

Introduction of an Efficient Method for the Observation of Living

Paramecium

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Abstract: Fast movement of living *Paramecium* makes its observation very difficult and low efficiency of taking pictures. Volume-fixing method being reported on Chinese local journals could solve this problem completely. To popularize this method, we will simply introduce this method here.

Keywords: *Paramecium*; Observation; Volume-fixing

Introduction

P. caudatum is the representative of ciliates (a huge group of unicellular eukaryotes) distributed globally, whose size is suitable for microscopic observation^[1]. Therefore, it has long been used in the experiments of Zoology teaching and life scientific research^[2-4]. *P. caudatum* has two types of nuclei, a small diploid micronucleus also called germinal nucleus and a big polygenomic macronucleus referred as vegetative nucleus^[5]. In addition to this nuclear dimorphism, like other ciliates, *P. caudatum* has some specialized organelles, such as oral organelles, contractile vacuoles and trichocysts^[5], most of which are required to be observed in living state^[3]. However, due to its high-speed movement, the students have to struggle in class to catch cells for observation and researchers have to try every possible way in the lab for obtaining high-quality fluorescent pictures of living *Paramecium*.

So far, many methods have been reported including cotton fibre blocking method^[3], squash method^[6], alcohol deciliation method^[7] and volume-fixing method^[8-10]. And volume-fixing is the best method to solve this problem thoroughly, which makes the observation easier and micro-photography more enjoyable. In fact, this method is benefited from micropipettes (one of the most important tools of molecular biology) making it possible to measure liquid or solution volume correctly at microliter level. Here we will introduce this method and hope it to be used by more related people.

Materials and Methods

Equipments and Materials

Microscope, 20 μ L micropipette and tips, *P. caudatum* culture, slide glasses, cover glasses.

Methods

A certain volume of cell suspension of *P. caudatum* is dropped onto a piece of clean slide glass, and then covered with a cover-glass of appropriate size. At this time point, a temporary slide is made being ready for observation and photography. Due to the different sizes of cover glass, the volume of cell suspension has to be adjusted^[8-10]. For the cover glasses of 18×18 mm², 20×20 mm², 22×22 mm² and 24×24 mm², the suggested volume is 6.5~7.0 μL, 10~11 μL, 11~12 μL and 14 μL, respectively.

At the very beginning of making the temporary slides, some cells are squashed, while about half of the cells are flattened and move very slowly, whose macronuclei, oral grooves, contractile vacuoles, trichocysts as well as cilia are observed clearly being suitable for observation and photography.

Precautions and suggestions

During the operation, attention should be paid to the following points.

1. Measure the volume of cell suspension correctly. Otherwise, the cells either are squashed completely or move too fast to be observed.
2. Do not move the cover glasses to avoid squashing cells after preparation.
3. Before observation, scan the slides briefly and choose intact cells without deformation.
4. If most of the cells move fast, pieces of absorbent paper could be used to suck the extra-solution away, but the operator must be very careful to avoid over-sucking.

Advantages

1. This method is simple, fast, effective and no need for experience.
2. According to the needs, the cells could be sealed in mineral oils, which could prevent the evaporation of water ensuring the survival of the cells for longer time^[8,10].
3. This method is not limited to *P. caudatum*^[11-14]. According to the size of the targeting cells, the volume of the cell suspension should be adjusted to the suitable volume.
4. Before the staining of cells, temporary slides could be made by this method ensuring perfect shape of the cells^[8,11].

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