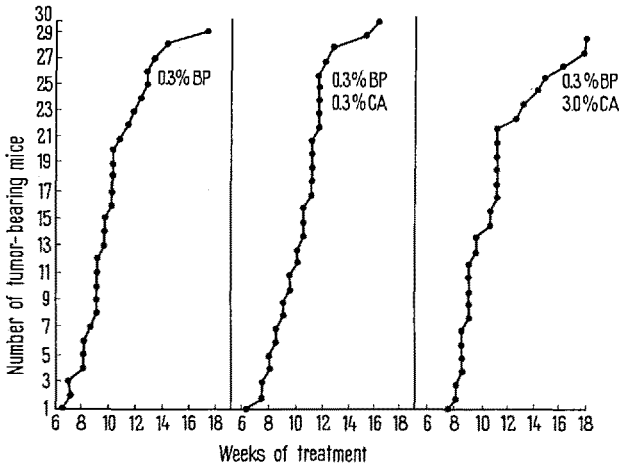
and CA solutions were prepared in freshly distilled acetone and were administered from a 1-ml tuberculin syringe to the surface of the skin of the interscapular region. This area of skin was shaved one day prior to the first treatment and subsequently when necessary. Each administration was of 0.1 ml of solution; BP was administered twice weekly on Tuesday and Thursday; CA was administered thrice weekly on Monday, Wednesday and Friday. All animals were examined for tumors prior to each treatment. A papilloma (1 mm minimum diameter) had to persist for 2 weeks to be considered as a true tumor, but it was recorded as appearing on the date it was first noted.

No tumors were observed in Group J receiving only 4.0% CA, nor in an additional group of 11 mice receiving only 0.1 ml distilled acetone twice weekly; both groups were treated and observed for 35 weeks. All other mice developed skin tumors. The Figure gives the results obtained with 0.3% BP alone (Group A) and together with 0.3% CA (Group D) or 3.0% CA (Group H). The manner of representation used is that of CRABTREE[2]. While our results with 0.3% BP alone are in excellent agreement with those found by CRABTREE, we find no inhibition with 0.3% CA as previously reported[2]. In fact none of the concentrations of CA used caused a statistically significant alteration in the rate of tumor incidence by BP, since there is an overlapping of the Confidence Interval of each group receiving both compounds (Groups B through I) with the Confidence Interval of the group receiving BP alone (Group A); the Confidence Interval equals the mean for the time (in weeks) of the first appearance of tumors $\pm 2\times$ standard error (see Table).



Mouse skin tumor induction by 3,4-benzpyrene in the presence and absence of mono-chloroacetone. Each point on the graph represents the time when a papilloma was first noted on an individual mouse. Animals were renumbered in the order of tumor appearance for the purpose of plotting the data.

Thus, in our hands, we were not able to demonstrate an inhibitory effect of CA on BP-induced skin tumorigenesis in mice as reported by CRABTREE[2]. In the present experiments the solvent (acetone) differed from that used previously[2] (98% ether, 2% medicinal liquid paraffin); however, this did not alter the rate of appearance of tumors induced by 0.3% BP alone (compare present results with those of CRABTREE[2]). Discussion of possible effects due to differences in the strain, sex or age of the mice is not possible since these variables were not defined in the previous report[2].

Résumé. Dans un mémoire paru en 1941, CRABTREE a décrit un cas unique de carcinogénèse chimique expérimentale dans lequel selon le degré de concentration ou selon l'ordre adopté pour le badigeonnage, le chloroacétone accélère ou inhibe l'apparition des tumeurs provoquées par le 3,4-benzpyrène (à 0,3%) sur la peau de la souris. Pour le confirmer, des expériences utilisant dix groupes de 30 souris femelles chacuns et huit concentrations croissantes de chloroacétone de 0,1 a 4,0%, n'ont pas montré de différences statistiquement significatives entre les groupes expérimentaux et celui du contrôle.

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Sekretomotorische Fasern und Endigungen in der Adenohypophyse

Obwohl die Innervation der Hypophyse und die Endigungen der innervierenden Fasern von mehreren Untersuchern eingehend beschrieben wurden[1-5], herrscht doch eine Unsicherheit bezüglich der sekretomotorischen Innervation der Adenohypophyse. Insofern stimmen die Forscher überein, als die Pars intermedia als Nervenfasern enthaltend beschrieben wird. Was die Nervenversorgung der Pars distalis des Hypophysenvorderlappens anbetrifft, herrschen stark abweichende Ansichten. Zahlreiche Forscher vertreten die Meinung, dass auch die Pars distalis innerviert sei[2-6], und diese Annahme wird durch die hervorragenden Untersuchungen von METUZALS[7] unterstützt, während GREEN et al.[8,9] auf Grund ihrer an verschiedenen Spezies durchgeführten Untersuchungen dies entschieden verneinen. Diese Auffassung wird von den meisten Endokrinologen akzeptiert[10]. Es scheint also berechtigt, einige Bemerkungen über diese Frage zu veröffentlichen.

Material und Methoden. Die Untersuchungen erfolgten an Gefrierschnitten bzw. Paraffinschnitten von in 10prozentigem Formalin fixierten Hypophysen von Ratten und Meerschweinchen und Schweinen. Die Schnitte wurden mit einem modifizierten Bodian-Protargol-Imprägnationsverfahren[11] gefärbt.

Ergebnisse. Aus dem reichhaltigen Fasernetz des Tractus hypothalamo-hypophysealis, dessen Fasern vorwiegend

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[9] S. OKAMOTO und I. JOSHIYUKI, Anat. Rec. *137*, 485 (1960).
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in der Neurohypophyse endigen, ziehen zahlreiche Nervenfassern unmittelbar in die Pars intermedia (Figur 1). Die in der Pars distalis gefundenen Fasern scheinen teilweise dem sympathischen Nervensystem anzugehören – diese Fasern ziehen den Gefässen entlang. Andererseits kommen aber – durch die Pars tuberalis hindurchtretend (Figur 2) – einige Fasern aus dem Tractus hypothalamo-hypophysealis-System in den Vorderlappen (Figur 3 und 4). Die Endigungen der Fasern im mittleren und vorderen Lappen haben grösstenteils Keulenform (Figur 5) und schmiegen sich entweder der Zelloberfläche an oder enden im intrazellulären Raum. Der Durchmesser dieser Endigungen beträgt 1–2 μ.

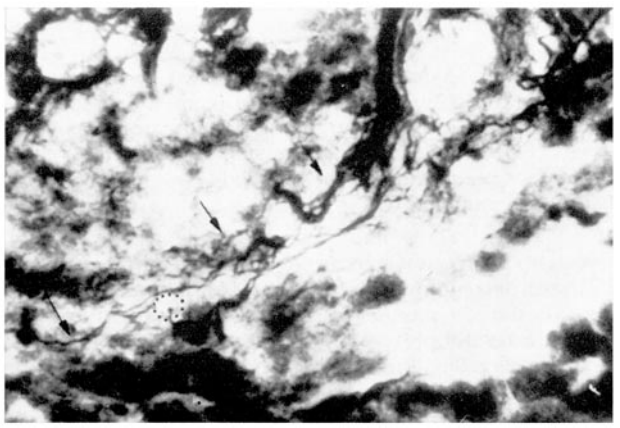

Fig. 1. Übertritt einer Nervenfaser aus der Neurohypophyse in die Pars intermedia. In der Mitte des mit einem Kreise umgebenen Gebietes eine kugelförmige Endigung. Gefrierschnitt, × 700.

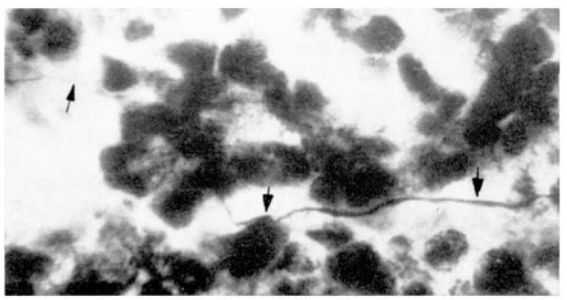

Fig. 2. Eine durch die Pars tuberalis hindurchtretende Nervenfaser. Gefrierschnitt, × 700.

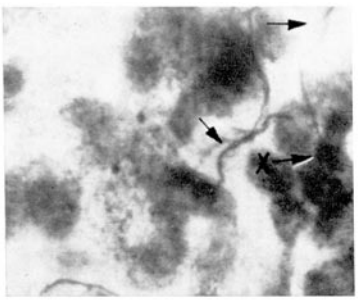

Fig. 3. Nervenfasern in der Pars distalis. x = perizelluläre Endigung. Gefrierschnitt, × 700.

Die innervierenden Fasern schlängeln in der Pars distalis zwischen den Drüsenzellen herziehend an die näherliegenden Drüsenzellen heran (Figur 5 und 6). Die Tatsache, dass die Nervenfasern ihren geraden Verlauf unterbrechen und in der Nähe einiger Drüsenzellen spezielle Windungen beschreiben, lässt vermuten, dass zwischen

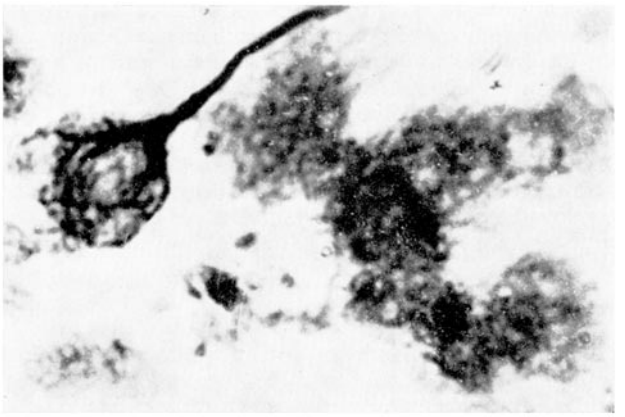

Fig. 4. Perizelluläres Netzwerk um eine Drüsenzelle in der Pars distalis. Gefrierschnitt, × 2000.

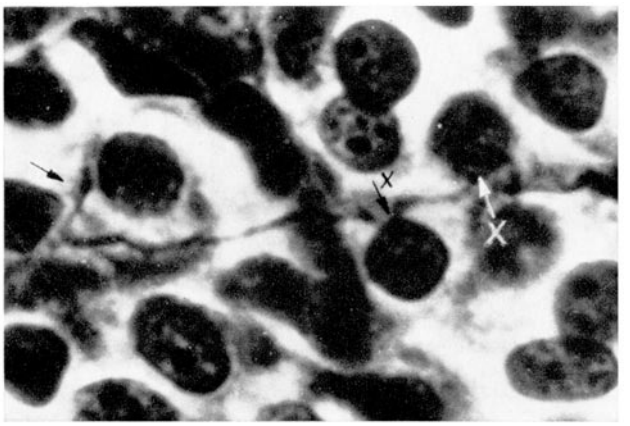

Fig. 5. Keulenförmige Nervenendigung in der Pars distalis der Ratte. x = Direktes Heranschwenken an die Zellen. Paraffineinbettung, × 1800.

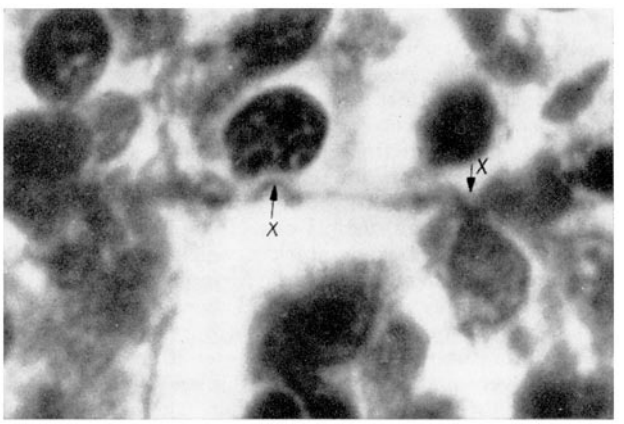

Fig. 6. Nervenfasern in der Pars distalis der Ratte. x = Herantritt der Faser an eine benachbarte Zelle. Paraffineinbettung, × 1800.

diesen Fasern und Zellen ein inniger Zusammenhang besteht (sekreto-motorischer Effekt?).

In der Pars distalis der Adenohypophyse fanden wir spezielle Nervenendigungen vor. Die innervierenden Fasern weisen hier nicht nur die beschriebenen keulen- bzw. ringförmigen Endigungen auf, sondern enden manchmal in einem, einige Drüsenzellen umgebenden, feinen Netzwerk (Figur 3 und 4). Ursprung und Wirkungsmechanismus dieser, spezielle Endigungsformen aufweisenden Fasern sind noch nicht hinreichend geklärt, obwohl ihr Verlauf zu der Annahme berechtigt, dass sie ebenfalls vom Tractus hypothalamo-hypophysealis stammen. Ob ihnen eine neurosekretorische Funktion zuzusprechen ist, soll im Laufe weiterer Untersuchungen entschieden werden.

Summary. We demonstrated, in the pituitary of the animals examined, fibres which emerge from the Tr. hypothalamo-hypophyseal system and enter partly into the Pars intermedia and partly *via* the Pars tubularis into the Pars distalis and take their course directly towards the glandular cells. In the Pars distalis the nerve fibres end around the glandular cells in a special pericellular net.

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Inhibition of Precipitin Formation in the Guinea-Pig with Cyclophosphamide

Evidence has been presented that cyclophosphamide, an alkylating agent, inhibits primary formation of anaphylactic antibody and induces a specific tolerance to anaphylactic sensitization in the guinea-pig[1,2]. In that work the crisp, but qualitative, mark of death-in-acute-anaphylaxis was the end point. In studies here reported, the ability of cyclophosphamide to inhibit the formation of precipitating antibody in the guinea-pig is evaluated using a quantitative method of double diffusion in gel for antibody determination[3]. Further, dosage schedules of antigen in saline solution as against similar amounts of antigen in complete Freund's adjuvant are compared and the antibody inhibiting abilities of two other cancer chemotherapeutic drugs, viz. vincaleukoblastine and 6-mercaptopurine, have been tested.

Methods. Antigen throughout was five times recrystallized egg albumin[4] prepared at a half-percent concentration in physiological saline. When adjuvant was used, equal parts of Freund's complete adjuvant[5] and egg-saline solution were emulsified immediately prior to injection. All antigen injections were intraperitoneal (IP). Animals were closed-colony randomly-bred Hartley strain, albino guinea-pigs, weighing approximately $^1/_2$ kg. Cyclophosphamide and vincaleukoblastine were prepared in physiological saline solution. 6-Mercaptopurine was suspended in cotton seed oil 150 mg/cm³. Blood was secured *via* intercardiac puncture under light ether anesthesia at 2, $4^1/_2$ and 8 weeks after first antigenic stimulation; and additionally, for the cyclophosphamide treated animals, at $2^1/_2$ weeks following second antigenic stimulation.

Antibody concentration was measured using the technique of PREER, which quantitates on the basis of the position of precipitation band in gel agar in capillary tubes[3].

Results. (1) *Saline cyclophosphamide:* Sensitizing schedule is shown in Figure 1. Serum from control animals showed good antibody concentration at 2 weeks (8/8), $4^1/_2$ weeks (7/7), and 8 weeks (5/5). The cyclophosphamide treated animals had no detectable antibody at 2 weeks (8/8) and 8 weeks (7/7); at $4^1/_2$ weeks 11 of 12 animals had no detectable antibody and the remaining guinea-pig had trace (but definite) antibody.

(2) *Failure of vincaleukoblastine and 6-mercaptopurine:* Sensitizing schedule is shown in Figure 1. Comparably timed and somewhat more toxic schedules of vincaleukoblastine and 6-mercaptopurine did not inhibit antibody formation; the antibody concentrations of these animals were indistinguishable from those of the control group. The comparative mortality of the several drugs is shown in the Table.

(3) *Immune tolerance induced by treatment with egg-cyclophosphamide:* A group of the cyclophosphamide treated animals that had been exposed to antigen $5^1/_2$ weeks previously and a new control group were injected with egg albumin (1 mg IP alternate days × 3). No additional cyclophosphamide was used. Blood was drawn $2^1/_2$

Comparative mortality
approximately $^1/_2$ kg guinea-pigs

	3 week survivors
Cyclophosphamide 10 mg/dx 14	16/20
Vincaleukoblastine 11 γ/dx 10	12/20
6-mercaptopurine 150 mg in oil day 1 and day 5	7/21

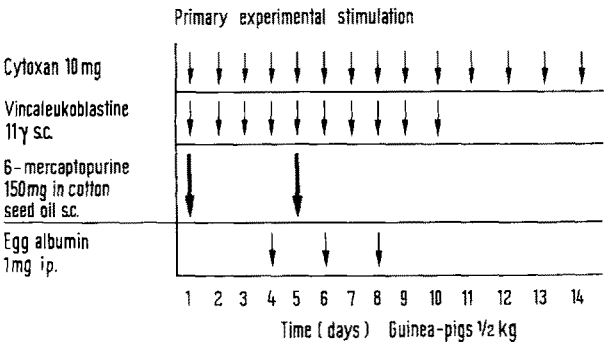

Fig. 1. Dose and time relationships of drugs and antigen are shown. Control animals received egg albumin and no drug. The three different experimental groups were given egg albumin and one of the 3 test drugs.

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