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AN ELECTRONIC PARTICLE SEPARATOR WITH POTENTIAL BIOLOGICAL APPLICATION (M. J. Fulwyler)

INTRODUCTION

The particle separator is a device which, in principle, is capable of separating microscopic particles according to an electronically measurable characteristic such as volume, optical density, or fluorescence. Separation is accomplished as follows. The particle, in liquid suspension, passes by a sensor which measures the characteristic of interest. The suspension is forced through a nozzle and emerges as a jet which is broken into droplets, thereby isolating the suspended particles. The droplets containing the particles of interest are charged and then deflected by an electrostatic field into a collection vessel.

METHOD

Shown in Fig. 1 is a device which is being developed to separate particles according to their volume. A cell suspension (under 4 atmospheres pressure) enters the droplet generator (C) via a tube (D) and emerges as a high-velocity fluid jet (E) [diameter 36 μ, velocity 15 m/sec]. A piezoelectric crystal (A), driven at a frequency of 72,000 c/sec, produces vibrations which pass down the Lucite rod (B) into the liquid within the droplet generator. The catenoidal shape of the rod amplifies the magnitude of the vibrations within the liquid, and the velocity fluctuations of the emerging liquid cause the jet to break into 72,000 very uniform droplets each second.

Droplets are charged as they pull away from the charged liquid column by applying a voltage at (K) relative to (M), which is in contact with the emerging stream. As the droplet separates, it carries away a charge proportional to the instantaneous charge on the column of liquid. In this way one or more droplets may be charged. The charged droplets are deflected (H) on entering the electrostatic field (7,000 volts/cm) between the deflection plates (G). A series of collection vessels (L) receives the deflected droplets.

The sequence of events leading to separation is as follows. Cell volume is sensed as the cell passes through a Coulter

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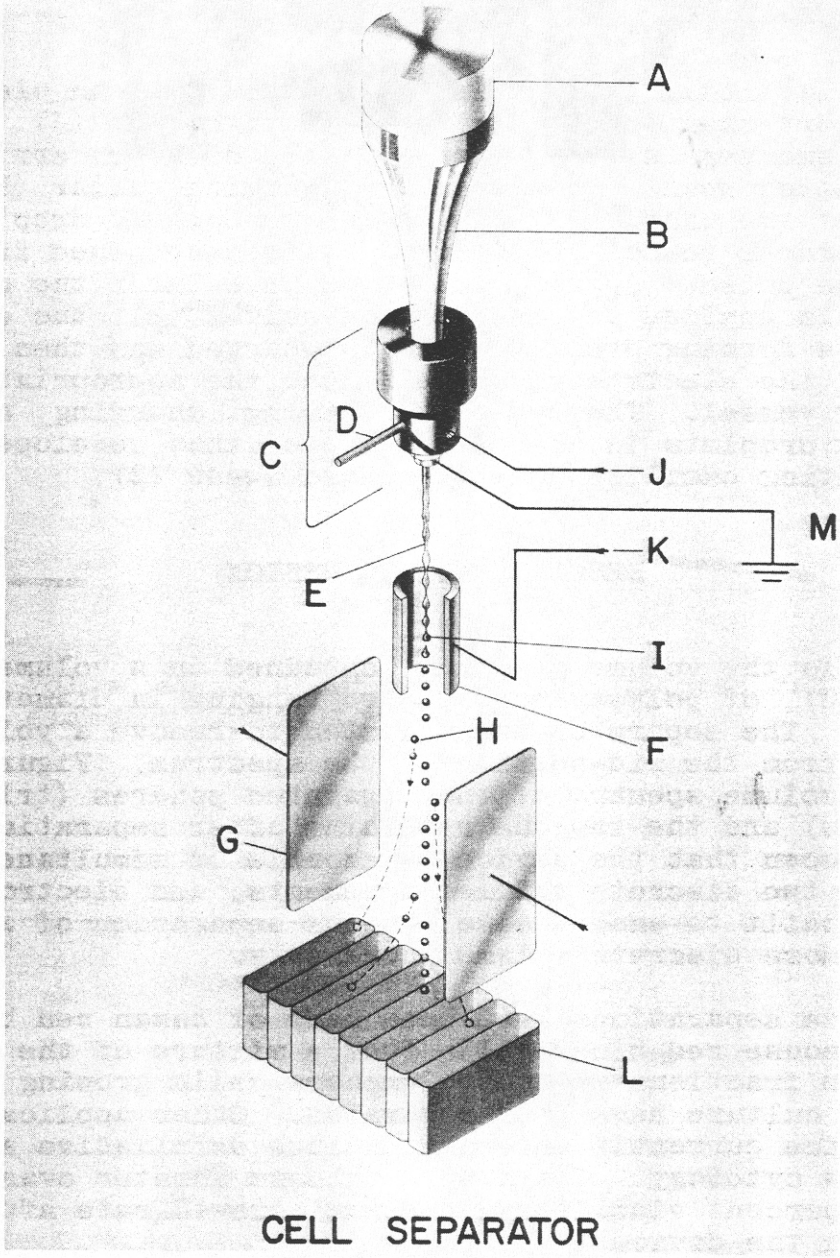

Fig. 1. Cell separator: (A) piezoelectric crystal; (B) acoustic coupling rod; (C) droplet generator; (D) fluid entry tube; (E) fluid jet; (F) charging collar; (G) electrostatic deflection plates; (H) deflected droplets; (I) droplet separation point; (J) cell volume signal contact; (K) charging collar contact; (L) droplet collection system; and (M) ground contact for emerging jet.

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aperture (1) within the droplet generator (C). An electric pulse proportional to cell volume is obtained at (J). The cell then emerges in the jet and, 250 μsec later, arrives at the separation point (I) within the charging collar (F). The size of the charging pulse needed to deflect droplets into the proper vessel is electronically determined from the cell volume pulse. Approximately 240 μsec later the charging pulse is applied to the charging collar (K); the cell is caught in a forming droplet which is charged and then deflected by the electrostatic field into the appropriate collection vessel. The method of forming, charging, and deflecting droplets is a modification of that developed as an ink-writing oscillograph by Richard Sweet (2).

RESULTS AND DISCUSSION

Figure 2A is the volume spectrum, obtained on a volume spectrometer (3), of polystyrene spheres ranging in diameter from 7 to 14 μ. The separator was adjusted to remove a volume increment from the mid-portion of the spectrum. Figure 2B shows the volume spectra of the separated spheres (triangular data points) and the residue remaining after separation. It has been shown that the device is capable of simultaneously separating two discrete volume increments, and electronics are being built to enable simultaneous separation of a sample into 6 or more discrete volume increments.

Quantitative separations have been made of human red blood cells and mouse red blood cells from a mixture of the two. Also volume fractions of mouse lymphoma cells growing in suspension culture have been separated. Other applications of the device currently underway include exfoliative and bone marrow cytology. Tests with Chinese hamster ovary cells showed 96 percent viability and normal growth rate after passing through the device.

Development and refinement of the device are proceeding. Separations under sterile conditions are expected in the near future. Investigation is underway of a sensor system able to measure optical characteristics of a particle. With such a system, it may be possible to measure simultaneously two (or more) characteristics of a cell and to make separation dependent on the relation of these two characteristics.

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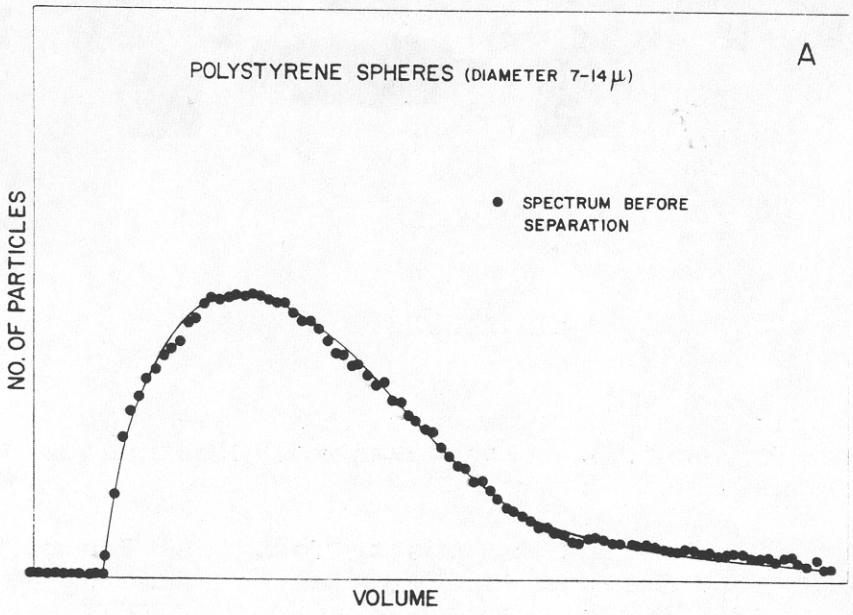

Fig. 2A. Volume spectrum of polystyrene spheres ranging in diameter from 7 to 14 μ.

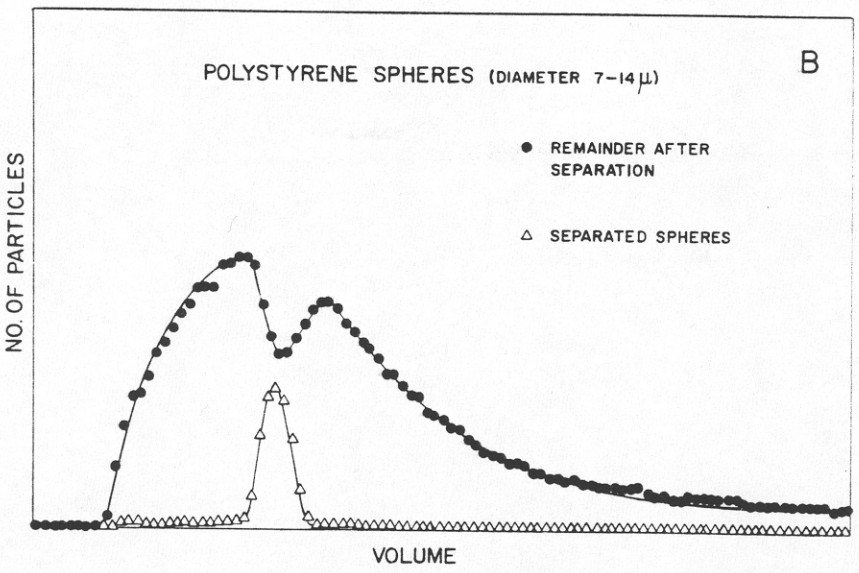

Fig. 2B. Volume spectrum of separated spheres (triangles) and the volume spectrum of the residue after separation (solid points).

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REFERENCES

(1) W. H. Coulter, U. S. Patent No. 2,656,508 (1953); W. H. Coulter, Proceedings of the National Electronics Conference 12 (1956), p. 1034.

(2) R. G. Sweet, Stanford University Technical Report 1722-1. Available from Defense Document Center, Washington, D. C., Report SU-SEL-64-004 (1964).

(3) M. A. Van Dilla, N. J. Basmann, and M. J. Fulwyler, Los Alamos Scientific Laboratory Report LA-3132-MS (1964), p. 182.

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