that of PP, throughout the hybrid development, there is a definite reduction in its turnover. Since the synthesis of DNA in this hybrid is also reduced, this indicates again the metabolic correlation between the two nucleic acids[14].

Zusammenfassung. Inkubation der Seeigelkeime mit ^{14}C-Adenin ergab, dass bei reinen Arten der Einbau der markierten Substanz in die RNS von der Mesenchymblastula bis zum Pluteus eindeutig zunimmt. Parallel zur DNS-Synthese verläuft die Zunahme bei *Paracentrotus* und *Sphaerechinus* viel rascher als bei *Arbacia*. Beim Bastard PA nimmt dieser Einbauprozess mit Beginn der morphogenetischen Hemmung (nach ca. 26 h) rasch ab.

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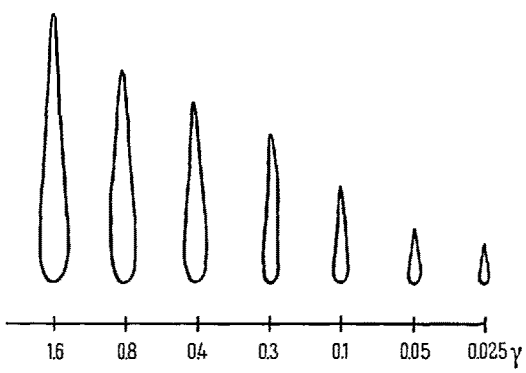
PRO EXPERIMENTIS

A Novel Quantitative Application of Thin Layer Chromatography: Assay of Tryptamine

Thin layer chromatography is a technique which is usually used for qualitative determinations. Previously recognized methods of quantitative analysis have been based on elution and subsequent UV analysis[1] and, more often, on the measurement of the area of the zone[2–4]. The method which we have used is based on the measurement of the distance between the spot of application and the front of the zone. (This may be proportional to the area but is considerably easier to measure.) It is not to be considered quantitative analysis of a chromatogram but rather as the use of a chromatogram for quantitative measurement.

A 20 cm square glass plate was covered with a layer (250 μ) of Kieselguhr-G (Merck, Germany), dried at 120°C for 1 to 3 h and stored at room temperature over $CaCl_2$ for 24 h or more. Tryptamine was spotted in 0.01 to 0.10 ml of chloroform, 1.5 cm from the bottom edge of the plate which was then developed with acetone (analytical reagent grade) and water (99 : 1, v/v) in a closed container. The solvent front reached the 10 cm end point in 10 to 15 min. The plate was then air-dried and sprayed with a freshly prepared 1 : 1 mixture of 1% aqueous ferric chloride and 1% aqueous potassium ferricyanide[5]. The tryptamine immediately turned a deep blue color, whereas the background developed a lighter blue color in about 2 to 3 h. The Figure is a photograph of such a plate (made after the blue streaks were outlined with a stylus) and shows the relationship between the quantity of compound and the distance of the movement of the front over the range of 0.025 μg to 1.6 μg. Greater quantities of tryptamine can be read, but as the gradient is steeper, accuracy is better at these lower levels. Since it is impractical to spot more than 0.1 ml of solution on a chromatoplate, the sensitivity of this method for tryptamine is 0.025 μg/0.1 ml. This sensitivity can be increased in application to analysis of biological tissues, if necessary, by the use of appropriate proportions of extractants or by concentration steps.



Relationship between quantity of tryptamine and the movement of its front on a thin layer chromatoplate. Kieselgur-G–Acetone-water 99:1 v/v.

The actual distance a given quantity of tryptamine moves from its point of application varies slightly from one plate to the next; thus, it is necessary to apply standard quantities to each assay plate. This variability is mainly the result of differences in the degree of hydration of the adsorbent[6]. Precision on a given plate, however, is good. One plate with 6 identical spots containing 0.1 μg tryptamine, gave streaks with a mean length of 29.4 $\pm$ 0.6 mm. Another plate with 6 identical spots containing 1.0 μg tryptamine gave a mean streak length of 49 $\pm$ 3.0 mm.

The application of this method to analysis involves standard preliminary procedures. For example, a 2.0 ml sample of canine plasma was made alkaline by the addition of 0.1 ml of 1 *N* NaOH and then extracted by shaking with 0.5 ml of chloroform. After centrifugation, the aqueous layer was aspirated and the remaining weak emulsion was broken by the addition of a small quantity of anhydrous sodium sulfate. A measured volume (0.05 or 0.10 ml) of the dried chloroform extract was then spotted on the thin layer chromatographic plate. Known amounts of tryptamine were added to plasma and yielded recoveries which were essentially quantitative. Thus, with this method, for each 0.10 μg of tryptamine per ml of plasma, each 0.1 ml of the chloroform extract will contain 0.04 μg, an amount easily measured on the plate.

The specificity of this method is not that usually expected with chromatography since streaks or trails

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[6] During the winter, the relative humidity in our laboratories was low, and satisfactory results could be obtained by airdrying the plates. However, with the advent of spring and rise in humidity, the tryptamine tended to follow the solvent front, so this procedure was no longer feasible. Therefore, all plates are now oven-dried.

such as these are generally considered undesirable because of the resulting limited resolution. Additional specificity is added by the solvent extraction step and by the color reaction. The chloroform extract of blank plasma in the method described gave only one blue coloring spot which moved with the solvent front. The addition of serotonin to the plasma did not interfere with the tryptamine analysis.

The use of thin layer chromatography for quantitative measurement has several advantages, when applicable. It would permit simplification of the preliminary extraction procedures, and although the sensitivity and accuracy of the method is comparable with that of fluorescence assay methods, it requires much less in the way of instrumentation. Although we have worked with only a few compounds, using only Kieselguhr and a number of solvents, there is no reason to believe that methods cannot be devised for many compounds.

Résumé. Les auteurs décrivent une technique microchromatographique d'adsorption sur couches minces de Kieselguhr, avec l'acétone comme éluant, dans laquelle le déplacement de la part de tryptamine est proportionnel à la quantité de ce corps dans le domaine compris entre 0,025 et 2,0 μg. On peut ainsi déterminer 0,10 μg de tryptamine par ml de plasma. Il est fort probable que cette méthode simple et quantitative pourra également être adaptée à de nombreux autres composés.

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An Experimental Method for Compressing the Sciatic Nerve

It has recently been recognized that certain nerve lesions are due to constriction or moderate compression and that symptoms from such lesions can be relieved by simple surgical measures[1–4]. The very simplicity and success of these operations have prevented study of the changes in the affected nerves. However, in animals the effects of continued constriction or moderate compression have been studied by Weiss[5–6], and Denny-Brown[7]. The methods used by these workers were imperfect, and a fresh study of this problem was therefore carried out.

Method. Under ether anaesthesia the right sciatic nerve in a rat was exposed in the thigh and an 8 mm length of No. 2 Sterivac (polythene) tubing slit longitudinally was then slipped over the nerve. The bore of this tubing (0.5 mm) was just small enough to constrict the nerve. This tubing was held in place by two 3 mm pieces of No. 3 Sterivac tubing also slit longitudinally to allow them to be applied over the narrower tubing. The internal diameter of the No. 3 tubing (1 mm) was the same as the external diameter of the No. 2 tubing.

Results. In a preliminary study 12 rats were used. In some animals immediately following the operation the foot was a little weak; however, all the rats were using the hind limbs normally when they were sacrificed three to six weeks later. Under urethane anaesthesia both sciatic nerves were exposed, a stimulating electrode was applied to the nerve opposite the ischio-coccygeus muscle (i.e. proximal to the constricting device on the right), and a recording electrode inserted into the muscles of the foot.

The oscillograph record from the foot showed a delay in conduction amounting to 2–4 msec on the constricted as compared with the normal side; in four animals repetitive firing occurred following a single shock.

Discussion. Simpson[8] regarded the delay in conduction and the repetitive firing as the characteristic electrical features of carpal tunnel compression and related syndromes. These electrical changes were produced in rats by the method of constriction described above and presumably the pathological changes in the sciatic nerve of the rat should be similar to those occurring in constricted or moderately compressed nerves of man. These changes are being studied and will be reported later[9].

Zusammenfassung. Ein leichter Druck wurde auf den Nervus ischiadicus der Ratte ausgeübt. Die electromyographischen Veränderungen sind den Veränderungen in pathologisch umklammerten menschlichen Nerven ähnlich. Die pathologische Anatomie dieser Nerven wird jetzt untersucht.

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[9] *Acknowledgments.* My thanks are due to Mr. G. V. Osborne for suggesting this topic. I am grateful to Professor F. W. Landgrebe and the Welsh National School of Medicine, Dr. K. N. Lloyd and the Governors of Cardiff Royal Infirmary for the use of research facilities. For technical assistance I am indebted to Messrs. R. J. Hillard, D. Jones, and I. Messenger. The Welsh Hospital Board provided the funds for the research.

STUDIORUM PROGRESSUS

Zur Ermittlung der Farben grösster Buntkraft

Einleitung[1–3]. Die *Buntkraft* erscheint in einer Reihe von Farben gleichen Buntstons[4] als eine unmittelbar auffallende und wichtige Eigenschaft, so dass auch ungeübte und mit Farbmessungen nicht vertraute Personen ohne Schwierigkeit aus solchen Reihen die nach ihrer Ansicht buntkräftigste Farbe auszusuchen vermögen. Da sich

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[4] Da zwischen bunten und unbunten Farben zu unterscheiden ist und nur die bunten in ihrem Ton differieren, so verwenden wir hier durchwegs den Ausdruck «Buntton» statt «Farbton» und dementsprechend «Buntkraft» statt «Farbkraft».