Bei experimenteller Somanvergiftung sind die Oxime HS 3, HS 6 und HS 9 unter Atropinschutz verwendet worden. HS 3 kann nur bei prophylaktischer Gabe, vermutlich aufgrund der raschen Alterung des Enzyms, eine Hemmung der AChE bis etwa 40 min nach Vergiftung ausschliessen. HS 6 vermag eine Inhibierung der Endplattenesterase durch Soman in keinem Fall zu beeinflussen. Abweichende Befunde ergeben sich bei Applikation von HS 9 gegenüber dem genannten Phosphonsäureester. Therapeutisch gegeben, blieb das Ferment bis 200 min nach Intoxikation intakt. Die prophylaktische Anwendung von HS 9 dagegen ergab nur eine Verzögerung der Esterasehemmung bis zu 100 min nach Vergiftung. Eine Applikation von Atropin allein ist ebenso wie die Kombination mit den Oximen 2-PAM und Toxogonin bei Somanvergiftung von geringer Bedeutung.

Die Esterasereaktivatoren sind Antidote, die den an das Ferment gebundenen Phosphorsäurerest wieder zu entfernen vermögen. Aus der Vielzahl der Arbeiten über dieses Problem sollen hier diejenigen von Loomis und Salafsky[11], Erdmann[12] sowie Hahn und Henschler[13] genannt werden. Eine Grundforderung an die Oxime ist, das Zentralnervensystem zu erreichen. 2-PAM gelingt dies auch in hohen Dosierungen nicht. Für Toxogonin® wiesen Erdmann[14] und Falb und Erdmann[15] – letztere mit ^{14}C markiertem Toxogonin® – Gehirnkonzentrationen nach. Oldiges[16] und Erdmann[12] haben das Oxim HS 6 auf seine pharmakologische Wirksamkeit untersucht. Jonecko[17] untersuchte die Blockierung der Endplattenesterase am M. quadriceps femoris und der Zunge von Mäusen, ohne jedoch den Zeitpunkt der Inhibierung und Restitution näher zu bestimmen. Die Auswirkungen auf die Endplattenesterase bei Somanvergiftung beschrieb Fischer[18]. Ariëns, Meeter, Wolthuis und Van Bentheim[19] beschrieben reversible Nekrosen an der Endplatte nach Verabreichung von Cholinesteraseinhibitoren. Ultrastrukturelle Veränderungen der Endplatte nach intraperitonealer Applikation von Soman beschrieb Preusser[20].

Summary. The inhibition of AChE by Paraoxon is overcome by the application of the oximes 2-PAM, Toxogonin® and HS 3. Soman inhibited AChE is affected only to a limited extent by HS 3 and HS 9.

G. Fischer

Institut für Aerobiologie der Fraunhofer-Gesellschaft zur Förderung der angewandten Forschung e.V., D-5949 Grafschaft (Sauerland, Deutschland), 30. Oktober 1969.

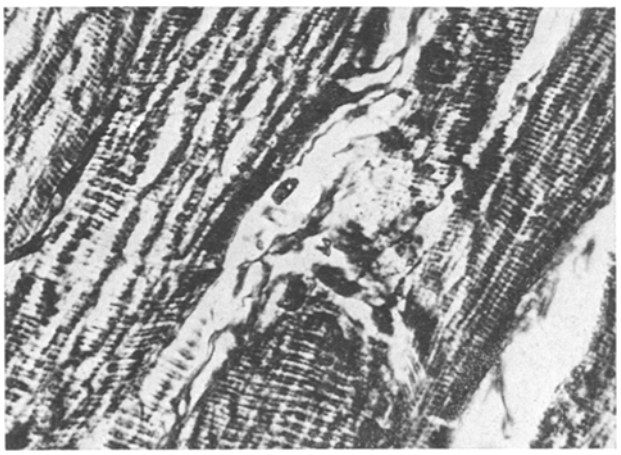

Fig. 2. Nekrose der Muskelfaser um die motorische Endplatte nach Vergiftung mit Paraoxon. × 250.

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Inhibition of Local Connective Tissue Repair in the Neoplastic Response to Water Soluble Carcinogens Administered Subcutaneously

Repeated s.c. injection in rats and mice is a recommended method for testing the carcinogenic activity of water-soluble chemical compounds. A wide variety of such substances have been shown capable of inducing sarcoma. These include glucose and sodium chloride administered in hypertonic solution[1,2], hydrochloric acid buffered at pH 5[3], and a number of surface active or amphipathic food colourings[2]. A multitude of experiments, some of which include detailed studies of the early lesion, indicate that the carcinogenic response appears to develop as a consequence of the local tissue reaction and as in the case of sarcoma induction by solid-state surfaces, is not a true index of carcinogenic activity.

Compared with this considerable amount of information on s.c. sarcoma induction by the agency of physical factors, little information is available on the tissue reaction and long-term results of repeated s.c. injection of 'true' carcinogens in aqueous solution, although such observations are extremely important in the study of the local mechanism of sarcoma production by carcinogens. Previous observations on the early effect of carcinogen injections used oily vehicles, and thus are not relevant comparisons, because of the formation of an oil granuloma and differences in the absorption rates of the vehicles.

In a study of the early local reaction and carcinogenic response to water-soluble carcinogens, 4-nitroquinoline-N-oxide (NQO), N-methyl-N-nitrosourea (MNU) and

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butyrylethyleneimine (BEI), were dissolved in water and administered in a dose of 0.2 ml of 0.1%, 0.1 ml of 0.5% and 0.5 ml of a 2% solution respectively, to a group of 40 rats per compound. Long-term experiments were terminated at 40 weeks. A high percentage of sarcomas were obtained with NQO and MNU. The BEI experiment had to be terminated earlier because the animals developed a high incidence of mammary tumours.

For a study of the early reaction, 2 rats were killed 24 h after the first injection of MNU, NQO or BEI, and then after the 2nd, 4th, 6th, 8th and 10th injection and several sections from each subcutaneous site examined histologically.

Local necrosis of the subcutaneous tissue, severe with NQO and BEI, less severe with MNU, was seen after the first injection. The necrotic area contained fibrin, fibrinoid and a variable lymphocytic and mononuclear cell infiltrate (Figure 1). A slight fibroblastic response was observed only after the 4th injection in all 3 experiments. No attempt at granulation tissue formation was observed, the newly formed cells were scattered haphazardly and were found principally at the peripheral areas of the zone of necrosis. A proportion of these fibroblasts were abnormal in size or nuclear structure, or in both. Enlarged fibroblasts 2–3 times normal size characterized the early connective tissue reaction to injections of MNU. Their nuclei were enlarged and exhibited characteristic heavy condensations of chromatin, demonstrable ultrastructurally close to the nuclear membrane (Figure 2). The fibroblasts that appeared at the site of NQO injections were not conspicuously enlarged, but contained enlarged nucleoli which ultrastructurally displayed bizarre shapes (Figure 4). Fibroblasts with large nucleoli were seen by light microscopy at the site of BEI injections.

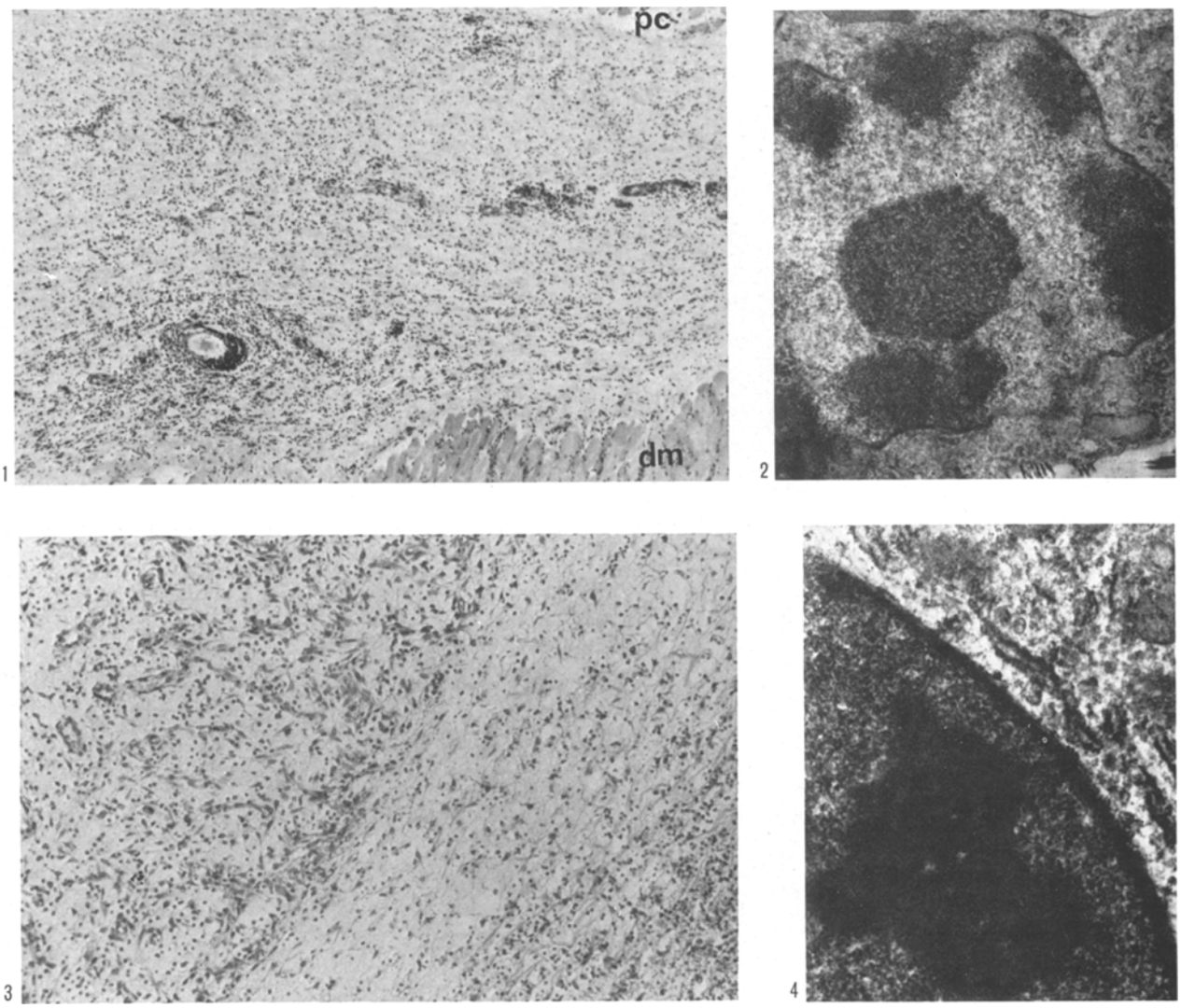

Fig. 1. Fibrin and inflammatory cells replacing subcutaneous fat. Granulation tissue absent after 2 weeks from first injection. Rat treated with 0.1 ml of 0.5% MNU twice weekly. pc, panniculus carnosus; dm, deep muscle; H. & E., × 80.

Fig. 2. Chromatin condensation at the periphery of an abnormally large fibroblast, after 6 injections of MNU. Pb citrate/uranyl acetate, × 12,500.

Fig. 3. Granulation tissue replacing fibrin at the site of 5 injections of Blue VRS, a surface active food colouring. Rat treated with 0.5 ml of 2% solution of Blue VRS twice weekly. H. & E., × 100.

Fig. 4. Uniform condensation of nuclear chromatin. Nucleolus enlarged and irregular in shape with microsegregation of fibrillar and granular components, after 4 injections of NQO. Pb citrate/uranyl-acetate, × 25,000.

With the further progress of the experiment (5th–10th injection) abnormal fibroblasts became less conspicuous. The site of injection and surrounding zones exhibited numerous normal-looking fibroblasts arranged in irregular bundles or foci, the latter suggesting pre-neoplasia. Throughout all 3 experiments a variable lymphocytic and histiocytic infiltrate was present. It was most marked in the case of BEI.

Temporary suppression of actively growing tissue appears to be the initial effect of carcinogenic agents. For example, 20-methylcholanthrene, 1,2,5,6-dibenzanthracene and other carcinogenic polycyclic hydrocarbons have been reported to inhibit the connective tissue response when implanted in paraffin wax pellets or injected s.c. in oil, in both rats and mice[4–7]. X-irradiation of a surgically inflicted wound in rats, 28 h after injury, reduces the fibroblastic response by 53%[8]. Oral administration of aromatic mustards, aminostilbenes and carcinogenic hydrocarbons retards the growth rate of young rats and mice[9] and inhibits the growth of malignant tissues[10], whilst N-2-fluorenylacetamide depresses mitotic activity in regenerating liver after hepatectomy[11]. Our results, indicating that the fibroblastic response to injury is inhibited by water-soluble carcinogens are in keeping with these related observations.

Nuclear abnormalities similar to those reported here have been observed in the nuclei of hepatocytes after treatment with a variety of chemical carcinogens[12, 13].

The evidence of a local inhibitory effect on fibroblastic proliferation and granulation tissue formation by carcinogens is in strong contrast to the proliferative lesions induced by surface-active model compounds, e.g. sodium lauryl sulphate, and food colourings, e.g. Blue VRS[14] (Figure 3), or by hypertonic solutions of glucose[1, 15] or by acidic solutions[3] and supports the hypothesis that sarcoma induction in rats could result from 2 distinct mechanisms: one representing a true carcinogenic response, the other leading to malignant change as a result of continued stimulation of fibroblastic proliferation in response to repeated trauma.

Zusammenfassung. Nach subkutaner Injektion einiger sogenannter wasserlöslicher Karzinogene (N-Methyl-N-Nitrosoharnstoff, Nitroquinolin-N-Oxyd und Butyryl-ethylenimin) erfolgt eine lokale Nekrose der subkutanen Gewebe mit verzögerter Regenerierung des Bindegewebes.

J. Hooson and P. Grasso

The British Industrial Biological Research Association, Woodmansterne Road, Carshalton (Surrey, England), 19 November 1969.

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Time and Dose Dependent Inhibition and Enhancement of Thymidine Incorporation into Fibroplasts by Prednisolone

Glucocorticoids have been found to inhibit growth of fibroblasts in vitro[1–7] as well as to stimulate proliferation[1, 8–12]. Fibroblasts have been used as an assay system for these hormones because the activity in depressing the rate of growth of fibroblasts reflects their clinical efficiency as anti-inflammatory agents[1]; moreover, fibroplasts may be considered as indicator cells for cytostatic effects on lympoid cells[3], effects which are exploited in leukaemia treatment and in immunosuppression.

In monolayers of L-cells we investigated the influence of Solu-Decortin-H®[13] (Prednisolone) on the incorporation of tritiated thymidine[14]. We employed a wide range of concentrations of the hormone at different time intervals. L-cells were grown in flasks (10 ml medium: 70% Hanks, 20% calf serum and 10% chicken embryo extract). They were harvested with trypsin and distributed in fresh medium into (80–90) roller rubes (1 ml/tube containing 500,000 cells). After 20–27 h the medium was replaced by TCM '199' (Difco) and 2 h later Prednisolone was added in saline to the final concentrations given in the Figure (moles/1.1 l). At the time points indicated, 2 μC thymidine were added in 20 μl saline and the L-cells were incubated for 2 h. The reaction was stopped by adding 0.1 ml 1.0*N* NaOH at ice temperature. The lysate was pipetted in 100 μl aliquots on to filter paper disks (3 disks per tube) and extracted with cold TCA according to Mans and Novelli[15]. The residual radioactivity was measured in a liquid scintillation counter.

The Figure represents a typical experiment out of four: At 10^{-4} and 10^{-5} *M* a pronounced inhibition of thymidine

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