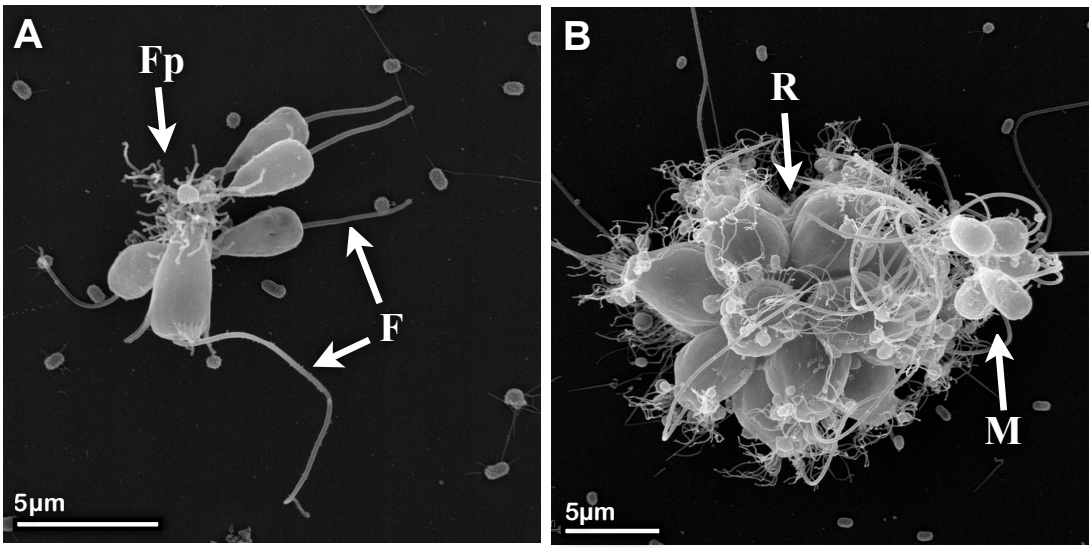


Figure S6: Minute cells form colonies that are distinct from rosette colonies. (a) Colony of tiny cells (b) Rosette colony `R' and a much smaller colony of minute cells `M.' F: flagellum, Fp: filopodia.

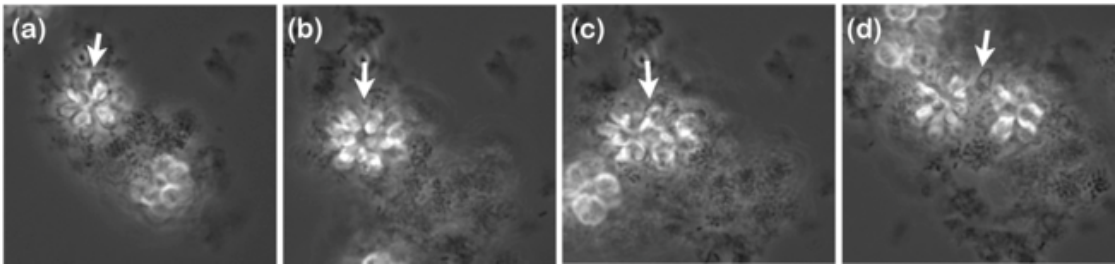


Figure S7: *S. rosetta* rosette colonies reproduce by fission. Cleavage plane indicated by arrow. See Supplemental Movie 6.

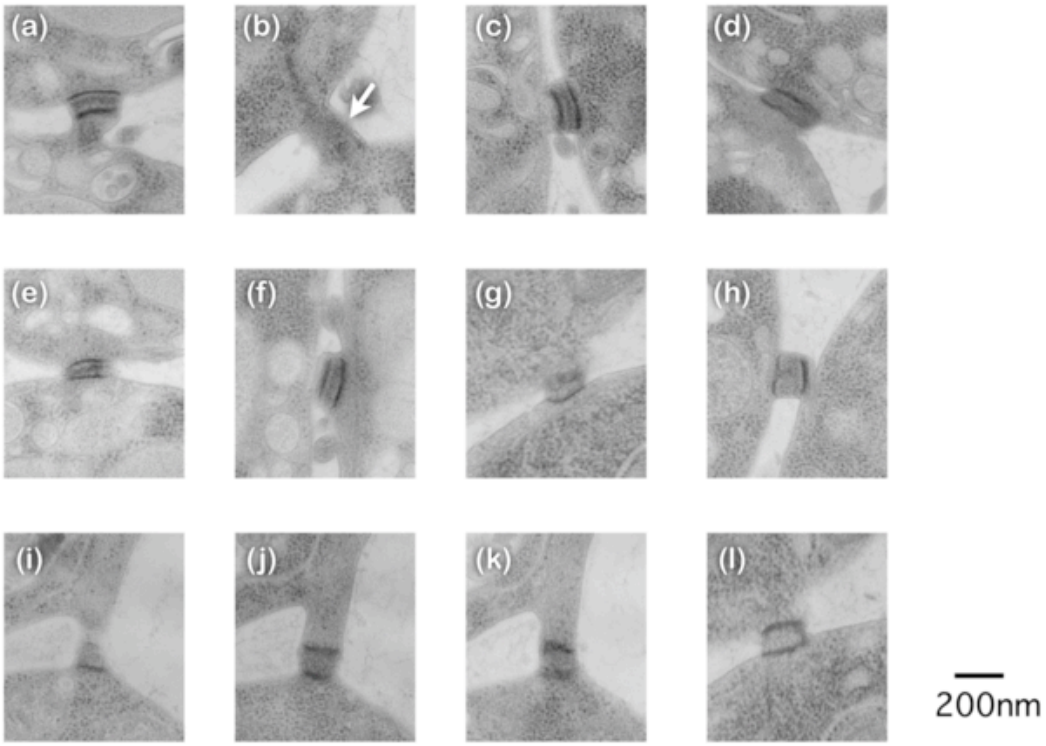


Figure S8: Examples of intercellular bridges. In all cases except (b), two plates were observed occluding the bridge

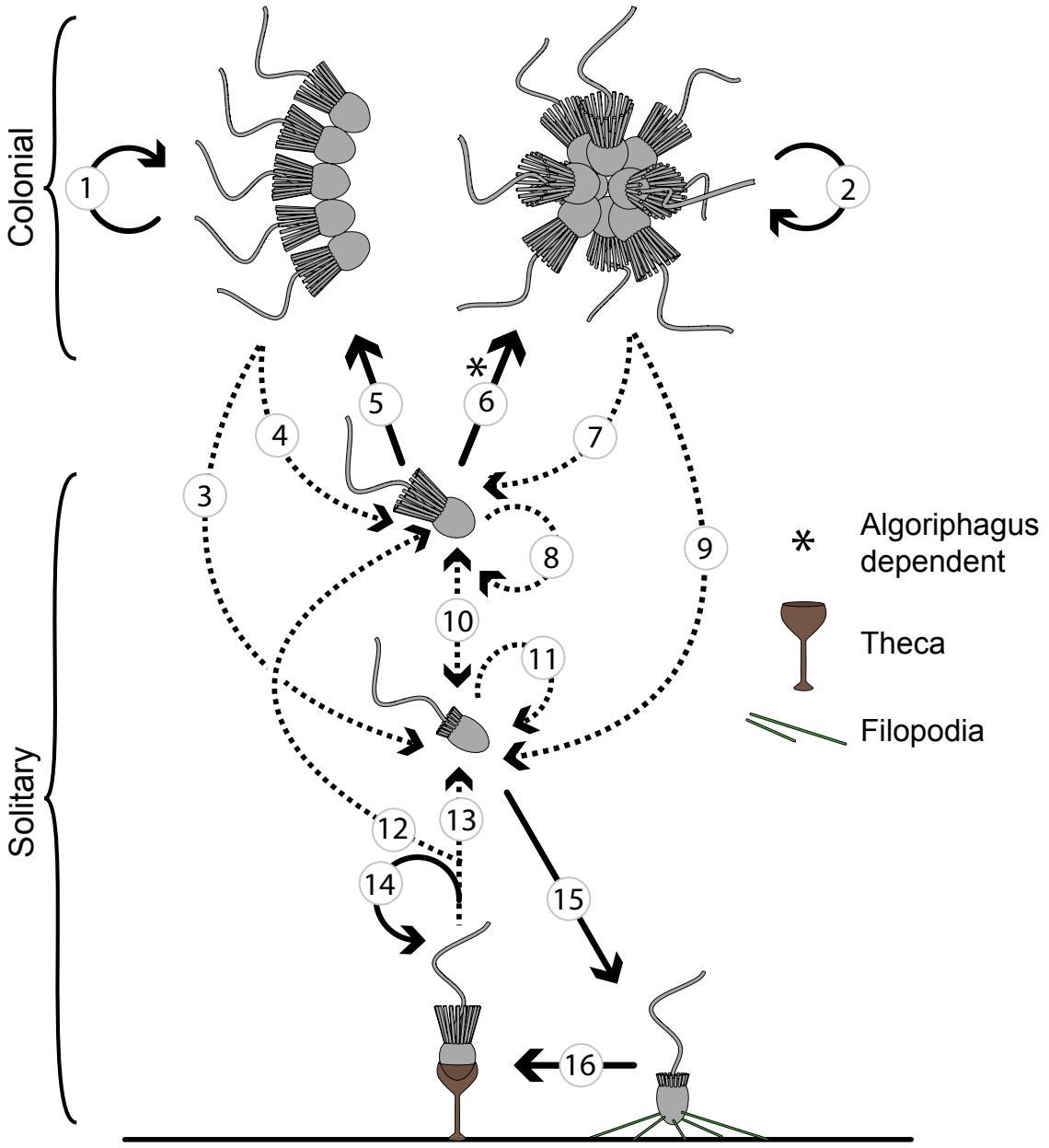


Figure S9: Model of transitions between cell morphs. The slow swimmer sits at the hub of the *S. rosetta* life history and can convert into chain colonies (5) or rosette colonies (6) through serial division coupled with incomplete cytokinesis. Rosettes only form in the presence of a signal from *Algoriphagus* bacteria. It is presumed that slow swimmers can also divide to produce more slow swimmers (8) or differentiate into fast swimmers (10), although neither process has been directly observed. Fast swimmers settle on surfaces (15) and differentiate into thecate cells (16) without requiring an intervening cell division. After thecate cells divide, one daughter cell remains in the theca (14) while the other (12 or 13) swims away. In addition, thecate cells can abandon their theca (12 or 13). It is not known whether thecate cells produce fast or slow swimmers following cell division or theca abandonment. While fast swimmers do not divide during differentiation into thecate cells, it is possible that they may divide in the water column (11). Chain and rosette colonies can each propagate through a process of fission (1 and 2, respectively). Chain and rosette

colonies are also observed to release individual swimming cells on occasion (3, 4, 7, 9), although it is not clear whether they are fast or slow swimmers. Solid lines indicate transitions that have been directly observed. Dotted lines indicate plausible transitions that have not been directly observed.