

De novo origin of multicellularity in response to predation



M. D. Herron^{1*}, J. C. Boswell¹, J. M. Borin¹, C. A. Knox²,
M. Boyd², W. C. Ratcliff¹, and F. Rosenzweig¹

¹Georgia Institute of Technology; ²University of Montana; *xprinceps@gmail.com

Georgia Tech School of Biological Sciences

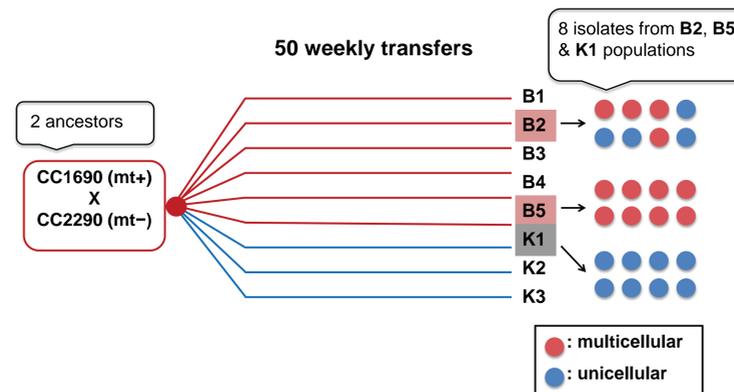
Abstract

The transition from unicellular to multicellular life was one of a few major events in the history of life that created new opportunities for more complex biological systems to evolve. Thus far, studying the proximate and ultimate causes of the resulting increases in complexity has been a major challenge in evolutionary biology. Traditionally, questions related to the emergence of multicellularity have been addressed retrospectively, through comparative studies of extant unicellular and multicellular lineages. Experimental microbial evolution allows for prospective studies that observe evolution in real time. In this study, we report the de novo origin of simple multicellularity in response to predation. We subjected outcrossed populations of the unicellular green alga *Chlamydomonas reinhardtii* to selection by the filter-feeding predator *Paramecium tetraurelia*. Two of five experimental populations evolved multicellular structures not observed in any of the three unselected control populations. Colonies consist of 4-16 cells enclosed within the cell wall of the maternal cell, and cells are encased within a transparent extracellular matrix. The highly structured, spheroidal colonies that evolved in this experiment are reminiscent of volvocine algae such as *Eudorina elegans*. These algae represent a completely novel origin of multicellularity, as *C. reinhardtii* has never had multicellular ancestors.^{1,2}

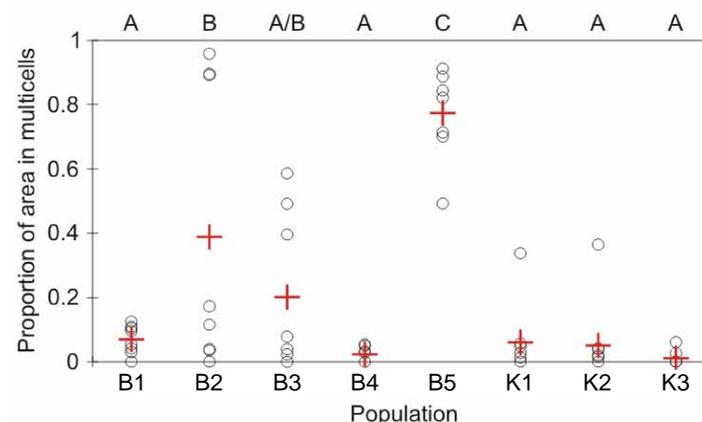
Introduction

- The evolution of multicellularity set the stage for unprecedented increases in biological complexity.
- Multicellularity has evolved independently in land plants, animals, fungi, red algae, green algae, brown algae, and several other taxa.
- Retrospective approaches to understanding the evolution of multicellularity have been productive but have limitations.
- Previous experiments have shown that multicellularity can evolve quickly under strong selection.³⁻⁵
- Experimental populations of the green alga *Chlamydomonas reinhardtii* were co-cultured with the filter-feeding predator *Paramecium tetraurelia*.
- After 50 weekly transfers, populations were plated and eight individual colonies isolated.

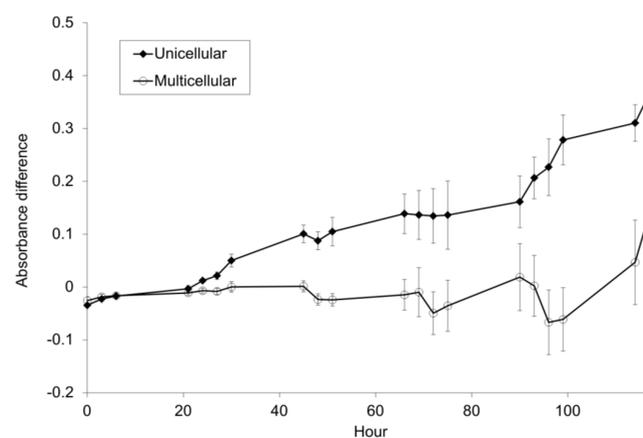
Methods



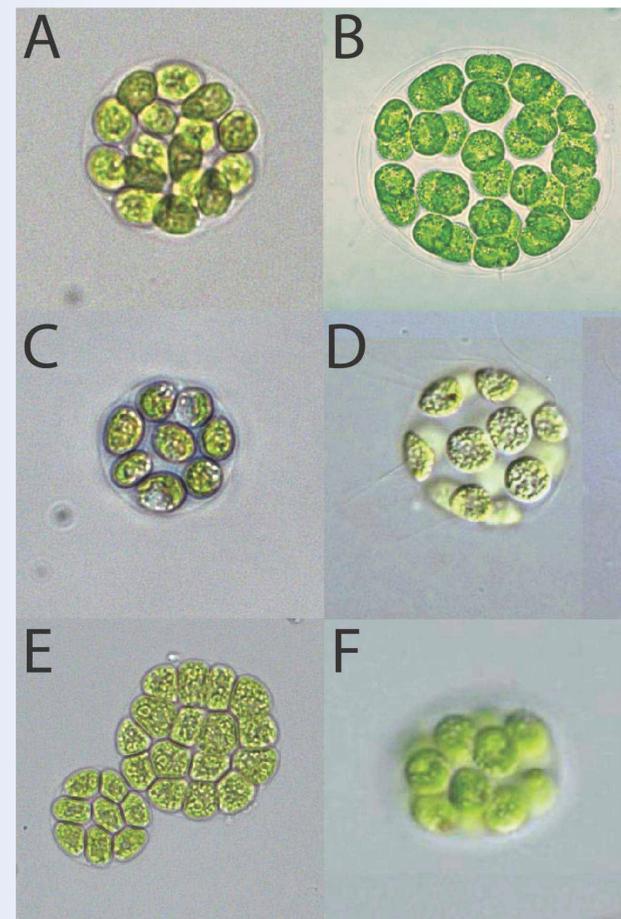
- Five experimental and three control populations of *Chlamydomonas reinhardtii* were founded from an F1 cross.
- Experimental populations were co-cultured with the filter-feeding predator *Paramecium tetraurelia*.
- After 50 weekly transfers, populations were plated and eight individual colonies isolated.



Proportion of cell image area in multicellular structures for experimental (E) and control (C) populations. Circles are individual clone measurements; red crosses are means of eight clones per population. Letters at top represent significant differences in population means (Tukey post-hoc comparisons, experiment-wise $\alpha = 0.05$).



Multicellularity protects algae from predation. Δ_{abs} values for evolved multicellular and unicellular *C. reinhardtii* strains under predation by the rotifer *Brachionus calyciflorus*. Multicellular strains averaged a much lower change in absorbance over the duration of the experiment than unicellular strains. Error bars = SEM of 4 replicates.



Naturally occurring and experimentally evolved volvocine algae. A, C, E: multicellular structures from the *Paramecium* predation experiment. B: *Eudorina elegans*. D: *Volvulina steinii*. F: *Yamagishiella unicocca*.

Conclusions

- Simple multicellular life cycles can evolve within a few hundred generations under strong selection.
- Evolved multicellular forms closely resemble extant volvocine algae.
- Predation is a plausible selective pressure that may have driven the evolution of multicellularity in some lineages.
- No chicken-and-egg problem exists: unicellular predators are sufficient to drive the evolution of multicellularity.

References

1. Herron MD, Michod RE (2008) *Evolution*, 62, 436–451.
2. Leliaert F et al. (2012) *Critical Reviews in Plant Sciences*, 31, 1–46.
3. Ratcliff WC et al. (2013) *Nature Communications*, 4, 2742.
4. Boraas ME et al. (1998) *Evolutionary Ecology*, 12, 153–164.
5. Becks L et al. (2012) *Ecology Letters*, 15, 492–501.