A Miniaturized Video System for Monitoring Drosophila Behavior

This method allows monitoring the movement of many fruit flies in space simultaneously.

Ames Research Center, Moffett Field, California

Long-term spaceflight may induce a variety of harmful effects in astronauts, resulting in altered motor and cognitive behavior. The stresses experienced by humans in space — most significantly weightlessness (microgravity) and cosmic radiation — are difficult to accurately simulate on Earth. In fact, prolonged and concomitant exposure to microgravity and cosmic radiation can only be studied in space.

Behavioral studies in space have focused on model organisms, including *Drosophila melanogaster*. Drosophila is often used due to its short life span and generational cycle, small size, and ease of maintenance. Additionally, the well-characterized genetics of Drosophila behavior on Earth can be applied to the analysis of results from spaceflights, provided that the behavior in space is accurately recorded.

In 2001, the BioExplorer project introduced a low-cost option for researchers: the small satellite. While this approach enabled multiple inexpensive launches of biological experiments, it also imposed stringent restrictions on the monitoring systems in terms of size, mass, data bandwidth, and power consumption. Suggested parameters for size are on the order of 100 mm³ and 1 kg mass for the entire payload. For Drosophila behavioral studies, these engineering requirements are not met by commercially available systems.

One system that does meet many requirements for behavioral studies in space is the actimeter. Actimeters use infrared light gates to track the number of times a fly crosses a boundary within a small container (3×3×40 mm). Unfortunately, the apparatus needed to monitor several flies at once would be larger than the capacity of the small satellite.

A system is presented, which expands on the actimeter approach to achieve a highly compact, low-power, ultra-low bandwidth solution for simultaneous monitoring of the behavior of multiple flies in space. This also provides a simple, inexpensive alternative to the current systems for monitoring Drosophila populations in terrestrial experiments, and could be especially useful in field experiments in remote locations. Two practical limitations of the system should be noted: first, only walking flies can be observed — not flying — and second, although it enables population studies, tracking individual flies within the population is not currently possible.

The system used video recording and an analog circuit to extract the average light changes as a function of time. Flies were held in a 5-cm diameter Petri dish and illuminated from below by a uniform light source. A miniature, monochrome CMOS (complementary metal-oxide semiconductor) video camera imaged the flies. This camera had automatic gain control, and this did not affect monitoring performance. The camera was positioned 5–7 cm above the Petri dish such that the imaging area was 2.25 cm². With this basic setup, still images and continuous video of 15 flies at one time were obtained. To reduce the required data bandwidth by several orders of magnitude, a band-pass filter (0.3–10 Hz) circuit compressed the video signal and extracted changes in image luminance over time. The raw activity signal output of this circuit was recorded on a computer and digitally processed to extract the fly movement “events” from the waveform. These events corresponded to flies entering and leaving the image and were used for extracting activity parameters such as inter-event duration. The efficacy of the system in quantifying locomotor activity was evaluated by varying environmental temperature, then measuring the activity level of the flies.

The system presented in this work for monitoring fly activity is an inexpensive, compact, and ultra-low bandwidth alternative to currently available options, making it a suitable candidate for biological experiments on small satellites or field experiments. In contrast to most actimeters, multiple flies can be imaged in a small apparatus. Additionally, the flies are contained in a larger chamber (20 cm² vs., e.g., 1.2 cm²), which permits motion in two axes rather than one. This may allow the fly behavior to more closely approximate the natural state.

This work was done by Sharmila Bhat-tacharya of Ames Research Center; Omer Inan, Gregory Kovacs, and Mozziyar Etemadi of Stanford University; Max Sanchez of Lockheed Martin; and Oana Marcu of National Space Grant Foundation. Further information is contained in a TSP (see page 1).

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Hydrofocusing Bioreactor Produces Anti-Cancer Alkaloids

Enhancement of production may be attributable to favorable aggregation of cells.

Lyndon B. Johnson Space Center, Houston, Texas

A methodology for growing three-dimensional plant tissue models in a hydrodynamic focusing bioreactor (HFB) has been developed. The methodology is expected to be widely applicable, both on Earth and in outer space, as a means of growing plant cells and aggregates thereof under controlled conditions for diverse purposes, including research on effects of gravitation and other environmental factors upon plant growth and utilization of plant tissue cultures to produce drugs in quantities greater and at costs lower than those of conventional methodologies.
The HFB was described in “Hydrofocusing Bioreactor for Three-Dimensional Cell Culture” (MSC-22358), NASA Tech Briefs, Vol. 27, No. 3 (March 2003), page 66. To recapitulate: The HFB offers a unique hydrofocusing capability that enables the creation of a low-shear liquid culture environment simultaneously with the “herding” of suspended cells and tissue assemblies and removal of unwanted air bubbles. The HFB includes a rotating cell-culture vessel with a centrally located sampling port and an internal rotating viscous spinner attached to a rotating base. The vessel and viscous spinner can be made to rotate at the same speed and direction or different speeds and directions to tailor the flow field and the associated hydrodynamic forces in the vessel in order to obtain low-shear suspension of cells and control of the locations of cells and air bubbles.

For research and pharmaceutical-production applications, the HFB offers two major benefits: low shear stress, which promotes the assembly of cells into tissue-like three-dimensional constructs; and randomization of gravitational vectors relative to cells, which affects production of medicinal compounds. Presumably, apposition of plant cells in the absence of shear forces promotes cell-cell contacts, cell aggregation, and cell differentiation. Only gentle mixing is necessary for distributing nutrients and oxygen.

It has been postulated that inasmuch as cells in the simulated microgravitation of an HFB do not need to maintain the same surface forces as in normal Earth gravitation, they can divert more energy sources to growth and differentiation and, perhaps, to biosynthesis of greater quantities of desired medicinal compounds. Because one can adjust the HFB to vary effective gravitation, one can also test the effects of intermediate levels of gravitation on biosynthesis of various products.

The potential utility of this methodology for producing drugs was demonstrated in experiments in which sandalwood and Madagascar periwinkle cells were grown in an HFB. The conditions in the HFB were chosen to induce the cells to form into aggregate cultures that produced anti-cancer indole alkaloids in amounts greater than do comparable numbers of cells of the same species cultured according to previously known methodologies. The observations made in these experiments were interpreted as suggesting that the aggregation of the cells might be responsible for the enhancement of production of alkaloids.

This work was done by Steve R. Gonda of Johnson Space Center and Jagan V. Valluri of Marshall University. Further information is contained in a TSP (see page 1).

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Dr. Jagan V. Valluri
Marshall University
6242 Highland Dr.
Huntington, WV 25705
E-mail: valluri@marshall.edu
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