Strain	Control		Bacteria	
	Total Adults	Range	Total Adults	Range
F2	70/100	56%-84%	73/100	60%-88%
S1	57/100	48%-68%	71/100	56%-96%
S1 repeat control	60/100	all 60%		

Again there was no significant difference between treatments, though all averages and ranges were higher with bacteria than without it.

Matings of flies from these cultures were unaffected by the bacteria and produced about the same mating propensities as the controls.

In conclusion, this Achromobacter has no detrimental effect on the life cycle under the conditions tested. It still could be detrimental via its mucoid by-product to mire the flies or to decrease their sensitivity to external stimuli after prolonged exposure, but those factors have not yet been tested. Briefly, the basis for unexpected variation between cultures and uncontrolled fluctuations in mating propensity is as yet undetermined.

References: Hendrix, N. and E. Ehrlich, 1965 DIS 40: 99.

Acknowledgments: Contract AT(11-1)-1652, U.S. Atomic Energy Commission.

Schneider, I. Walter Reed Army Institute of Research, Washington, D.C. Embryonic cell lines of D. melanogaster.

Within the past year, Echalier and Ohanessian (C.R. Acad. Sci. 268: 1771, 1969) and Kakpakov et al., (Genetika 5: 67, 1969) have independently reported the establishment in vitro of cell lines from embryos of D. melanogaster. These lines have, in general, not been available for study by individuals from other laboratories. This note reports the existence of 3 additional cell lines from D. melanogaster, subcultures of which are available upon request.

Timed embryos were collected, surface sterilized and allowed to develop almost to the point of hatching. They were then cut into 2 or 3 pieces and placed in 0.2% trypsin solution (1:250, DIFCO) in Rinaldini's salt solution for 30 minutes at room temperature. After a thorough washing the pieces were seeded into glass T-9 flasks with 1.25 ml Schneider's Drosophila medium (GIBCO) containing an additional 500 mg bacteriological peptone per 100 ml bottle and supplemented with 15% inactivated fetal bovine serum. The pH of the medium was 6.7 and the cultures maintained at 27 ± 0.5°C with a gaseous phase of ambient air.

Initially growth took place in the form of hollow spheres issuing from the cut ends of the embryonic fragments. The spheres, each of which consisted of a monolayer of cells, were allowed to grow to a diameter of 1 mm or more before being excised, teased apart (or alternatively treated with trypsin) and returned to the same flask together with the original embryonic fragments. Single cells as well as small cellular masses obtained in this manner usually attached to the bottom of the culture flask and began to multiply. This procedure was repeated until sufficient cells were present in the primary flask to make subculturing feasible. The primary cultures for the 3 lines were initiated in August 1969, December 1969 and February 1970.

The 3 lines are quite dissimilar in appearance. Cells of the 1st line are initially round and vary from 12 to 20μ in diameter when freshly seeded into a new flask. Within 3 to 5 days, however, the majority begin to differentiate in vitro as evidenced by a 2 to 6-fold increase in cell size (due primarily to an increase and/or flattening of the cell cytoplasm) with consequent alterations in cell shape. Once differentiated the cells cease to multiply. However, there are always sufficient numbers of "stem" cells present in each culture so that subsequent subcultures can be made at intervals of approximately 1 week. The chromosome number varies from 2n to possible 8n.

The other 2 lines are much more conventional in appearance and behavior. The cells of both lines are epithelial in shape, predominently diploid and form monolayers but differ from each other in size and generation time. Cells of the 2nd line are approximately 10μ in diameter and 20 to 30μ long and have a generation time of 22 hours at 27°C. Corresponding figures for the 3rd line are 6 to 10μ, 10 to 30μ and 18 hours, respectively.

Definitive identification of the cells' origin(s) has not been made but should be possible once sufficient numbers of cell spheres have been injected into 3rd instar larvae and the differentiated masses examined.