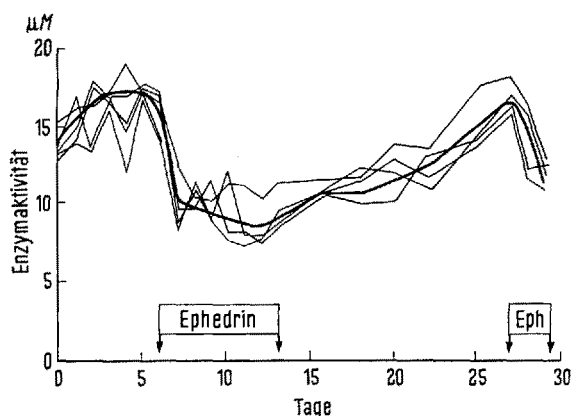


Iproniazid (Marsilid^R) hat *in vitro* in der Konzentration von $578 \cdot 10^{-3} \mu M/1$ ml keinen Einfluss auf die AdO-Aktivität¹. *In vivo* dagegen, einmal in der Dosis von 30 mg verabreicht, steigerte es deutlich die Enzymaktivität während 2 bis 3 Tagen, schon vom ersten Tage beginnend ($\bar{x}_1 = 13,64$, $\bar{x}_2 = 18,48$, $P < 0,001$) (Figur 1).

Reserpin (30 μg) verursacht 1 h nach der Verabreichung eine signifikante Steigerung der mittleren AdO-Aktivität ($\bar{x}_1 = 9,80$, $\bar{x}_2 = 12,14$, $P < 0,001$). Dieselbe Dosis, täglich während 5 Tagen verabreicht, führte zu einer sehr signifikanten Verminderung der mittleren Enzymaktivität (Kontrolltiere: $\bar{x}_1 = 12,89$ und $13,14$, $P < 0,7$; $\bar{x}_2 = 11,26$ und $8,33$, $P < 0,001$).



g. 2. Beeinflussung der AdO-Aktivität durch Ephedrin. — Einzelversuche, — Mittelwertelinie, ↓ — ↓ = Ephedrinverabreichung (150 μg täglich pro Ratte).

Ephedrin (150 μg täglich) bewirkt nach 1–2 Tagen eine deutliche, sehr signifikante Senkung der AdO-Aktivität (Kontrolltiere: $\bar{x}_1 = 13,28$, $\bar{x}_2 = 13,19$, $P < 0,9$; $\bar{x}_1 = 15,82$, $\bar{x}_2 = 9,90$, $P < 0,001$). Nach der Unterbrechung der Ephedrinverabreichung kehrt die Enzymaktivität allmählich zu den Anfangswerten zurück (Figur 2).

Unsere Resultate, die Adrenalin- und teilweise Reserpinwirkung betreffen, sind denen von NAKAJIMA und THUILLIER², die den Einfluss von neurotrophen Substanzen auf die Aktivität des Coeruloplasmins bzw. der *p*-Phenylen-diaminoxidase im Menschen- und Kaninchenserum untersucht haben, entgegengesetzt.

Summary. In rats adrenaline, noradrenaline, histamine and iproniazid induced in blood plasma a significant increase of the mean of the enzyme activity of 'adrenaline oxidases' (β -globulin-factors, different from coeruloplasmin, ferritin and catalase). Reserpine showed initially an increase and later a decrease and ephedrine caused a decrease of the enzymic activity.

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7. November 1962.

² H. NAKAJIMA und J. THUILLIER, *Neuropsychopharmacology* (Ed. by E. ROTHLIN, Elsevier, Amsterdam, London, New York, Princeton 1961), vol. 2, p. 500.

Phosphatase Activity During the Growth and Metamorphosis of *Tribolium confusum* Duval

Although extensive studies have been made on phosphatase activity in microorganisms and in plant and animal tissues, relatively few investigations have been made with insects. Several functions have been ascribed to this group of enzymes in biological processes. They are known to play an important role in intermediary metabolism. LAMBREMONT¹ found a positive correlation between age and decreased acid phosphatase activity in *Aedes aegypti*. A possible relationship of the alkaline phosphatase system to the detoxication mechanism in resistant flies was suggested by ROCKSTEIN and INASHIMA².

Since holometabolous insects undergo metamorphosis, it was thought that a study of phosphatase activity in *Tribolium confusum* might contribute towards a better understanding of the phenomena concerning morphogenesis and growth. The present paper includes the results of this investigation.

Experimental Procedure. The methods of rearing *T. confusum* have been described earlier³. For each determination, a suitable number of insects were weighed analytically and homogenized in a Elvehjem-Potter Homogenizer in 20 vol of ice-cold double-distilled water. The homogenate was passed through a 4-layered cheesecloth to remove the cellular debris and subsequently used for the assay of phosphatase activity. Since the total

homogenate showed higher phosphatase activity than any cellular or subcellular fractions (including the high-speed supernatant fraction obtained by centrifuging the homogenate at $105\,000 \times g$ in Spinco ultracentrifuge, Model L, for 2 h), it was used as the enzymatic source. The quantitative estimation of the phosphatase activity was based upon the techniques of BODANSKY^{4,5} with some modifications. The acid and alkaline phosphatase activity was determined by measuring the inorganic phosphorus released from citrate-buffered β -glycerophosphate (pH 5.4) and tris(hydroxymethyl aminoethane) buffered β -glycerophosphate (pH 8.3) respectively. The reaction mixture was incubated for 30 min at 37°C. Initial pre-incubation of each, the buffered substrate and the homogenate separately for 5 min, was found to have a favourable effect on the enzymatic activity.

Results and Discussion. Acid phosphatase: There appears to be a relatively higher activity of this enzyme during the embryonic and larval periods of *T. confusum*

¹ E. N. LAMBREMONT, *Ann. Ent. Soc. Amer.* 53, 87 (1960).

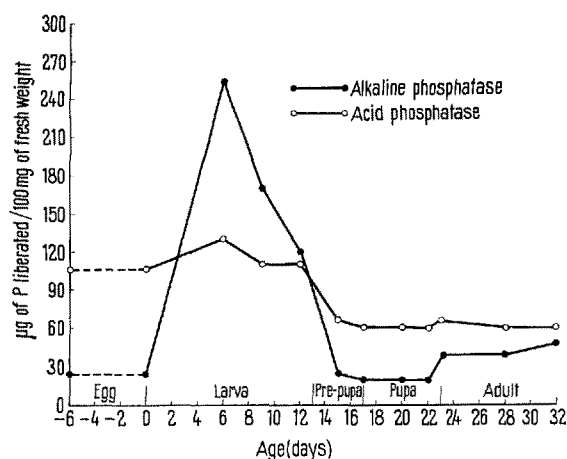
² M. ROCKSTEIN and M. INASHIMA, *Bull. Brooklyn Ent. Soc.* 48, 20 (1953).

³ K. D. CHAUDHARY and A. LEMONDE, *Canad. J. Zool.* 40, 375 (1962).

⁴ A. BODANSKY, *J. biol. Chem.* 101, 93 (1933).

⁵ A. BODANSKY, *Amer. J. clin. Path., Tech. Suppl.* 1, 51 (1937).

as compared to its correspondingly lower activity in the pupae and the adults (Figure). The highest acid phosphatase activity has also been reported in the egg stage of the stable fly and the house fly by ASHRAFI⁶ and BARKER and ALEXANDER⁷ respectively. It appears from these findings that increased acid phosphatase activity might be related to rapid cell division, especially during morphogenic changes in the embryonic development and during the larval growth of insects. At the time of pupation in *T. confusum*, a drop in the activity of this enzyme occurs, which seems to be in agreement with the fact that the insect ceases to grow during this period while it is undergoing an internal reorganization of the tissues necessitated by adult development. On adult emergence in *T. confusum*,



Phosphatase activity during the growth and metamorphosis of *T. confusum* Duval. Each point on the curves represents an average of 5 determinations. Dotted lines in the egg period indicate that only one stage during this period was analysed for phosphatase activity.

no major variation in the level of acid phosphatase activity occurs.

Alkaline phosphatase: While it is interesting to remark a tremendous increase in the activity of alkaline phosphatase activity in 7-day-old *T. confusum* larvae, the embryonic as well as the pupal stage seems to be deficient in the activity of this enzyme (Figure). Our results mostly seem to be in conformity with the findings of BARKER and ALEXANDER⁷ in *Musca domestica*. It therefore seems suggestive that alkaline phosphatase might play an important role in the phenomena concerning growth rather than morphogenesis. Alkaline phosphatase was also thought by YAO⁸ to be associated with growth in insects. Adult emergence in *T. confusum* was characterized by a slight rise in the activity of alkaline phosphatase⁹.

Résumé. Nous avons mesuré l'activité des phosphatases acide et alcaline au cours de la croissance et de la métamorphose de *T. confusum*. L'activité de la phosphatase acide est à son maximum au cours de l'embryogénèse et du stade larvaire. Il est donc possible que cette enzyme soit reliée aux phénomènes de la différenciation cellulaire et de la croissance. L'activité de la phosphatase alcaline est, au contraire, très élevée au cours de la croissance larvaire. Elle serait donc associée à la croissance seule.

K. D. CHAUDHARY and A. LEMONDE

Department of Biochemistry, School of Medicine, Laval University, Quebec (Canada), October 3, 1963.

⁶ S. H. ASHRAFI, Pakistan J. Sci. Ind. Res. 4, 70 (1961).

⁷ R. J. BARKER and B. H. ALEXANDER, Ann. Ent. Soc. Amer. 51, 255 (1958).

⁸ T. YAO, Quart. J. micr. Sci. 91, 89 (1950).

⁹ Acknowledgments. We acknowledge with thanks the technical assistance of Mr. S. UBERTELLI.

Effect of Adrenalectomy and Diet on the Activity of β -Galactosidase in the Small Intestine During the Postnatal Development of the Rat

Recently the development of β -galactosidase in the small intestine during postnatal development in the mouse (MAIO and RICKENBERG¹), calf (HUBER et al.²) and rat (ALVAREZ and SÁS³, DOELL and KRETCHMER^{4,5}) has again attracted attention. The activity of this enzyme is high at birth and falls rapidly during the weaning period. The reason for this loss of activity, however, is far from clear, in spite of efforts to show a correlation between changes of activity and changes in diet or in supply of lactose (ALVAREZ and SÁS³, DOELL and KRETCHMER⁴).

An attempt was therefore made to clarify the factors affecting the postnatal loss of β -galactosidase activity. Since the proximodistal distribution of some enzymes is different during the suckling period than later in life (non-specific esterase, KOLDOVSKÝ et al.⁶; alkaline phosphatase, KOLDOVSKÝ et al.⁶, VERNE and HEBERT⁷, MOOG⁸), this distribution was determined for β -galactosidase by the method of LEDERBERG⁹ (see Table I). From Table I it is evident that between day 15 and 20 postnatally, β -galactosidase activity decreases both in the proximal and

distal part of the small intestine. It can also be seen that activity is several times higher in the distal part than in the proximal part in rats 15-18 days old. In 20-day-old animals there is no significant difference between activity in the proximal and distal parts of the intestine. The distribution of β -galactosidase activity along the small intestine is thus very near to the distribution of non-specific esterase (KOLDOVSKÝ et al.⁶) and alkaline phosphatase

¹ J. J. MAIO and H. W. RICKENBERG, Biochim. biophys. Acta 37, 101 (1960).

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³ A. ALVAREZ and J. SÁS, Nature 190, 826 (1961).

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⁶ O. KOLDOVSKÝ, E. FALTOVÁ, P. HAHN, and Z. VACEK, in *The Development of Homeostasis*. Symposium ČSAV 1960 (Publ. House Czechoslov. Ac. Sci. Prague 1961, Academic Press 1962), p. 155.

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