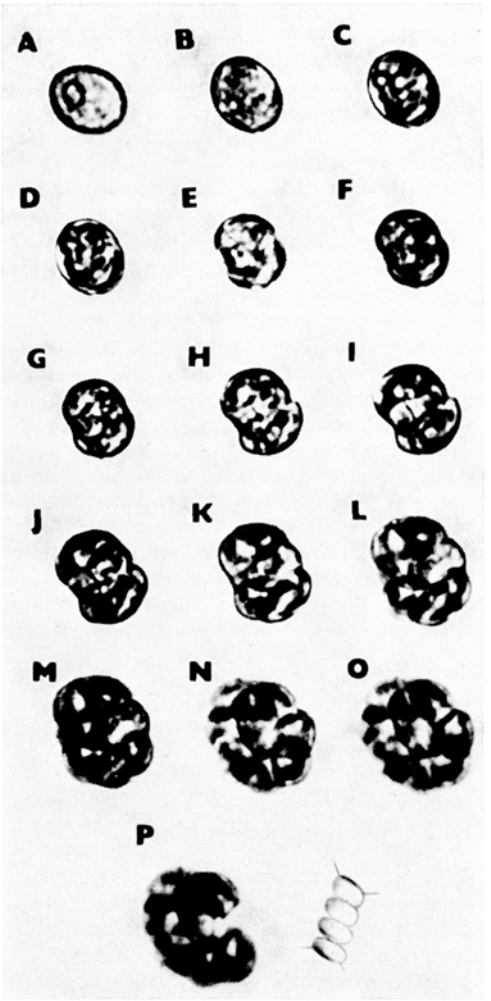

Morphogenese der Ruhezelle von *Scenedesmus quadricauda* (Turp., Bréb) nach der Übertragung in die optimalen Lebensbedingungen. A) Stellt die Ruhezelle nach der Übertragung in flüssiges Medium dar; B–G) die Entstehung eines zweizelligen Gebildes; H–K) die Bildung von zwei Tetraden; L–O) das Platzen der Muttermembrane und Entstehung des achtzelligen kugelförmigen Gebildes; P) die Entstehung des vierzelligen Coenobiums. Die einzelnen Aufnahmen wurden im Zeitabstand von einer Viertelstunde gemacht.

des vegetativen ontogenetischen Zyklus üblich ist. Die Entwicklung der Ruhezelle verläuft ähnlich wie das Keimen der Zygote bei coenobialen Volvocales. Zuerst schnürt sie sich ein, wodurch ein zweizelliges Gebilde (B–G) entsteht. Innerhalb jeder Zelle kommt es zur ersten und zweiten Kernteilung, wobei zwei Tetraden entstehen (H–K), die nach dem Platzen der Mutterzellwand ein achtzelliges kugelförmiges Gebilde (L–O) gestalten, aus dessen Zellen sich dann vierzellige Coenobien bilden. Auf diese Weise entstehen acht vierzellige Coenobien (P).

Seit der Übertragung der Ruhezelle aus dem Agar ins Medium der Kultivierkammer bis zur ersten Lösung des vierzelligen Coenobiums aus dem achtzelligen Gebilde sind 56 h vergangen. Die freigewordenen vierzelligen Coenobien wachsen und entwickeln sich dann weiter und bilden aus jeder Coenobialzelle Tochtercoenobien, was sich immerfort wiederholt. Es entsteht eine Generation von vegetativen Autosporen.

Summary. In the chlorococal alga *Scenedesmus quadricauda* (Turp., Bréb.) resting cells have been found resembling the akinete. The whole morphogenesis of the alternation of the resting cell into four cell autospores has been recorded by the method of microchamber cultivation in connection with time-lapse cinematography.

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PRO EXPERIMENTIS

Screening of Systemic Insecticides Through Artificial Feeding of Mealy Bugs

Salama and Saleh[1] developed a technique for artificial feeding of coccids. At present, 3 methods of artificial feeding are made use of in screening systemic insecticides in the citrus mealy bug, *Planococcus citri* Risso (Pseudococcidae). In the first method, the test unit consists of a round glass ring with 2 open ends and the artificial membrane used was 'Parafilm' (M). A small square of the membrane was stretched to $^1/_5$ of its thickness and pulled down over the upper end of the glass ring. 2 drops of the tested feeding solution, whether water, 10% sucrose or a mixture of sucrose with different concentrations of the tested insecticide, were placed over the stretched membrane with a pippette. A second stretched piece of membrane was then pulled down over the first one and thus encapsulating the fluid drops as a thin film between them. The experimental individuals were introduced through the other open end of the ring which was then covered with a cover slip. In the second method, the bugs were introduced into a glass tube with one open end which was then covered with Parafilm membrane. The tube was then inverted with the membrane down to adhere to a thin layer of the tested fluid contained in a small Petri-dish 5 cm in diameter. In the third

[1] H. S. Salama and M. Saleh, XIII Inter. Congress Entomology (Moskwa 1968).

method, 2 cm^3 of the tested fluid were placed in a small dish 3 cm in diameter which was then covered with the membrane. The dish was inverted over a glass tube of the same diameter confining the bugs.

In all cases, different stages of the mealy bugs were isolated from a standard culture maintained on potato sprouts at 30 °C. 10 replicates, each with 10 individuals, were made for each test. The insecticide to be tested was a dimethoate compound (Roger) 0,0-dimethyl-S (N-methyl carbamyl methyl) phosphorodithioate. The survival of adult insects was first determined on water and sucrose, and in screening the insecticide, the mortality of different insect stages was determined after 24 h access feeding time. Moribund individuals were considered dead. The mortality was corrected according to Abbott's formula. Prophylactic skin was also tried and proved to be efficient in feeding as well.

Results. The mealy bugs successfully ingest liquids accessible via artificial membranes of stretched Parafilm or prophylactic skin. Starved adults lived for 2–4 days and those fed on a water diet lived for 4–6 days. Adults fed on 10% sucrose survived for 7–9 days and they even laid eggs in small numbers.

In screening the toxicity of the dimethoate Roger through artificial feeding of *P. citri*, the EC50 were found to be 23, 50, 95 and 120 ppm for the 1st, 2nd, 3rd and adult stages respectively. The probability that the mortality was due to refusal to feed on the diet, or to a fumigant action, was ruled out, since the bugs can survive starved for 2–4 days and also the percent insects survival was high when they were kept in a small tube enclosed within a bigger one with a high concentration of the insecticide. This was also reported for aphids (MITTLER and PENNEL[2]).

The aforementioned method can be used as a standard test for screening insecticides, chemosterilants, derivatives produced in plants from the insecticide originally applied as well as chemicals in feeding behavioural studies of mealy bugs in general.

Zusammenfassung. Verschiedene Methoden zur Toxizitätsbestimmung von Insektiziden bei künstlicher Ernährung von *Planococcus citri* Risso mit Zuckerdiät und durch eine Parafilmmembran oder eine prophylaktische Haut werden beschrieben.

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Heart Cells in Culture: A Simple Method for Increasing the Proportion of Myoblasts

Cell cultures derived from trypsinized hearts of young rats, hamsters, or chick embryos contain a mixture of 2 kinds of cell: muscle cells and mesenchymal cells[1], with an initially high percentage of the latter[2–4]. These mesenchymal cells multiply more quickly than the myoblasts and pervade all the free space[2].

Methods so far reported for elimination of the mesenchymal cells[3,5–7] are not convenient when plates of confluent beating myoblasts are routinely needed in large numbers for electrophysiological, biochemical and electron microscopical investigations[8]. Therefore, we have tried to find whether the speed of attachment to the dish surface would be different for myoblasts and mesenchymal cells, and if so, whether this difference could be used to increase the proportion of myoblasts in culture.

We found that, following trypsinization of the hearts, most of the mesenchymal cells attach to the dishes before the myoblasts. The technique described here is based on this difference.

Although the elimination of the mesenchymal cells was never complete, the present technique gave better results than those achieved by modifying other culturing conditions such as temperature, centrifugation parameters or use of antimitotics.

Materials and methods. Preparation of cell suspension. Whole hearts of 2- to 3-day-old rats (not over 30 at a time) were cut into about 8 pieces and washed twice for 5 min in cold phosphate buffer solution (PBS)[9] with gentle stirring. The fragments of cardiac tissue were then subjected to 9 successive 8 min periods of trypsin digestion at 37 °C, again with gentle stirring (Difco Trypsin 1:250 – 0.25% in phosphate buffer without Ca^{++} and Mg^{++}), which generally sufficed to disaggregate them almost completely.

The first 3 supernatants were discarded because they mainly contained mesenchymal cells and debris. Each of the following supernatants was poured into half its volume of precooled medium in a centrifuge tube (usually 3 supernatants per tube) and kept at about 5 °C in iced water. (Medium was: MEM powder – GIBCO – plus 0.06% yeast extract and 0.25% lactalbumine hydrolysate, 10% calf serum, $NaHCO_3$ 14 m*M*, penicilline 400 U/ml and streptomycine 200 µg/ml.)

Once digestion of the tissue was complete, the cell suspensions were centrifuged for 3 min at 670 × *g*. The pellets of cells were resuspended in medium and the suspensions pooled into a beaker kept at 5 °C. To minimize pipetting and to prevent clumping of the cells the following procedure was used: about 20 ml of medium was added to each pellet and the tubes were gently shaken; as soon as part of the cells were resuspended 5 ml of the suspension was sucked off and transferred into the beaker and fresh medium added to the tube. This process was repeated until the pellets had been completely resuspended; at that time, the beaker contained the

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