

scopy; e.g. peaks of the open-chain mixed anhydrides around 5.47 and 5.67 μ disappeared during the reduction, and peaks of the Leuchs' anhydrides appeared around 5.38 and 5.58 μ ³. In a typical example the mixed anhydride of N-carbobenzoxy-DL-phenylalanine prepared with ethylchlorocarbonate in the usual way⁴ in dioxane solution was hydrogenated in the presence of 10% palladium on charcoal catalyst. After evaporation of the solvent, the Leuchs' anhydride of phenylalanine, which was identical with an authentic sample, was isolated in 60% yield. When L-phenylalanine was used in this reaction the Leuchs' anhydride polymerized immediately to poly-L-phenylalanine, however, its presence was always indicated by IR-spectroscopy. This is in agreement with earlier observations, that DL-Leuchs' anhydrides polymerize much slower than the L- or D-forms⁵. The catalytic hydrogenation of the active esters of carbobenzoxy-DL-phenylalanine, such as the pentachlorophenyl ester⁶, also gave poly-DL-phenylalanine through the Leuchs' anhydride which was detected by peaks at 5.38 and 5.58 μ in the IR-spectrum⁷.

Zusammenfassung. Durch katalytische Hydrierung von C-aktivierten Carbobenzoxyminosäurederivaten, z.B. von gemischten Anhydriden und aktivierten Estern, werden via die Carbaminsäurederivate die entsprechenden Leuchsschen Anhydride und Polypeptide erhalten.

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Investigation of Smooth-Muscle Extracts by Means of Immunodiffusion

It has recently been demonstrated¹⁻⁴ that there is a difference between striated and smooth muscles as regards the interproportion and composition of proteins extracted with concentrated salt solution. Present experiments were designed to register structural discrepancies between the proteins of the different muscles by means of their specific antigenic character.

The musculature of the urinary bladders and intestines of 5 dogs, freed from connective tissue and mucosa, was homogenized for 2 min with a 0.154M solution of KCl, and left standing in a solvent of fivefold volume for 20 h. The homogenate was then centrifuged, and its supernate used as myogen extract. We washed the sediment with a 0.154M KCl solution eight times and extracted it afterwards with a Weber-Edsall solution of sixfold volume during 17 h. The supernatant fluid served as *structure-protein* extract. All the chemical procedures were performed at +4°C. The extracts had a final K-concentration of 0.5M. They were freeze-dried and stored in ampoules. Striated muscles of the dogs (adductor femoris and rectus abdominis) and - as controls - pooled plasma and mixed fragments of renal and splenic tissue, were subjected to a similar procedure.

Groups of 4 rabbits each, were immunized with the extracts. Using Ouchterlony's double gel-diffusion method, a 1% agar gel was dissolved in a phosphate buffer of pH 7.6. The K-concentration of the agar was 0.5M.

The well, shown in the centre of Figure 1, was filled with the serum of a rabbit immunized with smooth-muscle (SM) myogen extract. This serum had previously been absorbed by mixed-tissue myogen extract. Two bands of precipitation appeared near the SM myogen extract indicating the presence of specific antigenic components.

The metabolism of SM is known to be different from that of striated ones. It remained, however, doubtful whether a quantitative difference of the sarcoplasmic enzymes may manifest itself through such a striking dis-

crepancy of antigenicity. JAISLE⁵ succeeded in extracting with water nearly all contractile proteins from human uterine tissue. We studied this problem in another series of experiments. The sera of rabbits immunized with SM myogen extract were absorbed by a Weber-Edsall extract of SM. This done, one of the above-described precipitation bands disappeared, and the other either disappeared also or became faint; it follows that the two antigenic components, observed in the myogen extract of the SM, may partly be identical with one or more SM structure proteins (StrProt) which dissolve even at such a low ionic strength.

It can be seen in Figure 2 that the rabbit serum produced against the StrProt extract of SM induced a multiple precipitation of the SM StrProt-s and the SM myogen extract but failed to precipitate the striated muscle structure extract. This is an additional proof that one or more of the proteins contained in the SM extract of low ionic strength are identical with those found in the extract of higher ionic strength. Such proteins occur in other tissues as well (mixed-tissue control). It is, at the same time, evident that there are no such antigenic components in the StrProt extract of striated muscles.

The central well in Figure 3 contained the serum of a rabbit immunized with SM StrProt extract that had previously been absorbed by a SM myogen extract. The band of precipitation between the central well and the SM structure extract indicates the presence of a specific antigen.

NEEDHAM et al.⁴ encountered in a uterine extract of 0.5M KCl-concentration, in addition to actomyosin and myosin, other StrProt-s and also readily soluble collagen.

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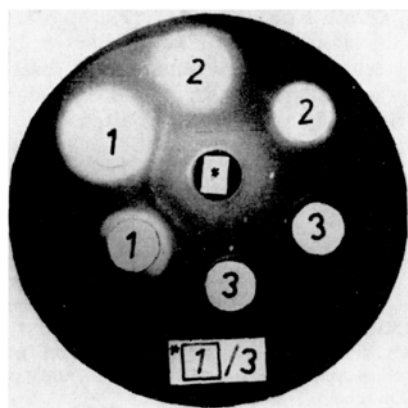


Fig. 1

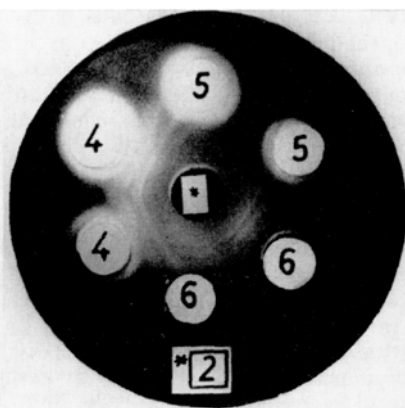


Fig. 2

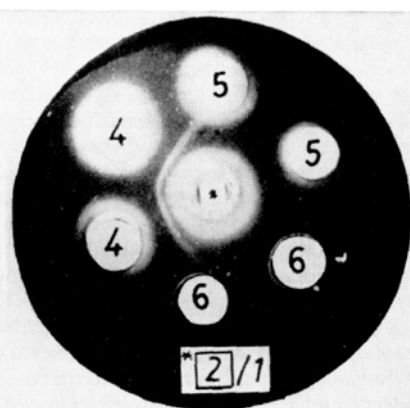


Fig. 3

Antigens: 1, smooth-muscle myogen extract; 2, striated-muscle myogen extract; 3, plasma; 4, smooth-muscle structure-protein extract; 5, striated-muscle structure-protein extract; 6, mixed-tissue myogen extract. Antibodies: [1], immune serum against antigen No. 1; [2], immune serum against antigen No. 2; [1]/3, supernate of immune serum No. [1] after absorption by antigen No. 3; [2]/1, supernate of immune serum No. [2] after absorption by antigen No. 1.

It has been noted in the foregoing that immune sera produced against SM StrProt-s do not precipitate antigens prepared from striated muscle StrProt-s (Figure 2), so that the latter are not identical with the specific antigenic component of the SM StrProt extract, nor is this SM structural component identical with that contained in the SM myogen extract (Figure 3). Considering that, according to our other investigations, the structure extracts contain not more than 10 to 15% actomyosin complex, it is possible that the specific factor remaining in the SM structure extract belongs to IVANOV's T-fraction, or else it may be a different structure component.

Zusammenfassung. Es gelang, zwei charakteristische Antigene im 0,154 M KCl-Extrakt glatter Hundemuskeln

durch Geldiffusion nachzuweisen, die möglicherweise eine Folge des spezifischen Stoffwechsels der glatten Muskulatur sind. Die weitere Möglichkeit besteht, dass ein Teil der Struktureiweiße der glatten Muskeln bereits bei niedriger Ionenkonzentration gelöst wird. Beim spezifischen Antigen des Weber-Edsall-Extrakts glatter Muskeln handelt es sich wahrscheinlich nicht um Aktomyosin bzw. Myosin, sondern um ein anderes Eiweiß oder Kollagen.

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Photosensitizing Furocoumarins: Interaction with DNA and Photo-Inactivation of DNA Containing Viruses

Some furocoumarins show a well-known photosensitizing activity on the human and guinea-pig skin¹⁻³; the lethal photosensitization of bacteria⁴⁻⁶ and of mammalian cells, adapted to in vitro growth⁷, was also studied.

The relationships between the photosensitizing activity of furocoumarins and their chemical structure are now well clarified^{2,3}.

The photosensitizing effects displayed on the skin are characteristic and different from those of many other photodynamic substances, such as hematoporphyrin, hypericin, methylene blue, fluoresceinic dyes, etc.⁸. The last compounds act by a photooxydative process. The furocoumarins, on the contrary, are lacking in photooxydative properties; the mechanism of their photosensitizing effect is at present not completely understood, in spite of much research done in this field^{2,3,9-11}.

Studies of the photoreactions between photosensitizing furocoumarins and flavin-monomonucleotide (FMN) seemed

to suggest a first approach to this problem; in fact only the active substances photoreact, the inactive ones do not form new compounds. The strict parallelism observed suggested the possibility of an explanation, through the photochemical in vitro reactivity with FMN, of the in vivo

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