Cloning of *Tetrahymena* cells using cell sorter

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SUMMARY

We developed a cloning method for *Tetrahymena* cells using fluorescence activated cell sorting (FACS). This new cloning method can isolate mutants due to the phenotype (using size, fluoresce intensity, etc.) quantitatively and achieve a high through-put. We previously developed a chemically defined medium (CDM mini15) that was modified from the CDM of Szablewski et al (1991). In this study, we found that *Tetrahymena* cells could proliferate from 10 cells/ml (= 1 cell/100 μl) in CDM mini15 containing BSA. By supplementing CDM mini15 with BSA, we can isolate single mutant cells based on their nutritional requirements. We tried to sort single cells into a broth medium of PPYG and CDM mini15 with BSA, and divide it over 96-well plates. The percentage of wells in which surviving cells were found were 93% and 44%, respectively.