können wir allerdings mit unseren Resultaten nicht geben und möchten auch die Frage offen lassen, ob nicht eine eventuelle postoperative Durchblutungsstörung des Splanchnicusgebietes an der Entstehung des Glykogenanstieges beteiligt ist, da, wie ebenfalls aus Arbeiten von BLOOM und RUSSELL hervorgeht, durch Hunger eine Erhöhung des Herzglykogens erzielt wird.

E. LÜTHY, B. BISCHOF und H. RÖSLI

Medizinische Universitäts-Poliklinik Zürich, 23. Juni 1959.

Summary

By an artificial stenosis of the abdominal aorta, hypertrophy of the left heart is produced. Different degrees of narrowing of the aorta lead to different degrees of heart hypertrophy. After a starvation of 24 h, the glycogen content of the heart that is in a hypertrophic condition is higher than in normal hearts.

Lead Reactive Substances in Peripheral Synapses

The subneural apparatus of motor end-plates was first demonstrated with a supravital Janus-Green B staining by COUTEAUX[1]. Histochemical investigations revealed that the palisade-like structure of the subneural apparatus exhibits cholinesterase activity[2–8].

Biochemical investigations concerning the function of the myoneural synapsis suggest, however, that a number of other factors play an essential rôle in the transmission of impulses. According to KOSHTOYANTZ *et al.*[9], especially –SH compounds must be taken into account. The participation of active –SH groups in the structure of Coenzyme A, an essential factor in the manufacturing of acetylcholine, is a well-known fact[10]. Considering the very sensitive reaction of heavy metal salts with –SH groups (mercaptide reaction), it was decided to attempt to trace the subneural apparatus and other synaptical structures by means of lead compounds.

Technique and Results. Anesthesia was induced in rats by an intraperitoneal injection of urethane (0·2–0·3 g). One ml of the following solution was injected percutaneously into the short flexor muscle of the hind pad:

$Pb(NO_3)_2$ cryst.	1·0 g
Urea (or guanidine)	1·0 g
Formalin 4 p.c.	20·0 ml

15 min later the animals were killed and the muscles excised. Frozen sections were cut immediately, washed briefly in distilled water and immersed for a few seconds in a 2 p.c. aqueous solution of sodium sulfide. This treatment resulted in the characteristic microscopical pattern of the subneural apparatus, as had been anticipated.

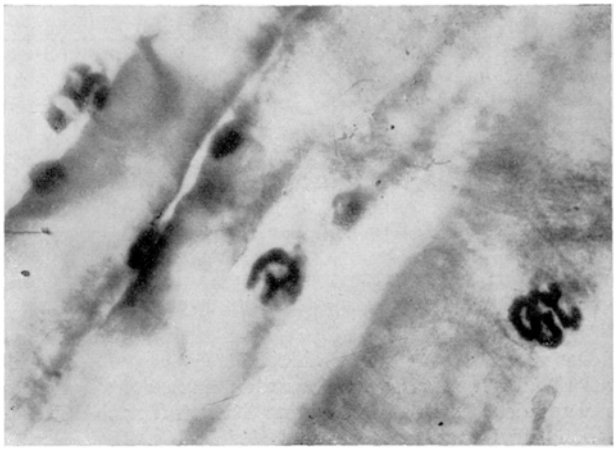

Fig. 1

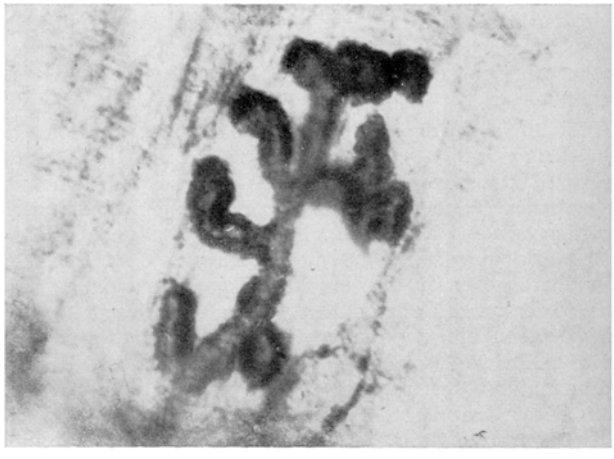

Fig. 2

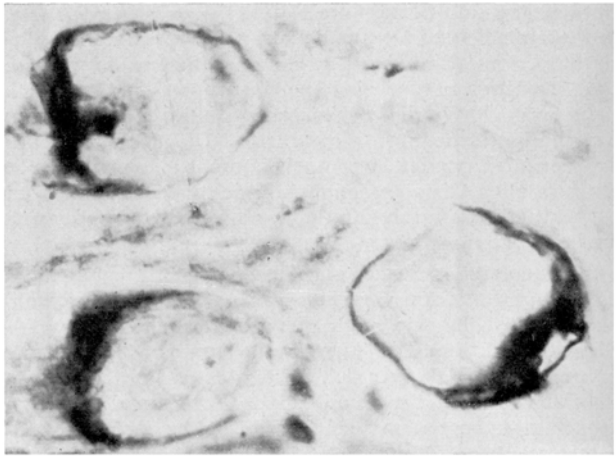

Fig. 3

Similar results were obtained, if fresh tissues were *immersed* in the aforementioned lead solution for 15 min, sectioned, and treated subsequently with Na_2S. Other muscles (e.g. diaphragm, intercostal muscle, gastrocne-

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mius) and vegetative ganglia were successfully investigated by means of this immersion technique.

Three photomicrographs illustrate our results. Figure 1 shows some subneural apparatus from the intercostal muscle of the rat. Note characteristic lining of the subneural apparatus and the slight background reaction of muscle fibres. Figure 2 (high power) demonstrates the double-contured arborisations of the apparatus; here and there also the palisade-like organisation of the 'organites' appear. Figure 3 illustrates three ganglionic cells from the ciliary ganglion of the hen. SZENTÁGOTHAI *et al.*[11] demonstrated that the goblet-like synaptical structures of these ganglionic cells exhibit cholinesterase activity. Our method reveals a very strong reaction in these goblets, while cell bodies react only faintly.

Discussion. The native condition of the tissues seems to be indispensable for a histological demonstration of lead-reactive synaptical substances. It was found that neither fixed tissues, nor fresh frozen sections exhibited any reaction with lead salts. Destructive action of fixation and/or freezing on fine cytochemical structures had been observed also in our previous investigations concerning the microscopical localisation of potassium in nerve fibres[12].

With regard to the formalin and urea content of the lead-solution employed, it should be noted that these substances are not essential for the reaction. Formalin, however, renders possible a better histological localisation, in so far as thinner sections may be obtained. On the other hand, urea liberates buried –SH groups[10] which seem to be necessary for a complete reaction. Employing a lead-solution without urea, the results are less satisfactory.

As demonstrated by the photomicrographs, morphological localisation of lead-reactive synaptical structures is identical with that of cholinesterase activity. The question may arise whether lead-reaction is not due to cholinesterase[13]. Although the lead nitrate solution employed contains no substrate to be hydrolysed by the enzyme, lead ions possibly combine with some part of the cholinesterase molecule, e.g. with the supposed –SH groups of this enzyme (NACHMANSOHN and LEDERER[14]). There are some observations, however, which oppose such an explanation. Cholinesterase activity is not affected by freezing, while lead-reaction is abolished by it. On the other hand, a pre-incubation in eserine (10^{-3} *M*, for 10 min) inhibits cholinesterase activity, while lead-reaction is not influenced by it. An isotonic solution of cadmium chloride, applied prior to the lead-reaction, abolishes it, while the cholinesterase activity remains unchanged. Finally, cholinesterase activity of motor end-plates may be demonstrated histochemically by means of indoxylacetate or thiolacetic acid in sections previously treated with a lead solution. It does not seem plausible, therefore, that lead reaction should be due to cholinesterase, but rather to another compound having the same histochemical localisation. Further investigations on this subject are in progress.

GY. SÁVAY and B. CSILLIK

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Zusammenfassung

Durch vitale bzw. supravitale Behandlung mit $Pb(NO_3)_2$ werden die subneuralen Apparate der quergestreiften Muskeln sowie die synaptischen Strukturen der vegetativen Ganglien dargestellt. Die Frage wird diskutiert, ob das bleireaktive Material mit den Sulfhydrylsubstanzen bzw. mit der Cholinesterase identisch sei.

Veränderungen der elektrischen Hirntätigkeit bei Katzen nach Röntgentotalbestrahlung[1]

Signifikante EEG-Veränderungen wurden bisher erst nach massiven Strahlendosen (4000–6000 r) gefunden[2–4], die ohnehin histologische Dauerschäden verursachen. Bei schwächeren Dosen konnten jedoch keine sicheren EEG-Veränderungen beobachtet werden[2]. Demgegenüber wird berichtet, dass die Strahlenempfindlichkeit bedingter Reflexvorgänge – insbesondere in komplexeren Testsituationen – auch bei schwachen Strahlendosen nachweisbar sei[5]. Es wurde deshalb untersucht, ob Veränderungen der elektrischen Hirntätigkeit nach Bestrahlung mit schwächeren Dosen erkennbar sind, wenn empfindlichere elektrophysiologische Messmethoden angewandt werden.

Methodik. Bei 5 frei beweglichen Katzen wurde die elektrische Hirntätigkeit mittels konzentrischer, stereotaktisch implantierter, chronischer Elektroden kortikal (sensomotorische und parietale Rinde) und subkortikal abgeleitet (mesenceph. subst. reticularis; thalamus: nucl. ventr. ant. (VA), centr. med. (CM) und centr. lat. (CL); hippocampus und nucl. amygdalae). Zugleich wurden nachfolgende Reizantworten auf elektrische Reizung untersucht und deren Schwellen bestimmt: 1. «Arousal reaction»[6] nach Reizung (150 Imp./sec, 1 msec) der mesencephalen subst. reticularis; 2. «Recruiting response»[7] nach Reizung (7 Imp./sec, 1 msec) des diffusen thalamischen Projektionssystems (VA, CM, CL); 3. Rhinencephale elektrische Nachentladung nach Reizung (150 Imp./sec, 1 msec) des Hippocampus oder des Mandelkerns. Alle Messungen wurden in einem schalldichten Käfig vorgenommen, wobei das Verhalten der Tiere durch eine Glasscheibe beobachtet werden konnte. Die Ableitung der elektrischen Hirntätigkeit erfolgte mit einem 8-Kanal-Offner-Tintenschreiber.

Gereizt wurde mit einem Grass-Impulsgenerator. Jeder Bestrahlung ging eine Kontrollperiode von 4 bis 6 Wochen voraus. Die Bestrahlung erfolgte mit zwei Picker-Einheiten simultan von oben und unten. Fokusabstand: 60 cm; Spannung: 250 KV, 15 mA; Dosisleistung: 43 r/min; Filterung: 0,5 mm Cu parabolisch und 1 mm Al; HWS: 1,7 mm; Gesamtdosis: 400 r Ganzkörperbestrahlung. Die Messungen setzten unmittelbar nach der Bestrahlung ein und wurden am gleichen Tag über 9 Stunden und weiterhin täglich zweimal fortgesetzt.

[1] This article is based on work performed under Contract No. AT-(04-1)-GEN-12 between the Atomic Energy Commission and the University of California at Los Angeles.

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