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### A Simple Gradient Maker

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## NOTE

### A Simple Gradient Maker

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#### Summary

A sugar gradient was formed directly in a centrifuge tube by means of a celluloid cone. This simple system does not require any pump or mixing reservoir.

Work on the isolation of plant cell organelles by centrifugation in a gradient solution indicated a need for a simple, rapid method of forming a gradient. The gradient maker devised is easy to make, does not require any pump or mixing reservoir, and a gradient can be formed directly in the centrifuge tube in less than 2 min. It consists of a truncated cone, having a volume equal to one half that of the centrifuge tube to be filled, and open at both ends. This cone is easily formed from a thin sheet of celluloid by wrapping it around a polycone universal adapter (Bel-Art Products Ltd., Pequannock, New Jersey) and gluing the seam with cellulose cement.

To prepare the gradient, the cone is inserted into the centrifuge tube with the large end resting on the bottom, and the two sucrose (or other gradient) solutions are introduced simultaneously with two pipets, the heavy and the light sucrose in the cone and in the tube, respectively

303

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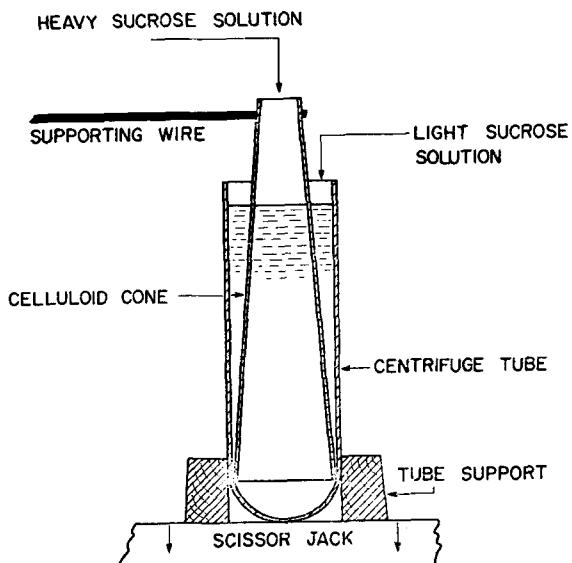


FIG. 1. Gradient maker assembly showing the celluloid cone held vertically by means of a wire and the centrifuge tube mounted on top of a scissor jack.

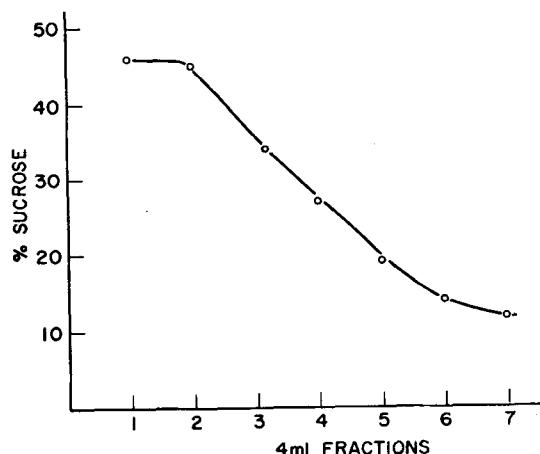


FIG. 2. Sucrose gradient obtained with 10 and 50% sucrose solutions. The fractions were taken from a hole punctured in the bottom of the tube.

(Fig. 1). The levels of the two sucrose solutions should be maintained the same during the filling operation. The cone is then withdrawn slowly at a constant rate during about 5 sec. The gradient obtained with 10 and 50% sucrose solutions is shown in Fig. 2.

The dimensions of a cone to be used with a centrifuge tube 1 in. in diameter  $\times$  3.5 in. long, are  $\frac{13}{16}$  in. at the large end,  $\frac{3}{8}$  in. at the small end, and  $3\frac{3}{4}$  in. long. The cone can be withdrawn by hand; however, securing the cone by means of a wire glued to the small end and lowering the centrifuge tube with a scissor jack was found most convenient. Cones of different dimensions can be made to fit tubes of different sizes. The gradient obtained can be increased to some extent by increasing the taper of the cone.

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