

quelques-unes des données fondamentales de la cytophysiologie:

Hétéroauxine { phosphatases
acide ribonucléique → synthèse protéique
Activateur Système activé

Nous tenons à exprimer notre profonde reconnaissance à M. le Prof. F. CHODAT, tant pour ses judicieux conseils que pour les facilités de travail qu'il nous offre dans son laboratoire.

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Summary

Heteroauxin (optimal concentration 10^{-5}) activates the germination of the zygotes and the dissolution of their RNA nuclear cap in *Allomyces javanicus*.

Alkaline phosphatase has been located cytochemically in the cytoplasm surrounding the RNA nuclear cap.

These new observations lead the author to postulate a relationship between the stimulating action of heteroauxin, activation of the phosphatases and awakening of the RNA-protein synthesis dynamic system during the germination of the zygotes in *Allomyces*.

Effect of Inanition and Adrenalectomy on the Electrophoretic Patterns of Soluble Lymphatic Tissue Proteins

In view of the remarkable modifications undergone by lymphatic tissue in some experimental conditions, the increase in weight and in lymphocyte number after adrenalectomy, and the marked loss of weight and of lymphocytes during fasting (see DOUGHERTY¹), it seemed interesting to ascertain if these morphological and functional changes were accompanied by modifications in the electrophoretic patterns of soluble cell proteins.

Experimental.—Male albino rats of the *Italo* strain bred in this Department, weighing 120–130 g and fed a standard diet (RANDOIN and CAUSERET²) were used throughout. They were divided into three groups: normals: 5 pools with a total of 22 rats; adrenalectomized: 5 pools with a total of 19 rats; fasted: 3 pools with a total of 18 rats.

Adrenalectomy was performed by the dorsal approach under light ether anaesthesia, and after the operation rats were given 1% NaCl solution to drink. If NaCl was withdrawn, the great majority of the adrenalectomized animals died in a few days. The weight decrease during 4 days of fasting was, on the average, 26%.

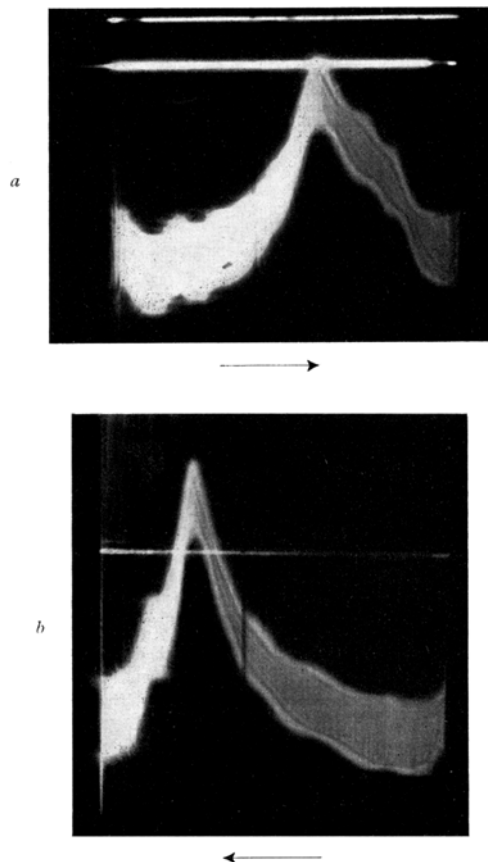
The animals were killed 4 days after the adrenalectomy or after 4 days of fast. Immediately lymphatic tissue was carefully dissected free of connective tissue and fat and kept at ice temperature during the collection of the rest of the tissue.

As representative of lymphatic tissue, the following organs were taken: 6 neck lymph nodes, 4 axillary ones, 4 inguinal ones, the whole mesenteric chain and the thymus.

Tissues from 3–6 rats, according to the experimental conditions, were pooled, weighed and dispersed in

diethyl-barbiturate buffer (pH 8.6, μ 0.1) with a glass homogenizer (POTTER and ELVEHJEM³) kept in cracked ice. Dilution of the dispersion was 1: 2.5. The tissue suspension was then centrifuged in the cold room for 15 min at 21,000 g in a Servall type SS-1 centrifuge.

The supernatant was carefully decanted and centrifuged again at 21,000 g for 1 h. The clear supernatant was then dialyzed for 16 h at 2° against 1 l of diethyl-barbiturate buffer. The electrophoresis was performed on the dialyzed fluid (centrifuged again if necessary) with the apparatus of TISELIUS (PERKIN-ELMER, Model 38) in a 2 ml rectangular cell. Time of electrophoresis was 5,400 s, with a current of 10 mA.



Electrophoretic pattern of soluble proteins of lymphatic tissue.
a Ascending limb; b descending limb.

Relative protein concentration was determined on photographic diagrams obtained with the scanning method of LONGSWORTH⁴. Enlarged patterns of the ascending cell limb were used. The conductivity of buffer and protein solutions was measured with a WHEATSTONE bridge (LEEDS and NORTHRUP).

Total nitrogen of the protein solutions was determined by the micro-Kjeldahl method.

Results. The electrophoretic pattern of soluble lymphatic tissue proteins was similar in all the analyses performed.

Very early in the electrophoretic run two small components appeared which rapidly traversed the cell and disappeared before the other slower components

¹ T. F. DOUGHERTY, *Physiol. Rev.* **32**, 379 (1952).

² L. RANDOIN and J. CAUSERET, *Bull. Soc. Sci. Hyg. Aliment. Paris* **14**, 1 (1947).

³ V. R. POTTER and C. A. ELVEHJEM, *J. biol. Chem.* **114**, 495 (1936).

⁴ L. G. LONGSWORTH, *J. Amer. chem. Soc.* **61**, 529 (1939).

Table I. — Relative percentage of the electrophoretic components of soluble proteins of lymphatic tissue in normal, adrenalectomized and fasted rats (means \pm S.E.M.).

Condition	Pools analyzed n.	Electrophoretic components					
		a	b	c	d	e	f
Normals	5	2.39 \pm 0.17	13.11 \pm 1.08	63.25 \pm 2.19	11.89 \pm 1.66	5.73 \pm 0.52	4.06 \pm 1.34
Adrenalectomized . . .	5	1.78 \pm 0.14*	8.97 \pm 0.81*	67.14 \pm 1.55	11.13 \pm 0.57	6.73 \pm 1.03	2.49 \pm 1.50
Fasted for 96 h.	3	1.70 \pm 0.26	11.43 \pm 0.27	62.66 \pm 2.15	16.40 \pm 0.52	5.97 \pm 1.12	3.20 \pm 1.01

* Differences with normals statistically significant ($P < 0.05$; Student's test).

were clearly separated. They had a mobility respectively of 14.4 and 9.6×10^{-5} cm² volt/s. Since at the end of the run, they were not present in the electrophoretic pattern, their percentage is not included in the data of Table I.

The final electrophoretic pattern is reproduced in the Figure. 6 components identified with the letters from *a* to *f* are apparent, of which *c* is quantitatively the most important, corresponding to more than 60% of the total proteins. The mobilities of these components are tabulated in Table II.

From Table I it is evident that no qualitative modifications took place in the patterns from adrenalectomized or fasted rats in comparison with the normals. From a quantitative point of view, the analysis of soluble proteins from adrenalectomized rats showed a statistically significant percentage decrease of components *a* and *b* in comparison with the normals.

Table II.—Mobilities of electrophoretic fractions of soluble proteins of lymphatic tissue

Fraction . . .	a	b	c	d	e	f
Mobility* . . .	6.07	5.58	4.59	3.28	2.54	1.64

* $\times 10^{-5}$ cm² volt⁻¹ s⁻¹.

Soluble proteins of normal lymphatic tissue have been studied by ABRAMS and COHEN⁵ and by ROBERTS and WHITE⁶, who found 6–7 components with a wide range of mobilities. In extracts of isolated lymphocytes, WHITE and DOUGHERTY⁷ and HARRIS, MOORE, and FARBER⁸ found 6–7 or respectively 3 components. Our results are therefore in broad agreement with those of the previous authors.

The main result of the present investigation is that in the experimental conditions employed, in which lymphatic tissue undergoes marked weight and morphological variations, very small or no modifications are appreciable in the electrophoretic patterns of the soluble proteins. This is particularly striking in the case of fasting, when the loss of lymphocytes is great and the lymphatic organs appear to be constituted mainly of reticulo-endothelial cells.

After adrenalectomy, where an increase of the lymphocytes content of the tissues is evident, two small fast components show a statistically significant decrease. It seems appropriate to recall here that WHITE and DOUGHERTY⁷ found, in lymphocytes, isolated from lymph nodes draining territories in which an antigen had been injected, an increase in the fast electrophoretic

components. These results seem, therefore, to indicate a correlation between these fast components and the functional activity of lymphocytes (presumably increased after the injection of the antigen and decreased after adrenalectomy).

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Zusammenfassung

Elektrophoresediagramme von löslichen Proteinen des lymphatischen Gewebes wurden geprüft unter zwei experimentellen Bedingungen, die Gewichtsveränderungen des lymphatischen Gewebes in entgegengesetzten Richtungen bewirkten (rascher Gewichtsverlust bei Hungertieren und Gewichtszunahme nach Adrenalektomie).

Im Vergleich mit Kontrolltieren zeigten die Elektrophoresediagramme unter diesen Versuchsbedingungen keine qualitativen Veränderungen. Nur nach Adrenalektomie war die Konzentration zweier kleiner Komponenten signifikant geringer als bei Normaltieren.

C₁₁-Substituted Corticosteroids. Some D Ring Transformations in the Compound S Series

The current interest in substituted analogs of cortisone and hydrocortisone as anti-inflammatory agents has recently been stimulated by the reported synthesis of several compounds displaying heightened glucocorticoid activity as well as markedly divergent mineralocorticoid properties¹. In searching for new therapeutic agents of this type, we have prepared as model compounds several C₁₁-substituted derivatives of Reichstein's Compound S. The synthesis of the corresponding 11-oxygenated compounds and their biological activities has been reported elsewhere².

The preparation of C₁₁-substituted derivatives of Compound S was accomplished from Δ^4 -pregnene-14 α , 17 α , 21-triol-3, 20-dione (14 α -hydroxy Compound S)

¹ J. FRIED *et al.*, J. Amer. chem. Soc. 75, 2273 (1953); 76, 1455 (1954); 77, 1068 (1955); 77, 4181 (1955). — A. NOBILE *et al.*, J. Amer. chem. Soc. 77, 4184 (1955). — R. F. HIRSCHMANN, R. MILLER, R. E. BEYLER, L. H. SARETT, and M. TISHLER, J. Amer. chem. Soc. 77, 3166 (1955). — J. A. HOGG *et al.*, J. Amer. chem. Soc. 77, 4438 (1955). — E. VISCHER, CH. MEYSTRE, and A. WETTSTEIN, Helv. chim. Acta 38, 835 (1955).

² E. J. AGNELLO, B. M. BLOOM, and G. D. LAUBACH, J. Amer. chem. Soc. 77, 4684 (1955); Abstracts of 128th Meeting, Amer. Chem. Soc., Minneapolis, Minn., September 11–16, 1955, p. 50.

⁵ A. ABRAMS and P. P. COHEN, J. biol. Chem. 177, 439 (1949).

⁶ S. ROBERTS and A. WHITE, J. biol. Chem. 178, 151 (1949).

⁷ A. WHITE and T. F. DOUGHERTY, Endocrinology 36, 207 (1945).

⁸ T. N. HARRIS, D. H. MOORE, and M. FARBER, J. biol. Chem. 179, 369 (1949).