

## Flow cytometric quantification of the number of *Escherichia coli* ingested by *Tetrahymena thermophila*.

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### SUMMARY

In order to investigate the origin of endosymbiosis, we have synthesized a mutualism composed of *Tetrahymena thermophila* and *Escherichia coli*. Previously, we found that *E.coli* cells entered *T. thermophila* cells by predation and that the number of intracellular *E.coli* cells increased during serial subculture of the mutualism, although it is still unclear whether the intracellular *E.coli* duplicate. Here, we quantified the number of intracellular *E. coli* cells by flow cytometry, and investigated their change in number during serial subculture. We found that the average number of intracellular *E. coli* cells per *T. thermophila* cell increased from 2 to 40 and that the standard deviation of the number among *T. thermophila* cells increased tenfold during serial subculture. These results suggest that *T. thermophila* increases its predation rate or decreases its digestion rate and that the extent of the change varies widely between individual cells. Moreover, we found that both the average cell volume of *T. thermophila* and the average number of intracellular *E.coli* cells per unit cell volume of *T. thermophila* increased during subculture. However, the cell volume was not correlated with the number of intracellular cells per unit volume among *T. thermophila* cells in a batch of the serial subculture.