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How We Raise *Paramecium*, Easily, in Large Numbers

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Introduction

We use more *Paramecium* spp. in our Physiological Protozoology course at the University of Kansas than any other cells, and use them in great numbers each week, so that we wish to avoid the cost of buying them commercially. Therefore, we raise our own cultures.

For over twenty years the senior author has used the easy method described here to raise large numbers of *Paramecium aurelia, P. caudatum,* or *P. multimicronucleatum,* with consistent success. It yields at least as good, and usually better, populations than most other methods (Needham, et al., 1937); avoids the necessity for the tedious, critical drying of and powdering of lettuce (Sonneborn, 1937) and other extraneous labors; and the cultures require very little attention over relatively long periods of time.

Materials and methods

We boil either a half-dozen alfalfa pellets, such as are manufactured and sold as rabbit food, or a few leaves and stems of dried alfalfa hay in about 100 ml of distilled water, long enough (about 10-15 min.) to soften and partly disperse the pellets or soften the leaves and stems. This produces a light-colored greenish-brown tea if pellets are used, a darker tea if the hay is used.

We add 25 to 50 ml of this tea to each 3-400 ml of previously boiled and cooled tap or pond water which has been placed in a 4-5 liter glass jar (pickle jar or battery jar) and stirred to aerate it. If the tap water or pond water is suspect because of possible ionic pollutions (especially copper or lead plumbing) we use glass distilled water with about 100 ml of Chalkley's solution (Chalkley, 1930) added to each liter to provide the principally needed ions.

If we wish a new clone of *Paramecium* we add one organism and its associated bacteria. It takes several weeks thus to get a large population in such a large jar. The population-density can be increased in a clonal population and the time to acquire a dense population shortened by reducing the volume of fluid in the culture by a factor of 10 or more, using a finger bowl instead of a battery jar to house the culture.

To further speed up growth, a few drops from a culture of bacteria

grown by Mote's method (Mote, 1968) may be added to the medium with the *Paramecium,* and larger numbers of *Paramecium* may also be inoculated into the medium. At intervals of a week to ten days a few more ml of the alfalfa tea is added.

Once the new culture is prepared it should be loosely covered so dust cannot enter, but gases may enter and exit. We find that sheets of aluminum foil are excellent for the covers, since they may be loosely crimped over the rim of the jars, but easily removed. Flat glass plate also serves well. The culture may then be left virtually untended for weeks while an abundant bacterial growth nourishes a numerous healthy colony of *Paramecium,* which maintains itself for many weeks to months.

Discussion

As any one culture declines in numbers a new one may be started. If several are started sequentially from any healthy culture at intervals of about three to four weeks, there will be a culture available at any time with hundreds of *Paramecium* per ml of fluid.

Some precautions are necessary.

While the three species mentioned grow in the same locales in nature, it is better to grow them separately in the laboratory. One or another of the species will competitively eliminate the others in a laboratory culture (Gause, et al., 1934), depending on the conditions that develop in the culture.

Also, while the ciliates tolerate light well, and can be placed on a north-facing window sill without detriment, they do not do well in direct sunlight. We usually keep the cultures on a laboratory bench or table about ten feet away from any window, and out of direct sunlight.

Glassware used should be cleaned with concentrated sulfuric acid (H_2SO_4) rather than detergents (e.g., Alconox) or chromate cleaning solutions. The detergents and chromates leave residues harmful to the protozoa. Scouring with soap and water, and steel wool pads will also suffice.

The advantages of this method of culturing *Paramecium* are the ease with which the cultures may be started and maintained, and the relatively long duration of the cultures. Even so, we find it advantageous to start a new series of cultures every two to three months to keep cultures with peak numbers of *Paramecium* available at all times.

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