Zusammenfassung

Bei Tenebrio-Larven werden cyclische Veränderungen der Prothoraxdrüsen beschrieben. Sie bestehen in Vergrösserung der Drüse, verbunden mit Bildung von Körnchen, Fibrillen und Sekrettröpfchen im Cytoplasma. Etwa zwei Tage vor der Häutung wird das Sekret ausgestossen und die Drüse wieder verkleinert. Im letzten Larvenstadium liegt diese kritische Phase 24 h nach Aufhören des Fressens. Die Drüse degeneriert in der Puppe bei Beginn der Augenpigmentierung.

Activation of Pancreatic Trypsinogen by Liver and Kidney Mitochondria

Following upon observations made in our Institute (in publications for the 2nd Italian Congress of Histochemistry, at Florence, 1959) on the inhibition of succinoxidasic activity of liver mitochondria by pancreatic granules and mitochondria, I decided to investigate whether such inhibition might be related to a tryptic action of the isolated pancreatic structures on liver mitochondria. For the first time, I was able to demonstrate that, like pancreatic granules, so also pancreatic mitochondria can develope tryptic activity when activated by enterokinase[1].

Afterwards SCHEPOWALNIKOW[2] demonstrated the existence of enterokinase, several discussions occurred between the various authors about its identity and action. PAVLOV, BAYLISS[3], ZUNZ, VERNON, HAMBURGER, STASSANO, and others concluded that there is more than one proteolytic enzyme of pancreatic production. VERNON stated that, to activate trypsinogen into trypsin, enterokinase was not indispensable, as the simple addition of trypsin to trypsinogen was enough to catalyse the reaction. Finally KUNITZ[4–6] observed that it was possible to obtain activation of trypsinogen by a kinase of Penicillium. NORTHROP and KUNITZ[7] concluded that reaction trypsinogen-trypsin can be catalysed by the same trypsin, by kinase of Penicillium, and by enterokinase. Considering that such a reaction could perhaps be catalysed by isolated structures of liver, and or of other organs, I considered the action of liver and other organs mitochondria on pancreatic trypsinogen.

Material and Method. For my research I used eight guinea pigs having an average weight of about 300 g each. Immediately after having killed each animal by beheading, I collected its pancreas and the other organs to be examinated; after having homogenized each organ in sucrose solution 0.25 *M*, I made fractionated centrifugation in cold room for 10 min at 2000 g on a first time, and then for 15 min at 10000 g; by this method I obtained isolated mitochondria of the various organs (liver, kidney, muscle, heart, lung, spleen, suprarenals) by 1 g of fresh organ.

In the meantime I homogenized pancreas, and by the method described by NOVELLI[8] I isolated in sucrose solution 0.88 *M* granules and mitochondria of pancreas that were suspended for a second time together in distilled water. While mitochondria isolated by 1 g of fresh organ were put into 2 ml of distilled water, a suspension of pancreas was prepared separately and total N_2 was titred by micro-Kjeldahl method. 1 ml of pancreas titred suspension was put with 1 ml of isolated mitochondria in test-tubes containing denaturated hemoglobin, and were incubated for 10 min at 25°C according to the method of ANSON[9]; in the meantime a control was prepared for each test. Complete description of this method has been already reported by myself in another work[1].

	Milliequivalents of tyrosin $\times 10^{-4}$ per 1 mg of pancreatic N_2	Tryptic Action
Liver mitochondria + pancreas	550.38 ± 103	Present
Kidney mitochondria + pancreas	284.80 ± 44	Present
Muscle mitochondria + pancreas	—	Absent
Heart mitochondria + pancreas	—	Absent
Suprarenals mitochondria + pancreas	—	Absent
Spleen mitochondria + pancreas	—	Absent
Lung mitochondria + pancreas	—	Absent

Tryptic activity was titrated by Beckman spectrophotometer on the basis of the tyrosin developed, and milliequivalents of tyrosin $\times 10^{-4}$ were calculated on the basis of 1 mg of pancreatic N_2.

Conclusions. The results of this research permit the conclusion that liver mitochondria contain an activator of pancreatic trypsinogen that acts like the activators previously described, that is by producing the reaction trypsinogen-trypsin. Kidney mitochondria were demonstrated to have the same capacity to activate trypsinogen, although at a lower rate.

As to what concerns the nature of the activator contained by liver and kidney mitochondria, the hypothesis that it could be the same enterokinase appears the most probable, although it still needs specific research for confirmation.

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Riassunto

Si dimostra la possibilità di catalizzare la trasformazione del tripsinogeno in tripsina mediante aggiunta di mitocondri isolati dal fegato e dal rene; i mitocondri del muscolo, polmone, cuore, surrene, milza non catalizzano tale reazione. La significatività dei risultati viene accertata calcolando il «*t*» di Fisher.

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