

Riassunto. È stata studiata l'incorporazione dell'acetato-1- ^{14}C nei fosfolipidi, nel colesterolo e negli acidi grassi di sezioni di fegato normale e vacuolizzato di ratto. Essa non è variata, in nessuna delle frazioni lipidiche considerate, nelle sezioni di fegato colpito da degenerazione vacuolare. Si discutono i risultati in rapporto alle precedenti osservazioni sulla inibizione dell'incorporazione

degli amino acidi marcati nelle proteine delle sezioni di fegato vacuolizzato.

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Inhibitory Effect *in vitro* of some Polysaccharides and of Horse Serum Albumin on Mitochondrial Swelling

It is well known that certain substances, such as ATP, Mg^{++} , EDTA, K^{+1-3} , protect mitochondria, which are suspended in an isotonic saccharose solution, from spontaneous swelling. This property has been demonstrated recently in this Institute by CASU¹ in the case of serotonin (5-hydroxytryptamine).

In this study, the possible inhibitory effect *in vitro* on mitochondrial swelling by certain polysaccharides is examined, such as rabbit liver glycogen, apple pectin, gum Arabic, and by horse serum albumin, the protective effect of which on old or damaged mitochondria has already been shown by other authors^{2,3}.

These substances were used at various concentrations, and solutions 70, 35, and 17.5 mg of glycogen, pectin, and gum Arabic in 100 ml of 0.25 M sucrose were prepared.

The concentrations of albumin in 0.25 M sucrose were $1 \cdot 10^{-5}$ M, $0.5 \cdot 10^{-5}$ M, $0.25 \cdot 10^{-5}$ M.

Substances were supplied by: British Drug Houses, Liverpool (England): glycogen; Nutritional Biochemicals

Corporation, Cleveland, U.S.A.: pectin and gum Arabic; Istituto Sieroterapico Toscano, Siena (Italy): albumin.

Assays were carried out by reading, on a DU model Beckmann spectrophotometer, the extinction values of the mitochondrial suspensions, judging the degree of swelling by the gradual decrease in optical density, according to CLELAND⁴.

Mitochondria were obtained by fractioned centrifugation of albino rat liver homogenates, according to SCHNEIDER⁵. They were then suspended in 4 ml of 0.25 M sucrose. 10% homogenates in 0.25 M sucrose were prepared from 1 g of the tissue.

Two containers were used for each experiment, one of which contained 3 ml 0.25 M sucrose solution, the other an equal quantity of sucrose solution with substance dissolved in it. All 0.25 M sucrose solutions were buffered

¹ A. CASU, Exper. 16, 489 (1960).

² B. SACKTOR, J. Gen. Physiol. 37, 343 (1952).

³ E. C. WEINBACH, J. biol. Chem. 234, 1580 (1959).

⁴ K. W. CLELAND, Nature 170, 497 (1952).

⁵ W. C. SCHNEIDER, J. biol. Chem. 165, 585 (1946).

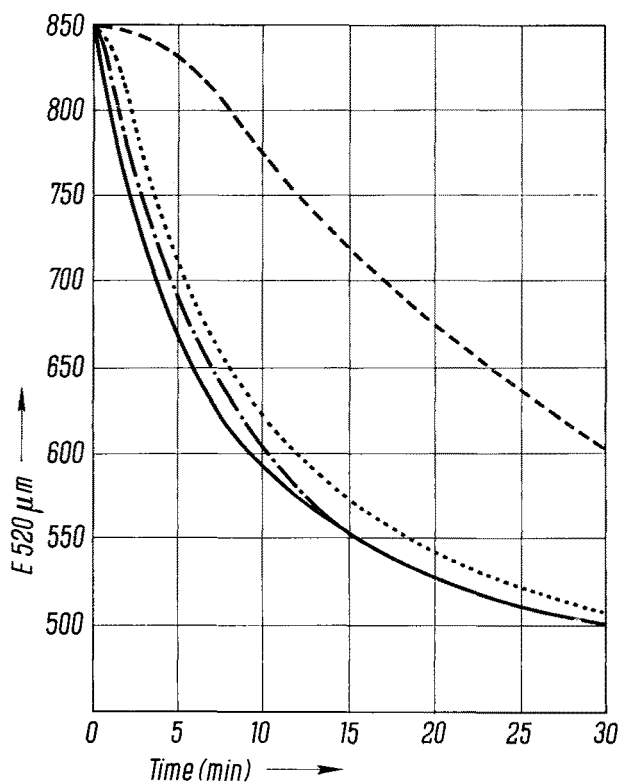


Fig. 1.

Inhibiting effect on mitochondrial swelling produced by glycogen.

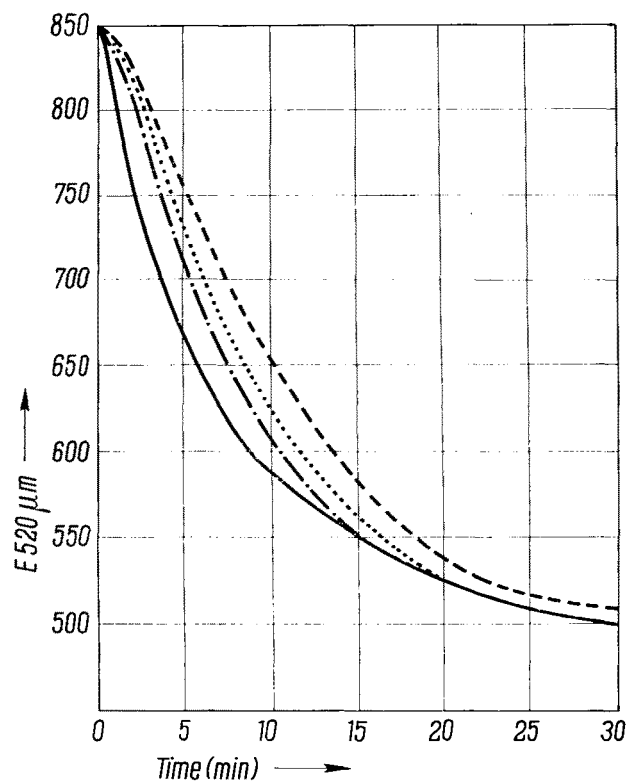


Fig. 2.

Inhibiting effect on mitochondrial swelling produced by pectin.

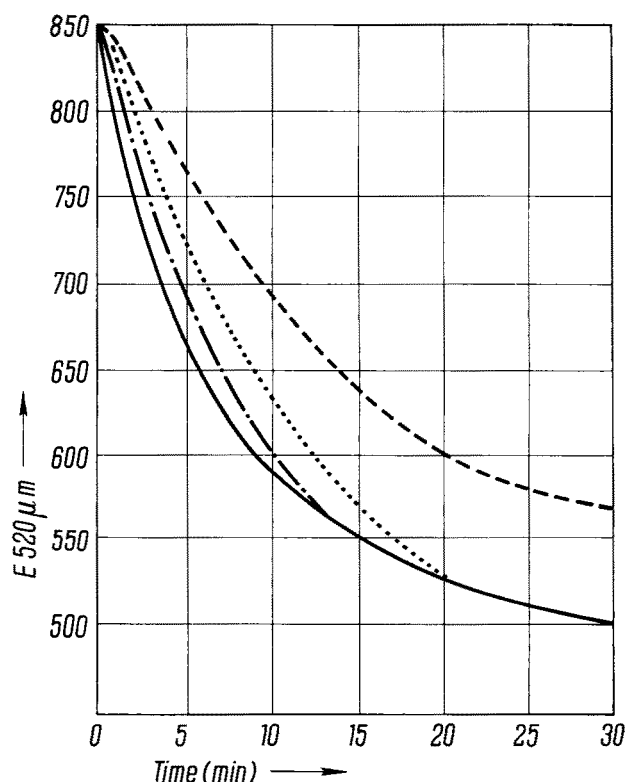


Fig. 3.

Inhibiting effect on mitochondrial swelling produced by gum Arabic.

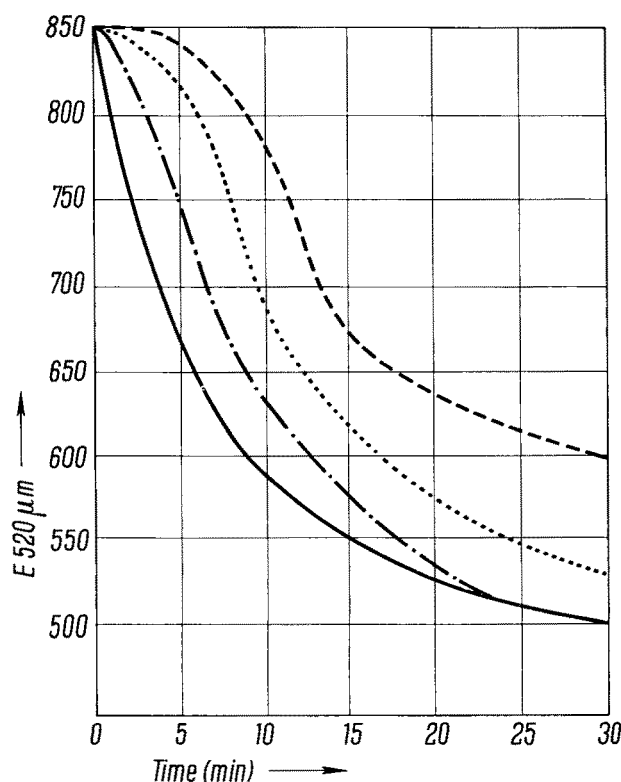


Fig. 4.

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.