

Thermal Exposures of Ectoderm Cells Isolated from Amphibian Gastrulae

The neutralizing effect of various unrelated chemical compounds acting upon gastrula ectoderm in amphibia can be understood if one accepts HOLTFRETER's<sup>1</sup> explanation of a sublethal cytolysis of the cells as the "common denominator" of all these reactions. HOLTFRETER's hypothesis gains additional support from the fact that almost all the artificial inductors so far tested produce a sublethal cytolysis. In order to explore the underlying mechanism of this sublethal cytolysis of cells, a study of the action of thermal exposures on the isolated ectoderm cells of gastrula has been made; an attempt has also been made to follow the basophilia after such a thermal treatment.

The presumptive ectoderm cells were removed from early gastrulae of *Triturus alpestris*. In a specially designed thermostat, the cells were exposed for the desired length of time, the temperature varying between 26° and 42°. Following the heat treatment, the cells were transferred to normal HOLTFRETER solution at 19° for culture. The total number of explants made was 150. The minimal period of culture was 7 days, the maximal 14 days. The explants were fixed in ZENKER's fluid with acetic acid and the sections were stained in Toluidin blue to follow the basophilia of the cells.

*Discussion.* The foregoing experiments make it clear that thermal shocks exerted on the isolated gastrula ectoderm are ineffective in producing neuralization. The changes produced by a sublethal cytolysis are thus not necessarily the same in heat treated and chemically treated cells. It looks as if a certain "type" of mild cytolysis, but not all cytolysis, can lead to neural differentiation.

BRACHET's claim that nucleoprotein granules play a part in the process of induction and differentiation of cells is in agreement with the failure of differentiation in our heat-treated ectoderm cells: whenever there is a weak or no cytoplasmic basophilia in the cells, there is invariably a blocking of development. That heat shocks result in the depolymerization of DNA with an increase in the maximum of ultraviolet absorption (THOMAS<sup>2</sup>), and produce a complete inhibition of ribonucleic acid synthesis (STEINERT<sup>3</sup>), is already known.

<sup>1</sup> J. HOLTFRETER, Symp. Soc. exp. Biol. 2, 17 (1948).  
<sup>2</sup> R. THOMAS, Exper. 7, 261 (1951).  
<sup>3</sup> M. STEINERT, Bull. Soc. Chim. Biol. 33, 549 (1951).

At the low range of temperature shocks, some of the cells may become revitalized and differentiate into epidermal structures. This confirms, on explants, what BRACHET<sup>1</sup> had observed after implantation of heat-treated cells into normal gastrulae. He has observed that this revitalization of the cells is linked with a reappearance of the basophilic granules; we have been able to observe cytochemically a similar phenomenon in our cells. But one should insist on the mode of differentiation shown by the revitalized cells: they become epidermal and not neural. Thus the cytolytic action of the heat, if there is any, does not initiate the neuralization of the isolated gastrula ectoderm cells, in contrast with the cytolytic actions of chemicals. It is thus clear that neuralization can only result from a certain "type" of sublethal cytolysis and that thermal exposures in the temperature range studied do not provide for such a tissue reaction in isolated condition. S. MOOKERJEE<sup>2</sup>

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Résumé

Des explantats ectodermiques de gastrulas de *Triturus alpestris* ont été soumis à des chocs thermiques dans des conditions variées; il ne s'est jamais produit de neuralisation des explantats, dont la basophilie était très faible. Il faut en conclure que la neuralisation des explantats ectodermiques ne se produit que lorsque les cellules ont été soumises à un type particulier de pré-cytolyse.

<sup>1</sup> J. BRACHET, Symp. Soc. exp. Biol. 63, 173 (1952).  
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Abhängigkeit der Katalaseaktivität von der H<sub>2</sub>O<sub>2</sub>-Konzentration

Die seit mehr als 20 Jahren beschriebene Abhängigkeit der Katalaseaktivität (monomolekulare Reaktionskonstante) von der verwendeten H<sub>2</sub>O<sub>2</sub>-Konzentration wird nach Untersuchungen von THEORELL<sup>1</sup>, BONNICHSEN, CHANCE und THEORELL<sup>2</sup> als Artefakt angesehen, das

<sup>1</sup> H. THEORELL, Exper. 4, 100 (1948).  
<sup>2</sup> R. K. BONNICHSEN, B. CHANCE und H. THEORELL, Acta chem. scand. 1, 685 (1947).

Results

Thermal exposure at °	No. of cases	Time exposed, in hours or minutes	Nature of basophilia	Type of differentiation
26	12	1 hour	low	blocked
28	12	1 hour	low	blocked
28	8	24 hours	low	blocked
20	12	1 hour	weaker	blocked
30	8	2 hours	weaker	blocked
32	8	3 hours	very weak	blocked
34	12	1 hour	very weak	blocked
34	8	2 hours	very weak	blocked
35	8	2 hours	very weak	blocked
37	12	1 hour	almost removed	blocked
37	8	3 hours	almost removed	blocked
40	12	5 minutes	almost removed	blocked
40	8	15 minutes	almost removed	blocked
42	12	3 minutes	removed	blocked
Control (19)	10	—	rich	epidermal