

Fig. 3.

Inhibiting effect on mitochondrial swelling produced by gum Arabic.

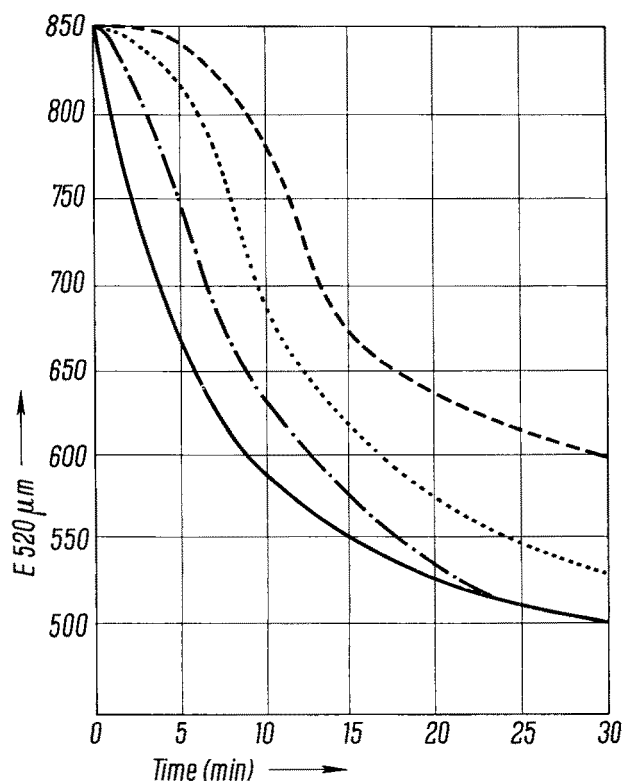


Fig. 4.

Inhibiting effect on mitochondrial swelling produced by albumin.

with Tris-HCl pH 7.4. 0.3 ml of the mitochondria suspension were added to each container, immediately after isolation of mitochondria. The first reading was taken about 30 sec after mixing the solutions, and the following ones 1 to 30 min later.

The curves representing the fall in optical density of the suspensions are shown. The curves, which express the mean values of five experiments, are drawn either as a continuous line or as lines composed of a series of dots, dashes and alternate dots and dashes. Dashes refer to substances used at the highest concentration, dots, and dots and dashes respectively, to substances used at the middle and lowest concentrations. The continuous line shows control values.

As seen from the results, all the substances used showed a protective action on mitochondrial swelling. The action of albumin and glycogen proved to be significant,

whereas that of pectin and gum Arabic was much less evident.

We propose to further this work by series of *in vivo* experiments.

Riassunto. Si è studiato l'eventuale effetto protettivo, esercitato da alcuni polisaccaridi: il glicogeno di fegato, la pectina di mele, la gomma arabica, e dell'albumina del siero di cavallo su mitocondri di fegato di ratto sospesi in soluzione isotonica di saccarosio. Si è osservato come le suddette sostanze esercitino un effetto inibitore sopra il rigonfiamento spontaneo mitocondriale.

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Differential Incorporation of Labeled Amino Acids in the Territories of the Sea Urchin Blastula

In this preliminary note we shall report on some differences in the rate of incorporation of labelled amino acids between the animal and the vegetal territory of the blastula of the sea urchin *Paracentrotus lividus*.

The embryos have been exposed to labelled amino acids (S^{35} -dl-methionine, C^{14} -dl-leucine, C^{14} -dl-alanine, 0.05–0.1 μ C/ml) as follows: (1) beginning immediately after fertilization and remaining in the radioactive solution throughout development; (2) for 2–3 h immediately after fertilization after which they are transferred to sea water

where development was allowed to proceed; (3) for 30 min at certain stages of development and processed immediately afterwards. The results of experiments of type (1) and (2) supply information about the over-all metabolic history of the various cells and territories from fertilization to the time of collection, whereas the information given by the 'pulse' experiments of type (3) refers to the metabolic activity of the cells during the 30 min of exposure. The embryos were fixed in Carnoy and embedded in paraffin; after removal of the paraffin, the sections (5 μ) were brought to water through alcohol and covered with AR 10 Kodak autoradiographic stripping film. The exposure usually lasted from 3 to 5 weeks.

It has been found that, when procedures (1) and (2) were used, the tracks were uniformly distributed throughout the embryo in all the stages considered (Figure 1). On the other hand, when procedure (3) was applied, the distribution of tracks was still uniform throughout the embryo until the early blastula stage, but at the time of the appearance of the primary mesenchyme, the vegetal territory of the embryo and the cells of the primary mesenchyme appeared to contain considerably less silver grains than the rest of the embryo (Figure 2). Counts of the silver grains in the individual cells of the vegetal territory of the

mesenchyme blastula are hardly feasible due to the very ill-defined cell limits. This, however, can be done in the cells of the ectoblast and of the primary mesenchyme. The results of such counts on sections of mesenchyme blastulae which had been exposed to C^{14} -dl-leucine for 30 min are the following:

	Silver grains per cell	
	Experiment I	Experiment II
Ectoblast	15.6 ± 4.08	15.6 ± 3.59
Prim. mes.	5.4 ± 1.14	5.9 ± 0.51

Autoradiograms of mesenchyme blastulae of *Paracentrotus lividus* treated with C^{14} -dl-leucine:

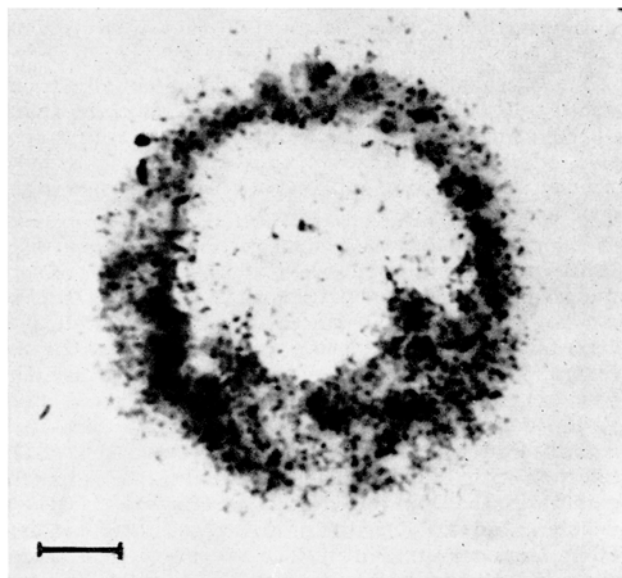


Fig. 1. From fertilization throughout development.

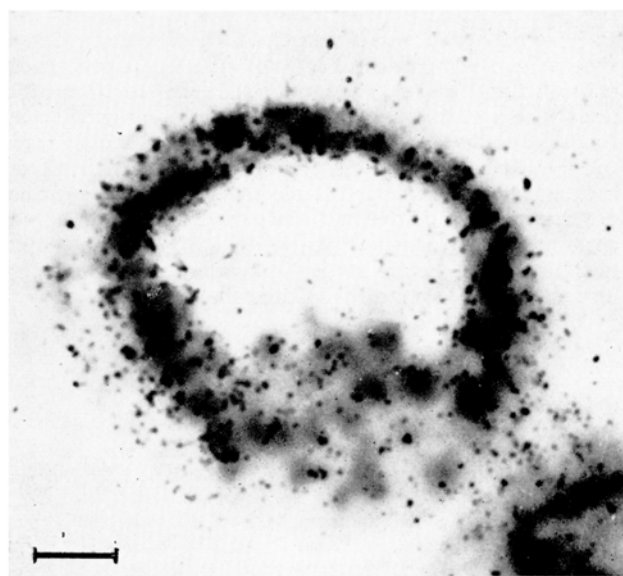


Fig. 2. For 30 min before fixation.

The reference line in the photographs corresponds to 10 μ .

No significant differences have been found among the various regions of the gastrula.

The results of the pulse experiments indicate that, in the vegetal territory of the mesenchyme blastula and in the cells of the primary mesenchyme, protein metabolism proceeds at a lower rate than in the animal region. This appears to be in agreement with the suggestion that vegetal proteins, and in particular proteins important for the differentiation of the primary mesenchyme, are synthesized during the early post-fertilization stages (Bosco and MONROY¹; MONROY, VITTORELLI, and GUARNERI²) and would also explain the uniform labelling when the exposure to the radioactive amino acids was started immediately after fertilization.

The present results seem to be somewhat at variance with those of MARKMAN³ and of IMMERS⁴. According to MARKMAN³, in the early blastula C^{14} -leucine is accumulated mainly in the animal region, whereas in the mesenchyme blastula it is the vegetal region that appears to be the most heavily labelled. In the gastrula, the strongest labelling is found in the archenteron. However, isolated animal halves 10 h after fertilization exhibit a greater incorporation than the vegetal halves. Also IMMERS⁴, who has used a C^{14} -algal protein hydrolysate, states that in the mesenchyme blastula the radioactivity 'is somewhat more concentrated in the vegetal part of the embryo'. At the present, we are unable to offer any satisfactory explanation of this discrepancy⁵.

Riassunto. Blastule con mesenchima primario di *Paracentrotus lividus* esposte a un trattamento di 30 min con S^{35} -metionina, leucina- C^{14} e alanina- C^{14} mostrano nel territorio vegetativo una densità di tracce considerevolmente minore che in quello animale. Se gli embrioni sono invece esposti all'aminoacido radioattivo durante tutto lo sviluppo o durante le prime ore dopo la fecondazione, tale differenza tra territori animale e vegetativo non si osserva. Si discute il significato di questi reperti.

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¹ M. BOSCO and A. MONROY, *Acta embryol. morphol. exp.* 3, 53 (1959).

² A. MONROY, M. L. VITTORELLI, and R. GUARNERI, *Acta embryol. morphol. exp.* 4, 77 (1961).

³ B. MARKMAN, *Exp. Cell Res.* 23, 197 (1961).

⁴ J. IMMERS, *Exp. Cell Res.* 18, 585 (1959); 24, 356 (1961).

⁵ This investigation has been supported by grants from the Consiglio Nazionale delle Ricerche and the Rockefeller Foundation.