

Evolution of reproductive strategies in incipient multicellularity

Electronic Supplementary Material

Yuanxiao Gao¹, Yuriy Pichugin^{1,2}, Chaitanya S. Gokhale³, and Arne Traulsen^{1,*}

¹Department of Evolutionary Theory, Max Planck Institute for Evolutionary Biology,
August-Thienemann-Str. 2, 24306 Plön, Germany

²Current affiliation: Department of Ecology and Evolutionary Biology, Princeton University,
Princeton, New Jersey, United States of America

³Research Group for Theoretical Models of Eco-evolutionary Dynamics, Department of
Evolutionary Theory, Max Planck Institute for Evolutionary Biology,
August-Thienemann-Str. 2, 24306 Plön, Germany

*Corresponding author: Arne Traulsen, traulsen@evolbio.mpg.de

S1: The probability distribution of newborn organisms

We show the calculation of the probabilities of producing different types of newborn organisms from a mature organism (n_A, n_B) , where $n_A + n_B = N$. The probability to produce the newborn organism type (n'_A, n'_B) ($n'_A + n'_B < N$) is calculated by

$$P(n'_A, n'_B) = \frac{\binom{n_A}{n'_A} \binom{n_B}{n'_B}}{\binom{N}{n'_A + n'_B}} \quad (1)$$

We take the mature organism $(1, 2)$ in a population with reproductive strategy $2 + 1$ as an example. There are five newborn organisms: $(1, 0)$, $(0, 1)$, $(2, 0)$, $(1, 1)$ and $(0, 2)$. The probability of reproducing each newborn organism is shown in Fig. S1.

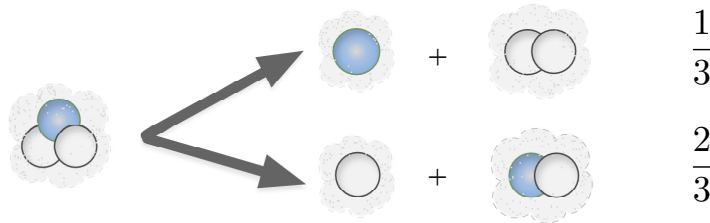


Figure S1: **The probability of producing each newborn organism from the mature organism $(1, 2)$ in the population with reproductive strategy $2 + 1$.** The organism $(1, 2)$ has the probability of $\frac{1}{3}$ to produce a newborn organism containing one A cell and a newborn organism containing two B cells. It has the probability of $\frac{2}{3}$ to produce a newborn organism containing one B cell and a newborn organism containing one B cell and one A cell. However, for small m , mixed mature groups occur only in small frequency in the long run.

S2: Population growth rate

We illustrate the calculation of population growth rates. For the reproductive strategy $n_1 + n_2 + \dots + n_M$ with maturity size N , its population consists of newborn organisms with size n_i , where $i = 1, \dots, M$, $0 < n_i < N$ and $\sum_{i=1}^M n_i = N$. As we consider two cell types, B cell and A cell, an organism with size n_i can have $0, 1, \dots, n_i$ B cells. Therefore, a newborn organism with n_i cells has $n_i + 1$ possible compositions. We denote the number of newborn organism types of a population by Ω . For example, a population with reproductive strategy $2 + 1$ can contain the newborn organisms $(1, 0)$, $(0, 1)$, $(2, 0)$, $(1, 1)$ and $(0, 2)$. Here, we would have $N = 3$, $n_1 = 1$, $n_2 = 2$, $M = 2$ and $\Omega = 5$ (see Fig.1D). The population growth rate depends on the growth rate of the newborn organisms. We assume that a population contains each type of newborn organisms initially. We track each newborn organism's growth time and the number of its offspring. We use T_{ij} to denote the growth time of a i type newborn organism until it produces a j type newborn organism, where $i, j = 1, \dots, \Omega$. We use N_{ij} to denote the number of offspring of type j offspring produced by the i type newborn organism. The growth time T_{ij} depends on the organism size and the organism composition via Eq.(1) in the main text. The number of newborn organism N_{ij} depends on the cell-type switching probability and the cell division probabilities of each cell type. Since organism growth is stochastic, T_{ij} and N_{ij} are different for different stochastic trajectories, see [Gao et al., 2019]. For example, for the strategy $1 + 1$, the newborn organism $(0, 1)$ could produce two $(1, 0)$, one $(1, 0)$ or zero $(1, 0)$ with different growth time. To capture the different development trajectories, we simulate the stochastic organism growth and average over Z replicates. Then the population growth rate is the largest root of the equation

$$\det(\mathbf{A}_{\Omega\Omega}(\lambda) - \mathbf{I}) = 0, \quad (2)$$

where $A_{\Omega\Omega}$ is a Ω by Ω matrix with elements $a_{ij} = \frac{\sum_{z=1}^Z N_{ij}^z e^{-\lambda T_{ij}^z}}{Z}$ [De Roos, 2008; Gao et al., 2019]. Here, T_{ij}^z and N_{ij}^z are the growth time and the number of offspring of the newborn organism of size i producing an j organism in z th replication.

The simulation of a population starts with newborn organisms. The newborn organisms differ in their composition, i.e. they have different (n_A, n_B) . For example, for the reproductive strategy $1 + 1$, the newborn organisms are of type $(1, 0)$ and $(0, 1)$. Organisms grow in the following way: In each single step, a cell (B cell or A cell) is selected to divide with its division probability, see Eq.(3) in the main text. The threshold component of growth time is $t_{gn} = \left(\frac{n_A e^{w_{FB}} + n_B e^{w_{FB}}}{n_A + n_B} \right)^{-1}$ based on Eq.(4) in the main text. The increment time for the single step is $t_{sn} \times t_{gn}$, where we assign values to t_{sn} according to different scenarios. With the cell division, two daughter cells are produced. Each daughter cell switches to another cell type with a probability m . After a single step, we update the number of B cells and A cells of the organism. Then, the organism repeats the above procedure to grow until reaching its maturity size. Organisms at maturity size produce offspring by random fragmentation. The probability of producing each newborn organism is calculated by Eq. (1) in S1. We obtain the number of offspring produced by the newborn organisms and the growth time (the sum of all time increments) in a single run. We make 5000 replicates of the life cycle of each newborn organism. In the z th replication, we record the growth time T_{ij}^z and the number of offspring N_{ij}^z for the j type newborn organism producing the i type newborn organism. Thus, we have $a_{ij} = \frac{\sum_{z=1}^Z N_{ij}^z e^{-\lambda T_{ij}^z}}{Z}$, where $Z = 5000$ for our simulations. We numerically recover our analytical results for maturity size $N \leq 3$, see Appendix . For $N \leq 3$, we show that that only the binary-splitting reproductive strategies are uniquely optimal under size effects only in Appendix . Our remaining conclusions are reached by numerical simulations.

S3: Analytical proof that smaller χ_n determines the optimal reproductive strategy when $N \leq 3$

For $N \leq 3$, there are only three reproductive strategies: $1 + 1$, $1 + 1 + 1$ and $2 + 1$. The optimal reproductive strategy is determined by the perturbation with the smaller χ_n . More precisely, the reproductive strategy $1 + 1$ is optimal when $\chi_1 < \chi_2$ (advantageous perturbation at $n = 1$) and $2 + 1$ is optimal when $\chi_1 > \chi_2$ (advantageous perturbation at $n = 2$). $1 + 1$, $1 + 1 + 1$ and $2 + 1$ are optimal when $\chi_1 = \chi_2$. The population growth rate of each reproductive strategy is denoted by a subscript. For example, λ_{1+1} describes the population growth rate of the reproductive strategy $1 + 1$. The three population growth rates λ_{1+1} , λ_{1+1+1} , and λ_{2+1} can be calculated by finding the largest eigenvalue of matrix A in Eq. (2) in S2. We obtain

$$\lambda_{1+1} = \frac{\ln 2}{\chi_1 t_{s1}^0} = \frac{1}{\chi_1} \quad (3)$$

$$\lambda_{1+1+1} = \frac{\ln 3}{\chi_1 t_{s1}^0 + \chi_2 t_{s2}^0} \quad (4)$$

$$0 = e^{-\lambda_{1+2}(\chi_1 t_{s1}^0 + \chi_2 t_{s2}^0)} + e^{-\lambda_{1+2}\chi_2 t_{s2}^0} - 1, \quad (5)$$

where $t_{sn}^0 = \ln \frac{n+1}{n}$ and $n = 1, 2$. Eq. (5) only provides an implicit solution for λ_{2+1} . The population growth rate is always positive, as there is no cell death in our model setting.

We first focus on $\chi_1 < \chi_2$ and prove that the reproductive strategy $1 + 1$ leads to faster growth than either $1 + 1 + 1$ or $2 + 1$. We start by comparing $1 + 1$ with $1 + 1 + 1$ for $\frac{\chi_1}{\chi_2} < 1$,

$$\begin{aligned} \frac{\lambda_{1+1}}{\lambda_{1+1+1}} &= \frac{\frac{\ln 2}{\chi_1 \ln 2}}{\frac{\ln 3}{\chi_1 \ln 2 + \chi_2 \ln \frac{3}{2}}} \\ &= \frac{1}{\ln 3} \frac{\chi_1 \ln 2 + \chi_2 \ln \frac{3}{2}}{\chi_1} \\ &= \frac{1}{\ln 3} \left(\ln 2 + \frac{\chi_2}{\chi_1} \ln \frac{3}{2} \right) \\ &> \frac{1}{\ln 3} \left(\ln 2 + \ln \frac{3}{2} \right) \\ &= 1. \end{aligned} \quad (6)$$

Thus $\lambda_{1+1} > \lambda_{1+1+1}$ for $\chi_1 < \chi_2$: The reproductive strategy $1 + 1$ leads to faster population growth than the reproductive strategy $1 + 1 + 1$.

Next we prove that $\lambda_{1+1} > \lambda_{2+1}$ for $\chi_1 < \chi_2$ by contradiction. If we would have $\lambda_{2+1} > \lambda_{1+1} = \frac{1}{\chi_1}$, then

$$\begin{aligned} 0 &= e^{-\lambda_{2+1}(\chi_1 t_{s1}^0 + \chi_2 t_{s2}^0)} + e^{-\lambda_{2+1}\chi_2 t_{s2}^0} - 1 \\ &= e^{-\lambda_{2+1}(\chi_1 \ln 2 + \chi_2 \ln \frac{3}{2})} + e^{-\lambda_{2+1}\chi_2 \ln \frac{3}{2}} - 1 \\ &< e^{-\ln 2 - \lambda_{2+1}\chi_2 \ln \frac{3}{2}} + e^{-\lambda_{2+1}\chi_2 \ln \frac{3}{2}} - 1 \\ &= \frac{3}{2} e^{-\lambda_{2+1}\chi_2 \ln \frac{3}{2}} - 1 \\ &= \frac{3}{2} \left(\frac{2}{3} \right)^{\lambda_{2+1}\chi_2} - 1. \end{aligned}$$

This can be simplified to $\left(\frac{2}{3} \right)^{\lambda_{2+1}\chi_2} > \frac{2}{3}$ and implies $\lambda_{2+1}\chi_2 < 1$ or

$$\lambda_{2+1} < \frac{1}{\chi_2} < \frac{1}{\chi_1} = \lambda_{1+1}.$$

which contradicts the assumption of $\lambda_{2+1} > \lambda_{1+1} = \frac{1}{\chi_1}$. Thus $\lambda_{1+1} > \lambda_{2+1}$ for $\chi_1 < \chi_2$. Thus the reproductive strategy 1 + 1 is optimal under $\chi_1 < \chi_2$.

Now we focus on $\chi_1 > \chi_2$ and prove that the reproductive strategy 2 + 1 leads to faster growth than either 1 + 1 or 1 + 1 + 1. We first compare 1 + 1 to 1 + 1 + 1. Since $\frac{\chi_2}{\chi_1} < 1$, we can revert the argument in Eq. (6) and obtain $\lambda_{1+1+1} > \lambda_{1+1}$.

Next we prove – again by contradiction – that $\lambda_{2+1} > \lambda_{1+1+1}$ for $\chi_1 > \chi_2$. If we would have $\lambda_{2+1} < \lambda_{1+1+1} = \frac{\ln 3}{\chi_1 \ln 2 + \chi_2 \ln \frac{3}{2}}$, then

$$\begin{aligned} 0 &= e^{-\lambda_{2+1}(\chi_1 t_{s1}^0 + \chi_2 t_{s2}^0)} + e^{-\lambda_{2+1} \chi_2 t_{s2}^0} - 1 \\ &= e^{-\lambda_{2+1}(\chi_1 \ln 2 + \chi_2 \ln \frac{3}{2})} + e^{-\lambda_{2+1} \chi_2 \ln \frac{3}{2}} - 1 \\ &> e^{-\ln 3} + e^{-\lambda_{2+1} \chi_2 \ln \frac{3}{2}} - 1 \\ &= \left(\frac{2}{3}\right)^{\lambda_{2+1} \chi_2} - \frac{2}{3}. \end{aligned}$$

This can be simplified to $\left(\frac{2}{3}\right)^{\lambda_{2+1} \chi_2} < \frac{2}{3}$ and implies $\lambda_{2+1} \chi_2 > 1$ or

$$\lambda_{2+1} > \frac{1}{\chi_2}.$$

On the other hand, we have for $\chi_1 > \chi_2$

$$\begin{aligned} \lambda_{1+1+1} &= \frac{\ln 3}{\chi_1 \ln 2 + \chi_2 \ln \frac{3}{2}} \\ &< \frac{\ln 3}{\chi_2 t_{s1}^0 + \chi_2 t_{s2}^0} \\ &= \frac{1}{\chi_2}, \end{aligned} \tag{7}$$

which implies $\lambda_{2+1} > \lambda_{1+1+1} > \lambda_{1+1}$. Thus the reproductive strategy 2 + 1 is optimal for $\chi_1 > \chi_2$.

The optimal reproductive strategy under a single size perturbation in the main text is the special case of $\chi_1 = 1$ or $\chi_2 = 1$. Thus, binary-splitting strategies are optimal for $N \leq 3$. Only for $\chi_1 = \chi_2$, all three reproductive strategies of 1 + 1, 1 + 1 + 1 and 2 + 1 have the same growth rate $\frac{1}{\chi_1}$. Thus, we have proven that the smaller χ_n determines the optimal strategy. In addition, we found the optimal strategy is either 1 + 1 or 2 + 1, which is consistent with the results that binary-splitting reproductive strategies are optimal under size effects, see Appendix .

S4: Only the binary-splitting reproductive strategies can be the optimal one under size effects

For size effects only, the number of newborn organism types is reduced as the cell composition does not impact the population growth rate. For example, a population with reproductive strategy 2 + 1 has only two types of newborn organisms: single-celled organisms and two-celled organisms. For the reproductive strategy $n_1 + n_2 + \dots + n_M$ with $N = \sum_{i=1}^M n_i$, the number of newborn organism types Ω is smaller or equal to M (since n_i may be equal to n_j). Therefore, Eq. (2) is reduces to

$$\begin{vmatrix} N_1 e^{-\lambda T_1} - 1 & N_1 e^{-\lambda T_2} & \dots & N_1 e^{-\lambda T_\Omega} \\ N_2 e^{-\lambda T_1} & N_2 e^{-\lambda T_2} - 1 & \dots & N_2 e^{-\lambda T_\Omega} \\ \vdots & \vdots & \ddots & \vdots \\ N_\Omega e^{-\lambda T_1} & N_\Omega e^{-\lambda T_2} & \dots & N_\Omega e^{-\lambda T_\Omega} - 1 \end{vmatrix} = 0. \tag{8}$$

Next, we simplify the determinant on the left hand side of Eq. (8) by changing lines 2 to Ω . We multiply the first row by $\frac{N_i}{N_1}$ and subtract the result from the i th row, where $i \in [2, \Omega]$. We obtain

$$\begin{vmatrix} N_1 e^{-\lambda T_1} - 1 & N_1 e^{-\lambda T_2} & \dots & N_1 e^{-\lambda T_\Omega} \\ \frac{N_2}{N_1} & -1 & \dots & 0 \\ \vdots & \vdots & \ddots & \vdots \\ \frac{N_\Omega}{N_1} & 0 & \dots & -1 \end{vmatrix} = 0. \quad (9)$$

Then we multiply the i th column by $\frac{N_i}{N_1}$ and add it to the first column, where $i \in [2, \Omega]$. We find

$$\begin{vmatrix} \sum_{i=1}^{\Omega} N_i e^{-\lambda T_i} - 1 & N_1 e^{-\lambda T_2} & \dots & N_1 e^{-\lambda T_\Omega} \\ 0 & -1 & \dots & 0 \\ \vdots & \vdots & \ddots & \vdots \\ 0 & 0 & \dots & -1 \end{vmatrix} = 0. \quad (10)$$

We finally obtain

$$\sum_{i=1}^{\Omega} N_i e^{-\lambda T_i} - 1 = 0, \quad (11)$$

where $i \in [1, \Omega]$. Since newborn organisms produce identical offspring, N_i is the number of the i th type offspring. For example, each organism produces 2 single-celled newborn organisms (the first type) and a two-celled newborn organism (the second type) under $1 + 1 + 2$. Thus $N_1 = 2$ and $N_2 = 1$. Thus, Eq. (11) can be written in the following equation

$$\sum_{i=1}^M e^{-\lambda T_{n_i}} - 1 = 0, \quad (12)$$

where T_{n_i} is the growth time for an organism from newborn size n_i to its maturity size N .

To prove that only binary-spitting reproductive strategies can be uniquely optimal, we use a similar method to [Pichugin and Traulsen, 2020]. We choose three reproductive strategies $S_1 = n_1 + n_2 + \dots + n_M$, $S_2 = (n_1 + n_2) + \dots + n_M$ and $S_3 = n_1 + n_2$, where $N = \sum_{i=1}^M n_i$. We use λ_1, λ_2 , and λ_3 to denote the growth rates of S_1, S_2 and S_3 , respectively. The growth rates can be calculated as roots of the equations

$$f_1(\lambda) = e^{-\lambda T_{(n_1, N)}} + e^{-\lambda T_{(n_2, N)}} + \sum_{i=3}^N e^{-\lambda T_{(n_i, N)}} - 1 = 0 \quad (13)$$

$$f_2(\lambda) = e^{-\lambda T_{(n_1+n_2, N)}} + \sum_{i=3}^N e^{-\lambda T_{(n_i, N)}} - 1 = 0 \quad (14)$$

$$f_3(\lambda) = e^{-\lambda T_{(n_1, n_1+n_2)}} + e^{-\lambda T_{(n_2, n_1+n_2)}} - 1 = 0. \quad (15)$$

Since the growth time T is positive, thus the above equations are monotonically decreasing functions. We multiply Eq. (15) by $e^{-\lambda T_{(n_1+n_2, N)}}$. Since $T_{(x,y)} + T_{(y,z)} = T_{(x,z)}$, we get

$$f_3'(\lambda) = e^{-\lambda T_{(n_1, N)}} + e^{-\lambda T_{(n_2, N)}} - e^{-\lambda T_{(n_1+n_2, N)}} = 0. \quad (16)$$

Thus, $f_1(\lambda) = f_2(\lambda) + f_3'(\lambda) = 0$. Hence, we have either $\lambda_1 = \lambda_2 = \lambda_3$, $f_2(\lambda_1) > 0 > f_3'(\lambda_1)$ or $f_2(\lambda_1) < 0 < f_3'(\lambda_1)$ at λ_1 . If $f_2(\lambda_1) < 0$ and $f_3'(\lambda_1) > 0$, we get $\lambda_2 < \lambda_1 < \lambda_3$. If $f_2(\lambda_1) > 0$ and $f_3'(\lambda_1) < 0$, we get $\lambda_3 < \lambda_1 < \lambda_2$. Thus, uniquely optimal reproductive strategies are always the binary-splitting ones.

S5: The effects of cell-type switching probability on reproductive strategies

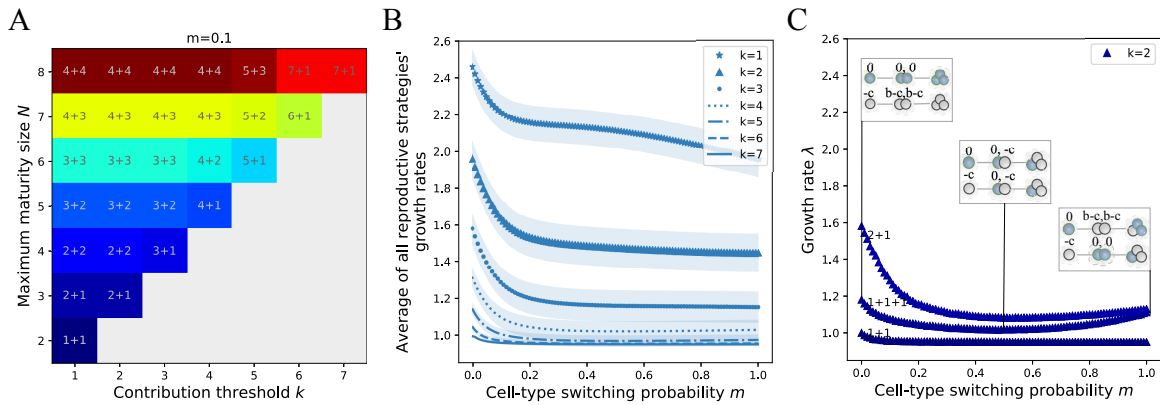


Figure S2: **The effects of the cell-type switching probability m on the optimal reproductive strategy.** **A.** The optimal reproductive strategy under high cell-type switching probability $m = 0.1$ is the binary-splitting one with maximum maturity size. **B.** The effects of cell-type switching probability on the average population growth rates of all reproductive strategies under different thresholds. **C.** The cell-type switching probability shows a complex effects on reproductive strategy 1+1+1 under $k = 2$. All reproductive strategies that have maturity size $N \leq 3$ are shown. Colours of reproductive strategies on panel A and panel C are consistent with Figure 3 in the main text.

References

- De Roos, A. M. (2008). Demographic analysis of continuous-time life-history models. *Ecology Letters*, 11(1):1–15.
- Gao, Y., Traulsen, A., and Pichugin, Y. (2019). Interacting cells driving the evolution of multicellular life cycles. *PLoS Computational Biology*, 15(5):e1006987.
- Pichugin, Y. and Traulsen, A. (2020). Evolution of multicellular life cycles under costly fragmentation. *PLOS Computational Biology*, 16(11):e1008406.