controls receiving only selenium. All rats survived the experimental period, and there were no statistically significant differences (P > 0.05), as measured by the t-test, in weight gain and in the amount of food and water consumed between the 2 groups of animals. It is well known that young rats drinking water containing 2-3 ppm of selenite-selenium develop typical symptoms of chronic selenium intoxication 3,5-7, whereas the addition of 50 ppm of fluoride to the water does not affect their health 8.

The present findings are contrary to those of the only earlier study conducted by Moxon and DuBois2. They reported that the addition of 5 ppm of fluoride to the drinking water of young rats eating a diet containing 11 ppm of selenium, as seleniferous wheat, increased mortality and caused considerable depression in weight gain and in food and water intake compared with controls receiving only selenium. These symptoms seem to suggest an increased retention of selenium in the body of the animals probably through the combined action of selenium and fluoride. However, it should be pointed out that although in the present experiment rats drank water containing 10 times more fluoride than in the earlier study², nevertheless, the symptoms of selenium toxicity were not intensified. The effects of trace elements on chronic selenosis have remained largely unexplored,

Effect of fluoride on selenium toxicity in rats

Variables	Treatment	
	Se+F	Se
No. of rats	15 (15) a	15 (15)
Initial weight (g)	50.4 ± 1.8	50.1 ± 1.7
Weight gain (g)	82.1 ± 5.7	73.7 ± 4.4
Food intake (g)	9.7 ± 0.3	9.1 ± 0.3
Water intake (ml)	10.0 ± 0.5	9.2 ± 0.4

Values are given as means ± S.E. a No. of survivors in parenthesis.

with the possible exception of arsenic which was found to alleviate the toxic action of selenium in experimental animals 9, 10. Recent studies with rats indicated that the combined administration of zinc or uranium and selenium increased the toxicity of selenium¹¹.

In view of the findings of this study, it is suggested that in high seleniferous areas where people may be exposed to increased consumption of dietary selenium through locally produced and consumed foodstuffs, intake of fluoride for prevention of caries, either from a water supply or as medication, is not likely to increase the retention of ingested selenium. It was also shown recently4, that the presence of selenium in the diet does not decrease the amount of fluoride deposited in bones and teeth.

Zusammenfassung. Wenn junge Ratten gleichzeitig Selen und Fluor erhalten, sind die toxischen Eigenschaften von Selen nicht grösser, als in den Kontrollen, die nur Selenium erhielten. Dieses Resultat steht im Widerspruch zu früheren Feststellungen.

D. M. HADJIMARKOS

Department of Preventive Dentistry, University of Oregon Dental School, Portland (Oregon 97201, USA), 31 December 1968.

- ⁵ W. Buttner, J. Dent. Res. 42, 453 (1963).
- ⁶ D. M. Hadjimarkos, Experientia 23, 930 (1967).
- ⁷ H. A. Schroeder, J. Nutr. 92, 334 (1967).
- 8 H. G. Hodge and F. A. Smith, in Fluorine Chemistry (Ed. J. H. Si-MONS; Academic Press, New York 1965), vol. IV, p. 112.
- 9 I. ROSENFELD and O. A. BEATH, Selenium, 2nd edn (Academic
- Press, New York 1964), p. 182.

 10 O. A. LEVANDER and C. A. BAUMANN, Toxic appl. Pharmac. 9, 98 (1966).
- ¹¹ I. Rosenfeld, Agric. Exp. Station Bull. 414 (University Wyoming
- 12 The study was supported by the General Research Support Grant from the National Institute of Dental Research, USPHS.

Effect of Continuous Illumination on Mitochondria of the Rat Pineal Body

The influence of constant light upon the pineal body has been extensively studied by several investigators. A marked increase in pineal 5-HTPD activity 1,2 and a decrease of the serotonin content by one-half in rats under diurnal lighting conditions³, are some of the effects already described. They indicate that the metabolism of serotonin in the pineal body is highly stimulated by continuous illumination. In a previous paper4 we described changes in the 2 hypothalamic neurohumors of the rat, norepinephrine and serotonin, following 30 days of continuous illumination as well as 30 days of total darkness. The participation of mitochondria was also mentioned at that time.

This paper deals with mitochondrial alterations in the pinealocytes as revealed by the electron microscope. For a period of 30 days, male Sprague-Dawley rats were exposed to light of 70 lux, as measured at the bottom of the cage. One group of control animals was kept under

total darkness (< 0.001 lux) and another group had 14 h of light (70 lux) to 10 h of darkness (< 0.001) for the same period of time.

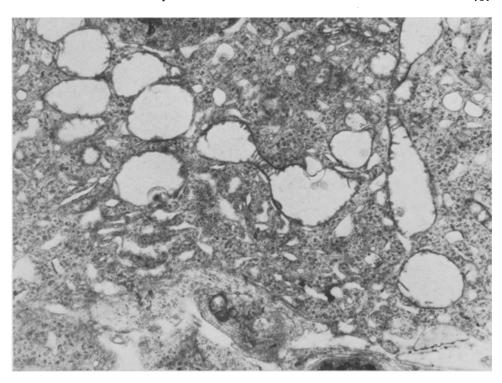
The Figure shows part of a pinealocyte. The striking feature of the electron micrograph are the enlarged and swollen mitochondria scattered over the cytoplasm. There is almost a complete absence of cristae mitochondriales. The matrix is pale and contains sparse flocculent material. Mitochondria are elongated with club-like deformities. The outer, double-membrane is intact.

¹ S. H. SNYDER and J. AXELROD, Biochem. Pharmac. 13, 803 (1964).

² S. H. Snyder, J. Axelrod, R. J. Wurtman and J. E. Fischer, J. Pharmac. exp. Ther. 147, 371 (1965).

³ W. B. Quay and A. Halevy, Physiol. Zool. 35, 1 (1962).

⁴ E. RÜTHER, A. HALARIS and N. MATUSSEK, Med. Pharmac. exp. 17, 139 (1967).



Part of a pinealocyte of the rat after 30 days of continuous illumination. Mitochondria are extremely swollen and deformed. × 18,000.

All animals kept under artificial lighting presented the typical appearance of swollen mitochondria. Swelling of mitochondria has been described by previous investigators both after in vitro and in vivo conditions in connection with cell injury. Animals kept in darkness or a light-dark cycle had intact mitochondria. Our electron micrographs of the rat hypothalamus provided findings similar to those found in the pineal body. These results will be reported in a later publication.

Since swelling of mitochondria could not be registered under conditions involving an increased metabolism of norepinephrine, we assume that the intensified metabolism of serotonin under continuous illumination is responsible for this effect. Mitochondrial swelling is a rather stereotypical reaction of the organelle under various conditions and in response to various agents. Absence of ADP (or ATP), a decreased P:0 ratio 6,7, or the uncoupling of oxydative phosphorylation8,9 are some of the factors leading to this ultrastructural alteration. Since serotonin has been proved to suppress phosphoryla $tion^{10, 11}$, it is possible that swelling of mitochondria can be accounted for by the increased serotonin metabolism induced by exposure to continuous illumination. A detailed discussion of this explanation, as well as of a possible physiological significance of the described effect, will be published elsewhere.

Zusammenfassung. Die Rattenepiphyse von Tieren, die 30 Tage lang einer ständigen Belichtung von 70 Lux Stärke ausgesetzt worden waren, wurde elektronenoptisch untersucht. Alle Tiere wiesen mitochondriale Veränderungen der Pinealozyten auf. Diese Veränderungen sind vermutlich auf den erhöhten Serotoninstoffwechsel zurückzuführen, der durch die ständige Belichtung induziert wird, da die Kontrolltiere keine derartigen Abartigkeiten zeigten.

A. Halaris and N. Matussek

Max-Planck-Institut für Psychiatrie, Biochemische Abteilung, 8 München 23 (Germany), 28 January 1969.

- ⁵ A. L. Lehninger, Physiol. Rev. 42, 467 (1962).
- ⁶ A. Fonnesu and C. Severi, J. biophys. biochem. Cytol. 2, 293 (1956).
- ⁷ M. U. DIANZANI, Biochem. J. 65, 116 (1957).
- 8 H. W. Altmann, Handbuch der allgemeinen Pathologie (Springer Verlag, Berlin, Göttingen, Heidelberg 1955), Vol. 2/1, p. 419.
- A. L. Lehninger, The Mitochondrion (Benjamin, New York 1964).
 E. Walaszek and L. G. Abood, Proc. Soc. exp. Biol. Med. 101, 37 (1959).
- ¹¹ S. G. ALIVISATOS, F. UNGAR, O. CALLAGHAN and C. BIAVA, Nature 201, 47 (1964).

On the Biosynthesis of Biliverdin-IX γ in Pieris brassicae

Recently¹ we identified pterobilin, the blue-green tegumental pigment of caterpillars of the cabbage butterfly *Pieris brassicae*, as biliverdin- $IX\gamma$ (Ia). Its biosynthesis is of particular interest because it is the first known natural bile pigment which has not the $IX\alpha$ structure. Bile pigments of vertebrates are formed from glycine and acetate via porphyrines and hemes²; glycine is also incorporated into biliverdin- $IX\alpha$ in Orthopteres³. The

question arose whether pterobilin is formed from small precursors in the caterpillars or whether it is derived from a hitherto unknown pyrrole compound ingested with food. We therefore tested the possible incorporation of labelled glycine into biliverdin-IX γ .

220 caterpillars (5th stage)⁴ are injected each with 1.36 μ Ci of 1.2 ¹⁴C-glycine (specific activity 90 mCi/mM) dissolved in 5 μ l of the Ringer solution for insects, and